EPA United States Environmental Protection Agency

August 2005
EPA/600/R-05/004aB

Air Quality Criteria for Ozone and Related Photochemical Oxidants (Second External Review Draft)

Volume I of III

EPA/600/R-05/004aB
August 2005

Air Quality Criteria for Ozone and Related Photochemical Oxidants

Volume I

National Center for Environmental Assessment-RTP Office
Office of Research and Development
U.S. Environmental Protection Agency
Research Triangle Park, NC

DISCLAIMER

This document is a second external review draft for review purposes only and does not constitute U.S. Environmental Protection Agency policy. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

PREFACE

National Ambient Air Quality Standards (NAAQS) are promulgated by the United States Environmental Protection Agency (EPA) to meet requirements set forth in Sections 108 and 109 of the U.S. Clean Air Act (CAA). Sections 108 and 109 require the EPA Administrator (1) to list widespread air pollutants that reasonably may be expected to endanger public health or welfare; (2) to issue air quality criteria for them that assess the latest available scientific information on nature and effects of ambient exposure to them; (3) to set "primary" NAAQS to protect human health with adequate margin of safety and to set "secondary" NAAQS to protect against welfare effects (e.g., effects on vegetation, ecosystems, visibility, climate, manmade materials, etc); and (5) to periodically review and revise, as appropriate, the criteria and NAAQS for a given listed pollutant or class of pollutants.

In 1971, the U.S. Environmental Protection Agency (EPA) promulgated National Ambient Air Quality Standards (NAAQS) to protect the public health and welfare from adverse effects of photochemical oxidants. The EPA promulgates the NAAQS on the basis of scientific information contained in air quality criteria issued under Section 108 of the Clean Air Act. Following the review of criteria as contained in the EPA document, Air Quality Criteria for Ozone and Other Photochemical Oxidants published in 1978, the chemical designation of the standards was changed from photochemical oxidants to ozone (O_3) in 1979 and a 1-hour O_3 NAAQS was set. The 1978 document focused mainly on the air quality criteria for O_3 and, to a lesser extent, on those for other photochemical oxidants (e.g., hydrogen peroxide and the peroxyacyl nitrates), as have subsequent revised versions of the ozone document.

To meet Clean Air Act requirements noted above for periodic review of criteria and NAAQS, the O_3 criteria document, *Air Quality Criteria for Ozone and Other Photochemical Oxidants*, was next revised and then released in August 1986; and a supplement, *Summary of Selected New Information on Effects of Ozone on Health and Vegetation*, was issued in January 1992. These documents were the basis for a March 1993 decision by EPA that revision of the existing 1-h NAAQS for O_3 was not appropriate at that time. That decision, however, did not take into account some newer scientific data that became available after completion of the 1986 criteria document. Such literature was assessed in the next periodic revision of the O_3 air quality criteria document (completed in 1996) and provided scientific bases supporting the setting by EPA in 1997 of an 8-h O_3 NAAQS that is currently in force together with the 1-h O_3 standard.

The purpose of this revised air quality criteria document for O_3 and related photochemical oxidants is to critically evaluate and assess the latest scientific information published since that assessed in the above 1996 Ozone Air Quality Criteria Document (O_3 AQCD), with the main focus being on pertinent new information useful in evaluating health and environmental effects data associated with ambient air O_3 exposures. However, some other scientific data are also presented and evaluated in order to provide a better understanding of the nature, sources, distribution, measurement, and concentrations of O_3 and related photochemical oxidants and their precursors in the environment. The document mainly assesses pertinent literature published or accepted for publication through 2004.

The present Second Draft O_3 AQCD (dated August 2005) is being released for public comment and review by the Clean Air Scientific Advisory Committee (CASAC) to obtain comments on the organization and structure of the document, the issues addressed, the approaches employed in assessing and interpreting the newly available information on O_3 exposures and effects, and the key findings and conclusions arrived at as a consequence of this assessment. Public comments and recommendations will be taken into account making any appropriate further revisions to this document for incorporation into the final version of the document to be completed and issued by February 28, 2006. Evaluations contained in the present document will be drawn on to provide inputs to associated PM Staff Paper analyses

prepared by EPA's Office of Air Quality Planning and Standards (OAQPS) to pose options for consideration by the EPA Administrator with regard to proposal and, ultimately, promulgation of decisions on potential retention or revision, as appropriate, of the current O_3 NAAQS.

Preparation of this document was coordinated by staff of EPA's National Center for Environmental Assessment in Research Triangle Park (NCEA-RTP). NCEA-RTP scientific staff, together with experts from other EPA/ORD laboratories and academia, contributed to writing of document chapters. Earlier drafts of document materials were reviewed by non-EPA experts in peer consultation workshops held by EPA. The document describes the nature, sources, distribution, measurement, and concentrations of O_3 in outdoor (ambient) and indoor environments. It also evaluates the latest data on human exposures to ambient O_3 and consequent health effects in exposed human populations, to support decision making regarding the primary, health-related O_3 NAAQS. The document also evaluates ambient O_3 environmental effects on vegetation and ecosystems, man-made materials, and surface level solar UV radiation flux and global climate change, to support decision making on secondary O_3 NAAQS.

NCEA acknowledges the valuable contributions provided by authors, contributors, and reviewers and the diligence of its staff and contractors in the preparation of this draft document.

Air Quality Criteria for Ozone and Related Photochemical Oxidants (Second External Review Draft)

VOLUME I

Air Quality Criteria for Ozone and Related Photochemical Oxidants (Second External Review Draft)

(cont'd)

VOLUME II

VOLUME III

Table of Contents

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

Table of Contents
(cont'd)

List of Tables

List of Tables
(cont'd)

List of Figures

List of Figures
(cont'd)

List of Figures
(cont'd)

List of Figures
(cont'd)

List of Figures
(cont'd)

List of Figures
(cont'd)

List of Figures
(cont'd)

List of Figures
(cont'd)

Authors, Contributors, and Reviewers

CHAPTER 1. INTRODUCTION

Principal Author

Dr. Lester D. Grant—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

CHAPTER 2 - PHYSICS AND CHEMISTRY OF OZONE IN THE ATMOSPHERE

Principal Authors

Dr. Joseph Pinto—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Russell Dickerson—University of Maryland, College Park, MD

Contributing Authors

Dr. Brooke Hemming—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Daniel Jacob—Harvard University, Cambridge, MA

Dr. William Keene—University of Virginia, Charlottesville, VA

Dr. Tadeusz Kleindienst—National Exposure Research Laboratory, U.S. Environmental Protection Agency, Research Triangle Park, NC

Dr. Jennie Moody—University of Virginia, Charlottesville, VA

Mr. Charles Piety—University of Maryland, College Park, MD

Dr. Sandy Sillman—University of Michigan, Ann Arbor, MI

Dr. Jeffrey Stehr—University of Maryland, College Park, MD

Dr. Bret Taubman—Pennsylvania State University, State College, PA

Contributors and Reviewers

Dr. Christoph Bruhl, Max Planck Institute for Atmospheric Chemistry, Mainz, Germany

Dr. Mohammed Elshahawy, Department of Meteorology and Astronomy, Cairo University, Giza, Egypt.

Dr. Arlene Fiore, NOAA/GFDL, Princeton, NJ

Mr. Chris Geron, NRML, U.S. EPA, Research Triangle Park, NC

Dr. David Golden, Stanford University, Palo Alto, CA

Dr. John Merrill, University of Rhode Island, Kingston, RI

Dr. Sam Oltmans, NOAA, CMDL, Boulder, CO

Dr. David Parrish, NOAA/AL, Boulder, CO

Dr. Perry Samson, Depart. Atmos. Ocean, and Space Sciences, University of Michigan, Ann Arbor, MI

Dr. Sandy Sillman, University of Michigan, Ann Arbor, MI

Dr. Melvin Shapiro, National Center for Atmospheric Research, Boulder, CO

CHAPTER 3 - ENVIRONMENTAL CONCENTRATIONS, PATTERNS, AND EXPOSURE ESTIMATES

Principal Authors

Ms. Beverly Comfort—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Joseph Pinto—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Arlene Fiore—NOAA/GFDL, Princeton, NJ

Dr. Daniel Jacob—Harvard University, Cambridge, MA

Authors, Contributors, and Reviewers
(cont'd)

Principal Authors
(cont'd)

Dr. Alan S. Lefohn—ASL &Associates, Helena, MT

Dr. Clifford Weisel—Rutgers University, New Brunswick, NJ

Contributing Authors

Dr. Jee-Young Kim——National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Dennis Kotchmar—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Timothy Lewis—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Mr. Thomas McCurdy—U.S. EPA, NERL U.S. EPA, Research Triangle Park, NC

Contributors and Reviewers

Dr. Christoph Bruehl—Max Planck Institute for Atmospheric Chemistry, Mainz, Germany

Dr. Russell Dickerson—University of Maryland, College Park, MD

Dr. Judith Graham—American Chemistry Council, Washington, D.C.

Dr. Laszlo Horvath—Hungarian Meteorological Service, Budapest, Hungary

Dr. Ted Johnson—TRJ Associates, Durham, NC

Dr. John Merrill—University of Rhode Island, Kingston, RI

Dr. Jennie Moody—University of Virginia, Charlottesville, VA

Dr. Sam Oltmans—NOAA CMDL, Boulder, CO

Dr. Michiel G.M. Roemer, TNO, The Netherlands

Dr. Sandy Sillman—University of Michigan, Ann Arbor, MI

Dr. Tamas Weidinger—University of Budapest, Budapest, Hungary

Authors, Contributors, and Reviewers
(cont'd)

CHAPTER 4 - DOSIMETRY, SPECIES HOMOLOGY, SENSITIVITY, AND EXTRAPOLATION

Principal Authors

Dr. John Overton—U.S. Environmental Protection Agency, National Health and Environmental Effects Research Laboratory-Research Triangle Park, NC 27711 (retired)

Dr. James S. Brown—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Lori White—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Contributors and Reviewers

Dr. Gary Hatch—U.S. Environmental Protection Agency, National Health and Environmental Effects Research Laboratory, NC

CHAPTER 5 - TOXICOLOGICAL EFFECTS IN LABORATORY ANIMALS AND IN VITRO TEST SYSTEMS

Principal Authors

Dr. Lori White—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Mr. James Raub—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

Dr. Deepak Bhalla—Wayne State University, Detroit, MI

Dr. Carroll Cross—University of California, Davis, CA

Dr. Mitch Cohen—NYU School of Medicine, New York University, New York, NY

Contributors and Reviewers

Dr. Steven Kleeberger—National Institute of Environmental Health Sciences, Research Triangle Park, NC 27711

Dr. George Liekauf—University of Cincinnati, Cincinnati, OH

Dr. David Basset—Wayne State University, Detroit, MI

Dr. E.M. Postlethwait—University of Texas Medical Branch, Galveston, TX

Dr. Kent Pinkerton—University of California, Davis, CA

Dr. Jack Harkema—Michigan State University, East Lansing, MI

Dr. Edward Schelegle—University of California, Davis, CA

Dr Judith Graham—American Chemical Council, Arlington, VA

CHAPTER 6 - CONTROLLED HUMAN EXPOSURE STUDIES

Principal Authors

Dr. James S. Brown—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Mr. James Raub—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

Dr. William C. Adams—University of California, Davis, CA (retired)

Dr. Milian J. Hazucha—University of North Carolina, Chapel Hill, NC

Dr. E. William Spannhake—Johns Hopkins University, Baltimore, MD

Contributors and Reviewers

Dr. Edward Avol—University of Southern California, Los Angeles, CA

Dr. Henry Gong—Ranchos Los Amigos Medical Center, Los Angeles, CA

Contributors and Reviewers
(cont'd)

Dr. Jane Q. Koenig—University of Washington, Seattle, WA

Dr. Michael Madden—National Health and Environmental Effects Research Laboratory,
U.S. Environmental Protection Agency, Chapel Hill, NC

Dr.William McDonnell—National Health and Environmental Effects Research Laboratory,
U.S. Environmental Protection Agency, Chapel Hill, NC

CHAPTER 7 - EPIDEMIOLOGICAL STUDIES OF HUMAN HEALTH EFFECTS

Principal Authors

Dr. Dennis Kotchmar—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Jee-Young Kim—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. David Svendsgaard—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr Kaz Ito—New York University, New York, NY

Dr. Pat Kinney—School of Public Health, Columbia University, New York, NY

Reviewers

Dr. Richard Burnett—Health Canada, Ottawa, CN

Dr. Vic Hasselblad—Duke University, Durham, NC

Dr. Lucas Neas—National Health and Environmental Effects Research Laboratory,
U.S. Environmental Protection Agency, Chapel Hill, NC

CHAPTER 8 - INTEGRATIVE SYNTHESIS: EXPOSURE AND HEALTH EFFECTS

Principal Authors

Dr. Srikanth Nadadur—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Lester Grant—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Reviewers

Dr. John Vandenberg—National Center for Environmental Assessment, Washington, DC

Dr. Daniel Costa—National Program Director for Air, Office of Research and Development, Research Triangle Park, NC 27711

CHAPTER 9 - ENVIRONMENTAL EFFECTS ON VEGETATION AND ECOSYSTEMS

Principal Authors

Dr. Jay Garner—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Timothy Lewis—National Center for Environmental Assessment (B243-01),
U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. William Hogsett—National Health and Environmental Effects Research Laboratory,
U.S. Environmental Protection Agency, Corvallis, OR

Dr. Christian Andersen—National Health and Environmental Effects Research Laboratory,
U.S. Environmental Protection Agency, Corvallis, OR

Dr. Allen Lefohn—ASL and Associates, Helena, MT

Dr. David Karnosky—Michigan Technological University, Houghton, MI

Dr. Michael Nannini—Ilinois State Water Survey, IL

Dr. Nancy Grulke—USDA Forest Service, Riverside, CA

Authors, Contributors, and Reviewers
(cont'd)

Principal Authors
(cont'd)

Dr. Richard Adams—Oregon State University., Corvallis, OR

Dr. Robert Heath—University of California, Riverside, CA,

Dr. Victor Runeckle—Vancouver, B.C., CN

Dr. Arthur Chappelka—Auburn University, School of Forestry, Auburn, AL

Dr. William Massman—USDA Forest Service, Ft. Collins, CO

Dr. Robert Musselman—USDA Forest Service, Fort Collins, CO

Dr. Peter Woodbury—Cornell University, Ithaca, NY (former USDA Forest Service)

Contributors and Reviewers

Dr. Boris Chevone—Department of Plant Pathology, Virginia Technological University, Blacksburg, VA 24061

Dr. Alan Davison—School of Biology, Newcastle University, Newcastle on Tyne, United Kingdom, NE1 7RU

Dr. Bruce L. Dixon—Department of Agricultural Economics, University of Arkansas, Fayetteville, AR 72701

Dr. David Grantz—Kearney Agricultural Center, University of California at Riverside, Parlier, CA 93648

Dr. Allen S. Heagle—1216 Scott Pl., Raleigh, NC 27511

Dr. Robert Horst, Jr.—121 Thorwald Dr., Plainsboro, NJ 08536

Dr. John Innes—Forest Sciences Centre, Department of Forest Resources, University of British Columbia, Vancouver, BC, Canada V6T 1Z4

Dr. Hans-Jürgen Jäger—Heinrich-Buff-Ring 26-32, Institute of Plant Ecology, Justus-Leibig University, Gessen, Germany D35392

Dr. Robert Kohut— Tower Road, Boyce Thompson Institute, Rm 131,Cornell University, Ithaca, NY 14853

Contributors and Reviewers
(cont'd)

Dr. Sagar Krupa—1519 Gortner Ave., Department of Plant Pathology, University of Minnesota, St. Paul, MN 55108

Dr. William Manning—203 Morrill, Department of Microbiology, University of Massachusetts, Amherst, MA 01003

Dr. Howard Neufeld—Rankin Science Bldg., Appalachian State University, Boone, NC 28608

Dr. Maria-Jose Sanz—Fundacion CEAM, c/Charles Darein, 14-Parque Te Valencia, Spain

Dr. James Shortle—Department of Ag Econ, Armsby, Pennsylvania State University, University Park, PA 16802

Dr. John Skelly—Department of Plant Pathology, Pennsylvania State University, University Park, PA 16803

CHAPTER 10 - TROPOSPHERIC OZONE EFFECTS ON UV-B FLUX AND CLIMATE CHANGE

Principal Authors

Dr. Brooke Hemming—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Jee-Young Kim—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Contributors and Reviewers

Dr. Sasha Madronich—Atmospheric Chemistry Division. National Center for Atmospheric Research (NCAR), Boulder, CO 80307

Dr. Daniel J. Jacob—Atmospheric Chemistry and Environmental Engineering, Division of Engineering & Applied Science, and Department of Earth & Planetary Sciences, Harvard University, Cambridge, MA 02138

Authors, Contributors, and Reviewers
(cont'd)

CHAPTER 11 - EFFECTS OF OZONE ON MAN-MADE MATERIALS

Principal Author

Mr. Bill Ewald—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

[Note: Any inadvertently omitted names of authors/reviewers will be inserted in the final draft of this O_3 AQCD, as will more complete addresses for all authors/reviewers.]

U.S. Environmental Protection Agency Project Team for Development of Air Quality Criteria for Ozone and Related Photochemical Oxidants

Executive Direction

Dr. Lester D. Grant (Director)—National Center for Environmental Assessment-RTP Division, (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Scientific Staff

Dr. Lori White(Ozone Team Leader)—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Joseph Pinto—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Ms. Beverly Comfort—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Brooke Hemming—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. James S. Brown—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Dennis Kotchmar—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Jee-Young Kim—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. David Svendsgaard—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Srikanth Nadadur—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Timothy Lewis—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Dr. Jay Garner—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

U.S. Environmental Protection Agency Project Team for Development of Air Quality Criteria for Ozone and Related Photochemical Oxidants
(cont'd)

Scientific Staff
(cont'd)

Dr. William Hogsett—National Health and Environmental Effects Research Laboratory, U.S. Environmental Protection Agency, Corvallis, OR

Dr. Christian Andersen—National Health and Environmental Effects Research Laboratory, U.S. Environmental Protection Agency, Corvallis, OR

Mr. Bill Ewald—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

Mr. James Raub—National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

Technical Support Staff

Ms. Nancy Broom—Information Technology Manager, National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Mr. Douglas B. Fennell—Technical Information Specialist, National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Ms. Emily R. Lee—Management Analyst, National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Ms. Diane H. Ray—Program Specialist, National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

Ms. Donna Wicker—Administrative Officer, National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711 (retired)

Mr. Richard Wilson—Clerk, National Center for Environmental Assessment (B243-01), U.S. Environmental Protection Agency, Research Triangle Park, NC 27711

U.S. Environmental Protection Agency Project Team for Development of Air Quality Criteria for Ozone and Related Photochemical Oxidants

(cont'd)

Document Production Staff

Ms. Carolyn T. Perry—Manager, Computer Sciences Corporation, 2803 Slater Road, Suite 220, Morrisville, NC 27560

Mr. John A. Bennett—Technical Information Specialist, Library Associates of Maryland, 11820 Parklawn Drive, Suite 400, Rockville, MD 20852

Mr. William Ellis—Records Management Technician, InfoPro, Inc., 8200 Greensboro Drive, Suite 1450, McLean, VA 22102

Ms. Sandra L. Hughey—Technical Information Specialist, Library Associates of Maryland, 11820 Parklawn Drive, Suite 400, Rockville, MD 20852

Mr. Matthew Kirk—Graphic Artist, Computer Sciences Corporation, 2803 Slater Road, Suite 220, Morrisville, NC 27560

Dr. Barbara Liljequist—Technical Editor, Computer Sciences Corporation, 2803 Slater Road, Suite 220, Morrisville, NC 27560

Ms. Faye Silliman—Word Processor, InfoPro, Inc., 8200 Greensboro Drive, Suite 1450, McLean, VA 22102

Mr. John A. Bennett—Technical Information Specialist, Library Associates of Maryland, 11820 Parklawn Drive, Suite 400, Rockville, MD 20852

Ms. Sandra L. Hughey—Technical Information Specialist, Library Associates of Maryland, 11820 Parklawn Drive, Suite 400, Rockville, MD 20852

Mr. William Ellis—Records Management Technician, InfoPro, Inc., 8200 Greensboro Drive, Suite 1450, McLean, VA 22102

U.S. Environmental Protection Agency Science Advisory Board (SAB)
Staff Office Clean Air Scientific Advisory Committee (CASAC)
Ozone Review Panel

Chair

Dr. Rogene Henderson*, Scientist Emeritus, Lovelace Respiratory Research Institute, 2425 Ridgecrest Drive SE, Albuquerque, NM, 87108, Phone: 505-348-9464, Fax: 505-348-8541, (rhenders@lrri.org) (FedEx: Dr. Rogene Henderson, Lovelace Respiratory Research Institute, 2425 Ridgecrest Drive SE, Albuquerque, NM, 87108, Phone: 505-348-9464)

Members

Dr. John Balmes, Professor, Department of Medicine, University of California San Francisco, University of California - San Francisco, San Francisco, California, 94143, Phone: 415-206-8953, Fax: 415-206-8949, (jbalmes@itsa.ucsf.edu)

Dr. Ellis Cowling*, University Distinguished Professor-at-Large, North Carolina State University, Colleges of Natural Resources and Agriculture and Life Sciences, North Carolina State University, 1509 Varsity Drive, Raleigh, NC, 27695-7632, Phone: 919-515-7564 , Fax: 919-515-1700, (ellis_cowling@ncsu.edu)

Dr. James D. Crapo*, Professor, Department of Medicine, National Jewish Medical and Research Center. 1400 Jackson Street, Denver, CO, 80206, Phone: 303-398-1436, Fax: 303- 270-2243, (crapoj@njc.org)

Dr. William (Jim) Gauderman, Associate Professor, Preventive Medicine, University of Southerm California, 1540 Alcazar #220, Los Angeles, CA, 91016, Phone: 323-442-1567, Fax: 323-442-2349, (jimg@usc.edu)

Dr. Henry Gong, Professor of Medicine and Preventive Medicine, Medicine and Preventive Medicine, Keck School of Medicine, University of Southern California, Environmental Health Service, MSB 51, Rancho Los Amigos NRC, 7601 East Imperial Highway, Downey, CA, 90242, Phone: 562-401-7561, Fax: 562-803-6883, (hgong@ladhs.org)

Dr. Paul J. Hanson, Senior Research and Development Scientist , Environmental Sciences Division, Oak Ridge National Laboratory (ORNL), Bethel Valley Road, Building 1062, Oak Ridge, TN, 37831-6422, Phone: 865-574-5361, Fax: 865-576-9939, (hansonpz@comcast.net)

Dr. Jack Harkema, Professor, Department of Pathobiology, College of Veterinary Medicine, Michigan State University, 212 Food Safety & Toxicology Center, East Lansing, MI, 48824, Phone: 517-353-8627, Fax: 517-353-9902, (harkemaj@msu.edu)

U.S. Environmental Protection Agency Science Advisory Board (SAB)
Staff Office Clean Air Scientific Advisory Committee (CASAC)
Ozone Review Panel
(cont'd)

Members
(cont'd)

Dr. Philip Hopke, Bayard D. Clarkson Distinguished Professor, Department of Chemical Engineering, Clarkson University, Box 5708, Potsdam, NY, 13699-5708, Phone: 315-268-3861, Fax: 315-268-4410, (hopkepk@clarkson.edu) (FedEx: 8 Clarkson Avenue, Potsdam, NY 136995708)

Dr. Michael T. Kleinman, Professor, Department of Community & Environmental Medicine, 100 FRF, University of California - Irvine, Irvine, CA, 92697-1825, Phone: 949-824-4765, Fax: 949-824-2070, (mtkleinm@uci.edu)

Dr. Allan Legge, President, Biosphere Solutions, 1601 11th Avenue NW, Calgary, Alberta, CANADA, T2N 1H1, Phone: 403-282-4479, Fax: 403-282-4479, (allan.legge@shaw.ca)

Dr. Morton Lippmann, Professor, Nelson Institute of Environmental Medicine, New York University School of Medicine, 57 Old Forge Road, Tuxedo, NY, 10987, Phone: 845-731-3558, Fax: 845-351-5472, (lippmann@env.med.nyu.edu)

Dr. Frederick J. Miller*, Consultant, 911 Queensferry Road, Cary, NC, 27511, Phone: 919-467-3194, (fjmiller@nc.rr.com)

Dr. Maria Morandi, Assistant Professor of Environmental Science & Occupational Health, Department of Environmental Sciences, School of Public Health, University of Texas - Houston Health Science Center, 1200 Herman Pressler Street, Houston, TX, 77030, Phone: 713-500-9288, Fax: 713-500-9249, (mmorandi@sph.uth.tmc.edu) (FedEx: 1200 Herman Pressler, Suite 624)

Dr. Charles Plopper, Professor, Department of Anatomy, Physiology and Cell Biology, School of Veterinary Medicine, University of California - Davis, Davis, California, 95616, Phone: 530-752-7065, (cgplopper@ucdavis.edu)

Mr. Richard L. Poirot*, Environmental Analyst, Air Pollution Control Division, Department of Environmental Conservation, Vermont Agency of Natural Resources, Bldg. 3 South, 103 South Main Street, Waterbury, VT, 05671-0402, Phone: 802-241-3807, Fax: 802-241-2590, (rich.poirot@state.vt.us)

Dr. Armistead (Ted) Russell, Georgia Power Distinguished Professor of Environmental Engineering, Environmental Engineering Group, School of Civil and Environmental Engineering, Georgia Institute of Technology, 311 Ferst Drive, Room 3310, Atlanta, GA, 30332-0512, Phone: 404-894-3079, Fax: 404-894-8266, (trussell@ce.gatech.edu)

U.S. Environmental Protection Agency Science Advisory Board (SAB)
Staff Office Clean Air Scientific Advisory Committee (CASAC)
Ozone Review Panel
(cont'd)

Members
(cont'd)

Dr. Elizabeth A. (Lianne) Sheppard, Research Associate Professor, Biostatistics and Environmental & Occupational Health Sciences, Public Health and Community Medicine, University of Washington, Box 357232, Seattle, WA, 98195-7232, Phone: 206-616-2722, Fax: 206 616-2724, (sheppard@u.washington.edu)

Dr. Frank Speizer*, Edward Kass Professor of Medicine, Channing Laboratory, Harvard Medical School, 181 Longwood Avenue, Boston, MA, 02115-5804, Phone: 617-525-2275, Fax: 617-525-2066, (frank.speizer@channing.harvard.edu)

Dr. James Ultman, Professor, Chemical Engineering, Bioengineering program, Pennsylvania State University, 106 Fenske Lab, University Park, PA, 16802, Phone: 814-863-4802, Fax: 814-865-7846, (jsu@psu.edu)

Dr. Sverre Vedal, Professor of Medicine, Department of Environmental and Occupational Health Sciences, School of Public Health and Community Medicine, University of Washington, 4225 Roosevelt Way NE, Suite 100, Seattle, WA, 98105-6099, Phone: 206-616-8285, Fax: 206-685-4696, (svedal@u.washington.edu)

Dr. James (Jim) Zidek, Professor, Statistics, Science, University of British Columbia, 6856 Agriculture Rd., Vancouver, BC, Canada, V6T 1Z2, Phone: 604-822-4302, Fax: 604-822-6960, (jim@stat.ubc.ca)

Dr. Barbara Zielinska*, Research Professor , Division of Atmospheric Science, Desert Research Institute, 2215 Raggio Parkway, Reno, NV, 89512-1095, Phone: 775-674-7066, Fax: 775-674-7008, (barbz@dri.edu)

Science Advisory Board Staff

Mr. Fred Butterfield, CASAC Designated Federal Officer, 1200 Pennsylvania Avenue, N.W., Washington, DC, 20460, Phone: 202-343-9994, Fax: 202-233-0643 (butterfield.fred@epa.gov) (Physical/Courier/FedEx Address: Fred A. Butterfield, III, EPA Science Advisory Board Staff Office (Mail Code 1400F), Woodies Building, 1025 F Street, N.W., Room 3604, Washington, DC 20004, Telephone: 202-343-9994)

*Members of the statutory Clean Air Scientific Advisory Committee (CASAC) appointed by the EPA Administrator

Abbreviations and Acronyms

α	alpha, probability value
AA	ascorbic acid
ACh	acetylcholine
ADSS	aged and diluted cigarette smoke
AER	air exchange rate
AEROCE	Atmospheric/Ocean Chemistry Experiment
AHR	airway hyperreactivity
AHSMOG	Adventist Health Study on Smog
AIRS	Aerometric Information Retrieval System
AM	alveolar macrophage
ANF	atrial natriuretic factor
AOP2	antioxidant protein 2
APHEA	Air Pollution on Health: European Approach (study)
AQCD	Air Quality Criteria Document
AQS	Air Quality System
ARIC	Atherosclerosis Risk in Communities (study)
ATLAS	atmospheric model by Kurucz
A/V	surface-to-volume ratio
β	beta-coefficient; slope of an equation
BAL	bronchioalveolar lavage
BALF	bronchioalveolar lavage fluid
BHR	bronchial hyperresponsiveness
BS	black smoke
BSA	body surface area
BMZ	basement membrane zone
BP	blood pressure
C	concentration
C × T	concentration × time; concentration times duration of exposure
CAA	Clean Air Act

CADS	Cincinnati Activity Diary Study
CAPs	concentrated ambient particles
CAR	centriacinar region
CASAC	Clean Air Scientific Advisory Committee
CASTNet	Clean Air Status and Trends Network
CC16	Clara cell secretory protein
CCSP	Clara cell secretory protein
CD96	Air Quality Criteria Document for Ozone and Related Photochemical Oxidants; O_3 AQCD
C_{dyn}	dynamic lung compliance
CE	continuous exercise
CFD	computational fluid dynamics
C_2H_5–H	ethane
C_5H_8	isoprene
C_6H_{16}	terpene
CHAD	Consolidated Human Activities Database
CH_3–CHO	acetaldehyde
CH_3–CCl_3	methyl chloroform
CH_3–CO	acetyl
CH_4	methane
CI	confidence interval
CIE	Commission Internationale de l'Eclaiarage (International Commission on Illumination)
CINC	cytokine-induced neutrophil chemoattractant
CLM	chemiluminescence method
CMAQ	Community Model for Air Quality
CO	carbon monoxide
CO_2	carbon dioxide
COD	coefficient of divergence
COP	Conference of Parties
COPD	chronic obstructive pulmonary disease

CRP	C-reactive protein
CTM	chemistry transport model
DHBA	2,3-dehydroxybenzoic acid
DNA	deoxyribonucleic acid
DOAS	differential optical absorption spectroscopy/spectrometry
DPPC	dipalmitoylglycero-3-phosphocholine
DU	Dobson units
ϵ	epsilon, convergence precision
EBC	exhaled breath condensate (fluid)
ECG	electrocardiographic
EDU	ethylenediurea
EEG	electroencephalographic
ELF	epithelial lining fluid
ENA-78	epithelial cell-derived neutrophil-activating peptide 78
ENSO	El Niño-Southern Oscillation
EPA	U.S. Environmental Protection Agency
ETS	environmental tobacco smoke
F	female
FA	filtered air
FACE	free-air carbon dioxide exposure
f_B	breathing frequency
FEF	forced expiratory flow
FEF_{25-75}	forced expiratory flow between 25 and 75% of vital capacity
FEV_1	forced expiratory volume in 1 second
FVC	forced vital capacity
GAM	Generalized Additive Model
GCM	general circulation model
GEE	Generalized Estimating Equation
GHGs	greenhouse gases
GLM	Generalized Linear Model

GM-CSF	granulocyte-macrophage colony stimulating factor
G6PD	glucose-6-phosphate dehydrogenase
GR	glutathione reductase
GSH	glutathione; reduced glutathione
GSHPx	glutathione peroxidase
GSTM1	glutathione S-transferase μ-1 (genotype)
H^+	hydrogen ion
H_2CO, HCHO	formaldehyde
HDMA	house dust mite allergen
HFCs	hydrofluorocarbons
HNE	4-hydroxynonenal
HNO_2, HONO	nitrous acid
HNO_3	nitric acid
HO	hydroxyl
HO_2	hydroperoxyl; hydroperoxy
H_2O_2	hydrogen peroxide
HR	heart rate
HRV	heart rate variability
H_2SO_4	sulfuric acid
IC	inspiratory capacity
ICAM	intracellular adhesion molecule
ICNIRP	International Commission on Non-Ionizing Radiation Protection
IE	intermittent exercise
Ig	immunoglobulin (e.g., IgA, IgE, IgG, IgM)
IL	interleukin (e.g., IL-1, IL-6, IL-8)
iNOS	inducible nitric oxide synthase; NOS-2
ip	intraperitoneal
IPCC	Intergovernmental Panel on Climate Change
IR	infrared
K_a	intrinsic mass transfer coefficient/parameter

K_g	mass transfer coefficient for gas phase
K_l	mass transfer coefficient for liquid phase
K_r	reaction rate constant
K_{TB}	terminal bronchiole region mass transfer coefficient
LDH	lactic acid dehydrogenase
LIDAR	LIght Detection And Ranging
LIS	lateral intercellular space
LLJ	low-level jet
LOESS	locally estimated smoothing splines
LOP	lipid ozonization products
LPS	lipopolysaccharide
LRT	lower respiratory tract; lower airways
LT	leukotriene (e.g., LTB_4, LTC_4, LTD_4, LTE_4)
LT	local time
M	gas molecule
M	male
M	maximum number of iterations
MAP	mean arterial pressure
MCP	monocyte chemotactic protein
MENTOR	Modeling Environment for Total Risk Studies
MI	myocardial infarction
MIP	macrophage inflammatory protein
MMEF	maximal midexpiratory flow
MONICA	Monitoring Trend and Determinants in Cardiovascular Disease (registry)
MPAN	peroxymethacryloyl nitrate; peroxy-methacrylic nitric anhydride
MPO	myeloperoxidase
mRNA	messenger ribonucleic acid
MSA	metropolitan statistical area
MSL	mean sea level
MT	metallothionein

n, N	number
NAAQS	National Ambient Air Quality Standards
NADPH	reduced nicotinamide adenine dinucleotide phosphate
NAS	Normative Aging Study
NCEA-RTP	National Center for Environmental Assessment Division in Research Triangle Park, NC
NCICAS	National Cooperative Inner-City Asthma Study
ND	not detectable; not detected
NEM	National Ambient Air Quality Standards Exposure Model
NF	national forest
NF-κB	nuclear factor kappa B
NH_4HSO_4	ammonium bisulfate
NHAPS	National Human Activity Pattern Survey
NIST	National Institute of Standards and Technology
NK	natural killer (cells)
NL	nasal lavage
NM	national monument
NMHCs	nonmethane hydrocarbons
NMMAPS	National Morbidity, Mortality and Air Pollution Study
NO	nitric oxide
NO_2	nitrogen dioxide
NO_3^-	nitrate
NOS	nitric oxide synthase
NOS-1	neuronal nitric oxide synthase
NOS-2	inducible nitric oxide synthase; iNOS
NOS-3	endothelial nitric oxide synthase
NO_x	nitrogen oxides
NO_y	reactive nitrogen system components; sum of NO_x and NO_z; odd nitrogen species
NO_z	difference between NOy and NOx
NP	national park

NPP	net primary productivity
NQO1wt	NAD(P)H-quinone oxidoreductase wild type (genotype)
NRC	National Research Council
NTP	National Toxicology Program
NTS	nucleus tractus solitarius
NWR	national wildlife refuge
$O(^1D)$	electronically excited oxygen atom
O_2	ground-state oxygen
O_3	ozone
O_3*	electronically excited ozone
$O(^3P)$	ground-state oxygen atom
OAQPS	Office of Air Quality Planning and Standards
8-OHdG	8-hydroxy-2′-deoxyguanosine
OH	hydroxyl; hydroxy
OTC	open-top chamber
OVA	ovalbumin
O_x	odd oxygen species
6PGD	6-phosphogluconate dehydrogenase
p	probability value
P_{90}	values of the 90th percentile absolute difference in concentrations
PAF	platelet-activating factor
PAN	peroxyacetyl nitrate; peroxyacetic nitric anhydride
PAR	proximal alveolar region
PBL	planetary boundary layer
PBPK	physiologically based pharmacokinetic (approach)
PCl	picryl chloride
PE	postexposure
PEF	peak expiratory flow
PEM	personal exposure monitor
P_{enh}	enhanced pause

PG	prostaglandin (e.g., PGD_2, PGE, PGE_1, PGE_2 $PGF_{1\alpha}$, $PGF_{2\alpha}$)
PI	probability interval
PM	particulate matter
$PM_{2.5}$	fine particulate matter (mass median aerodynamic diameter ≤2.5 μm)
PM_{10}	combination of coarse and fine particulate matter
$PM_{10\text{-}2.5}$	coarse particulate matter (mass median aerodynamic diameter between 10 and 2.5 μm)
PMNs	polymorphonuclear neutrolphil leukocytes; neutrophils
pNEM	Probabilistic National Ambient Air Quality Standard Exposure Model
ppb	parts per billion
ppbv	parts per billion by volume
ppm	parts per million
PPN	peroxypropionyl nitrate; peroxypropionic nitric anhydride
PRB	policy relevant background
PSA	picryl sulfonic acid
PUFA	polyunsaturated fatty acid
PWM	pokeweed mitogen
QCE	quasi continuous exercise
r	correlation coefficient
R	intraclass correlation coefficient
R^2	multiple correlation coefficient
R'CO	acyl
$R'C(O)–O_2$	acyl peroxy
RH	relative humidity
R_L	total pulmonary resistance
RO_2	organic peroxyl; organic peroxy
ROOH	organic peroxides
ROS	reactive oxygen species
RR	ribonucleotide reductase
RRMS	relatively remote monitoring sites
RT	respiratory tract

SAB	Science Advisory Board
SAC	*Staphylococcus aureus* Cowan 1 strain
SAMD	*S*-adenosyl methionine decarboxylase
SBUV	solar backscattered ultraviolet radiation
SC	stratum corneum
SD, S-D	Spraque-Dawley (rat)
SD	standard deviation
SES	socioeconomic status
sGAW	specific airways conductance
SHEDS	Simulation of Human Exposure and Dose System
SNPs	single nucleotide polymorphisms
SO_2	sulfur dioxide
SO_4^{2-}	sulfate
SOD	superoxide dismutase
SOS	Southern Oxidant Study
SP	substance P
SP	surfactant protein (e.g., SP-A, SP-D)
sRAW	specific airways resistance
STE	stratospheric-tropospheric exchange
STRF	Spatio-Temporal Random Field
SUM06	seasonal sum of all hourly average concentrations $\geq$ 0.06 ppm
SUM08	seasonal sum of all hourly average concentrations $\geq$ 0.08 ppm
SZA	solar zenith angle
t	*t*-test statistical value; t statistic
T_3	triiodothyronine
T_4	thyroxine
TAR	Third Assessment Report
TB	terminal bronchioles
TBARS	thiobarbituric acid reactive substances
Tc-DTPA	radiolabeled diethylenetriaminepentaacetic acid; ^{99m}Tc-DTPA

T_{CO}	core temperature
T_{CTL}	cytotoxic T-lymphocytes
TLC	total lung capacity
TNF	tumor necrosis factor
TNFR	tumor necrosis factor receptor
TOMS	Total Ozone Mapping Satellite; total ozone mapping spectrometer
TRIM	Total Risk Integrated Methodology (model)
TRIM Expo	Total Risk Integrated Methodology Exposure Event (model)
TSP	total suspended particulate
TWA	time-weighted average
UA	uric acid
UNFCCC	United Nations Framework Convention on Climate Change
URT	upper respiratory tract; upper airways
USGCRP	U.S. Global Change Research Program
UV	ultraviolet
UV-A	ultraviolet radiation of wavelengths 320 to 400 nm
UV-B	ultraviolet radiation of wavelengths 280 to 320 nm
UV-C	ultraviolet radiation of wavelengths 200 to 280 nm
VC	vital capacity
V_D	anatomic dead space
$\dot{V}_E$	minute ventilation; expired volume per minute
$\dot{V}O_{2max}$	maximal oxygen uptake (maximal aerobic capacity)
VOC	volatile organic compound
$\dot{V}_P$	volumetric penetration
$\dot{V}_{P50\%}$	volume at which 50% of an inhaled bolus is absorbed
V_T	tidal volume
V_{TB}	terminal bronchiole region volume
W126	cumulative integrated exposure index with a sigmoidal weighting function
WMO/UNEP	World Meteorological Organization/United Nations Environmental Program
WT	wild type

EXECUTIVE SUMMARY

E.1 INTRODUCTION

Tropospheric or "surface-level" ozone is one of six major air pollutants regulated by National Ambient Air Quality Standards (NAAQS) under the U.S. Clean Air Act. As mandated by the Clean Air Act, the U.S. Environmental Protection Agency (EPA) must periodically review the scientific bases (or "criteria") for the various NAAQS by assessing newly available scientific information on a given criteria air pollutant. This draft document, *Air Quality Criteria for Ozone and other Photochemical Oxidants*, is an updated revision of the 1996 Ozone Air Quality Criteria Document (O_3 AQCD) that provided scientific bases for the current O_3 NAAQS set in 1997.

E.1.1 Clean Air Act Legal Requirements

Clean Air Act (CAA) Sections 108 and 109 govern establishment, review, and revision of U.S. National Ambient Air Quality Standards (NAAQS).

- Section 108 directs the U.S. Environmental Protection Agency (EPA) Administrator to list ubiquitous (widespread) air pollutants that may reasonably be anticipated to endanger public health or welfare and to issue air quality criteria for them. The air quality criteria are to reflect the latest scientific information useful in indicating the kind and extent of all exposure-related effects on public health and welfare expected from the presence of the pollutant in the ambient air.

- Section 109 directs the EPA Administrator to set and periodically revise, as appropriate, two types of NAAQS: (a) *primary NAAQS* to protect against adverse health effects of listed criteria pollutants among sensitive population groups, with an adequate margin of safety, and (b) *secondary NAAQS* to protect against welfare effects (e.g., impacts on vegetation, crops, ecosystems, visibility, climate, man-made materials, etc.). Section 109 also requires peer review of the NAAQS and their underlying scientific bases by the Clean Air Scientific Advisory Committee (CASAC), a committee of independent non-EPA experts.

E.1.2 Chronology of Ozone NAAQS Revisions

In 1971, the U.S. EPA set primary and secondary standards for total photochemical oxidants. However, based on the criteria review completed in 1978, the original primary and secondary NAAQS set in 1971 were revised in 1979 to focus on O_3 as the indicator for new primary and secondary standards that would be attained when the expected number of days per calender year with maximum 1-h average O_3 concentrations >0.12 ppm did not exceed one. The NAAQS for ambient O_3 were revised in 1997 by replacing the 1-h standards with an 8-h primary standard that is met when the 3-year average of the annual fourth highest daily maximum 8-h average concentration is <0.08 ppm. The 1997 primary NAAQS was based on scientific data from controlled human exposure, laboratory animal, and epidemiological studies and associated analyses presented in the 1996 O_3 AQCD and in the 1996 O_3 Staff Paper (U.S. Environmental Protection Agency, 1996b).

- This revised O_3 AQCD is now being prepared by ORD's National Center for Environmental Assessment (NCEA) to support EPA's ongoing Congressionally-mandated periodic review of O_3 NAAQS under a consent decree (court-ordered) schedule that calls for issuance of the revised AQCD in final form by February 28, 2006. This document assesses the latest available scientific information (published mainly through December 2004) judged to be useful in deriving criteria as scientific bases for decisions on possible revision of the current O_3 NAAQS.

- A separate EPA O_3 Staff Paper will draw upon key findings/conclusions from this document, together with other analyses, to develop and present options for consideration by the EPA Administrator regarding review and possible revision of the O_3 NAAQS.

E.1.3 Document Organization and Structure

Volume I of this document consists of the present Executive Summary and eleven main chapters of this revised O_3 AQCD. Those main chapters focus primarily on interpretative evaluation of key information, whereas more detailed descriptive summarization of pertinent studies and/or supporting analyses are provided in accompanying annexes. Volume II contains the annexes for Chapters 4 through 7, whereas Volume III contains the annex for Chapter 9.

The topics covered in the main chapters are as follows:

- This Executive Summary summarizes key findings and conclusions from Chapters 1 through 11 of this revised O_3 AQCD.

- Chapter 1 provides a general introduction, including an overview of legal requirements, the chronology of past revisions of O_3-related NAAQS, and orientation to the structure of the document.

- Chapters 2 and 3 provide background information on atmospheric chemistry/physics of O_3 formation, air quality, and exposure aspects to help to place ensuing discussions of O_3 health and welfare effects into perspective.

- Chapters 4 through 7 then assess dosimetry aspects, experimental (controlled human exposure and laboratory animal) studies, and epidemiologic (field/panel; other observational) studies.

- Chapter 8 provides an integrative synthesis of key findings and conclusions derived from the preceding chapters with regard to ambient O_3 concentrations, human exposures, dosimetry, and health effects.

- Chapter 9 deals with effects of O_3 on vegetation, crops, and natural ecosystems, whereas Chapter 10 evaluates tropospheric O_3 relationships to alterations in surface-level UVB flux and climate change and Chapter 11 assesses materials damage (these all being key types of welfare effects of relevance to decisions regarding secondary O_3 NAAQS review).

E.2 ATMOSPHERIC CHEMISTRY AND PHYSICS OF TROPOSPHERIC OZONE FORMATION

Key findings/conclusions from Chapter 2 regarding the chemistry and physics of surface-level O_3 formation include the following:

- Ozone (O_3) is a secondary pollutant formed by atmospheric reactions involving two classes of precursor compounds, volatile organic compounds (VOCs) and nitrogen oxides (NO_x). Carbon monoxide also contributes to O_3 formation.

- The formation of O_3 and associated compounds is a complex, nonlinear function of many factors, including the intensity and spectral distribution of sunlight; atmospheric mixing and other atmospheric processes; and the concentrations of the precursors in ambient air.

- The photochemical oxidation of almost all anthropogenic and biogenic VOCs is initiated by reaction with hydroxyl (OH) radicals. At night, when they are most abundant, NO_3 radicals oxidize alkenes. In coastal and other select environments, Cl and Br radicals can also initiate the oxidation of VOCs.

- In urban areas, basically all classes of VOCs (alkanes, alkenes, aromatic hydrocarbons, carbonyl compounds, etc.) and CO are important for ozone formation. Although knowledge of the oxidative mechanisms of VOCs has improved over the past several years, gaps in knowledge involving key classes, such as aromatic hydrocarbons, still remain. For example, only about half of the carbon initially present in aromatic hydrocarbons in smog chamber studies form compounds that can be identified.

- In addition to gas phase reactions, reactions also occur on the surfaces of or within cloud droplets and airborne particles. Most of the well-established multiphase reactions tend to reduce the rate of O_3 formation in polluted environments. Direct reactions of O_3 and atmospheric particles appear to be too slow to reduce O_3 formation significantly at typical ambient PM levels.

- Oxidants other than O_3 are found in the gas phase and in particles. The chemistry occurring in particle bound-water and, hence, the mechanisms leading to the formation of reactive oxygen species in particles are largely unknown.

- Our basic understanding of meteorological processes associated with summertime O_3 episodes has not changed over the past several years. However, the realization is growing that long-range transport processes are important for determining O_3 concentrations at the surface . In addition to synoptic scale flow fields, nocturnal low-level jets are capable of transporting pollutants hundreds of km from their sources in either the upper boundary layer or the lower free troposphere. Turbulence then brings O_3 and other pollutants to the surface.

- Even in the absence of photochemical reactions in the troposphere, some O_3 would be found near the earth's surface due to its downward transport from the stratosphere. Intrusions of stratospheric O_3 that reach the surface are rare. Much more common are intrusions that penetrate to the middle and upper troposphere. However, O_3 transported to the middle and upper troposphere can still affect surface concentrations through various mechanisms that mix air between the planetary boundary layer and the free troposphere above.

- Chemistry transport models are used to improve understanding of atmospheric chemical and physical processes, as well as to develop air pollution control strategies. The performance of these models must be evaluated by comparison with field data as part of an iterative cycle of model improvement and subsequent evaluation. Discrepancies between model predictions and observations can be used to point out gaps in current understanding and thus to improve parameterizations of atmospheric chemical and physical processes.

- Model evaluation does not merely involve a straightforward comparison between model predictions and observed concentration fields of a pollutant of interest (e.g., O_3). Such comparisons may not be meaningful because it is difficult to determine if agreement between measurements and model predictions truly represents an accurate treatment of physical and chemical processes in the model or the effects of compensating errors in model routines.

- The main methods currently in use for routine monitoring of ambient ozone are based on chemiluminescence or UV absorption. Measurements at most ambient monitoring sites are based on UV absorption. Both of these methods are subject to interference by other

atmospheric components. Studies conducted in Mexico City and in a smog chamber have found positive interference, but a few studies conducted in urban plumes have not found evidence for significant positive interference in the UV absorption technique.

E.3 ENVIRONMENTAL DISPERSAL, AMBIENT CONCENTRATIONS, AND HUMAN EXPOSURE TO OZONE

Key findings/conclusions derived from Chapter 3 are as follows:

- Ozone is monitored in populated areas in the United States during "ozone seasons", which vary in length depending on location. All monitors should be operational from May to September. However, in many areas, O_3 is monitored throughout the year.

- The median of the mean daily maximum 8-h average O_3 concentration from May to September 2000 to 2004 across the U.S. was 0.049 ppm on a countywide average basis. Ninety five per cent of countywide mean daily maximum 8-h average O_3 concentrations were less than 0.057 ppm for the same period. Because most monitors are located in the East, these values should not be taken to represent conditions across the country.

- The daily maximum 1-h O_3 concentrations tend to be much higher in large urban areas or in areas downwind of large urban areas. For example, daily maximum 1-h O_3 concentrations in Houston, TX approached 0.20 ppm during the same period.

- Daily maximum 8-h average O_3 concentrations are lower than, but are highly correlated with, 1-h daily maximum O_3 concentrations. For example, in the Baltimore, MD area, the correlation coefficient between the two quantities was 0.98 for data obtained from May to September 1994 to 2004.

- Within individual MSAs, O_3 tends to be well correlated across monitoring sites. However, there can be substantial spatial variations in concentrations. Ozone in city centers tends to be lower than in regions either upwind or downwind of the center, because of titration by NO emitted by motor vehicles.

- Ozone concentrations tend to peak in early- to mid-afternoon in areas where there is strong photochemical production and later in the day in areas where transport is more important in determining O_3 abundance.

- Summertime maxima in O_3 concentrations occur in areas in the United States where there is substantial photochemical activity involving O_3 precursors emitted from human activities. Maxima can occur anytime from June through August.

- Springtime maxima are observed in relatively remote sites in the western United States and at various other relatively unpolluted sites throughout the Northern Hemisphere. Relatively high O_3 concentrations can also be found during winter in several cities throughout the southern United States.

- Long-term trends in O_3 concentrations reflect notable decreases over time throughout the United States, with decreases nationwide of approximately 29% in 2nd highest 1-h O_3 concentrations from 1980 to 2003 and of about 21% in 4th highest 8-h O_3 concentrations during the same time period.

- These trends include dramatic decreases from peak 1-h O_3 levels of 0.4 to 0.6 ppm seen in the Los Angeles area at times in the late 1950's to 1970's to current peak levels of 0.17 ppm and 0.15 ppm (1-h and 8-h avg, respectively) seen in the Los Angeles basin during 2000-2003.

- Downward trends in the upper tail of the O_3 concentration distribution do not reflect trends for O_3 values towards the center of the O_3 concentration distribution nationwide. These latter concentrations have remained more or less constant, and O_3 values in the lower tail of the distribution show some evidence of slight increases.

- Policy relevant background (PRB) O_3 concentrations are used for assessing risks to human health associated with O_3 produced from anthropogenic sources in the United States, Canada

and Mexico. Because of the nature of the definition of PRB concentrations, they cannot be derived from observations directly, instead they must be derived from model estimates.

- Current model estimates indicate that PRB O_3 concentrations in the United States surface air are generally 0.015 ppm to 0.035 ppm. Such concentrations decline from spring to summer and are generally <0.025 ppm under conditions conducive to high O_3 episodes. PRB Ozone concentrations may be higher, especially at elevated sites during the spring, due to enhanced contributions from (a) pollution sources inside and outside North America and (b) stratospheric O_3 exchange.

- Sufficient data for other oxidants (e.g., H_2O_2, PAN) and oxidation products (e.g., HNO_3, H_2SO_4) in the atmosphere are not available for use in epidemiologic time series studies. Limited data for oxidants besides O_3 in the gas and particle phases suggest that their combined concentrations are probably <10 % that of O_3.

- Relationships between O_3 and $PM_{2.5}$ are complex, in part because PM is not a distinct chemical species, but is a mix of primary and secondary species. For example, $PM_{2.5}$ concentrations were positively correlated with O_3 during summer, but negatively correlated with O_3 during the winter at Ft. Meade, MD. Similar relationships were found for PM_{10} and O_3 in data collected in a number of urban areas during the 1980s.

- Humans are exposed to O_3 either outdoors or in various microenvironments. Ozone in indoor environments results mainly from infiltration from outdoors. Once indoors, O_3 is removed by deposition on and reaction with surfaces and reactions with other pollutants. Hence, O_3 levels indoors tend to be notably lower than outdoor O_3 concentrations measured at nearby monitoring sites, although the indoor and ambient O_3 concentrations tend to vary together (i.e., the higher the ambient, the higher the indoor O_3 levels).

- Personal exposure to O_3 tends to be positively associated with time spent outdoors. Although O_3 concentrations obtained at stationary monitoring sites may not explain the

variance in individual personal exposures, they appear to serve reasonably well as surrogate measures for aggregate personal exposures.

- Atmospheric reactions between O_3 and certain other ambient airborne contaminants, e.g., terpenes emitted by vegetation or wood products, contribute to generation of ultrafine particles, with formation of such particles being observed in both urban and rural areas. These reactions also occur in indoor environments and involve O_3 infiltrating from outdoors and terpenes emitted by household products (e.g., air fresheners). Gaseous products resulting form such reactions may also be toxic.

E.4 DOSIMETRIC STUDIES

Chapter 4 discusses dosimetric issues, including factors that are important to consider in attempting animal-to-human extrapolations of experimentally-induced O_3 effects.

- Dosimetric studies seek to quantify dose and factors affecting the dose of O_3 and/or its active metabolites at specific lung regions, target tissues, or cells.

- In both humans and animals, the efficiency of O_3 uptake is greater in the nasal passages than the oral pathway. In the lower respiratory tract, increasing tidal volume increases O_3 uptake, whereas increasing flow or breathing frequency decreases O_3 uptake. As flow is increased, O_3 uptake shifts to the smaller peripheral airways.

- In adult human females relative to males, the smaller airways and associated larger surface-to-volume ratio enhance local O_3 uptake and cause somewhat reduced penetration of O_3 into the distal lung. However, it is not clear from these findings if the actual anatomical location of O_3 uptake differs between males and females.

- Similarly exposed individuals vary in the amount of actual dose received, but O_3 uptake is not predictive of intersubject variability in FEV_1.

- The efficiency of O_3 uptake is chemical-reaction rate dependent and the reaction products (hydrogen peroxide, aldehydes, and hydroxyhydroperoxides) created by ozonolysis of lipids in ELF and cell membranes appear to mediate O_3 toxicity.

- Ozone uptake in humans is increased by exposure to NO_2 and SO_2 and decreased during the O_3 exposure. This suggests that an inflammatory response during exposure to NO_2 and SO_2 may elicit increased production of O_3-reactive substrates in the epithelial lining fluid and that these substrates are depleted by O_3 exposure but not by NO_2 and SO_2 exposures.

- New experimental work in rats suggests that the primary site of acute O_3-induced cell injury is the conducting airways, whereas prior modeling studies suggested that the proximal alveolar and centriacinar regions may be principal O_3 target sites.

- In most clinical studies, humans are exposed to O_3 during exercise. Under these conditions, the switch from nasal to oral breathing, coupled with increases in respiratory flow (as occurs during exercise), causes a shift in the O_3 dose distribution, thusly allowing O_3 to penetrate deeper into the lung and thereby increasing the potential for damage to bronchiolar and alveolar tissues.

- Comparisons of acute exposures in rats and humans suggest that, though both species have similar qualitative responses to O_3 exposure, there are interspecies mechanistic disparities that necessitate careful comparisons of dose-response relationships. Currently available data suggest that lowest observable effect levels in resting rats are approximately 4- to 5-fold higher than for exercising humans for toxicological endpoints, including BAL protein and BAL PMNs.

E.5 ANIMAL TOXICOLOGY ASPECTS

Key toxicology findings/conclusions from laboratory animal studies discussed in Chapter 5 include:

E.5.1 Respiratory Tract Effects of Short-Term Exposures to Ozone

In general, O_3 concentration and duration of exposure (C and T), respectively, determine the dose and resultant health effects of O_3. Concentration usually dominates the response, with the impact of T being C-dependent (at higher Cs, the impact of T tends to be greater).

Effects on Pulmonary Function

- Rapid shallow breathing, which is not protective, but does cause a more evenly distributed injury pattern, is the most common change in pulmonary function induced by acute (1-8 h) O_3 exposure of ~0.2 ppm. Decreased lung volumes are observed in rats with acute exposures at levels of 0.5 ppm. Breathing mechanics (compliance and resistance) are affected at exposures of ~1.0 ppm.

- Attenuation of pulmonary function decrements occurs with 5 days of repeated acute O_3 exposures, which are not accompanied by concurrent attenuation of lung injury and morphological changes, indicating that the attenuation does not result in protection against all the effects of O_3.

- Ozone-induced airway hyperresponsiveness (AHR) occurs in laboratory animals with acute exposures ($\leq$1 h) in the range of 0.5 to 1.0 ppm. Animal studies have shown that O_3 exposure can augment OVA-induced AHR. A temporal relationship exists between inflammatory cell influx and O_3-induced AHR, but inflammation is not a prerequisite of AHR. Repeated O_3 exposures enhance AHR, possibly by modulating rapidly adapting airway receptors or by altering the structure of conducting airways. In human asthmatics, AHR appears to be due, in part, to chronic inflammation and airway remodeling.

- Studies using repeated O_3 exposure ($\leq$0.3 ppm) of nonsensitized laboratory animals have shown equivocal results. A few studies in sensitized laboratory animals are consistent with the O_3-induced exacerbation of AHR reported in atopic humans with asthma. However, extrapolation of these data is difficult due to interindividual and interspecies differences in responsiveness to bronchoprovocation and possible adaptation of airway responsiveness with long-term, repeated O_3 exposures.

Other Respiratory Tract Effects of Ozone

- Due to its high reactivity, O_3 penetrates only about 0.1 to 0.2 μm into the extracellular lining fluid (ELF) of the respiratory tract. Ozone interacts with a wide range of components in ELF that include polyunsaturated fatty acids, cholesterol, amino acid residues, reduced glutathione, uric acid, vitamins C and E, and free amino acids. Ozone's toxicity is dependent upon a cascade of reaction products, including ozonide, aldehyde, and hydroperoxide. Saturated phospholipids are thought to reduce the local dose and limit site-specific cell injury from O_3 exposure.

- Antioxidants present in ELF act to protect lung tissue from O_3-induced injury, but even with environmentally relevant exposures, the reactivity of O_3 is not quantitatively fully quenched. Thus, cell injury occurs in both the upper and lower respiratory tract. Short-term exposures to <1 ppm O_3 increase antioxidant metabolism. Previous O_3 exposure does not appear to be protective upon re-exposures.

- Both short- and long-term exposures to O_3 have been shown to enhance lung xenobiotic metabolism, possibly due to changes in the number and function of bronchiolar epithelial Clara cells and alveolar epithelial Type 2 cells. Elevations in enzyme activity appear to increase as a function of age, suggesting that O_3 exposure can cause greater lung injury in the older animal. Some studies found an effect on liver xenobiotic enzymes with exposure to O_3 concentrations as low as 0.1 ppm, whereas others did not detect alterations in metabolic enzymes even at 1 ppm, the effects appearing to be highly species-specific.

- Acute exposures of 0.1 ppm O_3 disrupt the barrier created by airway mucosa in the normal lung, resulting in an increase in serum proteins, bioactive mediators, and neurophils in the interstitium and air spaces of the lung. In rats, a single 3 h exposure to 0.5 ppm O_3 produces a significant increase in both lung permeability and inflammation. Ozone-induced permeability changes appear to occur predominantly in the trachea and bronchioalveolar regions compared to nasal passages. Species differences exist in responses, with guinea pigs being the most responsive; rabbits the least; and rats, hamsters, and mice intermediate. With continuing exposure, the increases in BALF protein and PMNs typically peak after a few

days and return toward control levels even with continuing exposure. Though inflammation and increased permeability occur somewhat concurrently, they are distinct events controlled by independent mechanisms.

- Important mechanisms of O_3-induced inflammation and injury involve inflammatory cytokines and chemokines, which are released as a result of stimulation or injury of macrophages, epithelial cells and PMNs. In vitro exposures to O_3 induce release of the cytokines IL-6, TNF-α, IL-1β, and IL-8. In vivo exposures of O_3 induce release of MIP-2, IL-6, MIP-1α, CINC, eotaxin and fibronectin. Studies utilizing antibodies to selected pro- or anti-inflammatory cytokines suggest a role of TNF-α, interleukin-10 (IL-10) and IL-1β in O_3-induced changes in permeability, inflammation and cytokine release.

- Cell adhesion molecules (e.g., ICAM-1) and extracellular matrix proteins (e.g., fibronectin) modulate O_3-induced lung inflammation and injury. Ozone exposure also affects macrophage functions by increasing their production of nitric oxide, superoxide anion and PGE_2.

- Mucociliary clearance is affected in most test species at just under 1 ppm, with lower levels (~0.1 ppm) increasing clearance and somewhat higher levels decreasing clearance. At O_3 exposures of 0.1 to 1.2 ppm, alveolar macrophage (AM) function is disrupted and the number of AM are increased. Ozone exposures are linked to decreased resistance to microbial pathogens.

- Ozone exposures can enhance or suppress immune responsiveness, depending on the species studied, the concentration of O_3, the route of exposure of allergen, and exposure timing. Continuous exposure to O_3 impairs immune responses for the first several days of exposure, followed by an adaptation to O_3 that allows a return of normal immune responses. Most species show little effect of O_3 exposures prior to immunization, but exhibit suppression of responses to antigen with O_3 exposures post-immunization. Ozone exposures are linked to a possible interaction between the innate and acquired immune system and a shift in the immune response towards a Th-2-like pattern. Surfactant proteins A and D, which have an

immunomodulatory function in protecting against oxidative stress, are affected by O_3 exposures. Ozone exposures at levels of 0.1 to 1 ppm O_3 for 1 week have been shown to cause, in general, increased mortality and morbidity, decreased clearance, increased bacterial growth, and increased severity of infection at exposure.

- Age, gender, nutritional status, genetic variability, exercise and exposure to co-pollutants are all factors which can impact the effects of O_3. Control of the ventilatory response to O_3 is determined, at least in part, by genetic factors. Genetic loci that modulate pulmonary responses to O_3 differ from each other and from loci controlling inflammatory responses. The effects of age and gender on lung inflammation are not well characterized, but exercise during O_3 exposure clearly generally increases susceptibility.

- Collagen increases with O_3 exposure and can persist after exposure stops. Rats exposed acutely or subchronically to 0.4 ppm O_3 showed centriacinar thickening of septa. Collagen content decreased with postexposure recovery time but not the structural fibrotic changes in ductular septa and respiratory bronchioles, suggesting that subchronic O_3 exposures in rats creates a progression of structural lung injury that can evolve to a more chronic form that likely includes fibrosis.

- Ozone-induced alterations in lung structure have been shown across a variety of species repeatedly exposed to O_3 concentrations as low as 0.15 ppm. Cells in the centriacinar region (CAR) are the primary targets of O_3, but ciliated epithelial cells in the nasal cavity and airways and Type 1 epithelial cells in the gas exchange region are also targeted. Ozone-induced fibrotic changes in the CAR are maximal at 3 d of exposure and recover 3 d postexposure with exposures to 0.2 ppm in rodents. Rats with induced allergic rhinitis are more susceptible to 0.5 ppm than are controls. The proximal respiratory bronchiole receives the most acute epithelial injury from exposures $\leq$1 ppm, while metabolic effects are greatest in the distal bronchioles and minor daughter airways.

E.5.2 Respiratory Tract Effects of Chronic (Long-Term) Exposures to Ozone

A variety of respiratory tract effects have been shown to occur as the result of more chronic, longer-term exposures of laboratory animals to O_3. Some of the more notable types of effects are as follow.

- Chronic O_3 exposures in a range of 0.5 to 1.0 ppm induce a pattern of epithelial hyperplasia which is similar to the pattern of inflammation, with a peak over the first few day, a drop, and then disappearance. In contrast, fibrotic changes in lung tissue increase very slowly over months of exposure, and, after exposure ceases, the changes sometimes persist or increase. Compared to continuous exposure regimens, seasonal episodic exposures demonstrated remodeling in the distal airways, abnormalities in tracheal basement membrane, eosinophil accumulation in conducting airways, and decrements in airway innervation. Also, long-term O_3 exposures have demonstrated that repeated daily exposure of rats to an episodic profile of O_3 caused small, but significant decrements in lung function that were consistent with early indicators of focal fibrogenesis in the proximal alveolar region, without overt fibrosis.

E5.3 Other Types of Ozone Exposure Effects Observed in Laboratory Animal Models

Systemic Effects of Ozone

- Decreased heart rate, core temperature, and blood pressure, all collectively termed the hypothermic response, are other types of effects observed at concentrations of 0.3 to 0.5 ppm. Concentrations of $O_3 \geq 0.5$ ppm cause tissue edema (possibly mediated by atrial natriuretic factor). Additionally, O_3-induced production of platelet-activating factor and oxysterols suggest mechanisms of cardiovascular injury.

- Neurobehavioral effects attributed to O_3 exposure (0.2 to 1.0 ppm) include decreased motor activity, short- and long-term memory deficits, increased freezing behavior, and decreased exploratory behaviors. Near-ambient exposures to O_3 elicit neuroendocrine effects, including morphological and hormonal changes in the pituitary-thyroid-adrenal axis and alterations of visual and olfactory neural pathways.

- No noticeable neurobehavioral or somatic effects have been observed with prenatal exposures of <1.0 ppm. Effects on neonatal mortality are observed with exposures of 1.0 to 1.5 ppm. Effects on spleen and thymus appear to only occur at high O_3 concentrations (>1.0 ppm), whereas relevant urban ambient exposures have no effect on systemic immune function in rats.

Genotoxicity Potential of Ozone

- The weight of evidence from new experimental studies, utilizing non-lifetime exposures, does not appear to support ambient O_3 as a pulmonary carcinogen in laboratory animal models. New data are in agreement with the 1994 National Toxicology Program evaluation of O_3 carcinogenicity. However, O_3 could possibly act as a co-carcinogen functioning to stimulate hyperplasia.

Interactions of Ozone with Other Co-occurring Pollutants

- Bases for toxic interactions of O_3 with co-occurring pollutants may include: adsorption of O_3 onto a co-pollutant with transport to another site; production of toxicologically active secondary products; biological or chemical alterations at target sites that affect response to O_3 or the co-pollutant; O_3- or co-pollutant-induced physiological change, such as alteration in ventilation pattern, resulting in changes in the penetration or deposition of one pollutant when another is present.

- Generalizations regarding interactions of O_3 and co-pollutants include: interactions of O_3-containing mixtures are generally synergistic; O_3 may produce more significant biological responses as a component of a mixture than when inhaled alone; and, although most studies have shown that interaction occurs only at higher than ambient concentrations with acute exposure, some have demonstrated interactions at more environmentally relevant levels (e.g., 0.05 to 0.1 ppm O_3 with NO_2) and with repeated exposures.

Effects of Other Photochemical Oxidants

- Ambient concentrations of the most abundant non-O_3 oxidants (peroxyacetyl nitrate, peroxypropionyl nitrate, and H_2O_2) have not been shown as being likely to cause adverse

health effects. However, as constituents of ambient air mixes, other ambient oxidants may contribute to some effects attributed to O_3.

E.6 CONTROLLED HUMAN EXPOSURE STUDIES

Key findings/conclusions derived from Chapter 6 assessment of experimental human studies include:

- Responses in humans exposed to ambient O_3 concentrations include decreased inspiratory capacity; mild bronchoconstriction; rapid, shallow breathing pattern during exercise; and symptoms of cough and pain on deep inspiration. Ozone exposure also results in airway hyperresponsiveness, inflammation, immune system activation, and epithelial injury.

- Young healthy adults exposed to O_3 concentrations of 0.08 ppm develop significant reversible, transient decrements in pulmonary function if minute ventilation or exposure duration are increased sufficiently. Healthy children experience similar spirometric responses but lesser symptoms from O_3 exposure relative to young adults. On average, spirometric and symptom responses to O_3 exposure appear to decline with increasing age beyond approximately 18 years of age.

- There is a tendency for slightly increased spirometric responses in mild asthmatics and allergic rhinitics relative to healthy young adults. Spirometric responses in asthmatics appear to be affected by baseline lung function.

- There is a large degree of intersubject variability in physiologic and symptomatic responses of adults exposed to O_3. However, responses tend to be reproducible within a given individual over a period of several months. With increasing O_3 concentration, the distribution of FEV_1 decrements becomes asymmetrical with a few individuals experiencing large decrements. An individual's innate susceptibility to ozone may be linked to the genetic background of an individual. Additional studies, however, are needed to ascertain the link between susceptibility and polymorphisms.

- Soluble mediators of inflammation such as the cytokines (IL-6, IL-8) and arachidonic acid metabolites (e.g., PGE2, PGF2", thromboxane, and leukotrienes [LTs] such as LTB4) have been measured in the BAL fluid of humans exposed to O_3. There appears to be no strong correlation between any of the measured cellular and biochemical changes and changes in pulmonary function. A limited number of studies suggest that inflammatory responses may be detected following O_3 exposures that are insufficient to cause decrements in pulmonary function.

- With repeated O_3 exposures over several days, spirometric and symptom responses become attenuated, but this tolerance is lost after about a week without exposure. Some markers of airway inflammation and small airways dysfunction may not be attenuated by repeated O_3 exposures.

- An initial phase of recovery from O_3 exposure in healthy individuals proceeds relatively rapidly, with acute spirometric and symptom responses resolving within about 2 to 4 h. Effects on the small airways, assessed by decrements in FEF_{25-75} and altered ventilation distribution at and possibly beyond 24 h, may be partly due to inflammation. Some inflammatory and cellular changes may persist for up to 48 h, but the time course for these parameters in humans has not been explored fully.

E.7 EPIDEMIOLOGIC STUDIES

Many epidemiologic studies, as discussed in Chapter 7, have shown associations of acute exposure to ambient O_3 with a variety of human health endpoints, including pulmonary function, respiratory symptoms, hospital admissions, and mortality. Key findings and conclusions regarding O_3 health effects drawn from the epidemiologic evidence and the issues that may affect the interpretation of the effect estimates can be briefly summarized as follows.

E.7.1 Health Effects Associated with Acute Ozone Exposures

- Field/panel studies of acute O_3 effects. Recent field/panel studies continue to confirm that short-term O_3 exposure is associated with acute decrements in lung function and increased

respiratory symptoms, particularly in children and asthmatics. There is also suggestive evidence that O_3 is related to increased asthma medication use. Taken together with the evidence from controlled human exposure studies, O_3 is likely causally related to the various respiratory health outcomes. The current evidence is limited but supportive of a potential effect of O_3 on heart rate variability, ventricular arrhythmias, and the incidence of myocardial infarctions.

- Acute O_3 effects on emergency department visits and hospitalizations. Large multicity studies, as well as many studies from individual cities have reported an association of short-term O_3 concentrations with respiratory and cardiovascular hospital admissions. Studies using year-round data noted some inconsistencies in the O_3 effect on daily hospitalizations. However, studies with data restricted to the summer or warm season, in general, indicated positive and robust associations between short-term (e.g., 1 h or 8 h) ambient O_3 concentrations and cardiopulmonary hospital admissions. Results for emergency department visits are less consistent.

- Acute O_3 effects on mortality. The majority of the studies suggest that an elevated risk of all-cause mortality is associated with acute exposure to O_3, especially in the summer or warm season when O_3 levels are typically high. Slightly greater O_3 effects were observed for cardiovascular mortality. Results from a recent, large U.S. multicity time-series study provide the strongest evidence to-date for acute O_3 exposure effects on mortality. Recent meta-analyses also showed consistent risk estimates that are unlikely to be confounded by PM; however, future work is needed to better understand the influence of model specifications on the risk coefficient.

- Age-related differences in O_3 health effects. Supporting evidence exists for heterogeneity in the effects of O_3 by age. The elderly population (>65 years of age) appear to be at greater risk of O_3-related hospitalizations and mortality compared to all age or younger populations. In addition, potentially adverse respiratory health outcomes were associated with O_3 exposure in children (<18 years of age).

- Ozone health effects in asthmatics. The effects of O_3 on asthmatics have been examined widely in both time-series studies and panel studies. Associations of O_3 with various respiratory health outcomes (including lung function declines, increased respiratory symptoms, and emergency department visits) were observed. These findings, along with the pathophysiologic understanding of asthma as a chronic inflammatory disease, indicate that asthmatics may be a notably susceptible population affected by O_3 exposures.

E.7.2 Issues Potentially Affecting Interpretation of Acute Exposure Studies

- Exposure assessment. Exposure misclassification may result from the use of stationary ambient monitors to determine exposure in population studies. Although central ambient monitors do not explain the variance of individual personal exposures, significant correlations are found between aggregate personal O_3 measurements and O_3 concentrations from ambient monitors. A simulation study indicated that the use of ambient monitor data will tend to underestimate the O_3 effect. Better understanding of the factors that affect the relationship between ambient concentrations and personal exposures should help to improve interpretation of the O_3 effect estimates.

- Ozone exposure indices. The three most commonly used daily O_3 exposure indices, 1-h max O_3, 8-max O_3, and 24-h avg O_3, were found to be highly correlated in studies conducted in various regions. In addition, effect-size estimates and significance of associations across all health outcomes were comparable when using standardized distributional increments of 40 ppb, 30 ppb, and 20 ppb for 1-h max O_3, 8-h max O_3, and 24-h avg O_3, respectively.

- Lag structures for O_3 exposure and effect. The lag time between O_3 exposure and effect may differ depending on various factors such as the specific health outcome of interest, the mechanism of effect, and preexisting health conditions. The majority of the studies found an immediate O_3 effect, with the strongest associations observed between health outcomes and O_3 exposure on the same day and/or previous day. Some studies found large cumulative effects of O_3 over longer lag periods, indicating that multiday lags also may be relevant for some health outcomes, including mortality.

- Sensitivity to model specifications for temporal trends. Ozone effect estimates that were reported in studies whose main focus was PM often were calculated using the same model specifications as PM. While the sensitivity of the O_3 risk estimates to alternative model specifications has not been throughly investigated, the limited available evidence indicates that O_3 effects appear to be robust to various model specifications for temporal trend adjustment.

- Influence of seasonal factors. An evaluation of the confounding effects of meteorologic factors and copollutants on O_3 risk estimates is complicated by their changing relationships with O_3 across seasons. In addition, seasonal or seasonally-modified factors (e.g., air conditioning use, time spent outdoors) complicate interpretation of all-year effect estimates, as they affect the relationship between ambient concentrations and personal exposures. Given the potentially significant influence of season, season-specific analyses are more informative in assessing O_3 health risks.

- Confounding by copollutants. Multipollutant regression models often are used to adjust for confounding by copollutants. Although there is some concern regarding the use of multipollutant models given the varying concurvity across pollutants, currently available results generally suggest that the inclusion of copollutants into the models do not substantially affect O_3 risk estimates. These findings indicate that effects of O_3 on various health outcomes are robust and independent of the effects of other copollutants.

- Concentration-response function. In the limited mortality and morbidity studies that have specifically examined the O_3 concentration-response relationship, the evidence is inconclusive regarding the detection of any clear effect threshold. Factors such as exposure measurement error may reduce the ability to detect a threshold in population studies.

- Heterogeneity of O_3 health effects. Consistent O_3 effect estimates have generally been observed for mortality, hospitalizations, and other respiratory health outcomes in multicity studies. Some other reported geographic heterogeneity in effect sizes may be attributable to differences in relative personal exposure to O_3, which is affected by variations in factors such

as air conditioning prevalence and human activity patterns as well as the varying concentrations and compositions of copollutants present by region.

E.7.3 Health Effects Associated with Chronic Ozone Exposure

Many fewer studies have investigated the effects of chronic O_3 exposure on morbidity and mortality. The strongest evidence is for negative seasonal effects of chronic O_3 exposure on lung function in adults and children. Less conclusive are longer-term studies investigating the association of chronic O_3 exposure on yearly lung function, asthma incidence, and respiratory symptoms. Studies of potential chronic O_3 exposure-mortality relationships have generally observed inconsistencies across exposure periods, cause-specific mortality outcomes, and gender.

E.8 INTEGRATIVE SYNTHESIS

This section summarizes key conclusions derived from the Chapter 8 integrated synthesis of information regarding health effects associated with ambient O_3 exposures. The conclusions were derived based on an integrated analysis of available laboratory animal, human clinical, and epidemiological studies that have evaluated health effects associated with short-term, repeated, and long-term exposures to O_3 alone or in combination with other ambient pollutants. The Chapter 8 synthesis utilized experimental evidence from dosimetric and human and animal toxicological studies presented in Chapters 4, 5, and 6 both to evaluate the biological plausibility of health effects observed in epidemiologic studies discussed in Chapter 7 and to inform delineation of O_3 exposure-dose-response relations and likely underlying mechanisms of action. These evaluations are also aimed at identifying susceptible populations that are at potentially greater risk for effects of O_3 exposure.

1. Health effects of acute (short-term) exposures to Ozone

Numerous field panel and time-series epidemiologic studies (using better weather models and adjustments to confounding copollutants than those assessed in the 1996 O_3 AQCD) have evaluated the effects of short-term exposure to O_3 on a wide range of health endpoints, from lung

function decrements to mortality. Results from the majority of studies continue to support the conclusions reported in the 1996 O_3 AQCD.

- Panel studies typically have evaluated the effects of short-term O_3 exposure on both healthy individuals and people with cardiopulmonary diseases. These evaluations included measurement of lung function changes, respiratory symptoms and use of asthma medication.

- Clinical controlled exposure studies in humans indicate changes in lung function and respiratory symptoms that vary as a function of exposure concentration, duration and level of exercise.

- Newer meta-analyses confirmed the interindividual differences in lung function decrements reported in the 1996 O_3 AQCD. Age-specific differences in the lung function responses were also observed. Spirometric responses (due to decrements in lung function) in healthy adults exposed to near ambient O_3 levels typically resolve to near baseline values within 4-6 h.

- Meta-analyses of four controlled human exposure studies (two new and two reported in the 1996 O_3 AQCD) reporting the effects of prolonged (6.6 h) exposures to 0.08 ppm O_3 during moderate exercise on pulmonary function in young healthy adults (M = 90, F = 30; mean age, 23 yrs) indicate an absolute FEV_1 decrease of approximately 6%, whereas FEV_1 increased by about 1% following free air (FA) exposures.

- Recent meta-analyses on numerous clinical studies indicate interindividual differences in lung inflammatory response to short-term O_3 exposures.

- Inflammatory and permeability responses also generally resolve (in some instances complete recovery) within a week or two after cessation of O_3 exposure, but exhibit differential attenuation profiles between normal healthy subjects and people with preexisting respiratory diseases. However, some lung inflammation markers may not completely resolve readily, and mild persistent inflammation has been reported.

- Field/panel studies of healthy individuals and asthmatics have found positive associations between short-term exposure to O_3 and decrements in lung function analogous to those shown by studies of controlled short-term (1 to 8 h) human exposures to O_3.

- Associations between short-term O_3 exposures and school absenteeism (due to respiratory illness) have also been suggested.

- With regard to cardiac impacts, a limited number of field studies that examined the relationship between short-term O_3 exposures and cardiovascular effects (heart rate variability, myocardial infarction) suggest an association.

- A large multicity and several single-city studies have indicated a positive association between increased O_3 levels (especially during the warm season) and increased risk for hospital admissions. On the other hand epidemiologic data on emergency department visits do not suggest such an association with increase in ambient O_3 levels.

- The results of two large multicity studies from the U.S. and several single-city studies suggest a positive association between increases in O_3 levels and all-cause (non-accidental) daily mortality. Meta-analyses on the influence of season suggest a causal association. Additional meta-analyses on cause-specific mortality are suggestive of a likely positive association between increases in ambient O_3 levels and cardiovascular mortality.

- Short-term O_3-induced lung function decrements, respiratory symptoms, inflammation and permeability changes observed in animal toxicology studies are consistent with human studies.

2. Health effects of repeated short-term exposures to Ozone

The results of new controlled human exposure studies of repeated short-term O_3 exposures continue to support the health effects findings/conclusions reported in the 1996 O_3 AQCD.

- Repeated exposure studies at higher concentrations typically show that FEV_1 response to O_3 is enhanced on the second of several days of exposure. Such an enhanced response was not

observed at lower O_3 concentrations. With repeated O_3 exposures over several days, spirometric and symptom responses become attenuated, but this tolerance is lost after about a week without exposure.

- In humans repeatedly exposed to 0.4 ppm O_3 for 5 consecutive days, several indicators of inflammation (e.g., PMN influx, IL-6, PGE_2, BAL protein, fibronectin) were attenuated after 5 days of exposure. However, lung injury and permeability markers (LDH, IL-8, total protein, epithelial cells) did not show attenuation, indicating that tissue damage probably continues to occur during repeated exposure. The recovery of the inflammatory response occurred for some markers after 10 days, but some responses were not normalized even after 20 days. The continued presence of cellular injury markers indicates a persistent effect that may not necessarily be recognized due to the attenuation of spirometric and symptom responses.

- Repeated daily exposure to lower concentrations of O_3 (0.125 ppm for 4 days) causes an increased response to bronchial allergen challenge in subjects with preexisting allergic airway disease, with or without asthma. In these subjects, changes in airway responsiveness after O_3 exposure appear to be resolved more slowly than changes in FEV_1 or respiratory symptoms.

3. Health effects of long-term exposures to Ozone

Assessment of human health effects associated with long-term O_3 exposures is hampered by the lack of pertinent data from human clinical and epidemiologic studies. Chronic animal toxicology studies continue to support structural alterations in several regions of the respiratory tract and identify the centriacinar region of the lung as the most affected region.

- Animal toxicology studies that utilized exposure regimens to simulate seasonal exposure pattern also report increased lung injury compared to conventional chronic stable exposures. One long-term study of infant rhesus monkeys exposed to simulated seasonal O_3 patterns (0.5 ppm 8h/day for 5 days, every 14 days for 11 episodes) demonstrated: (1) remodeling in the distal airways; (2) abnormalities in tracheal basement membrane; (3) eosinophil accumulation in conducting airways; (4) decrements in airway innervation. These findings

advance earlier information regarding possible injury-repair processes occurring with seasonal O_3 exposures.

- Effects of O_3 on the upper respiratory tract of F344 rats exposed to O_3 (0.12, 0.5, or 1.0 ppm for 20 months) included marked mucous cell metaplasia in the rats exposed to 0.5 and 1.0 pm O_3, but not at 0.12 ppm O_3. The persistent nature of the O_3-induced mucous cell metaplasia suggests that O_3 exposure may have the potential to induce similar long-lasting alterations in the airways of humans. Hyperplasia in the nasal epithelium of rats exposed to 0.25 and 0.5 ppm, 8h/day, 7 days/week, for 13 weeks has been reported.

- Pathophysiological changes associated with chronic O_3 exposures observed in animal studies suggest possible similar alterations in humans. The pulmonary function changes observed in children in polluted metropolitan areas and lung structural alterations reported in an autopsy study in Los Angeles suggest a role for long-term ambient O_3 exposure, but such possible effects need to be further evaluated with improved study design(s).

4. Susceptibility factors associated with exposure to ozone

Various factors such as age, gender, nutrition, socioeconomic, activity patterns, and disease status have been shown to influence the response to environmental air pollutants. Controlled human exposure studies clearly established differential biological response to O_3 based on physical activity (exertion) and age. These studies also demonstrated a large variation in sensitivity and responsiveness to O_3. The specific factors that contribute to this intersubject variability are yet to be identified.

- Increased hospital admissions for asthma and COPD in summer (with increased levels of ambient O_3) suggest that people with these respiratory diseases as potential sub-population for O_3-induced health effects.

- Similarly, based on O_3-induced differential responses in lung inflammation and in airway hyperresponsiveness, asthmatics (including children) appear to have potentially increased susceptibility to O_3. However, there is no supportive data from controlled human studies suggesting individuals with COPD are more sensitive to O_3-induced health effects.

- Animal toxicology studies provided supportive evidence to the observations of varied susceptibility. Various strains of mice and rats have demonstrated the importance of genetic background in O_3 susceptibility. Moreover, genetic and molecular characterization studies in laboratory animals identified genetic loci responsible for both sensitivity and resistance.

- Consistent with the 1996 O_3 AQCD, the scarcity of data prevents determination of the role of ethnic or racial background and nutrition status on O_3-induced health effects. However, as presented in this document, exercising (moderate to high physical exertion) healthy adolescents and asthmatics appear to demonstrate increased responsiveness to ambient concentrations of O_3 and may be susceptible for O_3-induced health effects.

5. Health effects of binary pollutant mixtures containing ozone

A limited number of controlled human exposure studies and a few animal toxicology studies of binary mixtures containing O_3 suggest potential interactions, depending on specific exposure regimens and copollutant constituents.

- Continuous exposure to SO_2 and NO_2 increased inhaled bolus O_3 absorption, while continuous exposure to O_3 decreased O_3 bolus absorption. Asthmatics exhibited enhanced airway reactivity to house dust mite following exposures to O_3, NO_2, and the combination of the two gases. Spirometric response, however, was impaired only by O_3 and O_3+NO_2 at higher concentrations.

- Animal toxicology studies with O_3 in mixture with NO_2, formaldehyde, and PM demonstrated additive, synergistic or antagonistic effects, depending on the exposure regimen and the endpoints evaluated.

- One controlled exposure study of children, designed to approximate exposure conditions of an epidemiologic study by matching the population and exposure atmosphere (0.1 ppm O_3, 0.1 ppm SO_2 and 101 $\mu g/^{m2}$ H_2SO_4), failed to support the findings of the epidemiologic study. This study points out difficulties in trying to link the outcomes of epidemiologic and controlled exposure studies by use of binary pollutant mixtures.

E.9 VEGETATION AND ECOLOGICAL EFFECTS

General

- Data published since 1996 continue to support the conclusions of previous O_3 AQCDs that there is strong evidence that ambient O_3 concentrations cause foliar injury along with growth and yield damage to numerous common and economically valuable plant and tree species.

- Research to date has continued its focus at the species level, with very few new studies at the ecosystem level. The lack of quantification of biotic and abiotic factors impinging on the individual to population organizational levels results in a limited ability to scale O_3 responses to the ecosystem level. Therefore, a high degree of uncertainty remains in our ability to assess ozone risk to ecological resources and the services they provide.

Methodologies

- Since the 1996 AQCD free-air exposure (FACE) systems have come into more frequent use. FACE systems eliminate many of the concerns raised about closed or open-top chamber (OTC) experiments including small plot size, altered microclimate within OTCs, and the effect of charcoal filtering on overall air quality within OTCs. One of the advantages of the application of plume systems to O_3 research is the ability to compare response of plants in open-field systems with results from OTCs. In particular, studies with quaking aspen (*Populus tremuloides* L.) performed in OTCs, FACE, and also at sites along an ambient O_3 gradient showed that O_3 symptom expression was generally similar, supporting the previously observed level of variation among aspen clones in OTC studies.

- The lack of rural monitors continues to be a major problem in the characterization of O_3 exposures in remote areas, as well as in linking effects to exposure in natural ecosystems. Since the 1996 O_3 AQCD, the use of passive samplers has expanded monitoring efforts to include remote areas that were previously uncharacterized.

- Advancements in biomonitoring have been made since the 1996 O_3 AQCD, primarily in the area of identification and symptom verification of sensitive species . The U.S. Department of Agriculture (USDA) Forest Service continues its program to monitor O_3 effects in forested

ecosystems throughout the United States. Although results are not useful for developing exposure-response relationships or for quantifying responses to O_3, they can provide an annual assessment and correlative information regarding the extent of O_3 injury occurring across many regions of the United States.

Mode of Action

- The new information available on the mode of action of O_3 is, in part, a result of improved molecular tools for following rapid changes that occur within the leaf. Many changes occur within hours or possibly days following O_3 exposure. Other O_3 effects take longer to occur and tend to be most obvious only under exposure to low O_3 concentrations for long periods. These low-exposure chronic effects have been linked to the senescence process or some physiological response very closely linked to senescence (e.g., translocation, reabsorption, allocation of nutrients and carbon).

Modification of Growth Response

- It has been known for decades that several factors, both biotic and abiotic, alter plant response to O_3. However, only a few studies reported since the 1996 O_3 AQCD have improved our understanding of the role of these interactions in modifying plant O_3 response.

- Recent studies have supported the earlier conclusion that O_3 often increases the likelihood and success of insect attacks, but only with respect to chewing insects. Although it seems likely that some insect problems could increase as a result of greater O_3 levels, we are still far from being able to predict the nature of any particular O_3-plant-insect interaction, its likelihood, or its severity.

- O_3 exposure generally increases plant diseases associated with facultative necrotrophic plant pathogens. Generally, pathogens that benefit from damage to cells are enhanced by O_3 stress of their hosts, whereas pathogens and pests that require healthy hosts are depressed by O_3 stress.

Exposure Indices

- Exposure indices are metrics that relate measured plant damage (i.e., reduced growth) to monitored ambient O_3 concentrations over time to provide a consistent metric for reviewing and comparing exposure-response effects obtained from various studies. Since the 1996 O_3 AQCD, there has been no direct experimental testing of the adequacy of exposure indices proposed in 1996; therefore, there is no new information to alter the basic conclusions put forth in the 1996 O_3 AQCD.

- The proposed indices in the 1996 O_3 AQCD (i.e., SUM06, W126, AOT40) included various functional and statistical summaries of monitored hourly O_3 concentrations over designated time periods. The few studies that have been published since the 1996 O_3 AQCD continue to support the earlier conclusions, including the importance of peak concentrations, and the duration and occurrence of O_3 exposures in altering plant growth and yield.

- A large body of new research, mostly out of Europe, addresses the need for an index related to the actual flux of O_3 into the plant. Despite additional research linking estimates of flux with plant response since 1996, information is still insufficient to identify a flux-based model that incorporates the necessary complexity across space and time to be non-site or non-species specific. Based on the current state of knowledge, exposure indices that cumulate and differentially weight the higher hourly average concentrations, but include the mid-level values (e.g., SUM06, W126, AOT40), still represent the best approach for relating vegetation effects to O_3 exposure in the United States.

Ozone Exposure-Plant Response Relationships

- Data published since 1996 continue to support the conclusions of previous O_3 AQCDs that there is strong evidence that ambient O_3 concentrations cause foliar injury and growth and yield damage to numerous common and economically valuable plant and tree species.

- In addition to reductions in crop yield, O_3 may also reduce the quality or nutritive value of annual species. Many recent studies have found O_3 effects on various measures of plant organs that affect quality, with most of those studies focusing on characteristics important for

food or fodder. These studies indicate that ambient O_3 may have economically important effects on the quality of crop and forage species.

- Results since 1996 support the conclusion of the 1996 O_3 AQCD that deciduous trees are generally less O_3 sensitive than are most annual plants, with the exception of a few very sensitive genera such as *Populus* and sensitive species such as black cherry. Various evergreen tree species and genotypes have widely varying O_3 sensitivities. Based on OTC studies with seedlings, major evergreen species in the United States are generally less sensitive than are most deciduous trees, and slower-growing evergreen species are less sensitive than are faster-growing species. For all types of perennial vegetation, cumulative effects over more than one growing season may be important; studies for only a single season may underestimate effects.

Ecosystem Effects

- There is evidence that tropospheric O_3 is an important stressor of ecosystems, with documented impacts on the biotic condition, ecological processes, and chemical/physical nature of natural ecosystems. Effects on individual keystone species and their associated microflora and fauna, which have been shown experimentally, may cascade through the ecosystem to the landscape level, although this has not yet been demonstrated.

- Systematic injury surveys (e.g., USDA Forest Service's ozone bioindicator plot network and Europe's ICP Forests) demonstrate that foliar injury occurs on O_3 sensitive species in many regions of the United States and Europe. Frequent lack of correspondence between foliar symptoms and growth effects means that other methods must be used to estimate the regional effects of O_3 on tree growth. Investigations of the radial growth of mature trees, combined with data from many controlled studies with seedlings and a few studies with mature trees suggest that ambient O_3 is reducing the growth of mature trees in some U.S. locations.

- The study of genetic aspects of O_3 impacts on natural ecosystems has been largely based on correlations, and it remains to be shown more conclusively whether O_3 affects biodiversity or genetic diversity.

Economics

- The physical and economic effects on agriculture are well documented and provide useful information for the consideration of establishing air quality standards for crops. Effects on forests and natural ecosystems remain problematic, due to limitations in biological response data and economic methods. The problem is even more acute for valuing natural ecosystem goods and services.

E.10 TROPOSPHERIC OZONE EFFECTS ON UV-B FLUX AND ITS ROLE IN CLIMATE CHANGE

The molecular properties specific to O_3 include a capacity for absorbing incoming ultraviolet (UV) and infrared (IR) radiation, and both incoming solar and outgoing terrestrial IR radiation. Consequently, O_3 plays an essential role in shielding the earth's surface from harmful levels of UV-B radiation, by way of the stratospheric O_3 layer. Its effectiveness as a screen for the residual UV-B flux that penetrates the stratosphere and passes into the troposphere and its role in reducing UV-induced human health effects are addressed in Chapter 10. The radiation-absorbing properties of O_3 also make it a greenhouse gas (GHG) having global and, more importantly, regional consequence for climate, as also addressed in Chapter 10. Important conclusions from Chapter 10 are summarized below.

- The distribution of O_3 within the atmosphere. Ozone is distributed very unevenly within the atmosphere, with ~90% of the total atmospheric burden present in the stratosphere. The remaining ~10% is distributed within the troposphere, with higher relative concentrations near the source of its precursors at the surface. Concentrations of O_3 at the mid- and upper-troposphere vary, depending upon meteorological conditions.

- Multiple factors govern the flux of UV-B radiation at the Earth's surface. Latitude and altitude are the two most important factors that define the residual UV-B flux at the surface. Natural variation in the total column density of stratospheric O_3 is also an important factor. All of these factors are followed in importance by tropospheric clouds, particulate matter

(PM) and O_3. The effect of natural stratospheric variation, clouds, PM and tropospheric O_3 on UV fluxes within the troposphere and at the surface are each very difficult to predict.

- A UV-B "climatology" is needed to predict human exposure levels. A UV-B climatology, representing patterns and trends in UV-B flux at the Earth's surface, must be based on extended in situ observations in order to adequately capture natural variability and the effects of human activities on atmospheric UV-B absorbers. At present, the body of UV-B measurements cannot support the development of a climatology.

- Human exposure to UV-B radiation. Quantitative evaluation of human exposure to UV-B radiation is necessary to perform health risk assessment of UV-B-related health effects. Individuals who participate in outdoor sports and activities, work outdoors, live in geographic areas with higher solar flux, and/or engage in high-risk behavior (e.g., extended sun bathing) can reasonably be projected to be at increased risk for higher UV radiation exposures. However, little is known about the impact of variability in these factors on individual exposure to UV radiation.

- Human health effects of UV-B radiation. Exposure to UV-B radiation is associated with increased risk of erythema, nonmelanoma and melanoma skin cancers, ocular damage, and immune system suppression. Some studies have attempted to estimate the potential effects of changes in surface-level UV flux resulting from stratospheric O_3 depletion on these health outcomes; however, the numerous simplifying assumptions made in the assessments limit the usefulness of the risk estimates. The effect of changes in surface-level O_3 concentrations on UV-induced health outcomes cannot yet be critically assessed within reasonable uncertainty.

- Vitamin D-related health benefits of UV-B radiation. A potential health benefit of increased UV-B exposure relates to the production of vitamin D in humans. Several studies have found that UV-B radiation, by increasing vitamin D production, is associated with reduced risks of various cancers. However, as with other impacts of UV-B on human health, this beneficial effect of UV-B has not been studied in sufficient detail to allow for a credible health benefits assessment.

- Ozone is a potent GHG. Ozone traps incoming solar radiation at both ends of the spectrum, as well as shortwave radiation that is scattered from high-albedo portions of the Earth's surface. Outgoing terrestrial IR is absorbed by O_3 within the range where water vapor does not absorb, so that natural variability in humidity does not alter its radiative impact. These effects directly force climate. By participating in the oxidative chemistry of the atmosphere, O_3 can indirectly and negatively force climate by the removal of other greenhouse gases.

- Multiple factors influence the forcing effect of tropospheric O_3. Estimates of present-day forcing by O_3 depend upon the available information on pre-industrial and current concentrations. Both are limited and, therefore, very uncertain. Other factors, including the albedo of underlying surface, altitude and co-occurrence of PM can also complicate the calculation of globally-averaged forcing.

- Globally-averaged direct forcing by O_3. On the basis of the best available information, a 2001 Intergovernmental Panel on Climate Change (IPCC) report offered an estimated value of 0.35 ± 0.15 Wm^{-2} for the annual, globally-averaged direct forcing by tropospheric O_3. Another recent estimate places this value at 0.5 ± 0.2 Wm^{-2}.

- Projections of forcing by O_3 into the future. A CTM-climate modeling intercomparison study carried out as part of the third assessment by the IPCC yielded an estimated 0.4 to 0.78 Wm^{-2} forcing by O_3 by the year 2100. The authors of this study concluded that O_3 can be expected to be an important contributor to climate forcing into the future.

- Climate forcing by O_3 at the regional scale may be its most import impact on climate. Satellites have detected high O_3 concentrations localized at the regional scale that are associated with large urban centers and extensive biomass burning. Climate forcing by these high, regional-scale O_3 concentrations have been estimated to be on the order of 1 Wm^{-2} (a substantial fraction of the direct, globally-averaged forcing due to well-mixed GHGs, including CO_2). The impact of climate forcing at this level depends upon the particular characteristics of the region in which it occurs. At present, regional-scale modeling studies

are not available that provide estimates of these effects. Research efforts to do so are underway.

E.11 MATERIALS DAMAGE

The Chapter 11 discussion of O_3 effects on man-made materials mainly summarizes key information from the 1996 O_3 AQCD, given that little new pertinent research information on O_3-related materials damage has been published since then. Key points include:

- Ozone and other photochemical oxidants react with many economically important man-made materials, decreasing their useful life and aesthetic appearance. Materials damaged by O_3 include elastomers; textiles and fibers; dyes, pigments, and inks; and paints and other surface coatings.

- Elastomeric compounds (natural rubber and synthetic polymers and copolymers of butadiene, isoprene, and styrene) are highly susceptible, even to low O_3 concentrations. Ozone damages these compounds by breaking the molecular chain at the carbon-carbon double bond and by adding a chain of three oxygen atoms directly across the double bond. This structure change promotes characteristic cracking of stressed/stretched rubber called "weathering." Tensile strain produces cracks on the surface of the rubber that increase in size and number with increased stress/stretching. The rate of crack growth is dependent on degree of stress, type of rubber compound, O_3 concentration, duration of exposure, O_3 velocity, and temperature. After initial cracking, further O_3 penetration results in additional cracking and, eventually, mechanical weakening.

- Ozone can damage textiles and fabrics by mechanisms similar to those associated with elastomers. Generally, synthetic fibers are less affected by O_3 than natural fibers. Overall, O_3 contribution to degradation of textiles and fabrics is not considered significant.

- Ozone fading of textile dyes is a diffusion-controlled process, with the rate of fading being controlled by diffusion of the dye to the fiber surface. Many textile dyes react with O_3. The rate and severity of the O_3 attack is influenced by the chemical nature of the textile fiber and the manner in which the dye has been applied.

- Paints applied to exterior surfaces of buildings and other structures (e.g., bridges), as well as several artists' pigments, are also sensitive to fading and oxidation by O_3 at concentrations found in urban areas.

1. INTRODUCTION

This is an update revision of the document, "*Air Quality Criteria for Ozone and Related Photochemical Oxidants*," published by the U.S. Environmental Protection Agency (EPA) in 1996 (U.S. Environmental Protection Agency, 1996). That 1996 Ozone Air Quality Criteria Document (O_3 AQCD) provided scientific bases for Congressionally-mandated periodic review by the EPA of the National Ambient Air Quality Standards for Ozone (O_3 NAAQS), which culminated in promulgation of new O_3 NAAQS by EPA in 1997.

The present document critically assesses the latest scientific information relative to determining the health and welfare effects associated with the presence of various concentrations of O_3 and related oxidants in ambient air. It builds upon the previous 1996 EPA O_3 AQCD, by focusing on evaluation and integration of information relevant to O_3 NAAQS criteria development that has become available since that covered by the 1996 criteria review; and it will provide scientific bases for the current periodic review of the O_3 NAAQS.

This introductory chapter of the revised O_3 AQCD presents: (a) background information on legislative requirements, the criteria and NAAQS review process, and the history of O_3 NAAQS reviews (including a chronology of changes in key elements of the O_3 standards); (b) an overview of the current O_3 criteria review process and projected schedule (including approaches and procedures used to prepare this document, as well as projected key milestones); and (c) an orientation to the general organizational structure and content of the document.

1.1 LEGAL AND HISTORICAL BACKGROUND

1.1.1 Legislative Requirements

Two sections of the Clean Air Act (CAA) govern the establishment, review, and revision of National Ambient Air Quality Standards (NAAQS). Section 108 (42 U.S.C. 7408) directs the Administrator of the U.S. Environmental Protection Agency (EPA) to identify ambient air pollutants that may be reasonably anticipated to endanger public health or welfare and to issue air quality criteria for them. These air quality criteria are to reflect the latest scientific

information useful in indicating the kind and extent of all identifiable effects on public health or welfare that may be expected from the presence of a given pollutant in ambient air.

Section 109(a) of the CAA (42 U.S.C. 7409) directs the Administrator of EPA to propose and promulgate primary and secondary NAAQS for pollutants identified under Section 108. Section 109(b)(1) defines a primary standard as one that, in the judgment of the Administrator, is requisite to protect the public health (see inset below) based on the criteria and allowing for an adequate margin of safety. The secondary standard, as defined in Section 109(b)(2), must specify a level of air quality that, in the judgment of the Administrator, is requisite to protect the public welfare (see inset below) from any known or anticipated adverse effects associated with the presence of the pollutant in ambient air, based on the criteria.

PUBLIC HEALTH EFFECTS

- Effects on the health of the general population, or identifiable groups within the population, who are exposed to pollutants in ambient air
- Effects on mortality
- Effects on morbidity
- Effects on other health conditions including indicators of:
  - pre-morbid processes,
  - risk factors, and
  - disease

PUBLIC WELFARE EFFECTS

- Effects on personal comfort and well-being
- Effects on economic values
- Deterioration of property
- Hazards to transportation
- Effects on the environment, including:
  - animals
  - climate
  - crops
  - materials
  - soils
  - vegetation
  - visibility
  - water
  - weather
  - wildlife

Section 109(d) of the CAA (42 U.S.C. 7409) requires periodic review and, if appropriate, revision of existing criteria and standards. If, in the Administrator's judgment, the Agency's review and revision of criteria make appropriate the proposal of new or revised standards, such standards are to be revised and promulgated in accordance with Section 109(b). Alternatively, the Administrator may find that revision of the standards is inappropriate and conclude the review by leaving the existing standards unchanged. Section 109(d)(2) of the 1977 CAA Amendments also requires that an independent scientific review committee be established to advise the EPA Administrator on NAAQS matters, including the scientific soundness of criteria (scientific bases) supporting NAAQS decisions. This role is fulfilled by the Clean Air Scientific Advisory Committee (CASAC) of EPA's Science Advisory Board (SAB).

1.1.2 Criteria and NAAQS Review Process

Periodic reviews by EPA of criteria and NAAQS for a given criteria air pollutant progress through a number of steps, beginning with preparation by EPA's National Center for Environmental Assessment Division in Research Triangle Park, NC (NCEA-RTP) of an air quality criteria document (AQCD). The AQCD provides a critical assessment of the latest available scientific information upon which the NAAQS are to be based. Drawing upon the AQCD, staff of EPA's Office of Air Quality Planning and Standards (OAQPS) prepare a Staff Paper that evaluates policy implications of the key studies and scientific information contained in the AQCD and presents EPA staff conclusions and recommendations for standard-setting options for the EPA Administrator to consider. The Staff Paper is intended to help "bridge the gap" between the scientific assessment contained in the AQCD and the judgments required of the Administrator in determining whether it is appropriate to retain or to revise the NAAQS. Iterative drafts of both the AQCD and the Staff Paper (as well as other analyses, such as exposure and/or risk assessments, supporting the Staff Paper) are made available for public comment and CASAC review. The final versions of the AQCD and Staff Paper incorporate changes made in response to CASAC and public review. Based on the information in these documents, the Administrator proposes decisions on whether to retain or revise the NAAQS, taking into account public comments and CASAC advice and recommendations. The Administrator's proposed decisions are published in the *Federal Register*, with a preamble that presents the rationale for the decisions and solicits public comment. The Administrator makes a final decision after considering comments received on the proposed decisions. The Administrator's final decisions are promulgated in a *Federal Register* notice that addresses significant comments received on the proposal.

NAAQS decisions involve consideration of the four basic elements of a standard: *indicator*, *averaging time*, *form*, and *level*. The indicator defines the pollutant to be measured in the ambient air for the purpose of determining compliance with the standard. The averaging time defines the time period over which air quality measurements are to be obtained and averaged, considering evidence of effects associated with various time periods of exposure. The form of a standard defines the air quality statistic that is to be compared to the level of the standard (i.e., an ambient concentration of the indicator pollutant) in determining whether an area attains the standard. The form of the standard specifies the air quality measurements that

are to be used for compliance purposes (e.g., the 98th percentile of an annual distribution of daily concentrations; the annual arithmetic average), the monitors from which the measurements are to be obtained (e.g., one or more population-oriented monitors in an area), and whether the statistic is to be averaged across multiple years. These basic elements of a standard are the primary focus of the staff conclusions and recommendations in the Staff Paper and in the subsequent rulemaking, building upon the policy-relevant scientific information assessed in the AQCD and on the policy analyses contained in the Staff Paper. These four elements taken together determine the degree of public health and welfare protection afforded by the NAAQS.

1.1.3 Regulatory Chronology[1]

On April 30, 1971, the EPA promulgated primary and secondary NAAQS for photochemical oxidants under Section 109 of the CAA (36 FR 8186). These were set at an hourly average of 0.08 ppm total photochemical oxidants, not to be exceeded more than 1 h per year. On April 20, 1977, the EPA announced (42 FR 20493) the first review and updating of the 1970 Air Quality Criteria Document for Photochemical Oxidants in accordance with Section 109(d) of the CAA. In preparing that AQCD, the EPA made two external review drafts of the document available for public comment, and these drafts were peer reviewed by the Subcommittee on Scientific Criteria for Photochemical Oxidants of EPA's Science Advisory Board (SAB). A final revised AQCD for ozone (O_3) and other photochemical oxidants was published on June 22, 1978.

Based on the 1978 revised AQCD and taking into account the advice and recommendations of the SAB Subcommittee and public comments, the EPA announced (44 FR 8202) a final decision to revise the NAAQS for photochemical oxidants on February 8, 1979. That final rulemaking revised the primary standard from 0.08 ppm to 0.12 ppm, set the secondary standard to be the same as the primary standard, changed the chemical designation of the standards from photochemical oxidants to O_3, and revised the definition of the point at which the standard is attained as indicated in Table 1-1.

[1]This following text is excerpted and adapted from the "Proposed Decision on the National Ambient Air Quality Standards for Ozone," 57 FR 35542, 35542-35557 (August, 10, 1992) and the "National Ambient Air Quality Standards for Ozone; Final Rule," 62 FR 38856, 83356-38896 (July 18, 1997).

Table 1-1. National Ambient Air Quality Standards (NAAQS) for Ozone

Date of Promulgation	Primary and Secondary NAAQS	Averaging Time
February 8, 1979	0.12 ppm[a] (235 $\mu g/m^3$)	1 h[b]
July 18, 1997	0.08 ppm[a] (157 $\mu g/m^3$)	8 h[c]

[a]1 ppm = 1962 $\mu g/m^3$, 1 $\mu g/m^3 = 5.097 \times 10^{-4}$ ppm @ 25 °C, 760 mm Hg.

[b]The standard is attained when the expected number of days per calendar year with a maximum hourly average concentration above 235 $\mu g/m^3$ (0.12 ppm) is equal to or less than one.

[c]Based on the 3-year average of the annual fourth-highest daily maximum 8-h average concentration measured at each monitor within an area.

Source: Federal Register (1979, 1997).

On March 17, 1982, in response to requirements of Section 109(d) of the CAA, the EPA announced (47 FR 11561) that it planned to revise the existing 1978 AQCD for O_3 and Other Photochemical Oxidants; and, on August 22, 1983, it announced (48 FR 38009) that review of the primary and secondary NAAQS for O_3 had been initiated. The EPA provided a number of opportunities for expert review and public comment on revised chapters of the AQCD, including two public peer-review workshops in December 1982 and November 1983. Comments made at both workshops were considered by EPA in preparing the First External Review Draft that was made available (49 FR 29845) on July 24, 1984, for public review. On February 13, 1985 (50 FR 6049) and then on April 2, 1986 (51 FR 11339), the EPA announced two public CASAC meetings, which were held on March 4-6, 1985 and April 21-22, 1986, respectively. At these meetings, the CASAC reviewed external review drafts of the revised AQCD for O_3 and Other Photochemical Oxidants. After these two reviews, the Chair summarized CASAC's consensus view in an October 1986 letter to the EPA Administrator, which stated that the document "represents a scientifically balanced and defensible summary of the extensive scientific literature." Taking into account public and CASAC comments on the two external review drafts, revisions were made by EPA and the final document was released by EPA in August 1986.

The first draft of the Staff Paper "Review of the National Ambient Air Quality Standards for Ozone: Assessment of Scientific and Technical Information" drew upon key findings and conclusions from the AQCD and was reviewed by CASAC at the April 21-22, 1986 public meeting. At that meeting, the CASAC recommended that new information on prolonged O_3

exposure effects be considered in a second draft of the Staff Paper. The CASAC reviewed the resulting second draft and also heard a presentation of new and emerging information on the health and welfare effects of O_3 at a December 14-15, 1987 public review meeting. The CASAC concluded that sufficient new information existed to recommend incorporation of relevant new data into a supplement to the 1986 AQCD (O_3 Supplement) and in a third draft of the Staff Paper.

A draft O_3 Supplement, "Summary of Selected New Information on Effects of Ozone on Health and Vegetation: Draft Supplement to Air Quality Criteria for Ozone and Other Photochemical Oxidants," and the revised Staff Paper were made available to CASAC and to the public in November 1988. The O_3 Supplement assessed selected literature concerning exposure- and concentration-response relationships observed for health effects in humans and experimental animals and for vegetation effects that appeared in papers published or in-press from 1986 through early 1989. On December 14-15, 1988, CASAC held a public meeting to review these documents and then sent the EPA Administrator a letter (dated May 1, 1989), which stated that the draft O_3 Supplement, the 1986 AQCD, and the draft Staff Paper "provide an adequate scientific basis for the EPA to retain or revise the primary and secondary standards of ozone." The CASAC concluded (a) that it would be some time before sufficient new information on the health effects of multihour and chronic exposure to O_3 would be published in scientific journals to receive full peer review and, thus, be suitable for inclusion in a criteria document and (b) that such information could be considered in the next review of the O_3 NAAQS. A final version of the O_3 Supplement was published in 1992 (U.S. Environmental Protection Agency, 1992).

On October 22, 1991, the American Lung Association and other plaintiffs filed suit to compel the Agency to complete the review of the criteria and standards for O_3 in accordance with the CAA. The U.S. District Court for the Eastern District of New York subsequently issued an order requiring the EPA to announce its proposed decision on whether to revise the standards for O_3 by August 1, 1992 and to announce its final decision by March 1, 1993.

The proposed decision on O_3, which appeared in the Federal Register on August 10, 1992 (57 FR 35542), indicated that revision of the existing 1-h NAAQS was not appropriate at that time. A public hearing on this decision was held in Washington, DC on September 1, 1992; and public comments were received through October 9, 1992. The final decision not to revise the 1-h NAAQS was published in the Federal Register on March 9, 1993 (58 FR 13008). However,

that decision did not take into consideration a number of more recent studies on the health and welfare effects of O_3 that had been published since the last of the literature assessed in the O_3 Supplement (i.e., studies available through 1985 and into early 1986).

The Agency initiated consideration of such studies as part of the next congressionally-mandated periodic review of criteria and NAAQS for Ozone. The new studies were assessed in revised draft O_3 AQCD chapters that were peer reviewed in July and September 1993 workshops, followed by public release of the First External Review Draft in February 1994 and CASAC review on July 20-21, 1994. Further drafts of the O_3 AQCD, revised in response to public comments and CASAC review, were reviewed by CASAC on March 21-25, 1995, and at a final CASAC review meeting on September 19-20, 1995. The scientific soundness of the revised O_3 AQCD was recognized by CASAC in a November 28, 1995 letter to the EPA Administrator; and the final AQCD for O_3 was published in July 1996.

The first draft of the associated Staff Paper, "Review of the National Ambient Air Quality Standards for Ozone: Assessment of Scientific and Technical Information," was also reviewed by CASAC at the March 21-22, 1995 public meeting. CASAC also reviewed subsequent drafts of the Staff Paper at public meetings on September 19-20, 1995 and March 21, 1996, with completion of CASAC review of the primary and secondary standard portions of the draft Staff Paper being communicated in letters to the EPA Administrator dated November 30, 1995 and April 4, 1996, respectively. The final O_3 Staff Paper was published in June 1996.

On December 13, 1996 EPA published its proposed decision to revise the O_3 NAAQS (61 FR 65716). EPA provided extensive opportunities for public comment on the proposed decision, including several public hearings and two national satellite telecasts. EPA's final decision to promulgate a new 8-h O_3 NAAQS (see Table 1-1) was published on July 18, 1997 (62 FR 38856).

Following promulgation of the new standards, numerous petitions for review of the standards were filed in the U.S. Court of Appeals for the District of Columbia Circuit (D.C. Circuit)[2]. On May 14, 1999, the Court remanded the O_3 NAAQS to EPA, finding that section 109 of the CAA, as interpreted by EPA, effected an unconstitutional delegation of legislative

[2] *American Trucking Associations* v. EPA, No. 97-1441

authority[3]. In addition, the Court directed that, in responding to the remand, EPA should consider the potential beneficial health effects of O_3 pollution in shielding the public from the effects of solar ultraviolet (UV) radiation. On January 27, 2000, EPA petitioned the U.S. Supreme Court for certiorari on the constitutional issue (and two other issues), but did not request review of the D.C. Circuit ruling regarding the potential beneficial health effects of O_3. On February 27, 2001 the U.S. Supreme Court unanimously reversed the judgment of the D.C. Circuit on the constitutional issue, holding that section 109 of the CAA does not delegate legislative power to the EPA in contravention of the Constitution, and remanded the case to the D.C. Circuit to consider challenges to the O_3 NAAQS that had not been addressed by that Court's earlier decisions[4]. On March 26, 2002, the D.C. Circuit issued its final decision, finding the 1997 O_3 NAAQS to be "neither arbitrary nor capricious," and denied the remaining petitions for review[5].

On November 14, 2001 EPA proposed to respond to the Court's remand to consider the potential beneficial health effects of O_3 pollution in shielding the public from the effects of solar UV radiation by leaving the 1997 8-h NAAQS unchanged. Following a review of information in the record and the substantive comments received on the proposed response, EPA issued a final response to the remand, reaffirming the 8 h O3 NAAQS (68 FR 614, January 6, 2003).

1.2 CURRENT OZONE CRITERIA AND NAAQS REVIEW

1.2.1 Key Milestones and Procedures for Document Preparation

It is important to note at the outset that development of the present document has and will continue to include substantial external expert review and opportunities for public input through (a) public workshops involving the general scientific community, (b) iterative reviews of successive drafts by CASAC, and (c) comments from the public on successive drafts. Extensive external inputs received through such reviews will help to ensure that the review of the O_3

[3] *American Trucking Associations* v. EPA, 175 F.3d 1027 (D.C. Cir., 1999)

[4] *Whitman v. American Trucking Associations*, 531 U.S. 457 (2001)

[5] *American Trucking Associations v. EPA*, 283 F.3d 355, (D.C. Cir. 2002)

standards will be based on critical assessment in this document of the latest available pertinent science.

The procedures for developing this revised O_3 AQCD build on experience derived from the other recent criteria document preparation efforts, with key milestones for development of this O_3 AQCD being listed in Table 1-2. Briefly, the respective responsibilities for production of the document and key milestones are as follows. An NCEA-RTP Ozone Team is responsible for the creation and implementation of a project plan for developing the O_3 AQCD, taking into account input from individuals in other EPA program and policy offices identified as part of the EPA Ozone Work Group. The resulting plan, i.e., the Project Work Plan for Revised Air Criteria for Ozone and Related Photochemical Oxidants (November 2002), was discussed with CASAC in January 2003. An ongoing literature search that was underway prior to initiation of work on this document has continued throughout its preparation to identify pertinent O_3 literature published since early 1996. Under the processes established in Sections 108 and 109 of the CAA, the EPA officially initiated the current criteria and NAAQS review by announcing the commencement of the review in the Federal Register (65 FR 57810, September, 2000) with a call for information. That Federal Register notice included (1) a request asking for recently available research information on O_3 that may not yet have been published and (2) a request for individuals with the appropriate type and level of expertise to contribute to the writing of O_3 AQCD materials to identify themselves. The specific authors of chapters or sections of the proposed document included both EPA and non-EPA scientific experts, who were selected on the basis of their expertise on the subject areas and their familiarity with the relevant literature. The project team defined critical issues and topics to be addressed by the authors and provided direction in order to focus on evaluation of those studies most clearly identified as important for standard setting.

As with other NAAQS reviews, critical assessment of relevant scientific information is presented in this updated O_3 AQCD. The main focus of this document is the evaluation and interpretation of pertinent atmospheric science information, air quality data, human exposure information, and health and welfare effects information newly published since that assessed in the 1996 O_3 AQCD. Draft versions of AQCD chapter materials were evaluated via expert peer-consultation workshop discussions (see Table 1-2) that focused on the selection of pertinent studies to be included in the chapters, the potential need for additional information to be added to

Table 1-2. Key Milestones for Development of Revised Ozone Air Quality Criteria Document [a]

Major Milestones	Target Dates
1. Literature Search	Ongoing
2. Federal Register Call for Information	September 2000
3. Draft Project Plan Available for Public Comment	Dec 2001 - March 2002
4. Revised Draft Project Plan Released for CASAC Review	December 2002
5. CASAC Review of Draft Project Work Plan	January 2003
6. Peer-Consultation Workshop on Draft Ecological Effects Materials	April 2003
7. Peer-Consultation Workshops on Draft Atmospheric Science/Exposure and Dosimetry/Health Chapters	July 2004
8. First External Review Draft of O_3 AQCD	January 2005
9. Public Comment Period (90 days)	Feb - April 2005
10. CASAC Public Review Meeting *(First External Review Draft)*	May 2005
11. Second External Review Draft of O_3 AQCD	August 2005
12. Public Comment Period (90 days)	Sept - Nov 2005
13. CASAC Public Review Meeting	December 2005
14. Final O_3 AQCD	February 2006

[a] Proposed schedule will be modified from time to time, as necessary, to reflect actual project requirements and progress.

the chapters, and the quality of the characterization and interpretation of the literature. The authors of the draft chapters then revised them on the basis of the workshop and/or other expert review comments[6]. These and other integrative materials were then incorporated into the First External Review Draft of this O_3 AQCD (January 2005), which was made available for public comment and CASAC review (see Table 1-2).

Following review of the First External Review Draft at a May 4-5, 2005 CASAC meeting, EPA incorporated revisions into the draft O_3 AQCD in response to comments from CASAC and the public and has made this Second External Review Draft (August, 2005) available for further

[6]It should be noted that materials contributed by non-EPA authors have, at times, been modified by EPA Ozone Team staff in response to internal and/or external review comments and that EPA is responsible for the ultimate content of this O_3 AQCD.

public comment and CASAC review according to the schedule projected in Table 1-2. More specifically, this Second External Review Draft is available for public comment (90 days) during September-November, 2005, and will be reviewed by CASAC at a public meeting in December 2005 (the site and specific dates to be announced in the Federal Register). The final O_3 AQCD is to be completed by February 28, 2006, and it is to be made publicly available electronically via an EPA website and then subsequently printed. Its availability will be announced in the Federal Register.

The EPA's Office of Air Quality Planning and Standards (OAQPS) staff has also prepared a first draft O_3 Staff Paper drawing upon key information contained in this Second External Review Draft O_3 AQCD, which presents recommendations regarding whether to retain or, if appropriate, to revise the O_3 NAAQS. After review of that draft Staff Paper (dated September, 2005) by the public and by CASAC, EPA will take public and CASAC comments into account in producing a Second Draft Staff Paper. That Second Draft Staff Paper (based on the final version of this O_3 AQCD) will also be made available for further public comment and CASAC review before EPA produces a final ozone Staff Paper by September 30, 2006.

1.3 ORGANIZATIONAL STRUCTURE OF THE DOCUMENT

1.3.1 General Document Format

The general format used in preparing this draft document is to open each new section for the updated document with concise summarization of key findings and conclusions from the previous 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). After presentation of such background information, the remainder of each section typically provides an updated discussion of newer literature and resulting key conclusions. In some cases where no new information is available, the summary of key findings and conclusions from the previous criteria document must suffice as the basis for current key conclusions. Increased emphasis is placed in the main chapters of this revised O_3 AQCD on interpretative evaluation and integration of evidence pertaining to a given topic than has been typical of previous EPA air quality criteria documents, with more detailed descriptions of individual studies being provided in a series of accompanying annexes.

A list of references published since completion of the 1996 criteria document was made available to the authors. The references were selected from information data base searches conducted by EPA. Additional references have been added to the list (e.g., missed or recently published papers or "in press" publications) as work has proceeded in creating the draft document materials. As an aid in selecting pertinent new literature, the authors were also provided with a summary of issues that need to be addressed in the revised air quality criteria document for O_3. These issues were identified by authors and reviewers of the previous documents and continue to be expanded, as appropriate, based on public discussions, workshops, or other comments received by EPA.

1.3.2 Organization and Content of the Document

This revised AQCD for O_3 and Related Photochemical Oxidants critically assesses scientific information on the health and welfare effects associated with exposure to the concentrations of these pollutants in ambient air. The document does not provide a detailed literature review; but, rather, discusses cited references that reflect the current state of knowledge on the most relevant issues pertinent to the NAAQS for O_3. Although emphasis is placed on discussion of health and welfare effects information, other scientific data are presented and evaluated in order to provide a better understanding of the nature, sources, distribution, measurement, and concentrations of O_3 and related photochemical oxidants in ambient air, as well as the measurement of population exposure to these pollutants.

The main focus of the scientific information discussed in the text comes from literature published since completion of the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). Emphasis is placed on studies conducted at or near O_3 concentrations found in ambient air. Other studies are included if they contain unique data, such as the documentation of a previously unreported effect or of a mechanism for an observed effect; or if they were multiple-concentration studies designed to provide exposure-response relationships. Generally, this is not an issue for human clinical or epidemiology studies. However, for animal toxicology studies, consideration is given mainly to those studies conducted at less than 1 ppm O_3. Key information from studies assessed in the previous O_3 AQCD and whose data impacted the derivation of the current NAAQS are briefly summarized in the text, along with specific citations to the previous document. Prior studies are also discussed if they (1) are open to reinterpretation in light of

newer data, or (2) are potentially useful in deriving revised standards for O_3. Generally, only information that has undergone scientific peer review and has been published (or accepted for publication) through December 2004 is included in this draft document. A few particularly pertinent and important new studies published or accepted for publication beyond the end of 2004 are also considered.

This document consists of three volumes. The first volume includes an Executive Summary and Conclusions, as well as Chapters 1 through 11 of the O_3 AQCD. This introductory chapter (Chapter 1) presents background information on the purpose of the document, legislative requirements, and the history of past O_3 NAAQS regulatory actions, as well as an overview of the organization and content of the document. Chapter 2 provides information on the physics and chemistry of O_3 and related photochemical oxidants in the atmosphere. Chapter 3 covers tropospheric O_3 environmental concentrations, patterns, and exposure estimates. The accompanying annexes to each of these background chapters are found in Volume II.

Health information pertinent to derivation of the primary O_3 NAAQS is then mainly covered in the next several chapters (Chapters 4 through 8). Chapter 4 discusses O_3 dosimetry aspects, and Chapters 5, 6, and 7 discuss animal toxicological studies, human health effects from controlled-exposure studies, and epidemiologic studies of ambient air exposure effects on human populations, respectively. Chapter 8 then provides an integrative and interpretive evaluation of key information relevant to O_3 exposure and health risks, of most pertinence to the review of primary O_3 NAAQS. The annexes to these health-related chapters are found in Volume II.

The remaining three chapters of the document assess welfare effects information pertinent to the review of secondary O_3 NAAQS. Chapter 9 deals with ecological and other environmental effects of O_3 and related photochemical oxidants. Chapter 10 assesses tropospheric O_3 involvement in climate change processes, including determination of solar UV flux in Earth's lower atmosphere. Lastly, Chapter 11 discusses O_3 effects on man-made materials as a third type of welfare effect of potential concern. Annex materials related to welfare effects (especially vegetation/ecological effects) are contained in Volume III.

REFERENCES

Federal Register. (1971) National primary and secondary ambient air quality standards. F. R. (April 30) 36: 8186-8201.

Federal Register. (1977) Review of the photochemical oxidant and hydrocarbon air quality standards. F. R. (April 20) 42: 20493-20494.

Federal Register. (1979) National primary and secondary ambient air quality standards: revisions to the national ambient air quality standards for photochemical oxidants. F. R. (February 8) 44: 8202-8237.

Federal Register. (1982) Air quality criteria document for ozone and other photochemical oxidants. F. R. (March 17) 47: 11561.

Federal Register. (1983) Review of the national ambient air quality standards for ozone. F. R. (August 22) 48: 38009.

Federal Register. (1984) Draft air quality criteria document for ozone and other photochemical oxidants. F. R. (July 24) 49: 29845.

Federal Register. (1985) Science Advisory Board; Clean Air Scientific Advisory Committee; open meeting. F. R. (February 13) 50: 6049.

Federal Register. (1986) Science Advisory Board; Clean Air Scientific Advisory Committee; open meeting. F. R. (April 2) 51: 11339.

Federal Register. (1992) National ambient air quality standards for ozone; proposed decision. F. R. (August 10) 57: 35542-35557.

Federal Register. (1993) National ambient air quality standards for ozone - final decision. F. R. (March 9) 58: 13008-13019.

Federal Register. (1996) National ambient air quality standards for ozone: proposed decision. F. R. (December 13) 61: 65,716-65,750.

Federal Register. (1997) National ambient air quality standards for ozone; final rule. F. R. (July 18) 62: 38856-38896.

Federal Register. (2000) Air Quality Criteria for Ozone and Related Photochemical Oxidants; notice; call for information. F. R. (September 26) 65: 57810.

Federal Register. (2003) National ambient air quality standards for ozone: final response to remand; final rule. F. R. (January 6) 68: 614-645.

U.S. Code. (2003a) Clean Air Act, §108, air quality criteria and control techniques.. U. S. C. 42: §7408.

U.S. Code. (2003b) Clean Air Act, §109, national ambient air quality standards. U. S. C. 42: §7409.

U.S. Court of Appeals for the District of Columbia. (1999a) American Trucking Associations, Inc. v. U.S. Environmental Protection Agency. 195 F.3d 4 (D.C. Cir. 1999).

U.S. Court of Appeals for the District of Columbia. (1999b) American Trucking Associations, Inc. v. U.S. Environmental Protection Agency. 175 F.3d 1027 (D.C. Cir. 1999).

U.S. Court of Appeals for the District of Columbia. (2002) American Trucking Associations, Inc. v. U.S. Environmental Protection Agency. 283 F.3d 355, 378-79 (D.C. Cir. 2002).

U.S. Environmental Protection Agency. (1992) Summary of selected new information on effects of ozone on health and vegetation: supplement to 1986 air quality criteria for ozone and other photochemical oxidants. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report no. EPA/600/8-88/105F. Available from: NTIS, Springfield, VA; PB92-235670.

U.S. Environmental Protection Agency. (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: http://cfpub2.epa.gov/ncea/.

U.S. Supreme Court. (2001) Whitman v. American Trucking Association. 531 U.S. 457 (nos. 99-1257 and 99-1426).

2. PHYSICS AND CHEMISTRY OF OZONE IN THE ATMOSPHERE

2.1 INTRODUCTION

Ozone (O_3) and other oxidants, such as peroxacyl nitrates and hydrogen peroxide (H_2O_2) form in polluted areas by atmospheric reactions involving two main classes of precursor pollutants, volatile organic compounds (VOCs) and nitrogen oxides (NO_x). Carbon monoxide (CO) is also important for ozone formation in polluted areas. Ozone is thus a secondary pollutant. The formation of O_3, other oxidants and oxidation products from these precursors is a complex, nonlinear function of many factors: the intensity and spectral distribution of sunlight; atmospheric mixing and processing on cloud and aerosol particles; the concentrations of the precursors in ambient air; and the rates of chemical reactions of the precursors. Information contained in this chapter and in greater detail in Annex AX2 describes these processes, numerical models that incorporate these processes to calculate O_3 concentrations, and techniques for measuring concentrations of ambient oxidants.

The atmosphere can be divided into several distinct vertical layers, based primarily on the major mechanisms by which they are heated and cooled. The lowest major layer is the troposphere, which extends from the earth's surface to about 8 km above polar regions and to about 16 km above tropical regions. The planetary boundary layer (PBL) is the lower sublayer of the troposphere, extending from the surface to about 1 or 2 km and is most strongly affected by surface conditions. The stratosphere extends from the tropopause, or the top of the troposphere, to about 50 km in altitude (Annex AX2.2.1). The emphasis in this chapter is placed on chemical and physical processes occurring in the troposphere, in particular in the PBL. The processes responsible for producing summertime O_3 episodes are fairly well understood, as outlined in the previous Air Quality Criteria Document for Ozone and Related Photochemical Oxidants (CD96). This chapter mainly considers topics for which there is substantial new information and on topics that form the basis for discussions in later chapters.

2.2 CHEMICAL PROCESSES INVOLVED IN OZONE FORMATION AND DESTRUCTION

Ozone occurs not only in polluted urban atmospheres but throughout the troposphere, even in remote areas of the globe. The same basic processes, involving sunlight-driven reactions of NO_x and VOCs contribute to O_3 formation throughout the troposphere. These processes also lead to the formation of other photochemical products, such as peroxyacetyl nitrate (PAN), nitric acid (HNO_3), and sulfuric acid (H_2SO_4), and to other compounds, such as formaldehyde (HCHO) and other carbonyl compounds, such as aldehydes and ketones.

The photochemical formation of O_3 in the troposphere proceeds through the oxidation of nitric oxide (NO) to nitrogen dioxide (NO_2) by organic (RO_2) or hydro-peroxy (HO_2) radicals. The photolysis of NO_2 yields nitric oxide (NO) and a ground-state oxygen atom, $O(^3P)$, which then reacts with molecular oxygen to form O_3. Free radicals oxidizing NO to NO_2 are formed during the oxidation of VOCs (Annex AX2.2.2).

The term VOC refers to all carbon-containing gas-phase compounds in the atmosphere, both biogenic and anthropogenic in origin, excluding carbon monoxide (CO) and carbon dioxide (CO_2). Classes of organic compounds important for the photochemical formation of O_3 include alkanes, alkenes, aromatic hydrocarbons, carbonyl compounds (e.g., aldehydes and ketones), alcohols, organic peroxides, and halogenated organic compounds (e.g., alkyl halides). This array of compounds encompasses a wide range of chemical properties and lifetimes: isoprene has an atmospheric lifetime of approximately an hour, whereas methane has an atmospheric lifetime of about a decade.

In urban areas, compounds representing all classes of VOCs, and CO are important for O_3 formation. In nonurban vegetated areas, biogenic VOCs emitted from vegetation tend to be the most important. In the remote troposphere, CH_4 and CO are the main carbon-containing precursors to O_3 formation. CO also can play an important role in O_3 formation in urban areas. The oxidation of VOCs is initiated mainly by reaction with hydroxyl (OH) radicals. The primary source of OH radicals in the atmosphere is the reaction of electronically excited O atoms, $O(^1D)$, with water vapor. $O(^1D)$ is produced by the photolysis of O_3 in the Hartley bands. In polluted areas, the photolysis of aldehydes (e.g., HCHO), nitrous acid (HONO) and hydrogen peroxide (H_2O_2) can also be significant sources of OH or HO_2 radicals that can rapidly be converted to OH (Eisele et al., 1997). Ozone can oxidize alkenes, and, at night, when they are

most abundant, NO_3 radicals also oxidize alkenes. In coastal environments and other selected environments, atomic Cl and Br radicals can also initiate the oxidation of VOCs (Annex AX2.2.3).

There are a large number of oxidized nitrogen containing compounds in the atmosphere including NO, NO_2, NO_3, HNO_2, HNO_3, N_2O_5, HNO_4, PAN and its homologues, other organic nitrates and particulate nitrate. Collectively these species are referred to as NO_y. Oxidized nitrogen compounds are emitted to the atmosphere mainly as NO which rapidly interconverts with NO_2 and so NO and NO_2 are often "lumped" together into their own group or family, or NO_x. NO_x can be oxidized to reservoir and termination species (PAN and its homologues, organic nitrates, HNO_3, HNO_4 and particulate nitrate). These reservoir and termination species are referred to as NO_z. The major reactions involving inter-conversions of oxidized nitrogen species are discussed in Annex AX2.2.4.

The photochemical cycles by which the oxidation of hydrocarbons leads to O_3 production are best understood by considering the oxidation of methane, structurally the simplest VOC. The CH_4 oxidation cycle serves as a model for the chemistry of the relatively clean or unpolluted troposphere (although this is a simplification because vegetation releases large quantities of complex VOCs, such as isoprene, into the atmosphere). In the polluted atmosphere, the underlying chemical principles are the same, as discussed in Annex AX2.2.5. The conversion of NO to NO_2 occurring with the oxidation of VOCs is accompanied by the production of O_3 and the efficient regeneration of the OH radical, which in turn can react with other VOCs. A schematic overview showing the major processes involved in O_3 production and loss in the troposphere and stratosphere is given in Figure 2-1.

The oxidation of alkanes and alkenes in the atmosphere has been treated in depth in CD96 and is updated in Annexes AX2.2.6 and AX2.2.7. In contrast to simple hydrocarbons containing one or two carbon atoms, detailed kinetic information about the gas phase oxidation pathways of many anthropogenic hydrocarbons (e.g., aromatic compounds, such as benzene and toluene), biogenic hydrocarbons (e.g., isoprene, the monoterpenes), and their intermediate oxidation products (e.g., epoxides, nitrates, and carbonyl compounds) is lacking. Reaction with OH radicals represents the major loss process for alkanes. Reaction with chlorine atoms is an additional sink for alkanes. Stable products of alkane photooxidation are known to include carbonyl compounds, alkyl nitrates, and *d*-hydroxycarbonyls. Major uncertainties in the

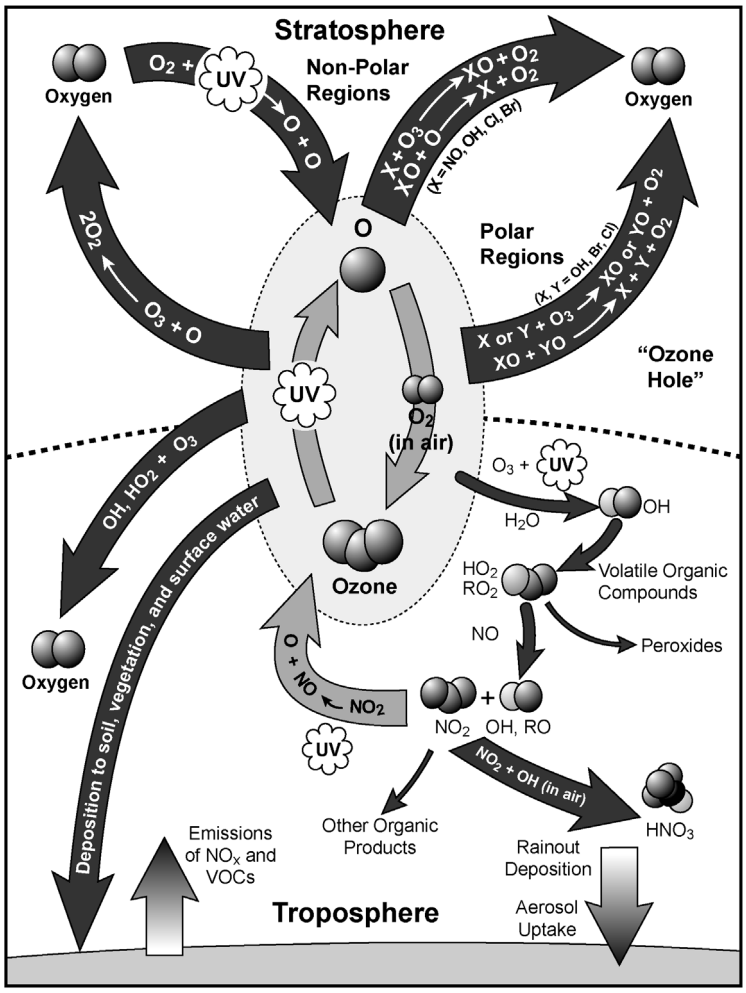


Figure 2-1. Schematic overview of O_3 photochemistry in the stratosphere and troposphere.

atmospheric chemistry of the alkanes concern the chemistry of alkyl nitrate formation; these uncertainties affect the amount of NO-to-NO_2 conversion occurring and, hence, the amounts of O_3 formed during photochemical degradation of the alkanes.

The reaction of OH radicals with aldehydes produced during the oxidation of alkanes forms acyl (R´CO) radicals, and acyl peroxy radicals (R´C(O)–O_2) are formed by the further addition of O_2. As an example, the oxidation of ethane (C_2H_5–H) yields acetaldehyde (CH_3–CHO). The reaction of CH_3–CHO with OH radicals yields acetyl radicals (CH_3–CO). The acetyl radicals will then participate with O_2 in a termolecular recombination reaction to form

acetyl peroxy radicals, which can then react with NO to form $CH_3 + CO_2$ or they can react with NO_2 to form PAN. PAN acts as a temporary reservoir for NO_2. Upon the thermal decomposition of PAN, either locally or elsewhere, NO_2 is released to participate in the O_3 formation process again.

Alkenes react in ambient air with OH, NO_3, and Cl radicals and with O_3. All of these reactions are important atmospheric transformation processes, and all proceed by initial addition to the >C=C< bonds. Products of alkene photooxidation include carbonyl compounds, hydroxynitrates and nitratocarbonyls, and decomposition products from the energy-rich biradicals formed in alkene-O_3 reactions. Major uncertainties in the atmospheric chemistry of the alkenes concern the products and mechanisms of their reactions with O_3, especially the yields of free radicals that participate in O_3 formation. Examples of oxidation mechanisms of complex alkanes and alkenes can be found in comprehensive texts such as Seinfeld and Pandis (1998).

The oxidation of aromatic hydrocarbons constitutes an important component of the chemistry of O_3 formation in urban atmospheres (Annex AX2.2.8). Virtually all of the important aromatic hydrocarbon precursors emitted in urban atmospheres are lost through reaction with the hydroxyl radical. Loss rates for these compounds vary from slow (i.e., benzene) to moderate (e.g., toluene), to very rapid (e.g., xylene and trimethylbenzene isomers). These loss rates are very well understood at room temperature and atmospheric pressure and numerous experiments have been conducted that verify this. However, the mechanism for the oxidation of aromatic hydrocarbons following reaction with OH is poorly understood, as evident from the poor mass balance of the reaction products. The mechanism for the oxidation of toluene has been studied most thoroughly and there is general agreement on the initial steps in the mechanism. However, at present there is no promising approach for resolving the remaining issues concerning the later steps. The oxidation of aromatic hydrocarbons also leads to particle formation which could remove gas-phase constituents that participate in O_3 formation. The chemistry of secondary organic aerosol formation from gaseous precursors was summarized in the latest AQCD for particulate matter.

The reactions of oxygenated VOCs are also important components of O_3 formation (Annex AX2.2.9). They may be produced either by the oxidation of hydrocarbons or they may be present in ambient air as the result of direct emissions. For example, motor vehicles and some industrial processes emit formaldehyde and vegetation emits methanol.

As much as 30% of the carbon in hydrocarbons in many urban areas is in the form of aromatic compounds. Yet, mass balance analyses performed on irradiated smog chamber mixtures of aromatic hydrocarbons indicate that only about one-half of the carbon is in the form of compounds that can be identified. The situation is not much better for some smaller anthropogenic hydrocarbons. For example, only about 60% of the initial carbon can be accounted for in the OH initiated oxidation of 1,3-butadiene. About two-thirds of the initial carbon can be identified in product analyses of isoprene oxidation. Adequate analytical techniques needed to identify and quantify key intermediate species are not available for many compounds. In addition, methods to synthesize many of the suspected intermediate compounds are not available so that laboratory studies of their reaction kinetics cannot be performed. Similar considerations apply to the oxidation of biogenic hydrocarbons besides isoprene.

In addition to reactions occurring in the gas phase, reactions occurring on the surfaces of or within cloud droplets and airborne particles also occur. Their collective surface area is huge implying that collisions with gas phase species occur on very short time scales. In addition to hydrometeors (e.g., cloud and fog droplets and snow and ice crystals) there are also potential reactions involving atmospheric particles of varying composition (e.g., wet [deliquesced] inorganic particles, mineral dust, carbon chain agglomerates and organic carbon particles) to consider. Most of the well-established multiphase reactions tend to reduce the rate of O_3 formation in the polluted troposphere. Removal of HO_x and NO_x onto hydrated particles will reduce the production of O_3. However, the photolysis of HONO formed in reactions such as these can increase the production of O_3. The reactions of Br and Cl containing radicals deplete O_3 in selected environments such as the Arctic during spring, the tropical marine boundary layer and inland salt lakes. Direct reactions of O_3 and atmospheric particles appear to be too slow to reduce O_3 formation significantly at typical ambient PM levels. In addition, the oxidation of hydrocarbons by Cl radicals could lead to the rapid formation of peroxy radicals and higher rates of O_3 production in selected coastal environments. It should be stressed that knowledge of multiphase processes is still evolving and there are still many questions that remain to be answered as outlined in Annex AX2.2.10.

The oxidants, other than O_3, that are formed from the chemistry described above could exert effects on human health and perhaps also on vegetation. Gas phase oxidants include PAN, H_2O_2 and CH_3OOH and other organic hydroperoxides (Annex AX2.2). In addition to

transfer from the gas phase, oxidants can be formed by photochemical reactions occurring in particles (Annex 2.2.10.6). However, the pathways leading to the formation of oxidants in the particle phase are not as well understood as they are in the gas phase.

2.3 METEOROLOGICAL PROCESSES AFFECTING OZONE

Since CD96, substantial new information about transport processes has become available from numerical models, field experiments and satellite-based observations. Ozone is produced naturally by photochemical reactions in the stratosphere as shown in Figure 2-1. Some of this O_3 is transported downward into the troposphere throughout the year, with maximum contributions during late winter and early spring mainly in a process known as tropopause folding. Figure 2-2a shows a synoptic situation associated with a tropopause folding event. A vertical cross section taken through the atmosphere from a to a′ is shown in Figure 2-2b. In this figure the tropopause fold is shown folding downward above and slightly behind the surface cold front, bringing stratospheric air with it. Although the tropopause is drawn with a solid line, it should not be taken to mean that it is a material surface, through which there is no exchange. Rather these folds should be thought of as regions in which mixing of tropospheric and stratospheric air is occurring (Shapiro, 1980). This imported stratospheric air contributes to the natural background of O_3 in the troposphere, especially in the free troposphere. It should be noted that there is considerable uncertainty in the magnitude and distribution of this potentially important source of tropospheric O_3. Stratospheric intrusions that reach the surface are rare. Much more common are intrusions which penetrate only to the middle and upper troposphere. However, O_3 transported to the upper and middle troposphere can still affect surface concentrations through various exchange mechanisms that mix air from the free troposphere with air in the planetary boundary layer. Substantial photochemical production of O_3 in the troposphere also begins in late winter and early spring; therefore, it cannot be assumed that O_3 present at these times is only stratospheric in origin. The basic atmospheric dynamics and thermodynamics of stratospheric-tropospheric exchange are outlined in Annex AX2.3.1.

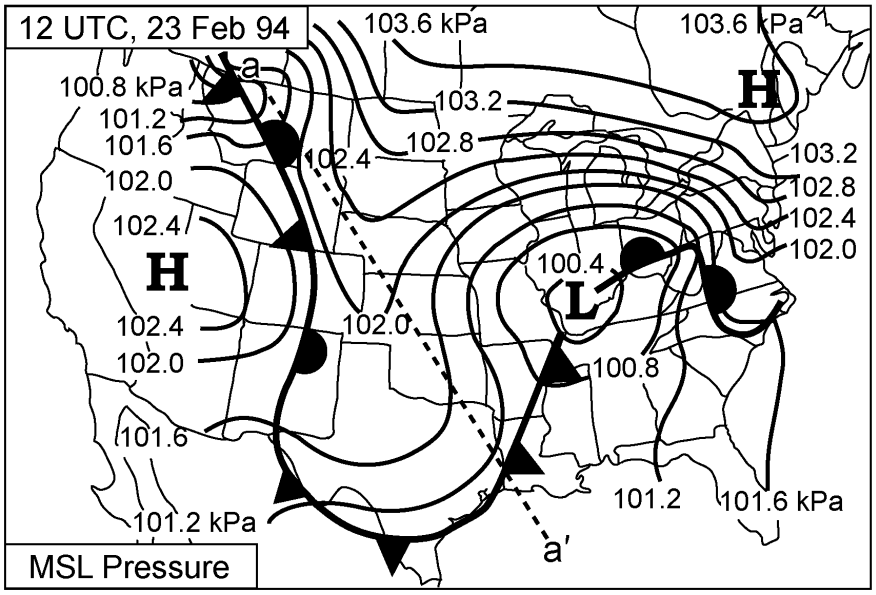


Figure 2-2a. Surface weather chart showing sea level (MSL) pressure (kPa), and surface fronts.

Source: Stull (2000).

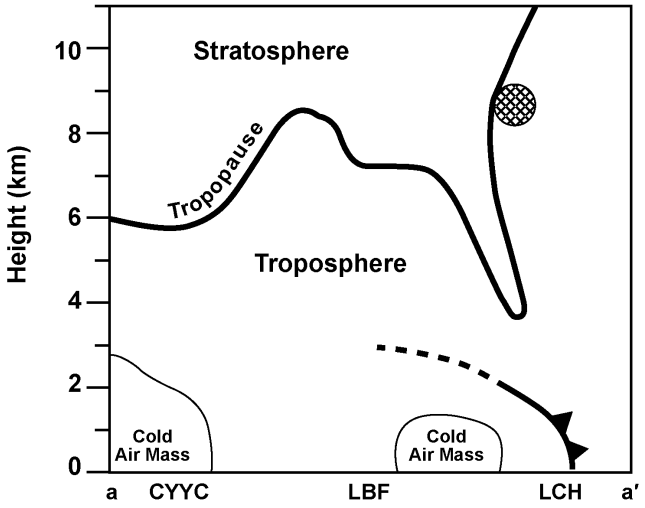


Figure 2-2b. Vertical cross section along dashed line (a-a′) from northwest to the southeast (CYYC = Calgary, Alberta; LBF = North Platte, NB; LCH = Lake Charles, LA). The approximate location of the jet stream core is indicated by the hatched area. The position of the surface front is indicated by the cold-frontal symbols and the frontal inversion top by the dashed line. Note: This is 12 h later than the situations shown in Figure 2-2a.

Source: Adapted from Stull (2000).

Our understanding of the meterological processes associated with summertime O_3 episodes remains basically the same as outlined in CD96. Major episodes of high O_3 concentrations in the eastern United States and in Europe are associated with slow moving, high pressure systems. High pressure systems during the warmer seasons are associated with the sinking of air, resulting in warm, generally cloudless skies, with light winds. The sinking of air results in the development of stable conditions near the surface which inhibit or reduce the vertical mixing of O_3 precursors. The combination of inhibited vertical mixing and light winds minimizes the dispersal of pollutants emitted in urban areas, allowing their concentrations to build up. Photochemical activity involving these precursors is enhanced because of higher temperatures and the availability of sunlight. In the eastern United States, high O_3 concentrations during a large scale episode can extend over hundreds of thousands of square kilometers for several days. These conditions have been described in greater detail in CD96. The transport of pollutants downwind of major urban centers is characterized by the development of urban plumes. However, the presence of mountain barriers limits mixing as in Los Angeles and Mexico City and will result in a higher frequency and duration of days with high O_3 concentrations. Ozone concentrations in southern urban areas, such as Houston, TX and Atlanta, GA tend to decrease with increasing wind speed. In northern cities such as Chicago, IL; New York, NY; Boston, MA; and Portland, ME, the average O_3 concentrations over the metropolitan areas increase with wind speed indicating that transport of O_3 and its precursors from upwind areas is important (Husar and Renard, 1998; Schichtel and Husar, 2001).

Ozone and other secondary pollutants are determined by meteorological and chemical processes extending typically over spatial scales of several hundred kilometers (e.g., Civerolo et al., 2003; Rao et al., 2003). An analysis of the output of regional model studies conducted by Kasibhatla and Chameides (2000) suggests that O_3 can be transported over a few thousand kilometers in the upper boundary layer of the eastern half of the United States during specific O_3 episodes. Convection is capable of transporting O_3 and its precursors vertically through the troposphere as shown in Annex AX2.3.2. Nocturnal low level jets (LLJs) can also transport pollutants hundreds of kilometers (Annex AX2.3.3). Schematic diagrams showing the atmospheric conditions during the formation of low level jets and the regions in which they are most prevalent are given in Figures 2-3 and 2-4. They have also been observed off the coast of

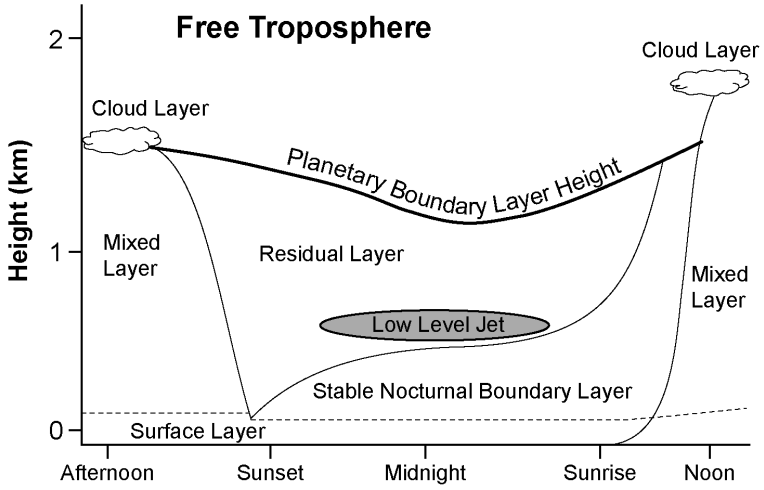


Figure 2-3. The diurnal evolution of the planetary boundary layer while high pressure prevails over land. Three major layers exist (not including the surface layer): a turbulent mixed layer; a less turbulent residual layer which contains former mixed layer air; and a nocturnal, stable boundary layer that is characterized by periods of sporadic turbulence.

Source: Adapted from Stull (1999) Figures 1.7 and 1.12.

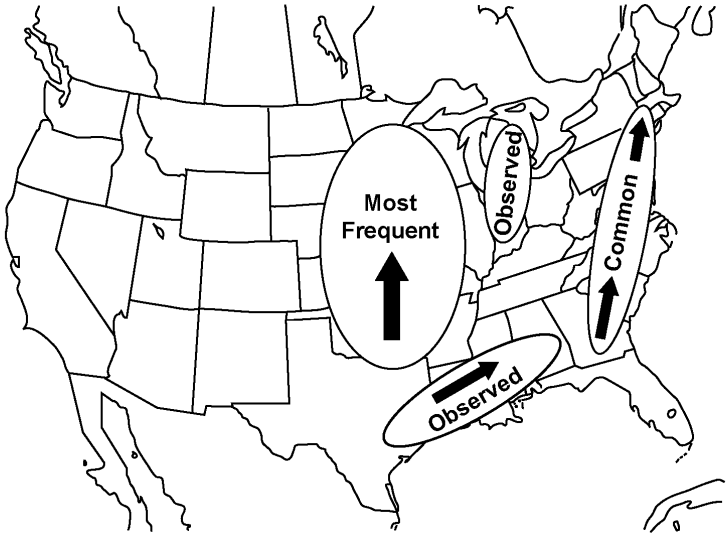


Figure 2-4. Locations of low level jet occurrences in decreasing order of prevalence (most frequent, common, observed). These locations are based on 2-years radiosonde data obtained over limited areas. With better data coverage, other low level jets may well be observed elsewhere in the United States.

Source: Bonner (1968).

California. Turbulence associated with LLJs can bring these pollutants to the surface and result in secondary O_3 maxima in the early morning in many locations (Corsmeier et al., 1997).

Aircraft observations indicate that there can be substantial differences in mixing ratios of key species between the surface and the atmosphere above (Fehsenfeld et al., 1996; Berkowitz and Shaw, 1997). In particular, mixing ratios of O_3 can be higher in the lower free troposphere (aloft) than in the planetary boundary layer during multiday O_3 episodes (Taubmann et al., 2004). These conditions are illustrated schematically in Figure 2-5. Convective processes and small scale turbulence transport O_3 and other pollutants both upward and downward throughout the planetary boundary layer and the free troposphere. Ozone and its precursors can be transported vertically by convection into the upper part of the mixed layer on one day, then transported overnight as a layer of elevated mixing ratios, and then entrained into a growing convective boundary layer downwind and brought back down to the surface. High concentrations of O_3 showing large diurnal variations at the surface in southern New England were associated with the presence of such layers (Berkowitz et al., 1998). Because of wind shear, winds several hundred meters above the ground can bring pollutants from the west, even though surface winds are from the southwest during periods of high O_3 in the eastern United States (Blumenthal et al., 1997). These considerations suggest that in many areas of the United States, O_3 formation involves processes occurring over hundreds if not thousands of square kilometers.

Although the vast majority of measurements are made near the Earth's surface, there is substantial photochemistry and transport of O_3 occurring above the boundary layer in the free troposphere. In the free troposphere, pollutants are chemically more stable and can be transported over much longer distances and O_3 is produced more efficiently than in the planetary boundary layer. Results from the Atmosphere/Ocean Chemistry Experiment (AEROCE) indicated that springtime maxima in surface O_3 over the western North Atlantic Ocean result from tropopause folding in close proximity to convective clouds (Annex AX2.3.4). The convection lifts O_3 and its precursors to the free troposphere where they mix with O_3 from the stratosphere and the mixture is transported eastward. Results from the North Atlantic Regional Experiment (Annex AX2.3.4) indicated that summertime air is transported along the East Coast northeastward and upward ahead of cold fronts. New England and the Maritime Provinces of Canada receive substantial amounts of O_3 and other pollutants through this mechanism.

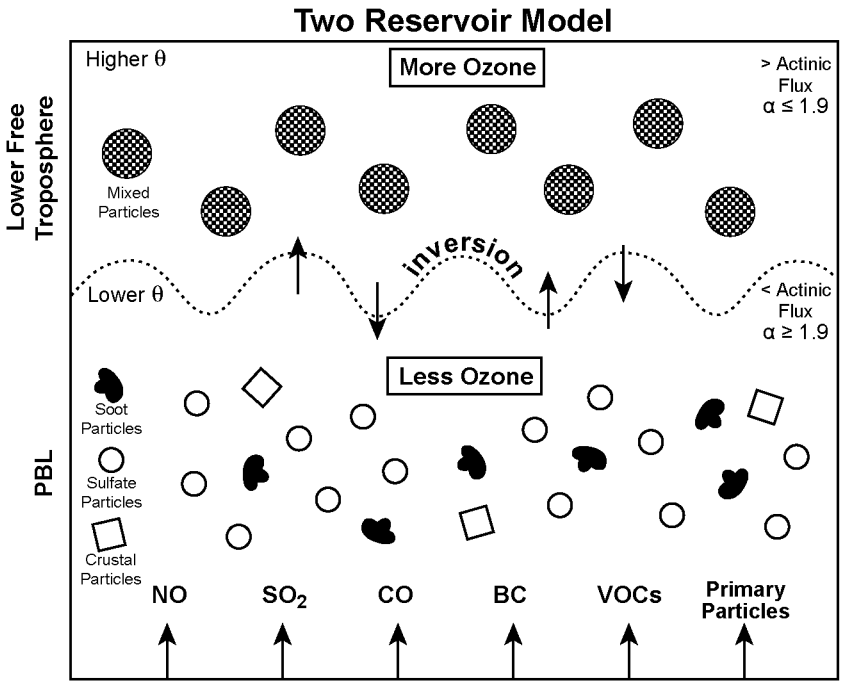


Figure 2-5. Conceptual two-reservoir model showing conditions in the PBL and in the lower free troposphere during a multiday O_3 episode. The dotted line represents the top of PBL. Emissions occur in the PBL, where small, unmixed black carbon, sulfate, and crustal particles in the $PM_{2.5}$ size range are also shown. Ozone concentrations as well as potential temperature (θ) and actinic flux are lower in the PBL than in the lower free troposphere, while relative humidity and the Angstrom exponent for aerosol scattering (α) are higher. Larger, internally mixed sulfate and carbonaceous particles (still in the $PM_{2.5}$ size range) and more O_3 exist in the lower free troposphere.

Source: Taubman et al. (2004).

Pollutants transported in this way can then be entrained in stronger and more stable westerly winds aloft and can travel across the North Atlantic Ocean. The pollutants can then be brought to the surface by subsidence in high pressure systems (typically behind the cold front in advance of the one mentioned above). Thus, pollutants from North America can be brought down either over the North Atlantic Ocean or in Europe. Pollutants can be transported across the North Pacific Ocean from Asia to North America in a similar way. Behind an advancing cold front, cold and dry stratospheric air is also being transported downward and southward. Stratospheric

constituents and tropospheric constituents can then mix by small-scale turbulent exchange processes. The results of these studies suggest that the mechanisms involved in the long-range transport of O_3 and its precursors are closely tied to the processes involved in stratospheric-tropospheric exchange.

2.4 RELATIONS OF OZONE TO ITS PRECURSORS

The local rate of O_3 formation depends on atmospheric conditions such as the availability of solar ultraviolet radiation capable of initiating photolysis reactions, air temperatures and the concentrations of chemical precursors (Annex AX2.3.5). The dependence of daily maximum 8-h O_3 concentrations on daily maximum temperature is illustrated in Figure 2-6 for the Baltimore, MD area. As can be seen, O_3 concentrations tend to increase with temperature ($r = 0.74$). However, this trend is absent in data from Phoenix, AZ as can be seen in Figure 2-7 ($r = 0.14$). These figures show that relations of O_3 to precursor variables are location-specific and relations observed in one area cannot be readily extrapolated to another. Factors that may be responsible for the differences in O_3 behavior in the two areas are discussed in Section AX2.3.5.3.

Rather than varying directly with emissions of its precursors, O_3 changes in a nonlinear fashion with the concentrations of its precursors (Annex AX2.4). At the low NO_x concentrations found in most environments, ranging from remote continental areas to rural and suburban areas downwind of urban centers (low - NO_x regime), the net production of O_3 increases with increasing NO_x. At the high NO_x concentrations found in downtown metropolitan areas, especially near busy streets and roadways, and in power plant plumes there is scavenging (titration) of O_3 by reaction with NO (high - NO_x regime). In between these two regimes there is a transition stage in which O_3 shows only a weak dependence on NO_x concentrations. In the high - NO_x regime, NO_2 scavenges OH radicals which would otherwise oxidize VOCs to produce peroxy radicals, which in turn would oxidize NO to NO_2. In this regime, O_3 production is limited by the availability of free radicals. The production of free radicals is in turn limited by the availability of solar UV radiation capable of photolyzing O_3 (in the Hartley bands) or aldehydes and/or by the abundance of VOCs whose oxidation produce more radicals than they consume. In the low-NO_x regime, the overall effect of the oxidation of VOCs is to generate (or

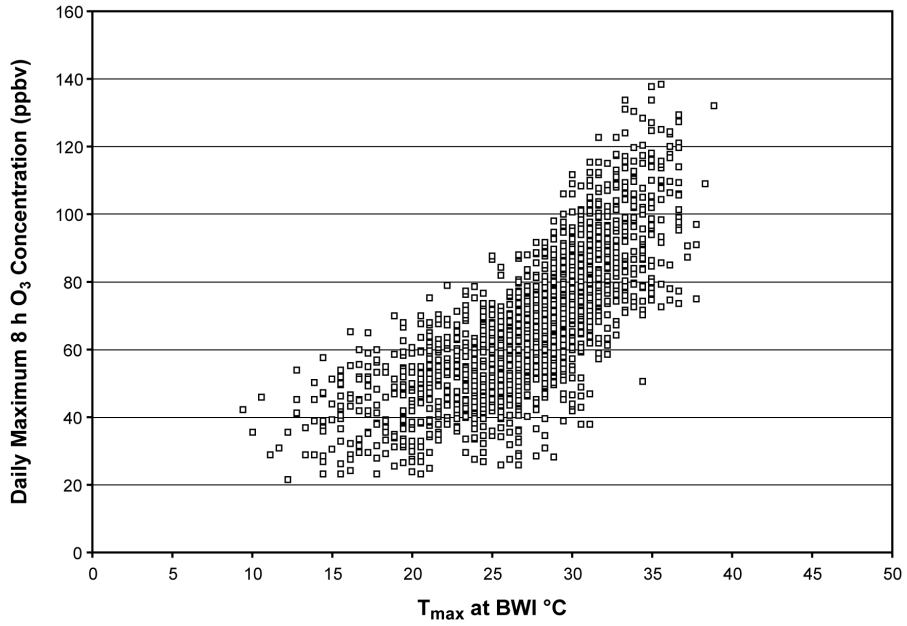


Figure 2-6. A scatter plot of daily maximum 8-h average O_3 concentrations versus daily maximum temperature for May through September 1994 to 2004 in the Baltimore, MD Air Quality Forecast Area.

Source: Piety (2005).

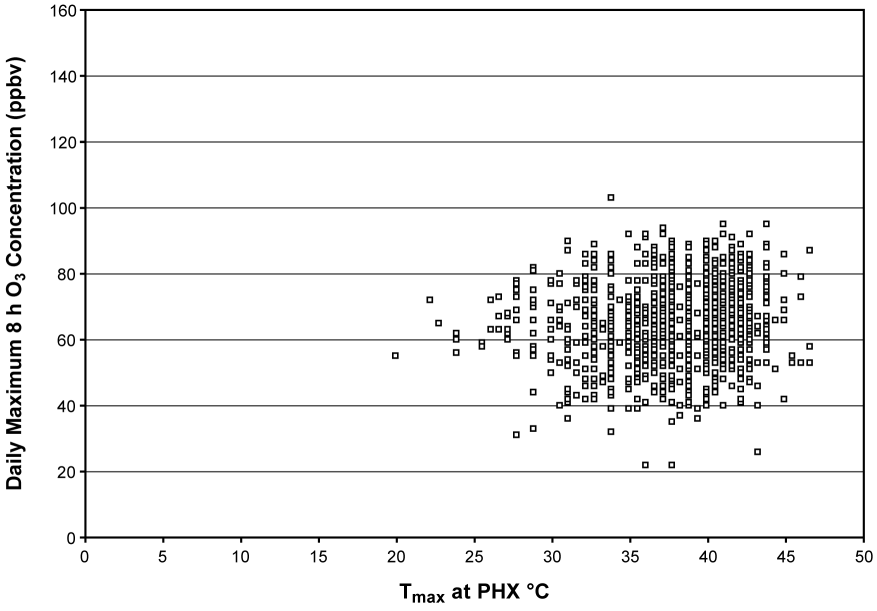


Figure 2-7. A scatter plot of daily maximum 8-h average O_3 concentrations versus daily maximum temperature for May through September 1996 to 2004 at sites downwind of Phoenix, AZ.

Source: Piety (2005).

at least not consume) free radicals, and O_3 production varies directly with NO_x. There are a number of ways to refer to the chemistry in these two chemical regimes. Sometimes the terms VOC-limited and NO_x-limited are used. However, there are difficulties with this usage because (1) VOC measurements are not as abundant as they are for nitrogen oxides, (2) rate coefficients for reaction of individual VOCs with free radicals vary over an extremely wide range, and (3) consideration is not given to CO nor to reactions that can produce free radicals without involving VOCs. The terms NO_x-limited and NO_x-saturated (e.g., Jaeglé et al., 2001) will be used wherever possible to more adequately describe these two regimes. However, the terminology used in original articles will also be used here.

The chemistry of OH radicals, which are responsible for initiating the oxidation of hydrocarbons, shows behavior similar to that for O_3 with respect to NO_x concentrations (Hameed et al., 1979; Pinto et al., 1993; Poppe et al., 1993; Zimmerman and Poppe, 1993). These considerations introduce a high degree of uncertainty into attempts to relate changes in O_3 concentrations to emissions of precursors. There are no definitive rules governing the levels of NO_x at which the transition from NO_x-limited to NO_x-saturated conditions occurs. The transition between these two regimes is highly spatially and temporally dependent and depends also on the nature and abundance of the hydrocarbons that are present.

Trainer et al. (1993) and Olszyna et al. (1994) have shown that O_3 and NO_y are highly correlated in rural areas in the eastern United States. Trainer et al. (1993) also showed that O_3 levels correlate even better with NO_z than with NO_y, as may be expected because NO_z represents the amount of NO_x that has been oxidized, forming O_3 in the process. NO_z is equal to the difference between measured total reactive nitrogen (NO_y) and NO_x and represents the summed products of the oxidation of NO_x. NO_z is composed mainly of HNO_3, PAN and other organic nitrates, particulate nitrate, and HNO_4.

Trainer et al. (1993) also suggested that the slope of the regression line between O_3 and NO_z can be used to estimate the rate of O_3 production per NO_x oxidized (also known as the O_3 production efficiency, or OPE). Ryerson et al. (1998, 2001) used measured correlations between O_3 and NO_z to identify different rates of O_3 production in plumes from large point sources. A number of studies in the planetary boundary layer over the continental United States have found that the OPE ranges typically from one to nearly ten. However, it may be higher in

the upper troposhere and in certain areas, such as the Houston-Galveston area. Observations indicate that the OPE depends mainly on the abundance of NO_x.

Various techniques have been proposed to use ambient NO_x and VOC measurements to derive information about the dependence of O_3 production on their concentrations. For example, it has been suggested that O_3 formation in individual urban areas could be understood in terms of measurements of ambient NO_x and VOC concentrations during the early morning (e.g., National Research Council, 1991). In this approach, the ratio of summed (unweighted) VOC to NO_x is used to determine whether conditions were NO_x-limited or VOC limited. This procedure is inadequate because it omits many factors that are important for O_3 production such as the impact of biogenic VOCs (which are typically not present in urban centers during early morning); important differences in the ability of individual VOCs to generate free radicals (rather than just total VOC) and other differences in O_3 forming potential for individual VOCs (Carter et al., 1995); and changes in the VOC to NO_x ratio due to photochemical reactions and deposition as air moves downwind from urban areas (Milford et al., 1994).

Photochemical production of O_3 generally occurs simultaneously with the production of various other species such as nitric acid (HNO_3), organic nitrates, and other oxidants such as hydrogen peroxide. The relative rate of production of O_3 and other species varies depending on photochemical conditions, and can be used to provide information about O_3-precursor sensitivity. Sillman (1995) and Sillman and He (2002) identified several secondary reaction products that show different correlation patterns for NO_x-limited and NO_x-saturated conditions. The most important correlations are for O_3 versus NO_y, O_3 versus NO_z, O_3 versus HNO_3, and H_2O_2 versus HNO_3. The correlations between O_3 and NO_y, and O_3 and NO_z are especially important because measurements of NO_y and NO_x are more widely available than for VOCs. Measured O_3 versus NO_z (Figure 2-8) shows distinctly different patterns in different locations. In rural areas and in urban areas such as Nashville, TN, O_3 is highly correlated with NO_z. By contrast, in Los Angeles, CA, O_3 is not as highly correlated with NO_z, and the rate of increase of O_3 with NO_z is lower and the O_3 concentrations for a given NO_z value are generally lower. The different O_3 versus NO_z relations in Nashville, TN and Los Angeles, CA reflects the difference between NO_x-limited conditions in Nashville versus an approach to NO_x- saturated conditions in Los Angeles.

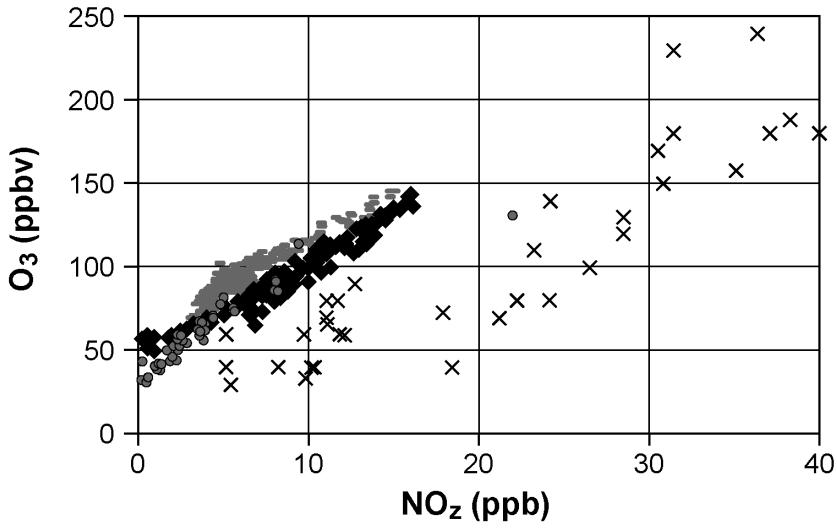


Figure 2-8. Measured values of O_3 and NO_z ($NO_y - NO_x$) during the afternoon at rural sites in the eastern United States (grey circles) and in urban areas and urban plumes associated with Nashville, TN (gray dashes); Paris, France (black diamonds); and Los Angeles CA (Xs).

Sources: Trainer et al. (1993), Sillman et al. (1997, 1998), Sillman and He (2002).

The difference between NO_x-limited and NO_x-saturated regimes is also reflected in measurements of hydrogen peroxide (H_2O_2). Hydrogen peroxide production is highly sensitive to the abundance of free radicals and is thus favored in the NO_x-limited regime. Measurements in the rural eastern United States (Jacob et al., 1995) Nashville, TN (Sillman et al., 1998), and Los Angeles, CA (Sakugawa and Kaplan, 1989), show large differences in H_2O_2 concentrations between likely NO_x-limited and NO_x-saturated locations.

2.5 THE ROLE OF CHEMISTRY-TRANSPORT MODELS IN UNDERSTANDING ATMOSPHERIC OZONE

Chemistry-transport models (CTMs) are used to improve understanding of atmospheric chemical processes and to develop control strategies (Annex AX2.5). The main components of a CTM are summarized in Figure 2-9. Models such as the CMAQ (Community Model for Air Quality) system incorporate numerical algorithms describing the processes shown in Figure 2-9.

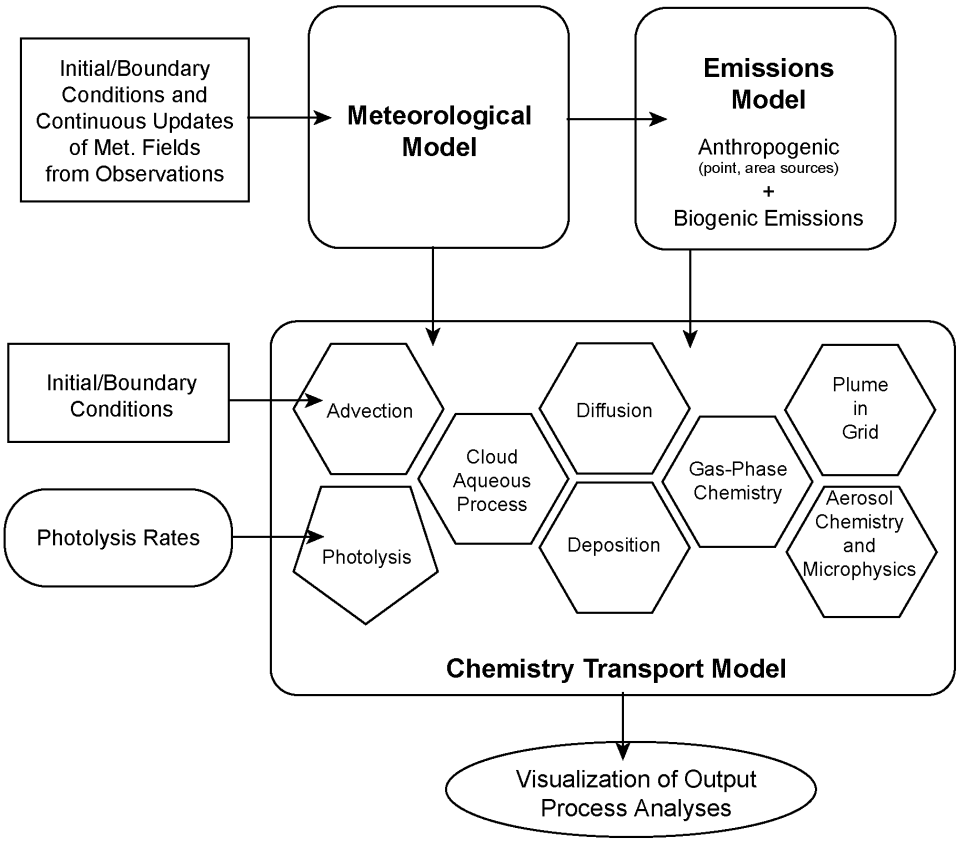


Figure 2-9. Main components of a comprehensive atmospheric chemistry modeling system, such as Models-3.

Also shown in Figure 2-9 is the meteorological model used to provide the inputs for calculating the transport of species in the CTM. Meteorological models, such as the MM5 model, which supply these inputs to the CTMs mentioned above, also provide daily weather forecasts. The domains of these models extend typically over areas of millions of square kilometers.

Because these models are computationally intensive, it is often impractical to run them over larger domains without sacrificing some features. For these reasons, both the meteorological model and the CTM rely on boundary conditions that allow processes occurring outside the model domain to influence their predictions. The entire system, consisting of meteorological model, emissions processor, and output processors shown in Figure 2-9 constitutes the framework of EPA's Models-3.

Because of the large number of chemical species and reactions that are involved in the oxidation of realistic mixtures of anthropogenic and biogenic hydrocarbons, condensed mechanisms must be used in atmospheric models. These mechanisms are tested by comparison with smog chamber data. However, the existing chemical mechanisms often neglect many important processes such as the formation and subsequent reactions of long-lived carbonyl compounds, the incorporation of the most recent information about intermediate compounds, and heterogeneous reactions involving cloud droplets and aerosol particles.

Emissions inventories are compiled for O_3 precursors (NO_x, VOCs, and CO). Recent estimates and more detailed discussions of the estimates are given in Annex AX2.5.2. Anthropogenic NO_x emissions are associated with combustion processes. Most emissions are in the form of NO, which is formed at high combustion temperatures from atmospheric nitrogen and oxygen and from fuel nitrogen. The two largest sources of NO_x are electric power generation plants and motor vehicles. Emissions of NO_x therefore are highest in areas having a high density of power plants and in urban regions having high traffic density. Natural NO_x sources include stratospheric intrusions, lightning, soils, and wildfires. Lightning, fertilized soils, and wildfires are the major natural sources of NO_x in the United States. Both nitrifying and denitrifying organisms in the soil can produce NO_x, mainly in the form of NO. Emission rates depend mainly on fertilization levels and soil temperature and moisture. Spatial and temporal variability in soil NO_x emissions leads to considerable uncertainty in emissions estimates. Nationwide, about 60% of lightning generated NO_x occurs in the southern United States and about 60% the total NO_x emitted by soils occurs in the central corn belt of the United States. The oxidation of NH_3 emitted mainly by livestock and soils, leads to the formation of a small amount of NO. Uncertainties in natural NO_x inventories are much larger than for anthropogenic NO_x emissions.

Hundreds of VOCs, containing mainly two to about twelve carbon atoms, are emitted by evaporation and combustion processes from a large number of anthropogenic sources. The two largest source categories in the U.S. EPA's emissions inventories are industrial processes and transportation. Emissions of VOCs from highway vehicles account for roughly two-thirds of the transportation-related emissions.

The accuracy of VOC emission estimates is difficult to determine, both for stationary and mobile sources. Evaporative emissions, which depend on temperature and other environmental

factors, compound the difficulties of assigning accurate emission factors. In assigning VOC emission estimates to the mobile source category, models are used that incorporate numerous input parameters (e.g., type of fuel used, type of emission controls, age of vehicle), each of which has some degree of uncertainty. Data for the ratio of CO to NO_x and NMHC to NO_x in traffic tunnels (e.g., Pierson et al., 1990) indicated that emissions of NMHCs and CO from motor vehicles have been underestimated by as much as a factor of two (based on the assumption that emissions of NO_x were reasonably well represented in the inventories). However, the results of more recent studies have been mixed, with many studies showing agreement to within ±50% (summarized in Air Quality Criteria for Carbon Monoxide [U.S. Environmental Protection Agency, 2000]). Remote sensing data (Stedman et al., 1991) indicate that about 50% of NMHC and CO emissions are produced by about 10% of the vehicles. These "super-emitters" are typically poorly maintained. Vehicles of any age engaged in off-cycle operations (e.g., rapid accelerations) emit much more than if operated in normal driving modes.

Vegetation emits significant quantities of VOCs such as terpenoid compounds (isoprene, 2-methyl-3-buten-2-ol, monoterpenes), compounds in the hexanal family, alkenes, aldehydes, organic acids, alcohols, ketones, and alkanes. The major chemicals emitted by plants are isoprene (35%), 19 other terpenoid compounds and 17 non-terpenoid compounds including oxygenated compounds (40%) (Guenther et al., 2000). Coniferous forests represent the largest source on a nationwide basis, because of their extensive land coverage. Most biogenic emissions occur during the summer, because of their dependence on temperature and incident sunlight. Biogenic emissions are also higher in southern states than in northern states for these reasons and because of species variations. The uncertainty in natural emissions is about 50% for isoprene under midday summer conditions and could be as much as a factor of ten higher for some compounds (Guenther et al., 2000). Uncertainties in both biogenic and anthropogenic VOC emission inventories prevent determination of the relative contributions of these two categories at least in many urban areas. On the regional and global scales, emissions of VOCs from vegetation are much larger than those from anthropogenic sources.

The performance of CTMs must be evaluated by comparison with field data as part of a cycle of model evaluations and subsequent improvements. Discrepancies between model predictions and observations can be used to point out gaps in current understanding of atmospheric chemistry and to spur improvements in parameterizations of atmospheric chemical

and physical processes. Model evaluation does not merely involve a straightforward comparison between model predictions and the concentration field of the pollutant of interest. Such comparisons may not be meaningful because it is difficult to determine if agreement between model predictions and observations truly represents an accurate treatment of physical and chemical processes in the CTM or the effects of compensating errors in complex model routines. Ideally, each of the model components (emissions inventories, chemical mechanism, meteorological driver) should be evaluated individually, however this is rarely done in practice. A comparison between free radical concentrations predicted by parameterized chemical mechanisms and observations suggests that radical concentrations were overestimated by current chemical mechanisms for NO_x concentrations <~5 ppb (Volz-Thomas et al., 2003).

In addition to comparisons between concentrations of calculated and measured species, comparisons of correlations between measured primary VOCs and NO_x and modeled VOCs and NO_x are especially useful for evaluating results from chemistry-transport models. Likewise, comparisons of correlations between measured species and modeled species can be used to provide information about the chemical state of the atmosphere and to evaluate model representations (including O_3 production per NO_x, O_3-NO_x-VOC sensitivity, and the general accuracy of photochemical representations). A CTM that demonstrates the accuracy of both its computed VOC and NO_x in comparison with ambient measurements and the spatial and temporal relations among the critical secondary species associated with O_3 has a higher probability of representing O_3-precursor relations correctly than one that does not.

2.6 TECHNIQUES FOR MEASURING OZONE AND ITS PRECURSORS

Several techniques have been developed for sampling and measurement of O_3 in the ambient atmosphere at ground level. Although the chemiluminescence method (CLM) using ethylene is designated as the Federal Reference Method for measuring O_3, monitoring in the NAMS/SLAMS networks is conducted mainly with UV absorption spectrometry using commercial short path instruments. The primary reference standard instrument is a relatively long-path UV absorption spectrometer maintained under carefully controlled conditions at NIST (e.g., Fried and Hodgeson, 1982). Episodic measurements are made with a variety of other

techniques based on the principles of chemiluminescence, electrochemistry, differential optical absorption spectroscopy (DOAS), and LIDAR.

In principle, each of these methods is subject to interference. Kleindienst et al. (1993) found that water vapor could cause a positive interference in the CLM with an average positive deviation of 3% ozone/% water vapor at 25 °C. The UV absorption spectrometers are subject to positive interference by atmospheric constituents, such as certain aromatic aldehydes that absorb at the 253.7 nm Hg resonance line and are at least partially removed by the MnO_2 scrubber. Parrish and Fehsenfeld (2000) did not find any evidence for significant interference (>1%) in flights through the Nashville urban plume. The same group tested the air of Houston, El Paso, Nashville, Los Angeles, San Francisco and the East Coast. They observed only one instance of substantive positive interference defined as the UV absorption technique showing more than a few ppb more than the CLM. This occurred in Laporte, TX under heavily polluted conditions and a low inversion, at night (Jobson et al., 2004). Leston et al. (2005) observed interference of from 20 to 40 ppb in Mexico City and in a separate smog chamber study. However, the concentrations of relevant compounds were many times higher than found in U.S. urban areas. Thus, it is not likely that such interference could be more than a few ppb under typical ambient conditions. However, Leston et al. (2005) suggested that the use of other materials in the scrubber could have eliminated the interference seen in their smog chamber study.

By far, most measurements of NO are made using the CLM, based on its reaction with O_3. Commercial instruments for measuring NO and NO_2 are constructed with an internal converter for reducing NO_2 to NO and then measuring NO by the CLM. In principle, this technique yields a measurement of NO_x with NO_2 found by difference between NO_x and NO. However, these converters also reduce NO_z compounds thereby introducing a positive interference in the measurement of NO_2. Other methods for measuring NO_2 are available, such as photolytic reduction followed by CLM, laser-induced fluorescence and DOAS. However, they require further development before they can be used for routine monitoring in the NAMS/SLAMS networks. More detailed descriptions of the issues and techniques discussed above and techniques for measuring HNO_3 and VOCs can be found in Annex AX2.6.

2.7 SUMMARY

Ozone (O_3) is formed by atmospheric reactions involving two classes of precursor compounds, volatile organic compounds (VOCs) and nitrogen oxides (NO_x). Ozone is thus a secondary pollutant. Ozone is ubiquitous throughout the atmosphere; it is present even in remote areas of the globe. The photochemical oxidation of almost all anthropogenic and biogenic VOCs is initiated by reaction with hydroxyl (OH) radicals. At night, when they are most abundant, NO_3 radicals also oxidize alkenes. In coastal and other select environments, Cl and Br radicals can also initiate the oxidation of VOCs.

In urban areas, basically all classes of VOCs (alkanes, alkenes, aromatic hydrocarbons, carbonyl compounds, etc.) and CO are important for O_3 formation. Although knowledge of the oxidative mechanisms of VOCs has improved over the past several years, gaps in knowledge involving key classes, such as aromatic hydrocarbons, still remain. For example, only about half of the carbon initially present in aromatic hydrocarbons in smog chamber studies form compounds that can be identified.

In addition to gas phase reactions, reactions also occur on the surfaces of, or within cloud droplets and airborne particles. Most of the well-established multiphase reactions tend to reduce the rate of O_3 formation in polluted environments. Reactions of Cl and Br containing radicals deplete O_3 in selected environments such as the Arctic during spring, the tropical marine boundary layer and inland salt lakes. Direct reactions of O_3 with atmospheric particles appear to be too slow to reduce O_3 formation significantly at typical ambient PM levels.

Our basic understanding of the meteorological processes associated with summertime O_3 episodes has not changed over the past several years. However, the realization that long-range transport processes are important for determining O_3 concentrations at the surface is growing. In addition to synoptic scale flow fields, nocturnal low-level jets are capable of transporting pollutants hundreds of km from their sources in either the upper boundary layer or the lower free troposphere. Turbulence then brings O_3 and other pollutants to the surface. On larger scales, important progress has been made in identifying the mechanisms of intercontinental transport of O_3 and other pollutants.

Some O_3 would be found near the earth's surface as the result of its downward transport from the stratosphere, even in the absence of photochemical reactions in the troposphere. Intrusions of stratospheric O_3 that reach the surface are rare. Much more common are intrusions

that penetrate to the middle and upper troposphere. However, O_3 transported to the middle and upper troposphere can still affect surface concentrations through various mechanisms that mix air between the planetary boundary layer and the free troposphere above.

The formation of O_3 and associated compounds is a complex, nonlinear function of many factors including the intensity and spectral distribution of sunlight; atmospheric mixing and other atmospheric processes; and the concentrations of the precursors in ambient air. At the lower NO_x concentrations found in most environments, ranging from remote continental areas to rural and suburban areas downwind of urban centers, the net production of O_3 increases with increasing NO_x. At the higher concentrations found in downtown metropolitan areas, especially near busy streets and highways, and in power plant plumes there is net destruction of O_3 by reaction with NO. In between these two regimes there is a transition stage, in which O_3 production shows only a weak dependence on NO_x concentrations. The efficiency of O_3 production per NO_x oxidized is generally highest in areas where NO_x concentrations are lowest and decrease with increasing NO_x concentration.

Chemistry transport models are used to improve understanding of atmospheric chemical and physical processes as well as to develop air pollution control strategies. The performance of these models must be evaluated by comparison with field data as part of a cycle of model evaluations and subsequent improvements. Discrepancies between model predictions and observations can be used to point out gaps in current understanding and thus to improve parameterizations of atmospheric chemical and physical processes. Model evaluation does not merely involve a straightforward comparison between model predictions and the concentration fields of a pollutant of interest (e.g., O_3). Such comparisons may not be meaningful because it is difficult to determine if agreement between measurements and model predictions truly represents an accurate treatment of physical and chemical processes in the model or the effects of compensating errors in model routines.

The main methods in use for routine monitoring of ambient O_3 are based on chemiluminescence or UV absorption. Measurements at most ambient monitoring sites are based on UV absorption. Both of these methods are subject to interference by other atmospheric components. One study found large positive interference in Mexico City and in a smog chamber, but few studies conducted in urban plumes did not find significant positive interference in the UV absorption technique.

REFERENCES

Berkowitz, C. M.; Shaw, W. J. (1997) Airborne measurements of boundary layer chemistry during the Southern Oxidant Study: a case study. J. Geophys. Res. [Atmos.] 102: 12,795-12,804.

Berkowitz, C. M.; Fast, J. D.; Sprinston, S. R.; Larsen, R. J.; Spicer, C. W.; Doskey, P. V.; Hubbe, J. M.; Plastridge, R. (1998) Formation mechanisms and chemical characteristics of elevated photochemical layers over the northeast United States. J. Geophys. Res. [Atmos.] 103: 10,631-10,647.

Blumenthal, D. L.; Lurmann, F. W.; Kumar, N.; Dye, T. S.; Ray, S. E.; Korc, M. E.; Londergan, R.; Moore, G. (1997) Transport and mixing phenomena related to ozone exceedances in the northeast U.S. (analysis based on NARSTO-northeast data). Available: http://capita.wustl.edu/otag/reports/otagrept/otagrept.html (30 October 2003).

Bonner, W. D. (1968) Climatology of the low level jet. Mon. Weather Rev. 96: 833-850.

Carter, W. P. L. (1995) Computer modeling of environmental chamber studies of maximum incremental reactivities of volatile organic compounds. Atmos. Environ. 29: 2513.

Civerolo, K. L.; Mao, H. T.; Rao, S. T. (2003) The airshed for ozone and fine particulate pollution in the eastern United States. Pure Appl. Geophys. 160: 81-105.

Corsmeier, U.; Kalthhoff, N.; Kolle, O.; Motzian, M.; Fiedler, F. (1997) Ozone concentration jump in the stable nocturnal boundary layer during a LLJ-event. Atmos. Environ. 31: 1977-1989.

Eisele, F. L.; Mount, G. H.; Tanner, D.; Jefferson, A.; Shetter, R.; Harder, J. W.; Williams, E. J. (1997) Understanding the production and interconversion of the hydroxyl radical during the tropospheric OH photochemistry experiment. J. Geophys. Res. 102: 6457-6465.

Fehsenfeld, F. C.; Trainer, M.; Parrish, D. D.; Volz-Thomas, A.; Penkett, S. (1996) North Atlantic Regional Experiment (NARE) 1993 summer intensive: foreword. J. Geophys. Res. [Atmos.] 101: 28,869-28,875.

Fried, A.; Hodgeson, J. (1982) Laser photoacoustic detection of nitrogen dioxide in the gas-phase titration of nitric oxide with ozone. Anal. Chem. 54: 278-282.

Guenther, A.; Geron, C.; Pierce, T.; Lamb, B.; Harley, P.; Fall, R. (2000) Natural emissions of non-methane volatile organic compounds, carbon monoxide, and oxides of nitrogen from North America. Atmos. Environ. 34: 2205-2230.

Hameed, S.; Pinto, J. P.; Stewart, R. W. (1979) Sensitivity of the predicted CO-OH-CH_4 perturbation to tropospheric NO_x concentrations. J. Geophys. Res. C: Oceans Atmos. 84: 763-768.

Husar, R. B.; Renard, W. P. (1998) Ozone as a function of local wind speed and direction: Evidence of local and regional transport. Presented at: 91st annual meeting and exhibition of the Air & Waste Management Association; June; San Diego, CA. Pittsburgh, PA: Air & Waste Management Association; online paper no. 98-A922. Available: http://capita.wustl.edu/capita/CapitaReports/REPORTS1.HTML (13 November 2003).

Jacob, D. J.; Horowitz, L. W.; Munger, J. W.; Heikes, B. G.; Dickerson, R. R.; Artz, R. S.; Keene, W. C. (1995) Seasonal transition from NO_x- to hydrocarbon-limited conditions for ozone production over the eastern United States in September. J. Geophys. Res. 100: 9315-9324.

Jaeglé, L.; Jacob, D. J.; Brune, W. H.; Wennberg, P. O. (2001) Chemistry of HOx radicals in the upper troposphere. Atmos. Environ. 35: 469-489.

Jobson, B. T.; Berkowitz, C. M.; Kuster, W. C.; Goldan, P. D.; Williams, E. J.; Fehsenfeld, F. C.; Apel, E. C.; Karl, T.; Lonneman, W. A.; Riemer, D. (2004) Hydrocarbon source signatures in Houston, Texas: influence of the petrochemical industry. J. Geophys. Res. 109: D24305: 10.1029/2004JD004887.

Kasibhatla, P.; Chameides, W. L. (2000) Seasonal modeling of regional ozone pollution in the eastern United States. Geophys. Res. Lett. 27: 1415-1418.

Kleindienst, T. E.; Hudgens, E. E.; Smith, D. F.; McElroy, F. F.; Bufalini, J. J. (1993) Comparison of chemiluminescence and ultraviolet ozone monitor responses in the presence of humidity and photochemical pollutants. Air Waste 43: 213-222.

Leston, A.; et al. (2005) J. Air Waste Manage. Assoc.: in press.

Milford, J. B.; Gao, D.; Sillman, S.; Blossey, P.; Russell, A. G. (1994) Total reactive nitrogen (NO_y) as an indicator of the sensitivity of ozone to reductions in hydrocarbon and NO_x emissions. J. Geophys. Res. 99: 3533-3542.

National Research Council. (1991) Rethinking the ozone problem in urban and regional air pollution. Washington, DC: National Academy Press. Available: http://www.nap.edu/books/0309046319/html/ [26 March, 2004].

Olszyna, K. J.; Bailey, E. M.; Simonaitis, R.; Meagher, J. F. (1994) O_3 and NO_y relationships at a rural site. J. Geophys. Res. [Atmos.] 99: 14,557-14,563.

Parrish, D. D.; Fehsenfeld, F. C. (2000) Methods for gas-phase measurements of ozone, ozone precursors and aerosol precursors. Atmos. Environ. 34: 1921-1957.

Pierson, W. R.; Gertler, A. W.; Bradow, R. L. (1990) Comparison of the SCAQS tunnel study with historical data. Presented at: 83rd annual meeting & exhibition of the Air and Waste Management Association; June; Pittsburgh, PA. Pittsburgh, PA: Air and Waste Management Association; paper no. 90-175.3.

Piety, C. A. (2005) The relation between daily maximum ozone and daily maximum temperature [memorandum to Dr. Joseph Pinto]. Research Triangle Park, NC: U.S. Environmental Protection Agency; July 18.

Pinto, J. P.; Bruhl, C.; Thompson, A. M. (1993) The current and future envirionmental role of atmospheric methane. In: Khalil, M. A. K., ed. Atmospheric methane sources, sinks, and role in global change, p. 514-531. (NATO ASI Series, v. 113).

Poppe, D.; Wallasch, M.; Zimmermann, J. (1993) The dependence of the concentration of OH on its precursors under moderately polluted conditions: a model study. J. Atmos. Chem. 16: 61-78.

Rao, S. T.; Ku, J.-Y.; Berman, S.; Zhang, K.; Mao, H. (2003) Summertime characteristics of the atmospheric boundary layer and relationships to ozone levels over the eastern United States. Pure Appl. Geophys. 160: 21-55.

Ryerson, T. B.; Buhr, M. P.; Frost, G. J.; Goldan, P. D.; Holloway, J. S.; Hübler, G.; Jobson, B. T.; Kuster, W. C.; McKeen, S. A.; Parrish, D. D.; Roberts, J. M.; Sueper, D. T.; Trainer, M.; Williams, J.; Fehsenfeld, F. C. (1998) Emissions lifetimes and ozone formation in power plant plumes. J. Geophys. Res. 103(D17): 22,569-22,583.

Ryerson, T. B.; Trainer, M.; Holloway, J. S.; Parrish, D. D.; Huey, L. G.; Sueper, D. T.; Frost, G. J.; Donnelly, S. G.; Schauffler, S.; Atlas, E. L.; Kuster, W. C.; Goldan, P. D.; Hübler, G.; Meagher, J. F.; Fehsenfeld, F. C. (2001) Observations of ozone formation in power plant plumes and implications for ozone control strategies. Science (Washington, DC) 292: 719-723.

Sakugawa, H.; Kaplan, I. R. (1989) H_2O_2 and O_3 in the atmosphere of Los Angeles and its vicinity: factors controlling their formation and their role as oxidants of SO_2. J. Geophys. Res. [Atmos.] 94: 12,957-12,973.

Schichtel, B. A.; Husar, R. B. (2001) Eastern North American transport climatology during high- and low-ozone days. Atmos. Environ. 35: 1029-1038.

Seinfeld, J. H.; Pandis, S. N. (1998) Atmospheric chemistry and physics: from air pollution to climate change. New York, NY: John Wiley & Sons, Inc.

Shapiro, M. A. (1980) Turbulent mixing within tropopause folds as a mechanism for the exchange of chemical constituents between the stratosphere and troposphere. J. Atmos. Sci. 37: 994-1004.

Sillman, S. (1995) The use of NO_y, H_2O_2 and HNO_3 as indicators for ozone-NO_x-hydrocarbon sensitivity in urban locations. J. Geophys. Res. 100: 14,175-14,188.

Sillman, S.; He, D.-Y. (2002) Some theoretical results concerning O_3-NO_x-VOC chemistry and NO_x-VOC indicators. J. Geophys. Res. (Atmos.) 107: 10.1029/2001JD001123.

Sillman, S.; He, D.; Cardelino, C.; Imhoff, R. E. (1997) The use of photochemical indicators to evaluate ozone-NO_x-hydrocarbon sensitivity: case studies from Atlanta, New York, and Los Angeles. J. Air Waste Manage. Assoc. 47: 1030-1040.

Sillman, S.; He, D.; Pippin, M. R.; Daum, P. H.; Imre, D. G.; Kleinman, L. I.; Lee, J. H.; Weinstein-Lloyd, J. (1998) Model correlations for ozone, reactive nitrogen, and peroxides for Nashville in comparison with measurements: implications for O_3-NO_x-hydrocarbon chemistry. J. Geophys. Res. [Atmos.] 103: 22,629-22,644.

Stedman, D. H.; Bishop, G.; Peterson, J. E.; Guenther, P. L. (1991) On-road CO remote sensing in the Los Angeles Basin: final report. Sacramento, CA: California Air Resources Board, ARB Contract No. A932-189.

Stull, R. B. (1999) An introduction to boundary layer meteorology. Dordrecht, The Netherlands: Kluwer Academic Publishers; pp. 9-15, 500-505.

Stull, R. B. (2000) Meteorology for scientists and engineers: a technical companion book with Ahrens' Meteorology Today. 2nd ed. Pacific Grove, CA: Brooks/Cole.

Taubman, B. F.; Marufu, L. T.; Piety, C. A.; Doddridge, B. G.; Stehr, J. W.; Dickerson, R. R. (2004) Airborne characterization of the chemical, optical, and meteorological properties, and origins of a combined ozone-haze episode over the eastern United States. J. Atmos. Sci. 61: 1781-1793.

Trainer, M.; Parrish, D. D.; Buhr, M. P.; Norton, R. B.; Fehsenfeld, F. C.; Anlauf, K. G.; Bottenheim, J. W.; Tang, Y. Z.; Wiebe, H. A.; Roberts, J. M.; Tanner, R. L.; Newman, L.; Bowersox, V. C.; Meagher, J. F.; Olszyna, K. J.; Rodgers, M. O.; Wang, T.; Berresheim, H.; Demerjian, K. L.; Roychowdhury, U. K. (1993) Correlation of ozone with NO_y in photochemically aged air. J. Geophys. Res. [Atmos.] 98: 2917-2925.

U.S. Environmental Protection Agency. (2000) Air quality criteria for carbon monoxide. Research Triangle Park, NC: National Center for Environmental Assessment; report no. EPA/600/P-99/001F. Available: http://www.epa.gov/ncea/pdfs/coaqcd.pdf (7 May 2003).

Volz-Thomas, A.; Geiss, H.; Hofzumahaus, A.; Becker, K.-H. (2003) Introduction to special section: photochemistry experiment in BERLIOZ. J. Geophys. Res. [Atmos.] 108(D4): 10.1029/JD002029.

Zimmermann, J.; Poppe, D. (1993) Nonlinear chemical couplings in the tropospheric NO_x–HO_x gas phase chemistry. J. Atmos. Chem. 17: 141-155.

3. ENVIRONMENTAL CONCENTRATIONS, PATTERNS, AND EXPOSURE ESTIMATES

3.1 INTRODUCTION

Identification and Use of Existing Air Quality Data

Topics discussed in this chapter include the characterization of ambient air quality data for ozone (O_3), the uses of these data in assessing the exposure of vegetation to O_3, concentrations of O_3 in microenvironments, and a discussion of the currently available human exposure data and exposure model development. The information contained in this chapter pertaining to ambient concentrations is taken primarily from the U.S. Environmental Protection Agency (EPA) Air Quality System (AQS; formerly the AIRS database). The AQS contains readily accessible detailed, hourly data that has been subject to EPA quality control and assurance procedures. Data available in AQS were collected from 1979 to 2001. As discussed in previous versions of the O_3 Air Quality Criteria Document or AQCD (U.S. Environmental Protection Agency, 1986, 1996), the data available prior to 1979 may be unreliable due to calibration problems and uncertainties.

As noted in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996), O_3 is the only photochemical oxidant other than nitrogen dioxide (NO_2) that is routinely monitored and for which a comprehensive database exists. Data for peroxyacetyl nitrate (PAN), hydrogen peroxide (H_2O_2), and other oxidants either in the gas phase or particle phase typically have been obtained only as part of special field studies. Consequently, no data on nationwide patterns of occurrence are available for these non-O_3 oxidants; nor are extensive data available on the relationships of levels and patterns of these oxidants to those of O_3. However, available data for gas phase and particle phase oxidants will be discussed.

Characterizing Ambient Ozone Concentrations

The "concentration" of a specific air pollutant is typically defined as the amount (mass) of that material per unit volume of air. However, most of the data presented in this chapter are expressed as "mixing ratios" in terms of a volume-to-volume ratio (parts per million [ppm] or parts per billion [ppb]). Data expressed this way are often referred to as concentrations, both in

the literature and in the text, following common usage. Human exposures are expressed in units of mixing ratio times time.

Several different types of indicators are used for evaluating exposures of vegetation to O_3. The peak-weighted, cumulative exposure indicators used in this chapter for characterizing vegetation exposures are SUM06 and SUM08 (the sums of all hourly average concentrations $\geq$0.06 and 0.08 ppm, respectively) and W126 (the sum of the hourly average concentrations that have been weighted according to a sigmoid function that is based on a hypothetical vegetation response [see Lefohn and Runeckles, 1987]). Further discussion of these exposure indices is presented in Chapter 9.

The EPA has established "ozone seasons" during which measurement of ambient O_3 concentrations for different locations within the United States and the U.S. territories is required (CFR, 2000). Table AX3-1 shows the O_3 seasons during which continuous, hourly averaged O_3 concentrations must be monitored. Monitoring is optional outside of these O_3 seasons and indeed is conducted during the winter in a number of areas.

Data for O_3 in ambient air across the United States are summarized in Section 3.2. The data are summarized for urban, rural, and relatively remote sites. Relatively remote monitoring sites (RRMS) are sites that are not strongly influenced by nearby pollution sources and are located mainly in national parks in the West. However, this does not mean that they are free of the effects of regional or local pollution, especially during tourist seasons. Data for the spatial variability of O_3 within urban areas are summarized in Section 3.3. Data for the diurnal and seasonal variability of O_3 concentrations are given in Section 3.4. The long term temporal variability of O_3 concentrations is discussed in Section 3.5. Relationships among O_3 and other species are discussed in Section 3.6. Information about the occurrence of other oxidants and their relationship to O_3 is given in this section. A discussion of Policy Relevant Background (PRB) O_3 concentrations is presented in Section 3.7. PRB O_3 concentrations are background O_3 concentrations used for the purposes of setting the O_3 NAAQS. They are used by the EPA to assess risks to human health. Indoor sources and emissions of O_3 are discussed in Section 3.8. Issues related to evaluating human exposure to O_3 are summarized in Section 3.9. Finally, a summary of key points in Chapter 3 is given in Section 3.10.

3.2 AMBIENT AIR QUALITY DATA FOR OZONE

Ozone Air Quality at Urban, Suburban, and Nonurban Sites

Figure 3-1 shows the mean daily maximum 8-h O_3 concentrations and Figure 3-2 shows the 95th percentile values of the daily maximum 8-h O_3 concentrations, based on countywide averages across the United States from May to September 2000 to 2004. The period from May to September was chosen because, although O_3 is monitored for different lengths of time across the country, all O_3 monitors should be operational during these months. Data flagged because of quality control issues were removed with concurrence by the local monitoring agency. Only days with data for 18 of 24 hours were kept, and a minimum of 115 of 153 days were required in each year. Cut points for the tertile distributions on each map were chosen at the median and 95th percentile values. These cut points were chosen as they represent standard metrics for characterizing important aspects of human exposure used by the EPA. Any other percentiles or statistics that are believed to be helpful for characterizing human exposures could also be used. Blank areas on the maps indicate no data coverage. It should be noted that county areas can be much larger in the West than in the East, but monitors are not spread evenly within a county. As a result, the assigned concentration range might not represent conditions throughout a particular county and so large areas in western counties where there are no monitors were blanked out.

As shown in Figure 3-1, the median of the countywide, mean daily maximum 8-h O_3 concentration across the United States is 49 ppb, and the corresponding 95th percentile value is 57 ppb. Though the median and 95th percentile values are fairly close, these results cannot be taken to imply that average O_3 concentrations lie in a relatively narrow range throughout the United States, because data coverage is not as complete in the West as it is in the East. High mean daily maximum 8-h O_3 concentrations are found in California and states in the Southwest as well as in several counties in the East. As shown in Figure 3-2, the nationwide median of the countywide, 95th percentile value of the daily maximum 8-h O_3 concentration is 73 ppb and 5% of these values are above 85 ppb. High values for the 95th percentiles are found in California, Texas, and some counties in the East, but not necessarily in the same counties in the East as shown for the mean daily maximum 8-h concentrations in Figure 3-1.

Although mean O_3 concentrations in Houston, TX were below the nationwide median, its 95th percentile value ranks in the highest 5% nationwide. Conversely, mean O_3 concentrations in southwestern states are among the highest in the United States, but values at the upper end of

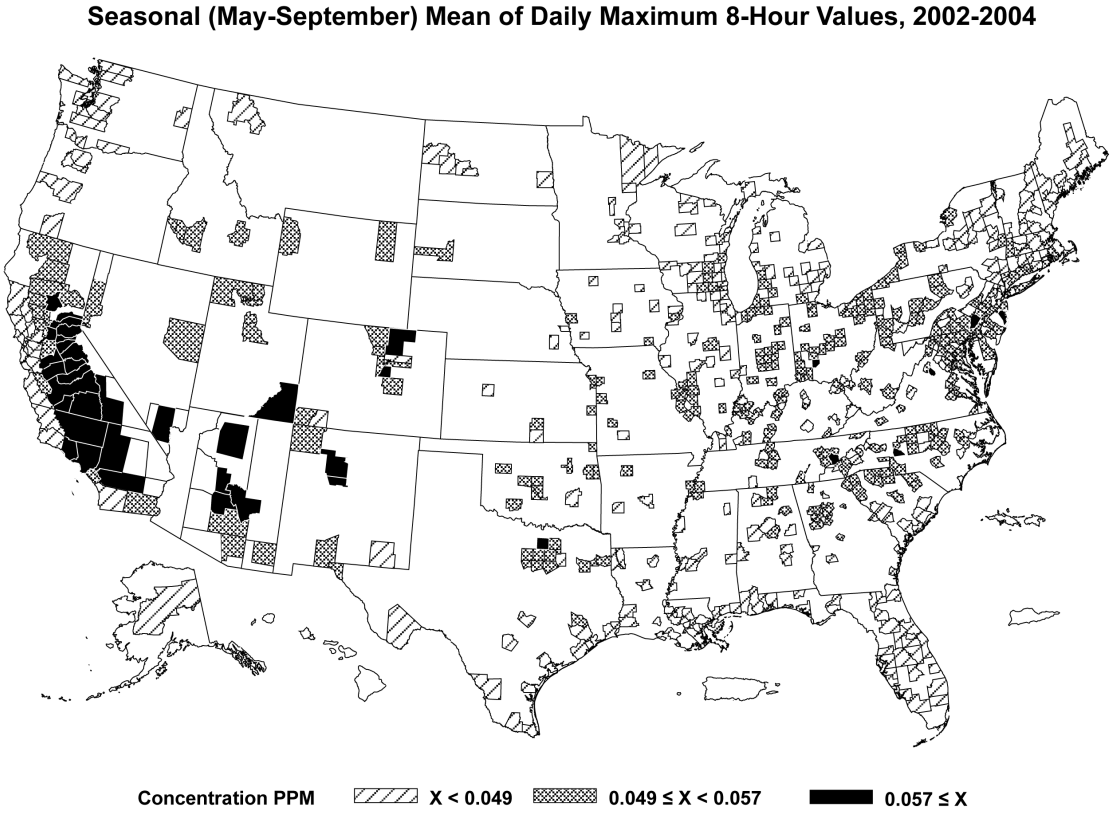


Figure 3-1. Countywide mean daily maximum 8-h O_3 concentrations, May to September 2000 to 2004.

Source: Fitz-Simons et al. (2005).

the distribution (e.g., the 95th percentile value) in these states are not among the highest peak values in the United States. In other areas where the highest mean O_3 concentrations occurred, such as California; Dallas-Fort Worth, TX; and the Northeast Corridor, the highest peak values were also observed.

Although countywide averages are shown, it should be noted that considerable spatial variability can exist within a county, especially within urban areas as described in Section 3.3. In addition, there can also be differences in the diurnal profile of O_3 among monitors within counties.

Box plots showing the percentile distribution of nationwide O_3 concentrations for different averaging periods (1-h daily maximum, 8-h daily maximum and 24-h daily average) are given in

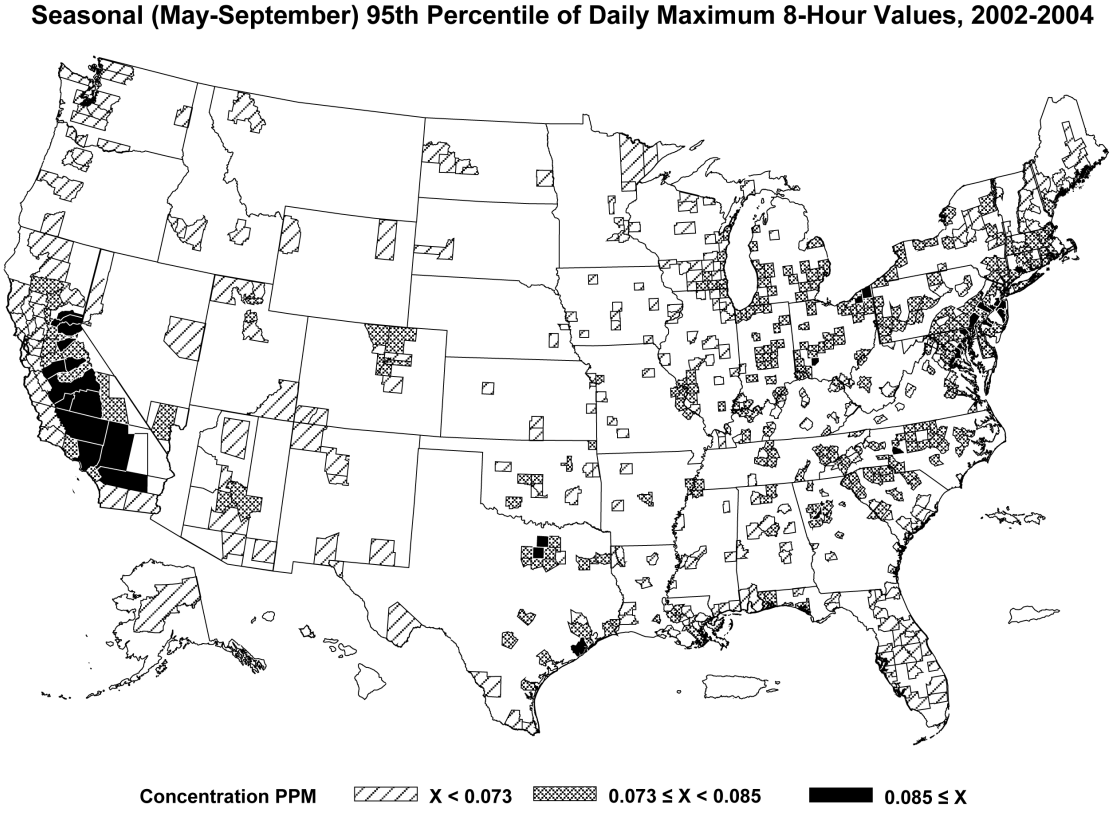


Figure 3-2. Countywide 95th percentile value of daily maximum 8-h O_3 concentrations, May to September 2000 to 2004.

Source: Fitz-Simons et al. (2005).

Figures AX3-4 to AX3-6 and numerical values are given in Table AX3-2. The differences between the 50th and 95th percentile values can be used to provide indications of differences in O_3 levels between "typical" O_3 days and "high" O_3 days. These differences are approximately 40, 30, and 25 ppb for the daily 1-h and 8-h daily maxima and 24-h average O_3 concentrations. As might be expected, the daily maximum 1-h and 8-h O_3 concentrations are highly correlated.

Lehman et al. (2004) have shown that the eastern United States can be divided into five regions, each of which exhibit spatial, relatively coherent patterns of O_3 properties at nonurban sites. Only sites classified as being rural or suburban and with land usage of forest, agriculture, or residential were included in the analyses. These criteria were chosen to avoid sites where O_3 is scavenged by NO that can be found in high concentrations near major sources, such as traffic

in urban cores. The five regions, shown in Figure 3-3, are characterized by different patterns of O_3 properties such as temporal persistence and seasonal variability. Figure 3-3 shows nonurban, monthly average, daily maximum 8-h O_3 concentrations in the five regions in the eastern United States from April to October 1993 to 2002.

Regional differences are immediately apparent. Highest concentrations among all the regions are generally found in the Mid-Atlantic region (mean of 52 ppb) with highest values throughout the percentile distribution except for the overall maximum. Lowest mean concentrations (42 ppb) are found in Florida. In the northern regions (the Northeast, Great Lakes) and the Mid-Atlantic region, highest median and peak concentrations are found in July, whereas in the Southwest region, highest median concentrations are found in August, with highest peaks in June and September, i.e., outside the warmest summer months. In Florida, highest monthly averaged median and peak concentrations are found during the spring. High O_3 concentrations tend to be most persistent (3-4 days of persistence) in the southern regions, less persistent in the Mid-Atlantic region (2-3 days) and least persistent in the northern regions (1 or 2 days). Analyses, such as these, are not available for the western United States, in part because of the difficulty in defining regions with relatively coherent O_3 properties.

Box plots showing the percentile distribution of hourly average O_3 concentrations for different types of rural sites for 2004 are given in Figures 3-4a (rural-agricultural), 3-4b (rural-forest) and 3-4c (rural-residential or commercial). Some associated metrics for vegetation exposures are given in Figures AX3-8 to AX3-10. Note that high O_3 concentrations are found at sites that are classified as rural, such as Anne Arundel Co., MD; Yosemite NP, CA; and Crestline, CA. Land use designations do not usually give an accurate picture of exposure regimes in rural areas, because the land use characterization of "rural" does not imply that a specific location is isolated from anthropogenic influences. Rather, the characterization refers only to the current use of the land, not to the presence of sources. Since O_3 produced from emissions in urban areas is transported to more rural downwind locations, elevated O_3 concentrations can occur at considerable distances from urban centers. In addition, major sources of O_3 precursors such as power plants and highways are located in nonurban areas and also produce O_3 in these areas. Due to lower chemical scavenging in nonurban areas, O_3 tends to persist longer in nonurban than in urban areas also tending to lead to higher exposures in nonurban areas influenced by anthropogenic precursor emissions.

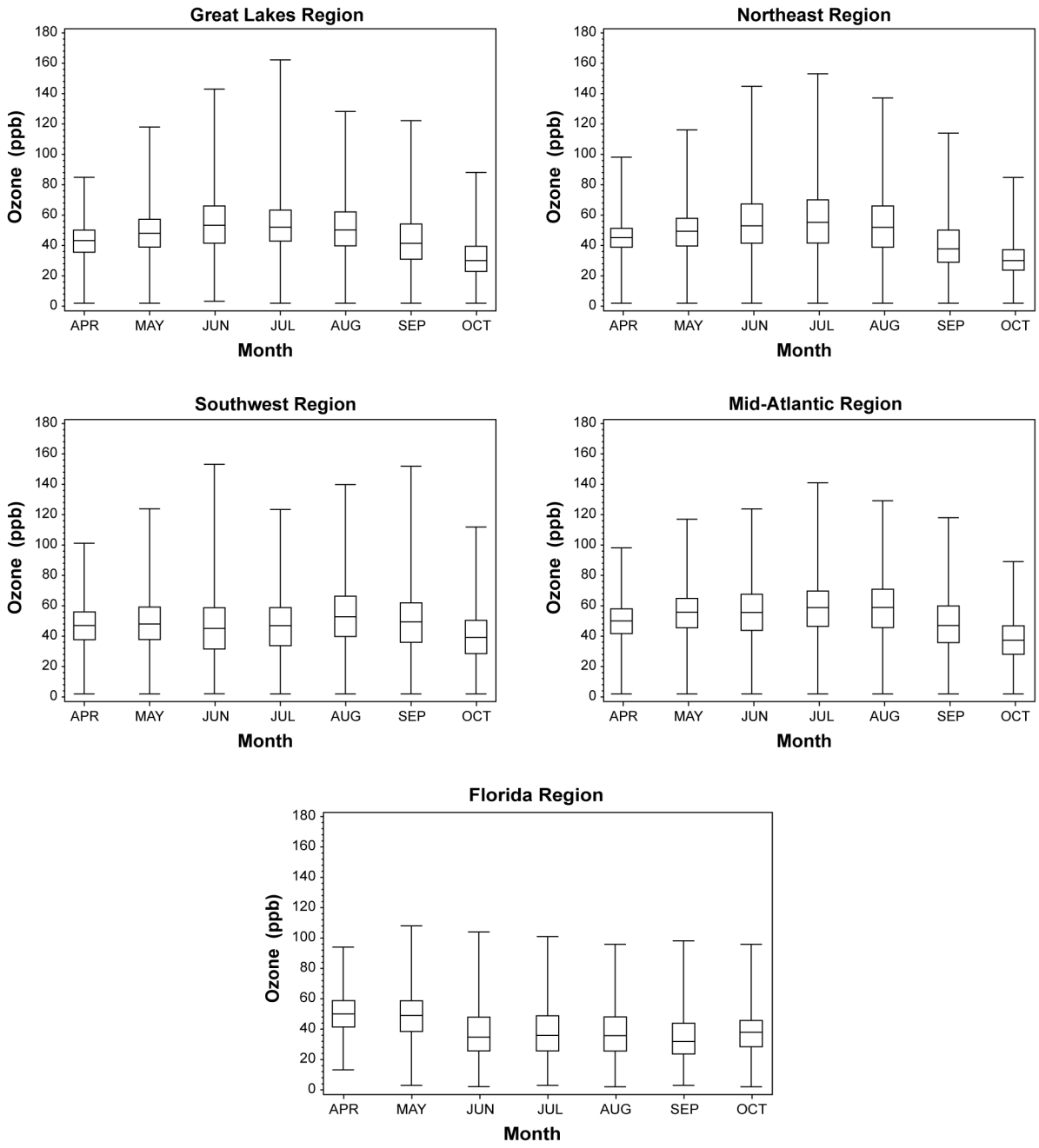


Figure 3-3. Box plots showing daily maximum 8-h O_3 averaged by month over 1993 to 2002 in the five regions in the eastern United States derived by Lehman et al. (2004). The boxes define the interquartile range and the whiskers, the extreme values.

Source: Lehman et al. (2004).

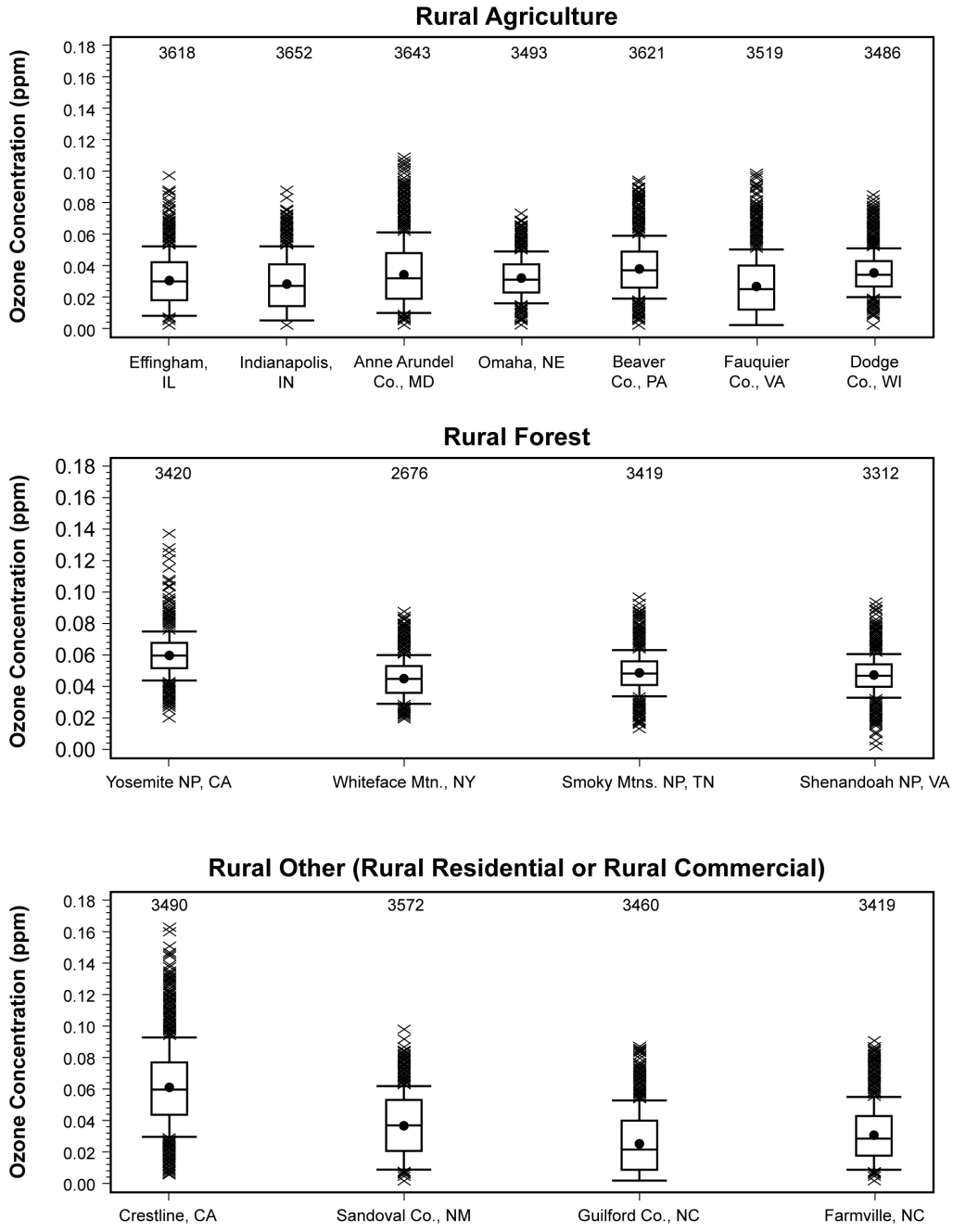


Figure 3-4a-c. Hourly average O_3 concentrations observed at selected (a) rural-agricultural (b) rural-forested, and (c) rural-residential or commercial sites for 2004. The whiskers on the box plot represent the 10th and 90th percentile concentrations. The "X"s above and below the whiskers are the values that fall below and above the 10th and 90th percentile concentrations. The dots inside the box represent the mean, for the statistic, at all sites. The number of observations is shown above each box plot.

Source: Fitz-Simons et al. (2005).

Ozone Air Quality Data at Relatively Remote Monitoring Sites (RRMS)

RRMS are sites that are located in the national parks that tend to be less affected by obvious pollution sources than other sites. This does not mean that they are completely unaffected by local pollution, as evidenced by the number of visitors to these national parks.

Box plots showing the percentile distribution of annual hourly averaged O_3 concentrations at four relatively remote monitoring sites (RRMS) are given in Figures 3-5a-d. It is important to characterize hourly average O_3 concentrations at RRMS so that assessments of the possible effects of O_3 on human health and vegetation use ranges of concentrations in their experiments that span the range of O_3 concentrations found in the U.S. In many controlled exposure studies examining vegetation, O_3 is filtered out of ambient air before it is admitted into the exposure chambers. As a result, O_3 levels of only a few ppb are used as controls.

As can be seen from Figures 3-5a-d, annual mean values of the daily maximum 8-h O_3 concentration have not changed much over the past 10 years of available data. Mean values typically range from about 0.020 ppm to about 0.040 ppm. Concentrations only rarely exceed 0.080 ppm, in contrast to observations at other "rural" sites shown in Figures 3-4a-c.

The extent to which distributions found at sites with low maximum hourly average concentrations in the western United States are representative of sites in the eastern and midwestern United States is debatable because of regional differences in sources of precursors and transport patterns. Given the high density of sources in the eastern and midwestern United States, it is unclear whether a site could be found in either of these regions that would not be influenced by the transport of O_3 from nearby urban areas. Thus, with the exception of the Voyageurs NP site in Minnesota, observations at RRMS are limited to those obtained in the western United States. However, not all national park sites in the West can be considered to be free of strong regional pollution influences, e.g., Yosemite NP (CA) as shown in Figure 3-4b. Maps showing the nationwide distribution of various metrics for vegetation exposures are given in Section AX3.2, Figures AX3-13 to AX3-27.

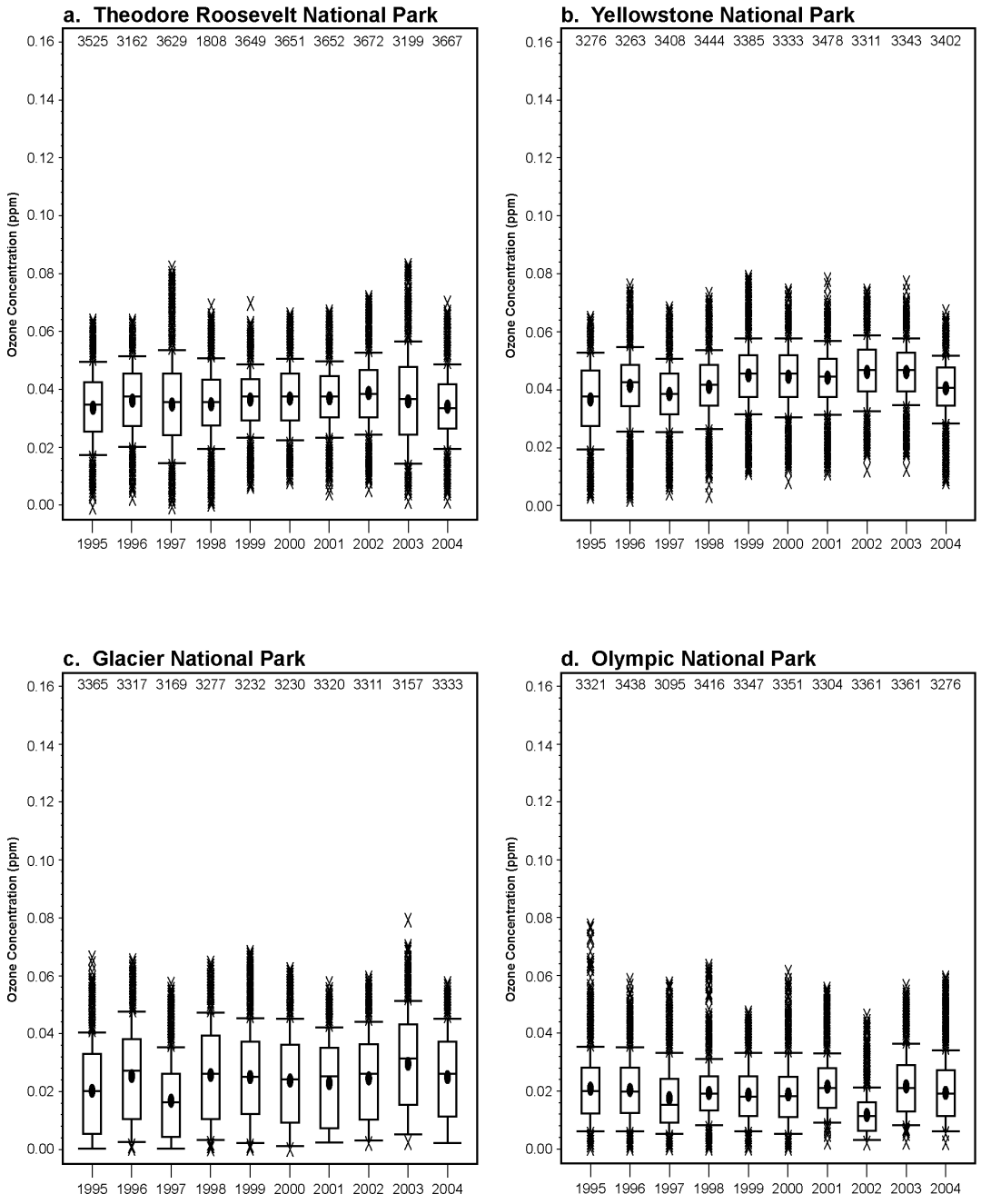


Figure 3-5a-d. Daily 8-h maximum O_3 concentrations observed at selected national park sites. The whiskers on the box plot represent the 10th and 90th percentile concentrations. The "X"s above and below the whiskers are the values that fall below and above the 10th and 90th percentile concentrations. The dots inside the box represent the mean. The number of observations is shown above each box plot.

Source: Fitz-Simons et al. (2005).

3.3 SPATIAL VARIABILITY OF OZONE IN URBAN AREAS

The spatial variability in O_3 concentrations in 24 MSAs across the United States was examined. These MSAs were selected to provide (1) information helpful for risk assessments, (2) a general overview of the spatial variability of O_3 in different regions of the country, and (3) insight into the spatial distribution of O_3 in cities where health outcome studies have been conducted. Statistical analyses of the human health effects of airborne pollutants based on aggregate population time-series data have often relied on ambient concentrations of pollutants measured at one or more central sites in a given metropolitan area. In the particular case of ground-level O_3 pollution, central-site monitoring has been justified as a regional measure of exposure mainly on the grounds that correlations between concentrations at neighboring sites measured over time are usually high. In MSAs with multiple monitoring sites, averages over the monitors have often been used to characterize population exposures. However, substantial differences in concentrations between monitors can exist even though concentrations measured at the monitoring sites are highly correlated, thus leading to the potential for exposure misclassification error.

Metrics for characterizing spatial variability include the use of Pearson correlation coefficients (r), values of the 90th percentile absolute difference in O_3 concentrations (P_{90}), and coefficients of divergence (COD)[1]. These methods of analysis follow those used for characterizing $PM_{2.5}$ and $PM_{10-2.5}$ concentrations in Pinto et al. (2004) and in the latest edition of the Particulate Matter (PM) AQCD (U.S. Environmental Agency, 2004a). However, the calculations were performed on an hourly basis rather than on a 24-h basis. Data were aggregated over the local O_3 season as indicated in Table AX3-1. The length of the O_3 season varies across the country. In several southwestern states, it lasts all year long. In other areas, such as in New England, the mid-Atlantic states, the Midwest and the Northwest, it can be 6 months long, but typically it lasts from April through October.

[1] The COD is defined as follows:

$$COD_{jk} = \sqrt{\frac{1}{p}\sum_{i=1}^{p}\left(\frac{x_{ij} - x_{ik}}{x_{ij} + x_{ik}}\right)^2} \qquad \text{(AX3-1)}$$

where x_{ij} and x_{ik} represent the 24-h average $PM_{2.5}$ concentration for day i at site j and site k and p is the number of observations.

Table 3-1 shows the urban areas chosen, the range of 24-h average O_3 concentrations over the O_3 season, the range of intersite correlation coefficients, the range of P_{90} differences in O_3 concentrations between site pairs, and the range in COD values. A COD of zero implies that values in both data sets are identical, and a COD of one indicates that two data sets are completely different. In general, statistics were calculated for partial MSAs. This was done so as to obtain reasonable lower estimates of the spatial variability that is present, as opposed to examining the consolidated MSAs. However, this could not be readily done for Boston, MA and New York, NY, so statistics were calculated for those consolidated MSAs. More detailed calculations for a subset of nine MSAs are given in Figures AX3-28 through AX3-36 in Section AX3.3.

As can be seen from Table 3-1, no clearly discernible regional differences were found in the ranges of parameters analyzed. Additional urban areas would need to be examined to discern broadscale patterns. The data indicate considerable variability in the concentration fields. Mean O_3 concentrations vary within individual urban areas by factors of 1.4 to 4. Intersite correlation coefficients show mixed patterns (i.e., in some urban areas all pairs of sites are moderately to highly correlated, while other areas show a larger range of correlations). As may be expected, those areas showing a smaller range of seasonal mean concentrations also show a smaller range of intersite correlation coefficients. However, there are a number of cases where sites in an urban area may be moderately to highly correlated, but show substantial differences in absolute concentrations. In many cases, P_{90} values can equal or exceed seasonal mean O_3 concentrations.

It is instructive to compare the metrics for spatial variability shown in Table 3-1 to those calculated for $PM_{2.5}$ and $PM_{10-2.5}$ in the PM AQCD (U.S. Environmental Agency, 2004). The values for concentrations and concentration differences are unique to the individual species, but the intersite correlation coefficients and the COD values can be directly compared. In general, the variability in O_3 concentrations is larger than for $PM_{2.5}$ concentrations and comparable to that obtained for $PM_{10-2.5}$. Intersite correlation coefficients in some areas (e.g., Philadelphia, PA; Atlanta, GA; Portland, OR) can be very similar for both $PM_{2.5}$ and for O_3. However, there is much greater variability in the concentration fields of O_3 as evidenced by the much higher COD values. Indeed, COD values are higher for O_3 than for $PM_{2.5}$ in each of the urban areas examined. In all of the urban areas examined for O_3, some site pairs are always very highly

Table 3-1. Summary Statistics for the Spatial Variability of O_3 (in ppm) in Selected Urban Areas in the United States

Urban Area	Number of Sites	Minimum Mean Conc.	Maximum Mean Conc.	Minimum Corr. Coeff.	Maximum Corr. Coeff.	Minimum P_{90} [a]	Maximum P_{90}	Minimum COD [b]	Maximum COD
Boston, MA	18	0.021	0.033	0.46	0.93	0.012	0.041	0.17	0.45
New York, NY	29	0.015	0.041	0.45	0.96	0.0080	0.044	0.17	0.55
Philadelphia, PA	12	0.020	0.041	0.79	0.95	0.011	0.036	0.23	0.46
Washington, DC	20	0.022	0.041	0.72	0.97	0.010	0.032	0.17	0.45
Charlotte, NC	8	0.031	0.043	0.48	0.95	0.012	0.038	0.17	0.32
Atlanta, GA	12	0.023	0.047	0.63	0.94	0.013	0.045	0.24	0.55
Tampa, FL	9	0.024	0.035	0.74	0.94	0.011	0.025	0.20	0.35
Detroit, MI	7	0.022	0.037	0.74	0.96	0.0090	0.027	0.19	0.36
Chicago, IL	24	0.015	0.039	0.38	0.96	0.0080	0.043	0.16	0.50
Milwaukee, WI	9	0.027	0.038	0.73	0.96	0.0090	0.025	0.18	0.33
St. Louis, MO	17	0.022	0.038	0.78	0.96	0.0090	0.031	0.15	0.41
Baton Rouge, LA	7	0.018	0.031	0.81	0.95	0.0090	0.029	0.23	0.41
Dallas, TX	10	0.028	0.043	0.67	0.95	0.011	0.033	0.16	0.36
Houston, TX	13	0.016	0.036	0.73	0.96	0.0090	0.027	0.20	0.38
Denver, CO	8	0.022	0.044	0.60	0.92	0.013	0.044	0.16	0.46
El Paso, TX	4	0.022	0.032	0.81	0.94	0.012	0.023	0.24	0.31
Salt Lake City, UT	8	0.029	0.048	0.52	0.92	0.012	0.043	0.13	0.51
Phoenix, AZ	15	0.021	0.058	0.29	0.95	0.011	0.057	0.15	0.61
Seattle, WA	5	0.015	0.038	0.63	0.94	0.0080	0.024	0.16	0.46
Portland, OR	5	0.015	0.036	0.73	0.91	0.011	0.025	0.20	0.50
Fresno, CA	6	0.030	0.047	0.90	0.97	0.0090	0.027	0.17	0.40
Bakersfield, CA	8	0.028	0.047	0.23	0.96	0.013	0.052	0.20	0.58
Los Angeles, CA	14	0.010	0.042	0.42	0.95	0.010	0.053	0.22	0.59
Riverside, CA	18	0.018	0.054	0.38	0.95	0.013	0.057	0.15	0.64

[a] P_{90} = 90th percentile absolute difference in concentrations.
[b] COD = coefficient of divergence for different site pairs.

correlated with each other (i.e., r >0.9) as seen for $PM_{2.5}$. These sites also show less variability in concentration and are probably influenced most strongly by regional production mechanisms.

The above considerations indicate that caution should be observed in using data from the network of ambient O_3 monitors to approximate community-scale human exposures. A similar conclusion was reached for PM using data from the $PM_{2.5}$ FRM network, as indicated in Section 3.4 of the PM AQCD (U.S. Environmental Protection Agency, 2004a).

3.3.2 Small-scale Horizontal and Spatial Variability in Ozone Concentrations

Ozone concentrations near roadways

Apart from the larger scale variability in surface O_3 concentrations, there is also significant variability on the micro-scale (< a few hundred meters), especially near roadways and other sources of emissions that react with O_3. These sources are not confined to urban areas. Sources of emissions that react with O_3 such as highways and power plants are also found in rural areas. Johnson (1995) described the results of studies examining O_3 upwind and downwind of roadways in Cincinnati, OH. In these studies, O_3 upwind of the roadway was about 50 ppb and these values were not found again until distances of about 100 m downwind. The O_3 profile varied inversely with that of NO, as might be expected. For peak NO concentrations of 30 ppb immediately downwind of the road, the O_3 mixing ratio was about 36 ppb, or about 70% of the upwind value. The magnitude of the downwind depletion of O_3 depends on the emissions of NO, the rate of mixing of NO from the roadway and ambient temperature and so depletions of O_3 downwind of roadways are expected, but with variable magnitude. Guidance for the placement of O_3 monitors (U.S. Environmental Protection Agency, 1998) states a separation distance that depends on traffic counts. For example, a minimum separation distance of 100 m from a road with 70,000 vehicles per day (about 3,000 vehicles per hour) is recommended for siting an O_3 monitor to avoid interference that would mean a site is no longer representative of the surrounding area. An average rate of about 3,000 vehicles per hour passing by a monitoring site implies a road with rather heavy traffic. As noted in Section AX3.3.1 for the Lakewood, CA monitoring, O_3 levels are lower at sites located near traffic than those located some distance away and the scavenging of O_3 by emissions of NO from roadways is a major source of spatial variability in O_3 concentrations. It should also be noted that scavenging of O_3 by NO near roadways was more pronounced before the implementation of stringent NOx emissions controls.

Vertical Variations in Ozone Concentrations

In addition to horizontal variability in O_3 concentrations, there are also variations in the vertical profile of O_3 in the lowest layers of the atmosphere to consider. The planetary boundary layer consists of an outer and an inner portion. The inner part of the planetary boundary layer extends from the surface to about one-tenth the height of the planetary boundary layer. Winds and transported properties, such as O_3, are especially susceptible to interactions with obstacles, such as buildings and trees in the inner boundary layer (atmospheric surface layer) (e.g., Garratt, 1992). Inlets to ambient monitors (typically at heights of 3 to 5 meters) are located in, and human and vegetation exposures occur in this part of the boundary layer.

Photochemical production and destruction of O_3 occur throughout the planetary boundary layer. However, O_3 is also destroyed on the surfaces of buildings, vegetation, etc. On most surfaces, O_3 is destroyed with every collision. In addition, O_3 is scavenged by NO emitted by motor vehicles and soils. These losses imply that the vertical gradient of O_3 should always be directed downward. The magnitude of the gradient is determined by the intensity of turbulent mixing in the surface layer.

Most work characterizing the vertical profile of O_3 near the surface has been performed in nonurban areas with the aim of calculating fluxes of O_3 and other pollutants through forest canopies and to crops and short vegetation, etc. Corresponding data are sparse for urban areas. However, monitoring sites are often set up in open areas such as parks and playgrounds where surface characteristics may be more similar to those in rural areas than to those in the surrounding urban area. The vertical profiles of O_3 measured over low vegetation are shown in Figure 3-6. These measurements were obtained as part of a field campaign to measure the fluxes of several gas and aerosol phase pollutants using the gradient-flux technique (Horváth et al., 1995). The labels stable and unstable in the figure refer to atmospheric stability conditions and average represents the overall average. Ozone concentrations were normalized to their values at 4 m height. As can be seen from the figure, there was a decrease of about 20% in going from a height of 4 m down to 0.5 m above the surface during stable conditions, but O_3 decreased by only about 7% during unstable conditions. The average decrease was about 10% for all measurements. As might be expected, O_3 concentrations at all heights were very highly correlated with one another. Of course, these values represent averages and there is scatter about them. Under strongly stable conditions, they fall off toward the surface. However, these

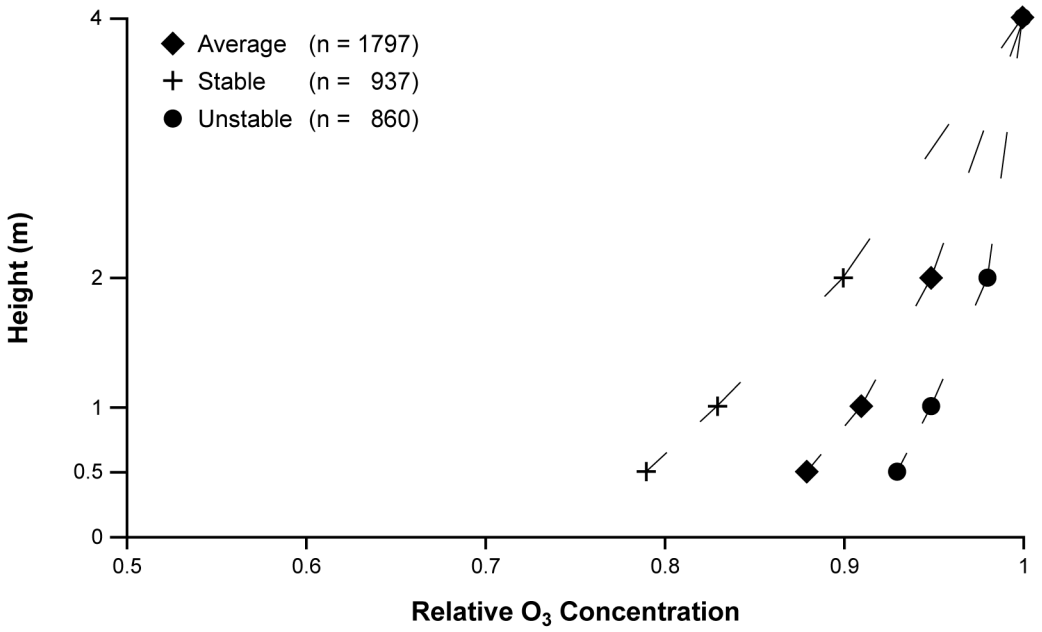


Figure 3-6. Vertical profile of O_3 obtained over low vegetation. Values shown are relative to concentrations at 4 m above the surface. Ozone concentrations for unstable and unstable conditions were 41.3 and 24.1 ppb, and average O_3 concentration weighted by stability class was 33.1 ppb at 4 m.

Source. Horváth et al. (1995).

conditions tend to occur mainly during night and the stability regime during the day in urban areas tends more toward instability because of the urban heat island effect. Figure 3-7 shows the vertical profile of O_3 measured in a spruce forest by the same group (Horváth et al., 2003). The fall off of O_3 in this case is due to uptake by trees, reaction with ambient NO and with NO emitted by the soil in the forest, and reaction with hydrocarbons emitted by the trees in addition to deposition on the surface.

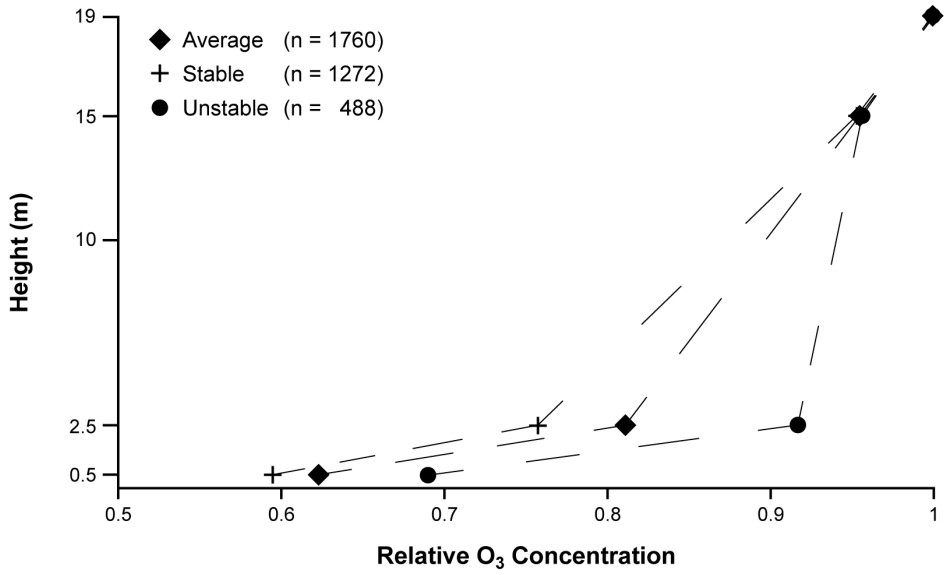


Figure 3-7. Vertical profile of O_3 obtained in a spruce forest. Values shown are relative to concentrations at 19 m above the surface. Mean tree height is 14.5 m. Ozone concentrations for unstable and unstable conditions were 36.7 and 33.8 ppb, and the average O_3 concentration weighted by stability class was 34.6 ppb at 19 m.

Source: Horváth et al. (2003).

3.4 DIURNAL AND SEASONAL VARIABILITY OF OZONE

Diurnal Variability

Diurnal variations in O_3 at a given location are controlled by a number of factors such as the relative importance of transport versus local photochemical production and loss rates, the timing for entrainment of air from the nocturnal residual boundary layer and the diurnal variability in mixing layer height.

Diurnal Patterns in the Nationwide Data Set

Composite urban, diurnal variations in hourly averaged O_3 for April through October 2000 to 2004 are shown in Figure 3-8. As can be seen from Figure 3-8, daily 1-h maxima tend to occur in mid-afternoon and daily 1-h minima tend to occur during the early morning. However, there is also considerable spread in these times. Therefore, some caution must be exercised in

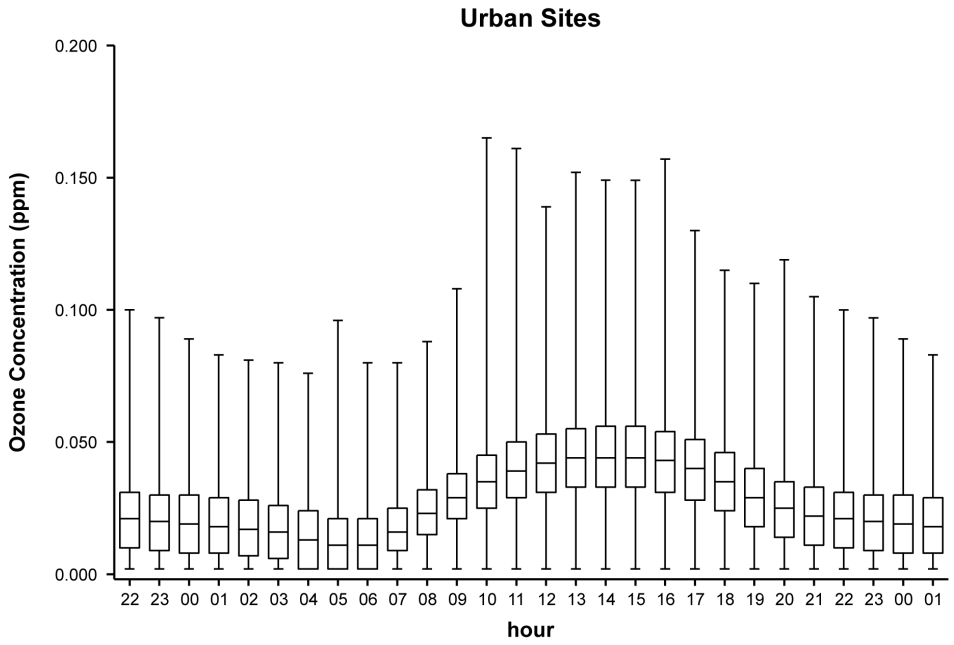


Figure 3-8. Composite, nationwide diurnal variability in hourly averaged O_3 in urban areas. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima.

Source: Fitz-Simons et al. (2005).

extrapolating results from one city to another and when attempting to judge the time of day when the daily 1-h maximum occurs.

Corresponding data for 8 hour average O_3 data are shown in Figure 3-9. As can be seen from Figure 3-9, daily maximum eight hour O_3 concentrations tend to occur from about 10 a.m. to about 6 p.m. As can be seen from Figure 3-9, they can also occur at slightly different times and the variation in the 8-h averages is smoother than for the 1-h averages. The minima in the 8 h averages tend to occur starting at about midnight.

Diurnal Patterns in EPA's 12 Cities

The diurnal variability of hourly averaged O_3 in the twelve urban areas considered for inclusion in EPA's human health exposure assessment risk assessment for the current review is illustrated in Figures 3-10a-l for April to October. Daily maximum 1-h concentrations tend to

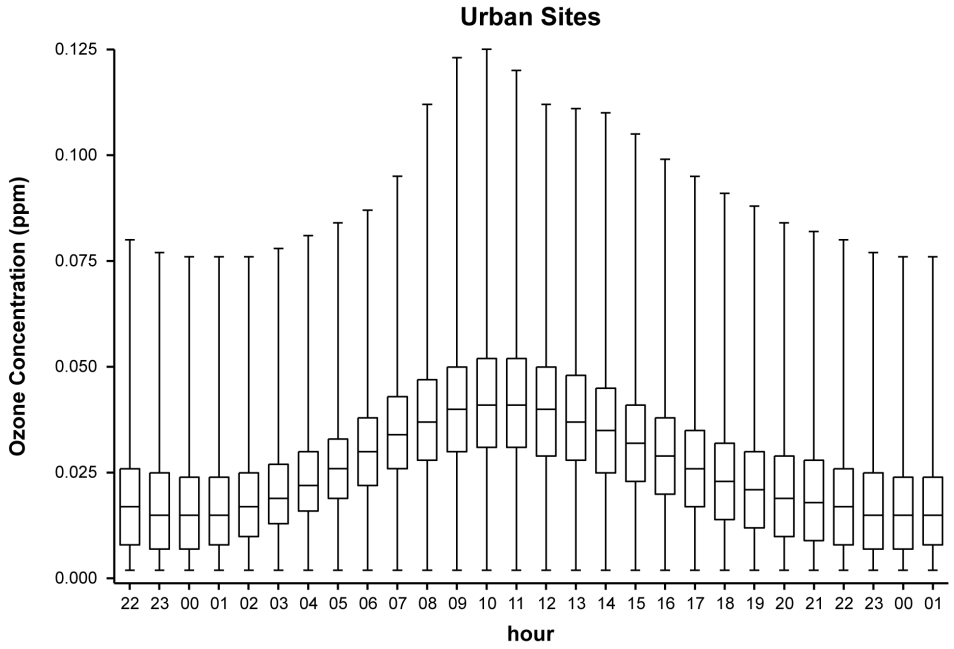


Figure 3-9. Composite, nationwide diurnal variability in 8 hour average O_3 in urban areas. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

occur in mid-afternoon. However, as can be seen from the figures, the diurnal patterns vary from city to city, with high values (≥0.100 ppm) also occurring either late in the evening as in Boston, past midnight as in Los Angeles and Sacramento, or midmorning as in Houston. Typically, high values such as these are found during the daylight hours in mid to late afternoon. The reasons for the behavior of O_3 during the night at the above-mentioned locations are not clear. Measurement issues may be involved or there may be physical causes such as transport phenomena, as discussed in Chapter 2. As discussed in Chapter 2, and in greater detail in Section AX2.3.3, nocturnal low level jets are capable of producing secondary O_3 maxima at night.

The diurnal variability of O_3 averaged over 8 hours in the same twelve urban areas is shown in Figures 3-11a-l. The diurnal patterns of O_3 are broadly similar between 1-h averages and 8-h averages. A typical pattern shows the 8-h daily maximum occurring from about 10 a.m.

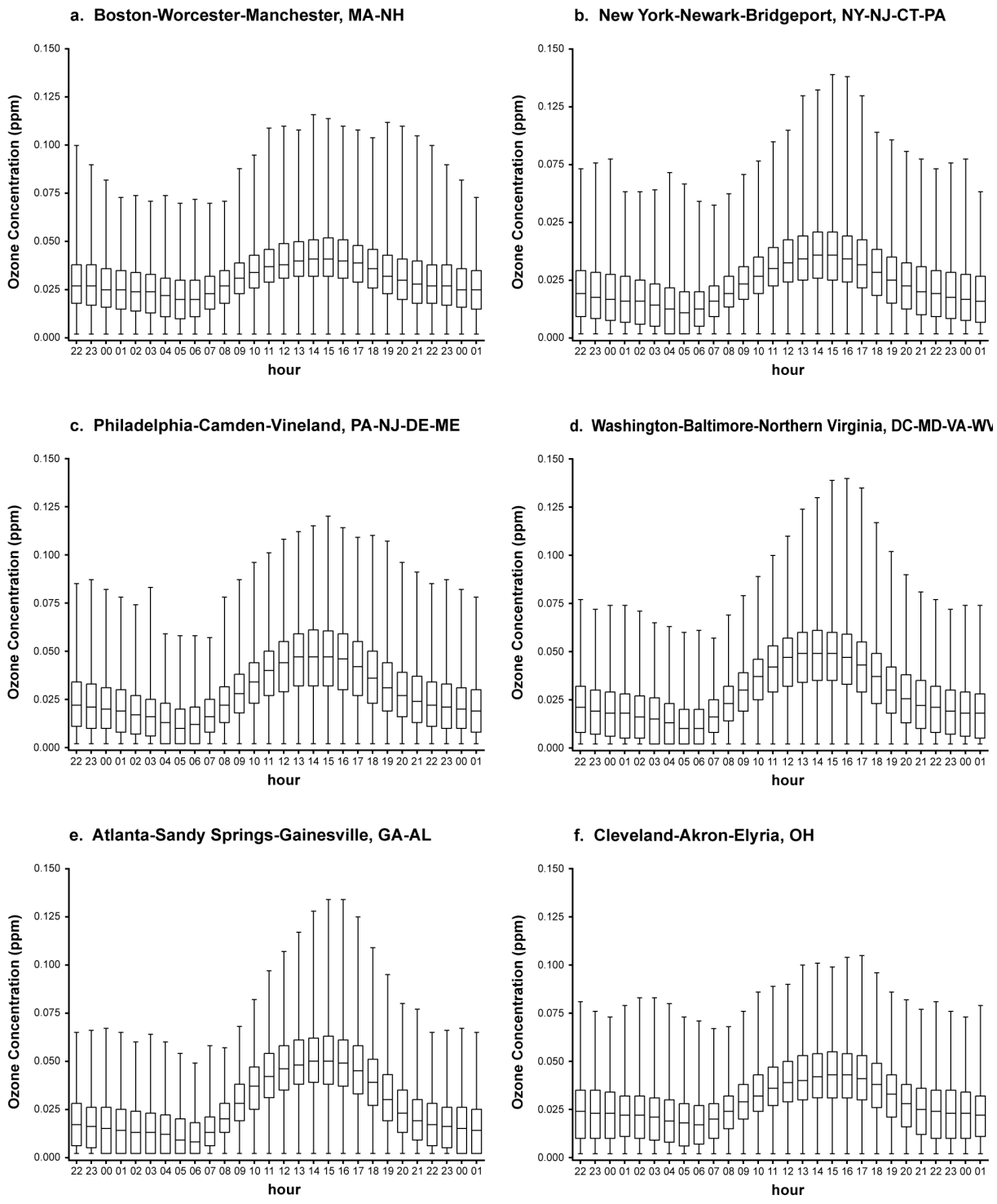


Figure 3-10a-f. Diurnal variability in hourly averaged O_3 in selected urban areas. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima.

Source: Fitz-Simons et al. (2005).

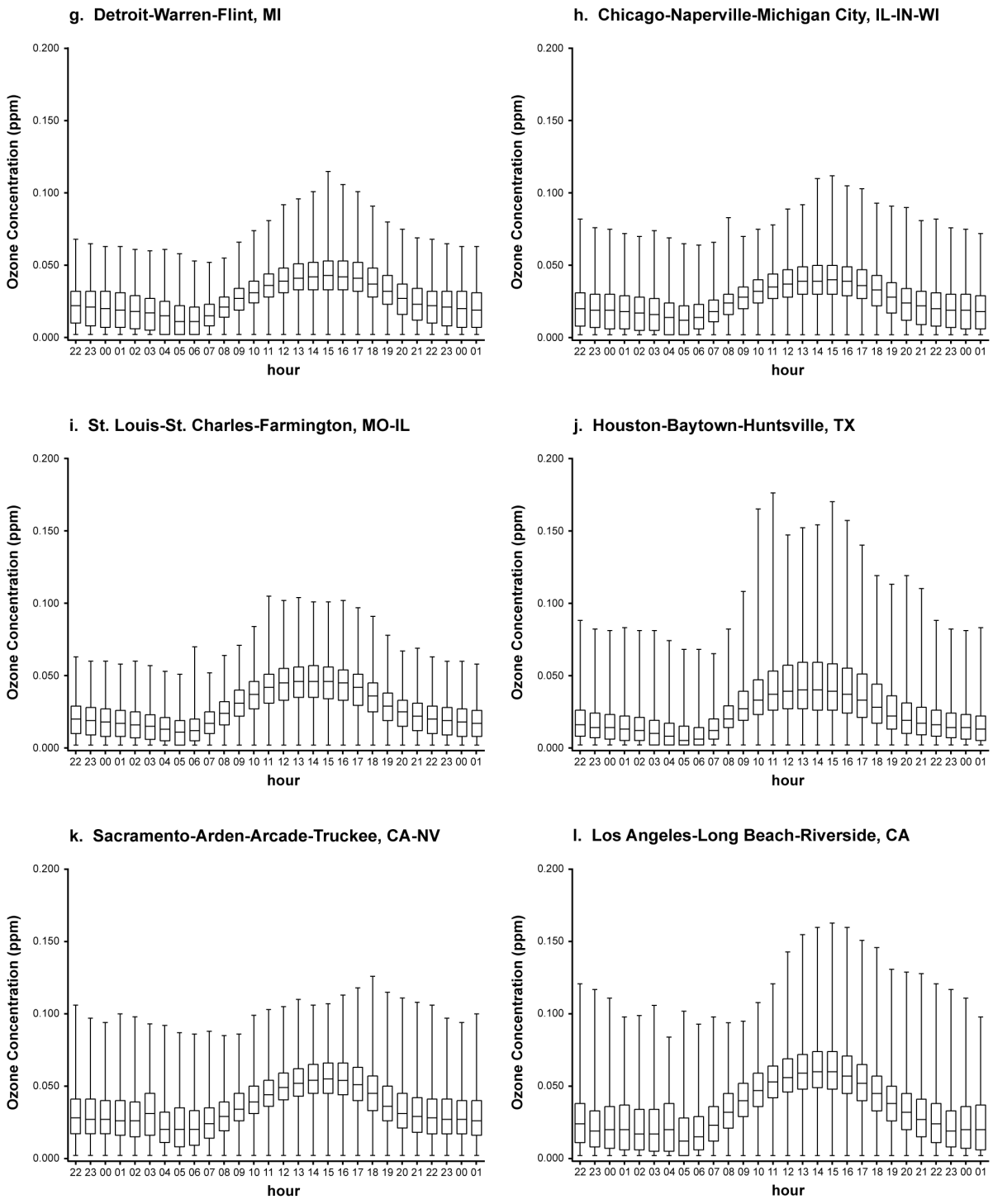


Figure 3-10g-l. Diurnal variability in hourly averaged O_3 in selected urban areas. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima.

Source: Fitz-Simons et al. (2005).

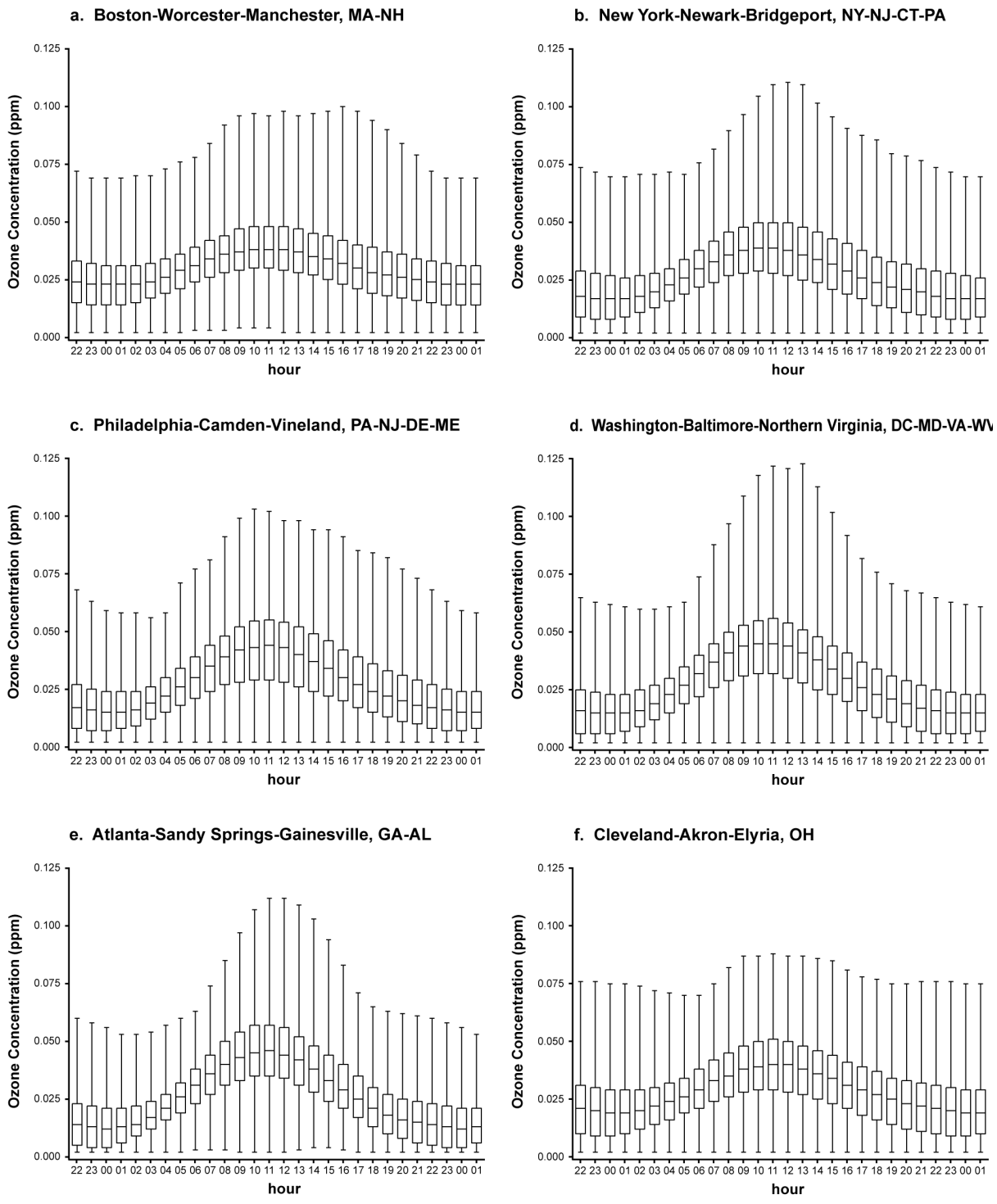


Figure 3-11a-f. Diurnal variability in 8 hour averaged O_3 in selected urban areas. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

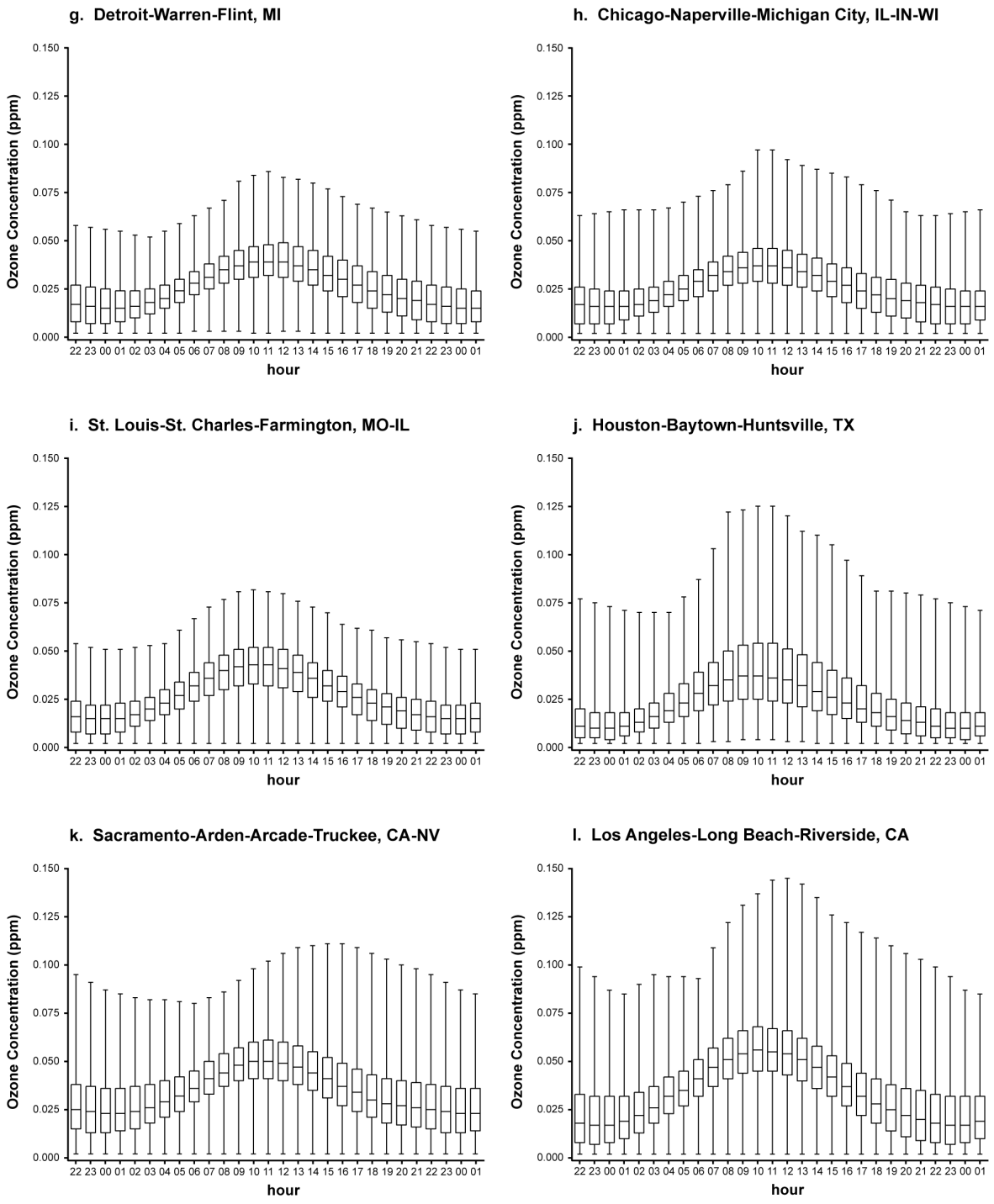


Figure 3-11g-l. Diurnal variability in 8 hour averaged O_3 in selected urban areas. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

to about 6 p.m., with some deviations from these times. For example, as shown in Figures 3-11a for Boston and 3-11k for Sacramento, the highest 8-h daily maximum values occur starting in mid-afternoon and extending into late evening. These results suggest that transport processes are playing the dominant role in determining the timing of the highest daily maxima in these areas.

On days with high 1-h daily maximum concentrations (e.g., $\geq$0.12 ppm) the maxima tend to occur in a smaller time window centered in the middle of the afternoon, compared to days in which the maximum is lower. For example, on the high O_3 days the 1-h maximum occurs from about 11 a.m. to about 6 p.m. However, on days in which the 1-h daily maximum is $\leq$0.080 ppm, the daily maximum can occur at any time during the day or night, with only a 50% probability that it occurs between 1 and 3 p.m., in each of the 12 cities. (The time of day when the daily maximum 1-h O_3 concentration occurs is illustrated for four of the cities in Figures AX3-45a-d.). Photochemical reactions in combination with diurnal emissions patterns are expected to produce mid-afternoon peaks in urban areas. These results suggest that transport from outside the urban airshed plays the major role for determining the timing of the daily maxima for low peak O_3 levels. This pattern is typical for the Los Angeles-Long Beach-Riverside, CA area even on high O_3 days.

The same general patterns emerge for the timing of the 1-h daily maximum O_3 concentration as are found for the daily maximum 8-h average O_3 concentration. As mentioned above, the daily maximum 8-h O_3 concentrations are generally found between the hours of 10 a.m. and 6 p.m. However, there are significant fractions of the time when this is not the case, e.g., for high values in Houston, TX and Los Angeles, CA, or in general for lower values at any of the cities examined. (The time of day when the daily maximum 8-h average O_3 concentrations occurs is shown for four cities in Figures AX3-46a-d.). Although the 8-h average O_3 concentration is highly correlated with the daily maximum 1-h average O_3 concentration, there are situations where the daily maximum 8-h average O_3 concentration might be driven by very high values in the daily maximum 1-h average O_3 concentration as illustrated in Figure 3-10j for Houston, TX. In cases such as these, the predicted 8-h average may overestimate the short-term O_3 concentration later in the day.

The patterns of diurnal variability for both 1-h and 8-h averages have remained quite stable over the 15-year period from 1990 to 2004, with times of occurrence of the daily maxima varying by no more than an hour from year to year in each of the 12 cities.

Weekday/Weekend Differences

Differences in the diurnal behavior of O_3 have been observed in a number of cities (e.g., Heuss et al., 2003). Figures 3-12a-h show the contrast in the patterns of hourly averaged O_3 in the greater Philadelphia, Atlanta, Houston and Los Angeles areas from weekdays to weekends. Daily maximum concentrations occur basically at the same time on either weekdays or weekends. Differences are apparent in the hourly concentrations, especially in the extreme values. Weekday/weekend differences in 8-h average O_3 concentrations are shown in Figures 3-13a-h. As can be seen from a comparison of the weekend versus weekday patterns, there is a tendency for the lowest values to be higher on weekends than on weekdays. Lower traffic volumes, in particular diesel truck traffic, lead to less NO emissions and titration of O_3 on weekends. The spike in values shown for Houston in midmorning shown in Figure 3-12f resulted from the release of highly reactive hydrocarbons from the petrochemical industry (which could occur on any day of the week). Otherwise, the maximum O_3 concentrations could be seen to occur on the weekdays as they do in Philadelphia and Atlanta, in contrast to Los Angeles. Indeed, the diurnal pattern in Houston is similar to that observed in Atlanta on weekdays, indicating some overall similarity in the sources of O_3.

Spatial Variability in Diurnal Patterns in Urban Areas

Daily maxima in either the 1-h or 8-h averages do not necessarily occur at the same time of day at each site in the 12 cities, and the diurnal pattern observed at individual sites can vary from the composites shown in Figures 3-8 and 3-9. Differences between sites are not only related to distance; they also depend on the presence of nearby sources, such as highways. For example, in the Los Angeles basin, daily 1-h maxima are reached in the late afternoon in Riverside relatively close to sites in which the maximum is reached much earlier.

The general pattern that emerges from the site-to-site variability within the urban areas examined is that peaks in 1-h average concentrations are higher and tend to occur later at downwind sites than in the urban cores. To the extent that monitoring sites are either near to or remote from sources of precursors in urban/suburban areas, the behavior of O_3 will follow these basic patterns. Similar relations are found for the 8-h average O_3 concentrations. Differences in diurnal patterns between sites in urban cores and sites downwind of urban cores are illustrated

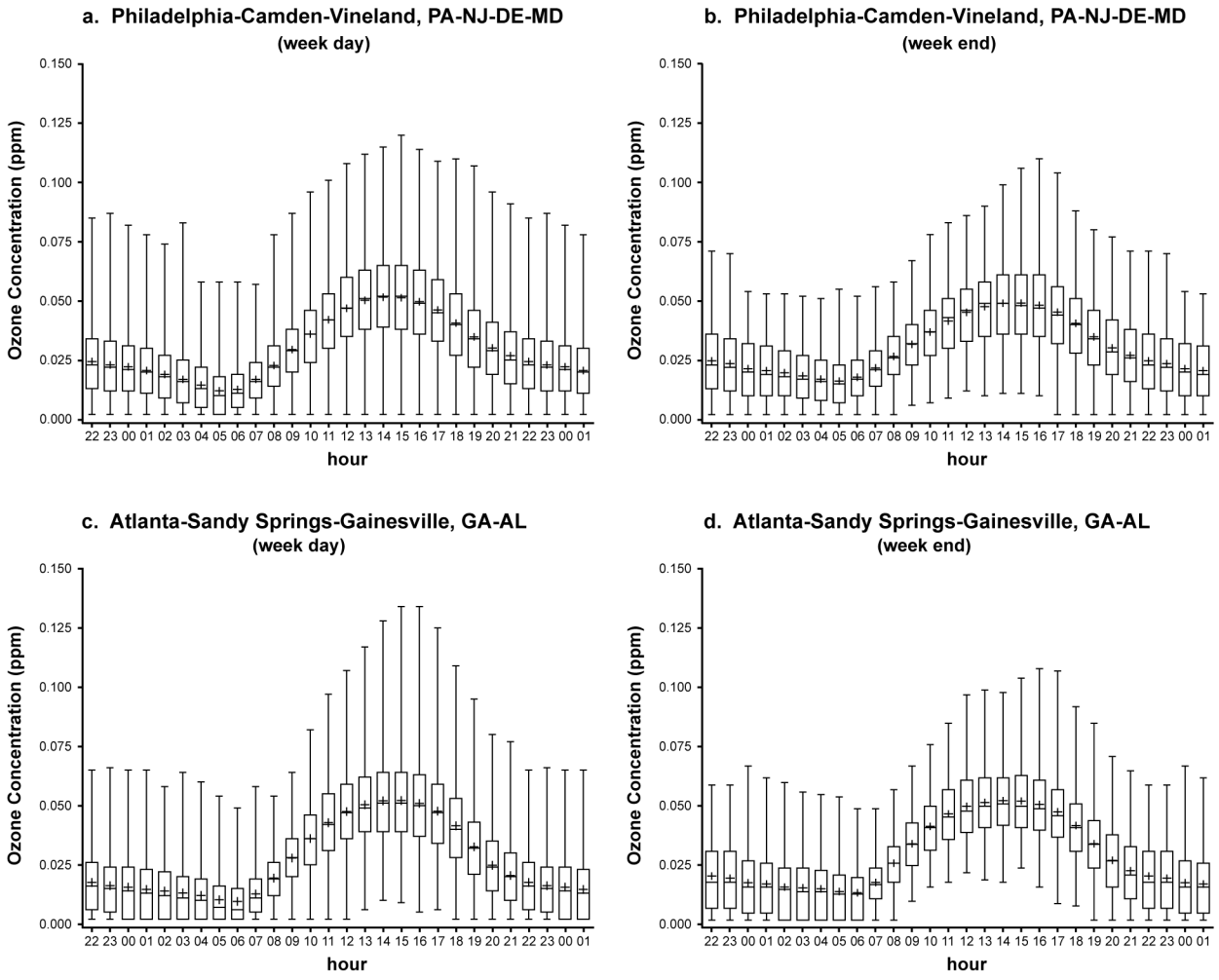


Figure 3-12a-d. Diurnal variations in hourly averaged O_3 on weekdays and weekends in four cities. Values shown represent averages from May to September of 2004.

Source: Fitz-Simons et al. (2005).

in Figures AX3-49a-b to AX3-51a-b for 1-h average O_3 and in Figures AX3-52a-b to AX3-54a-b for Detroit, MI, St. Louis, MO, and Riverside, CA areas.

Seasonal Variability

It should not be assumed that highest O_3 levels are confined to the summer. Highest average O_3 concentrations generally occur at background monitoring sites at midlatitudes in the

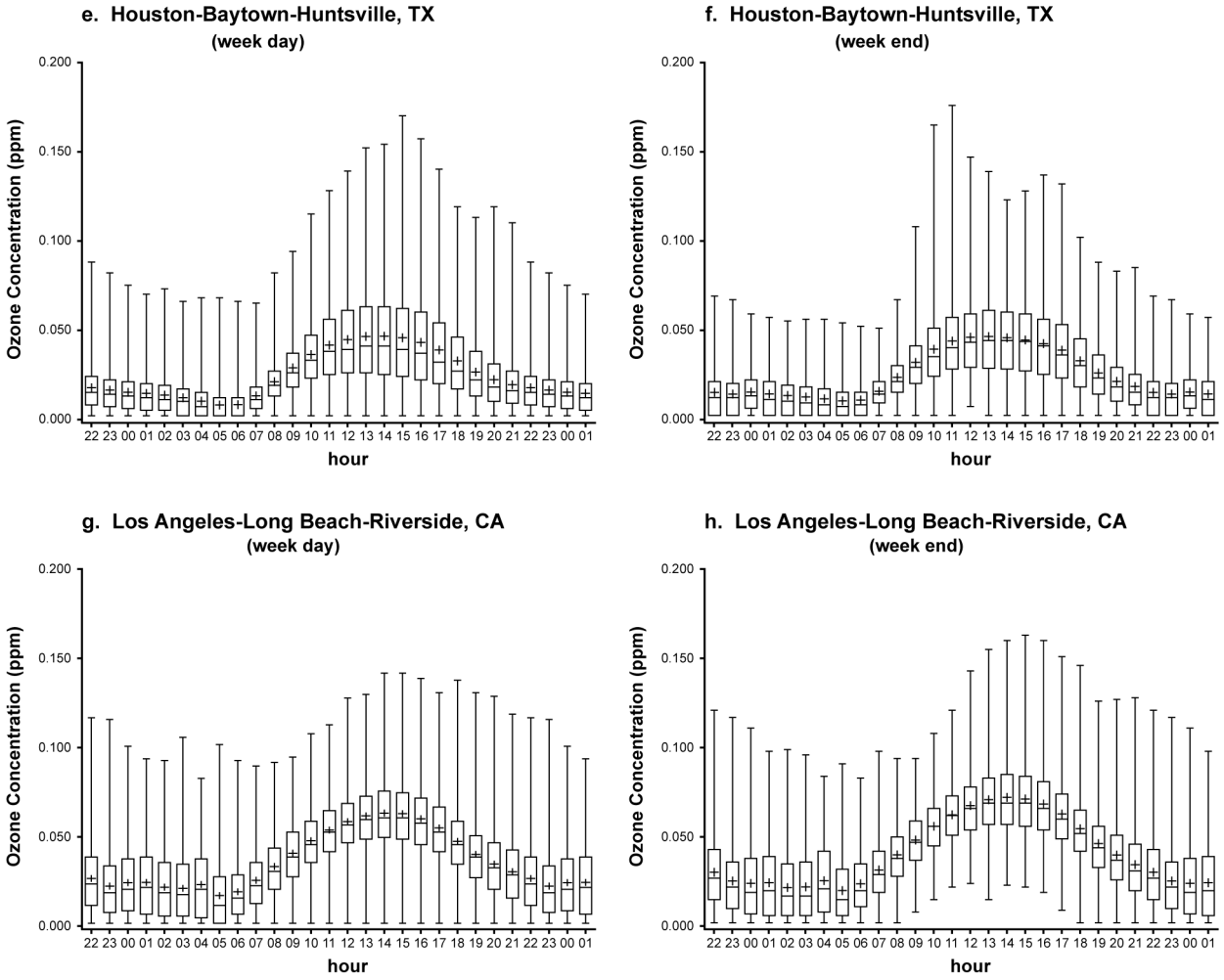


Figure 3-12e-h. Diurnal variations in hourly averaged O_3 on weekdays and weekends in four cities. Values shown represent averages from May to September of 2004.

Source: Fitz-Simons et al. (2005).

Northern Hemisphere during late winter and spring versus summer as for urban sites or for nonurban sites heavily affected by regional pollution sources.

High O_3 values are also found at some of the 12 cities outside of summer. The seasonal behavior of O_3 varies across the 12 cities. In most northern cities, the extreme values of the daily maximum 8-h average O_3 concentration are a little more than half of those during the warm season, the ratios of the medians are more similar as can be judged by comparison of

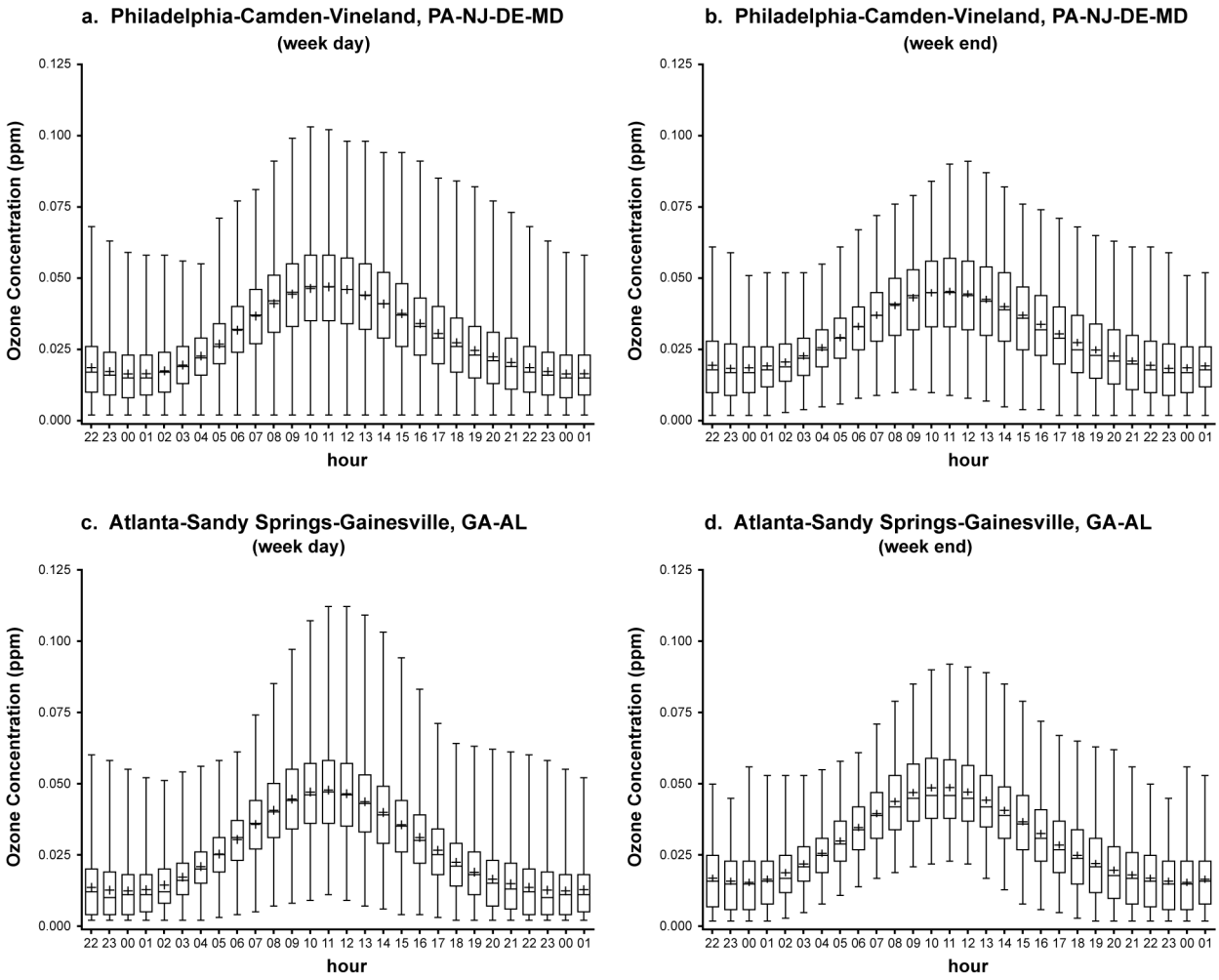


Figure 3-13a-d. Diurnal variations in 8-h average O_3 on weekdays and weekends in four cities. Values shown represent averages from May to September of 2004. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

Figures 3-11a-l with Figures 3-14a-l. Differences are even smaller for the southern cities. Indeed, some of the highest O_3 values are found in the Houston CSA outside of summer (Figure 3-14j).

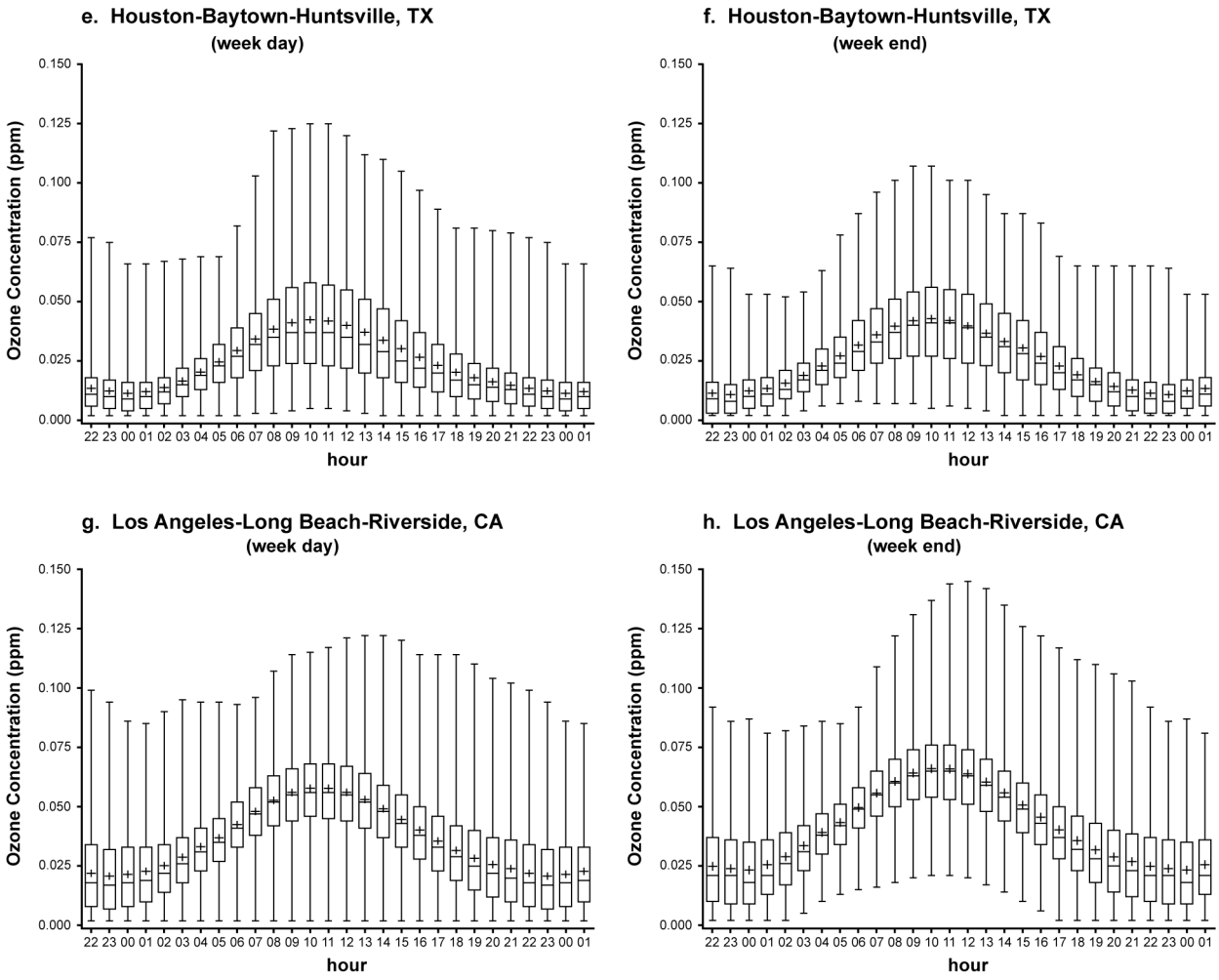


Figure 3-13e-h. Diurnal variations in 8-h average O_3 on weekdays and weekends in four cities. Values shown represent averages from May to September of 2004. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

Diurnal Patterns in Nonurban Areas

Composite diurnal patterns of O_3 are shown in Figure 3-15 for hourly averaged O_3, and in Figure 3-16 for 8 hour average O_3 at rural (CASTNET) sites. As can be seen from a comparison of Figures 3-15 and 3-16 with Figures 3-8 and 3-9, diurnal patterns of O_3 are smoother and shallower at the rural sites than at the urban sites. Maxima in hourly average O_3 also tend to occur in afternoon. However, highest concentrations observed during any particular hour at night at the CASTNET sites (~0.130 ppm) are substantially higher than observed in urban areas

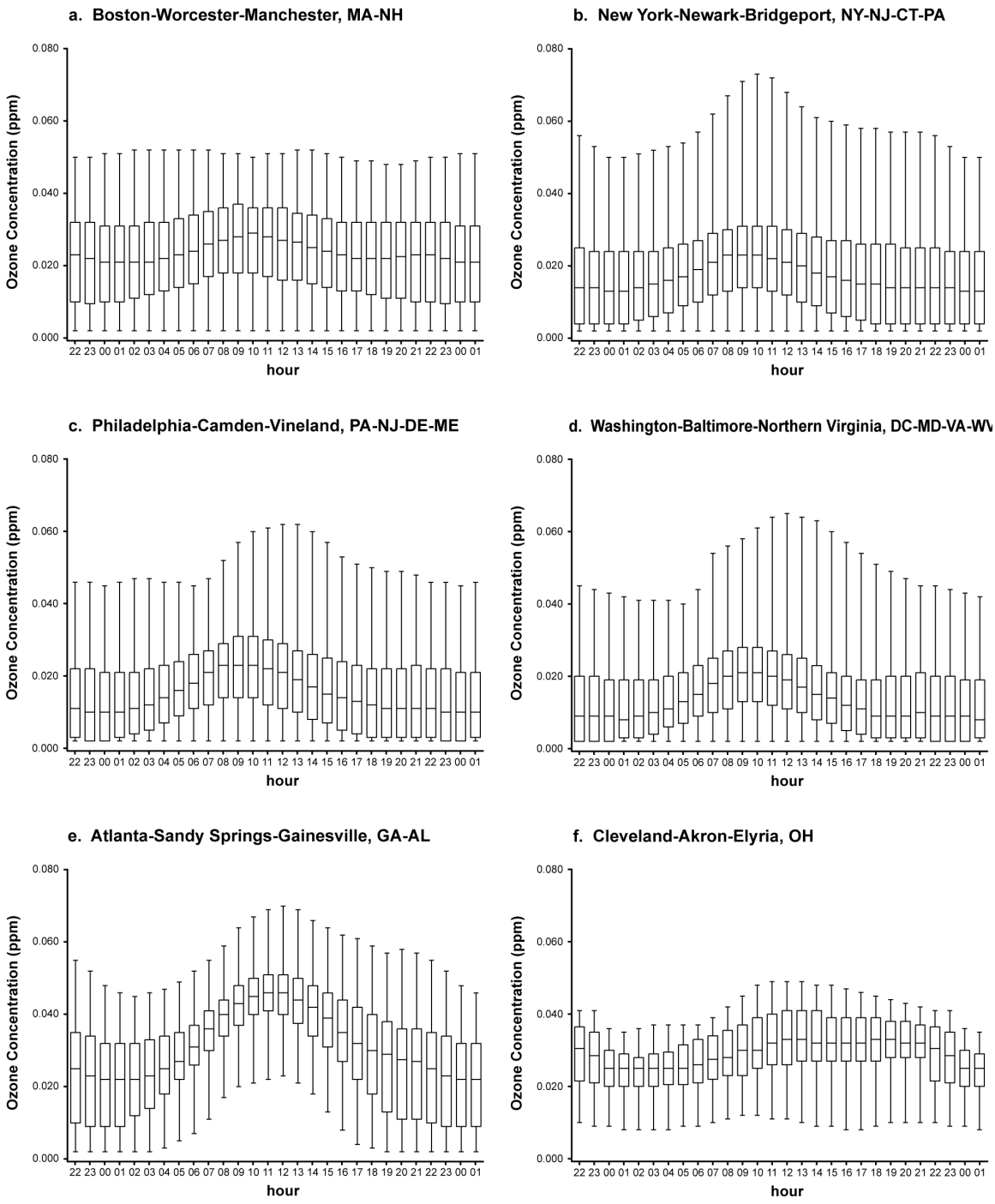


Figure 3-14a-f. Diurnal variability in 8 hour averaged O_3 in selected urban areas. Values shown are averages from November to March 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

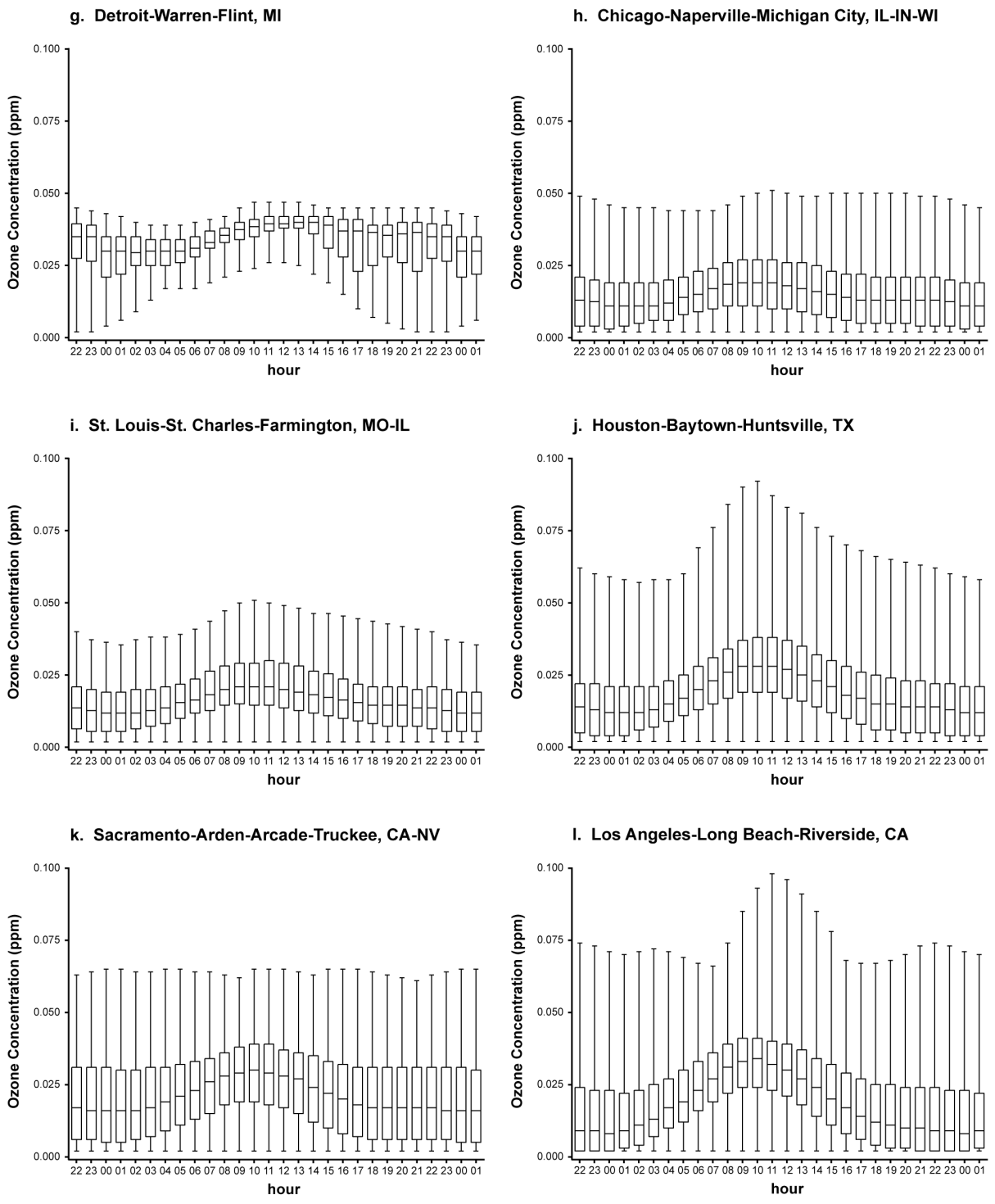


Figure 3-14g-l. Diurnal variability in 8 hour averaged O_3 in selected urban areas. Values shown are averages from November to March 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

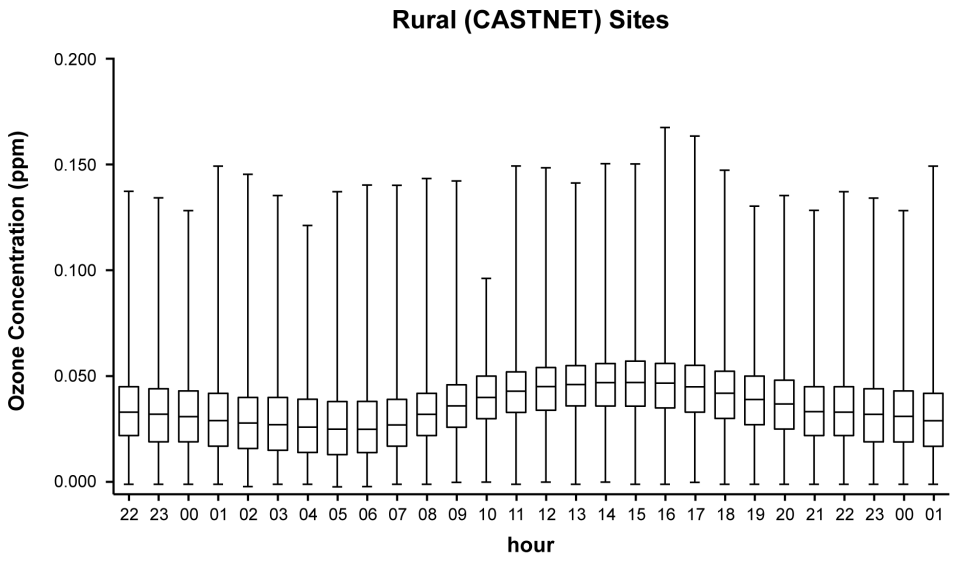


Figure 3-15. Composite diurnal variability in hourly O_3 concentrations observed at CASTNET sites. Values shown are averages from April to October 200 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima.

Source: Fitz-Simons et al. (2005).

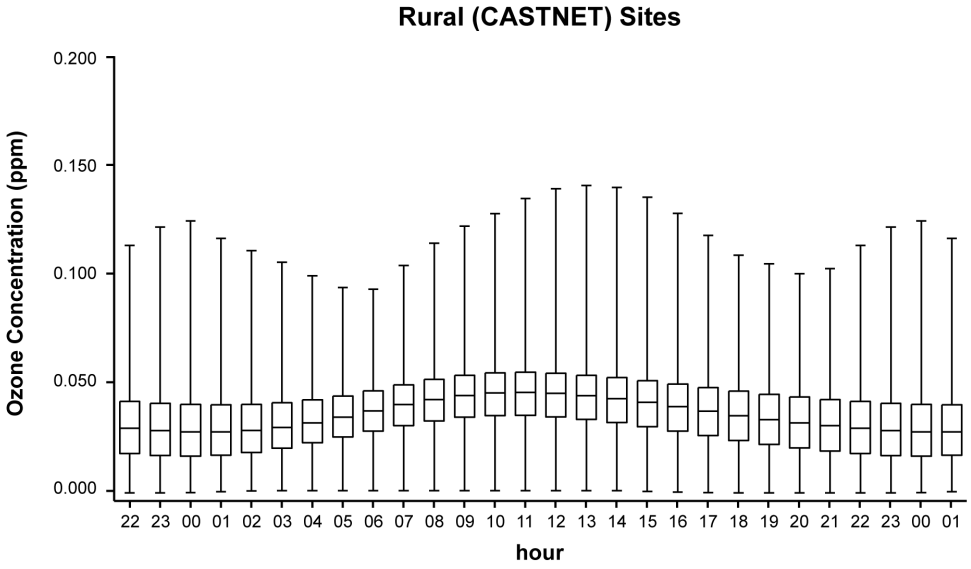


Figure 3-16. Composite diurnal variability in 8-h O_3 concentrations observed at CASTNET sites. Values shown are averages from April to October 2000 to 2004. Boxes define the interquartile range and the whiskers the minima and maxima. The hour refers to the start of the 8-h averaging period.

Source: Fitz-Simons et al. (2005).

(<0.100 ppm) and daily 1-h maxima at CASTNET sites have exceeded 0.150 ppm. The diurnal variations in 8-h average O_3 concentrations are also much smaller at the CASTNET sites than at the urban sites. Note also that the maxima in 8-h average O_3 concentrations are higher at the CASTNET sites than at the urban sites.

3.5 TRENDS IN OZONE CONCENTRATIONS

Year-to-year variability in the nationwide May to September, mean daily maximum 8-h O_3 concentrations are shown in Figure 3-17. The corresponding year-to-year variability in the 95th percentile concentrations is shown in Figure 3-18. Data flagged because of quality control issues were removed with concurrence by the local monitoring agency. Only days with data for 18 of 24 hours were kept, and a minimum of 115 of 153 days were required in each year. Missing years were filled in using simple linear interpolation, as done in EPA Trends reports. Year-to-year variability in the 95th percentile values of the daily maximum 8-h O_3 concentrations are shown in Figure 3-18. Sites considered in this analysis are shown in the map in Figure AX3-3. As was shown in Figures 3-1 and 3-2, most sites are located in the East. As can be seen from Figure 3-17, the highest O_3 concentrations have tended to decrease over the past 15 years, while there has been little change in O_3 concentrations near the center of the distribution. This is consistent with observations in Europe (Volz-Thomas et al., 2003). Mean O_3 concentrations were slightly lower in 2003 and 2004 than in earlier years. The summer of 2003 was slightly cooler than normal in the East (Levinson and Waple, 2004) and the summer of 2004 was much cooler than normal in the East (Levinson, 2005) accounting in part for the dip in O_3 during these two years. Observations of O_3 at a number of sites in the Northern Hemisphere likewise do not show convincing evidence of strong upward trends during the 1990s (Oltmans et al., 1998). There may even have been a slight increase in O_3 concentrations at the lower end of the distribution throughout the monitoring period. This would be consistent with data obtained in Europe, showing that O_3 minima increased during the 1990s. Reduced titration of O_3 by reaction with NO in response to reductions in NO_x emissions may be responsible in large measure for this finding. The concentration of O_X ($NO_2 + O_3$) shows little if any increase at all (Volz-Thomas et al., 2003).

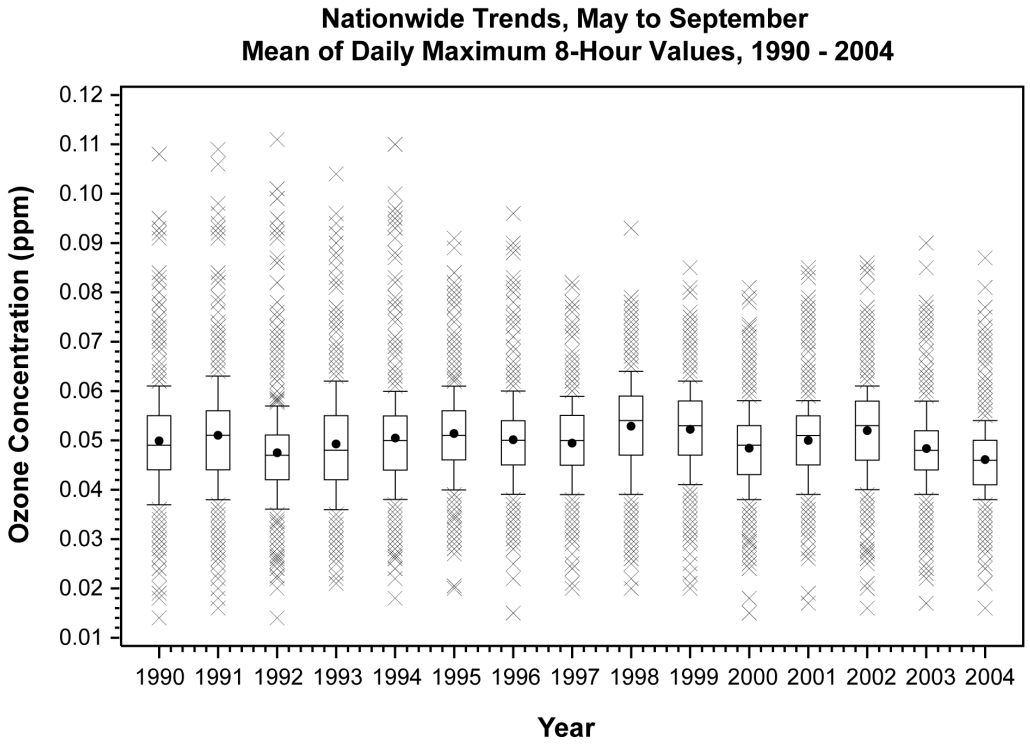


Figure 3-17. Year-to-year variability in nationwide mean daily maximum 8-h O_3 concentrations. The whiskers on the box plot represent the 10th and 90th percentile concentrations. The "X"s above and below the whiskers are the values that fall below and above the 10th and 90th percentile concentrations. The dots inside the box represent the mean, for the statistic, at all sites.

Source: Fitz-Simons et al. (2005)

Trends in compliance metrics such as the fourth highest daily maximum 8-h O_3 concentration can be found in the EPA Trends reports.

Figures 3-19a-h show year-to-year variability in mean daily 8-h O_3 concentrations observed at selected national park sites across the United States. Figures 3-20a-h show year-to-year variability in the 95th percentile value of daily maximum 8-h O_3 concentrations at the same sites shown in Figures 3-19a-h. The same criteria used for calculating values in Figures 3-17 and 3-18 were used for calculating the May to September seasonal averages for the national parks shown in Figures 3-19a-h and 3-20a-h. Sites at 22 national parks met these criteria, and data for all 22 sites are given in Appendix AX3 in Figures AX3-66a-v and AX3-67a-v.

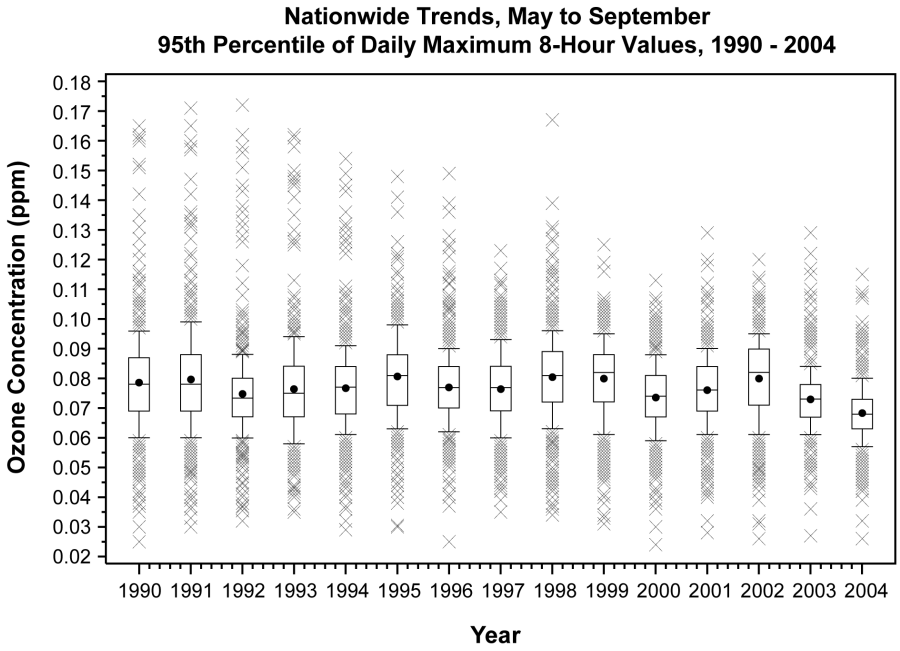


Figure 3-18. Year-to-year variability in nationwide 95th percentile value of the daily maximum 8-h O_3 concentrations. The whiskers on the box plot represent the 10th and 90th percentile values for the statistic. The "X"s above and below the whiskers are the values that fall below and above the 10th and 90th percentile values. The dots inside the box represent the mean, for the statistic, at all sites.

Source: Fitz-Simons et al. (2005).

However, several monitoring sites were moved during the period from 1990 to 2004. Sites were moved at Acadia NP in 1996, Joshua Tree NP in 1993, Mammoth Cave NP in 1996, Voyageurs NP in 1996, and Yellowstone NP in 1996. These moves often resulted in offsets in O_3 and so trends for these locations have not been calculated (cf., Section AX3.6, Table AX3-9). As noted in The Ozone Report—Measuring Progress through 2003 (U.S. Environmental Protection Agency, 2004b), O_3 trends in national parks in the South and the East are similar to nearby urban areas and reflect the regional nature of O_3 pollution. For example, O_3 trends in Charleston, SC and Charlotte, NC track those in nearby Cowpens NP and Cape Romaine NP in South Carolina; O_3 in Knoxville and Nashville, TN tracks O_3 in Great Smoky NP; O_3 in Philadelphia, PA and Baltimore, MD tracks Brigantine NP in New Jersey; and New York, NY and Hartford, CT track O_3 in Cape Cod NS. The situation is not as clear in the West, where national parks are affected

differently by pollution sources that are located at varying distances away (e.g., Lassen Volcanic National Park and Yosemite National Park, CA). However, data obtained at these sites still provide valuable information about the variability in regional background concentrations, especially since the West has not been broken down into regions as has been done by Lehman et al. (2004) for the East and shown in Figure 3-3. Comparison of Figures 3-19a-h and 3-20a-h (in conjunction with Table AX3-9) shows that O_3 concentrations near the center of the distribution do not necessarily track those at the upper end, as pointed out earlier for nationwide composite data.

Caution should be exercised in using trends calculated at national parks to infer contributions from distant sources either inside or outside of North America, because of the influence of regional pollution. For example, using a 15-year record of O_3 from Lassen Volcanic NP and data from two aircraft campaigns, and observations spanning 18 years from five U.S. west coast marine boundary layer sites, Jaffe et al. (2003) have estimated that the amount of O_3 in air arriving from the Eastern Pacific in spring has increased by approximately 10 ppb from the mid-1980s to the present. This positive trend might be due to increases of emissions of O_3 precursors in Asia. Positive trends in O_3 were found during all seasons. Although the Lassen Volcanic NP site is not close to any major emission sources or urban centers, maximum hourly average O_3 concentrations of >0.080 ppm (during April-May) and >0.100 ppm (during the summer) occur at Lassen Volcanic NP. Thus, although there is evidence that O_3 levels may be increasing at some rural locations, there is also evidence that O_3 levels at other locations have either not increased or have decreased over the same period.

3.6 RELATIONSHIPS BETWEEN OZONE AND OTHER SPECIES

Correlations between Ozone and other Species

In order to understand relationships among atmospheric species, an important distinction must be made between primary (directly emitted) species and secondary (photochemically produced) species. In general, it is likely that primary species will be highly correlated with other primary species, and that secondary species will be highly correlated with other secondary species. By contrast, primary species are less likely to be correlated with secondary species. Secondary reaction products tend to correlate with each other, but there is considerable variation.

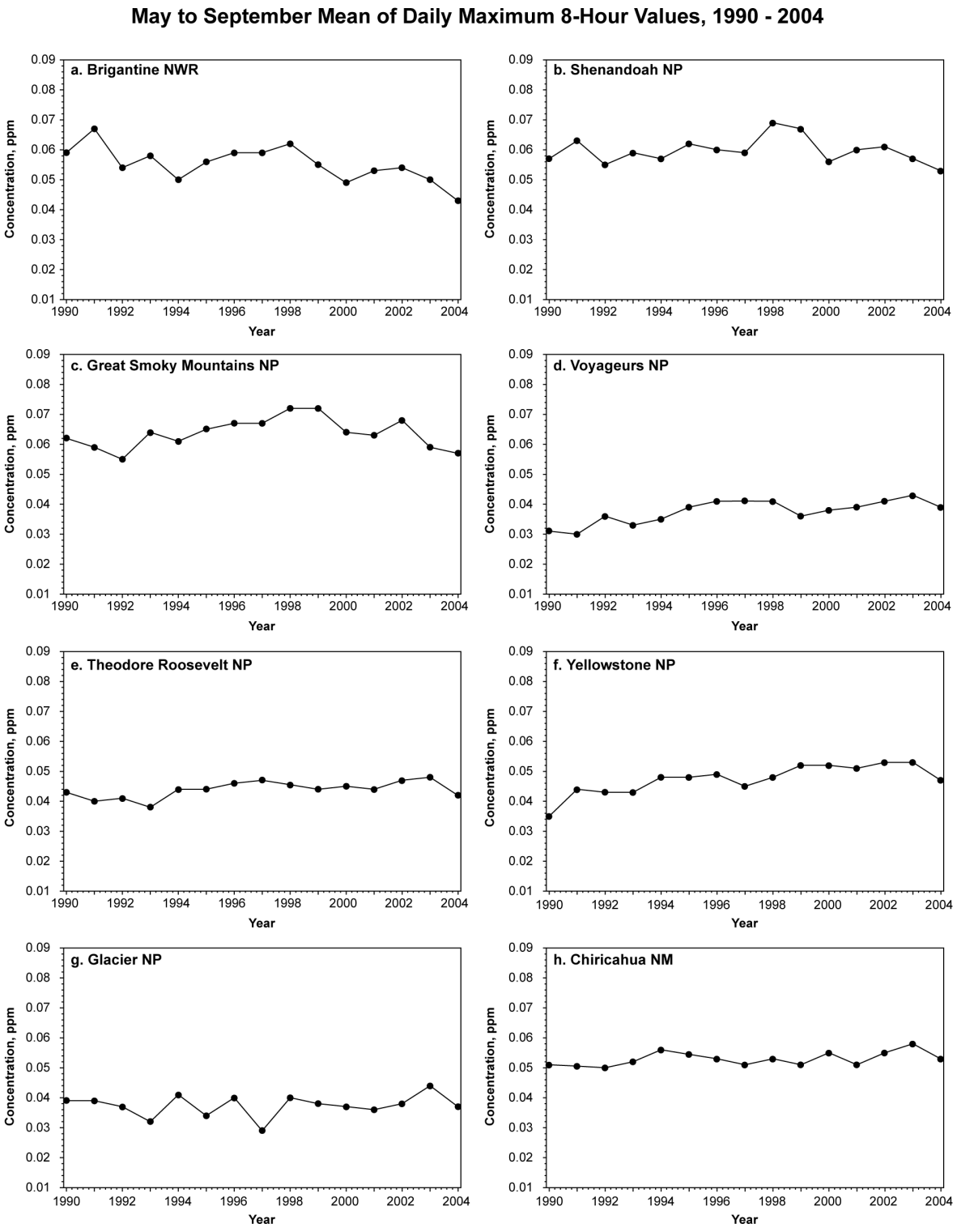


Figure 3-19a-h. Year-to-year variability in mean daily maximum 8-h O_3 concentrations at selected national park (NP), national wildlife refuge (NWR), and national monument (NM) sites.

Source: Fitz-Simons et al. (2005)

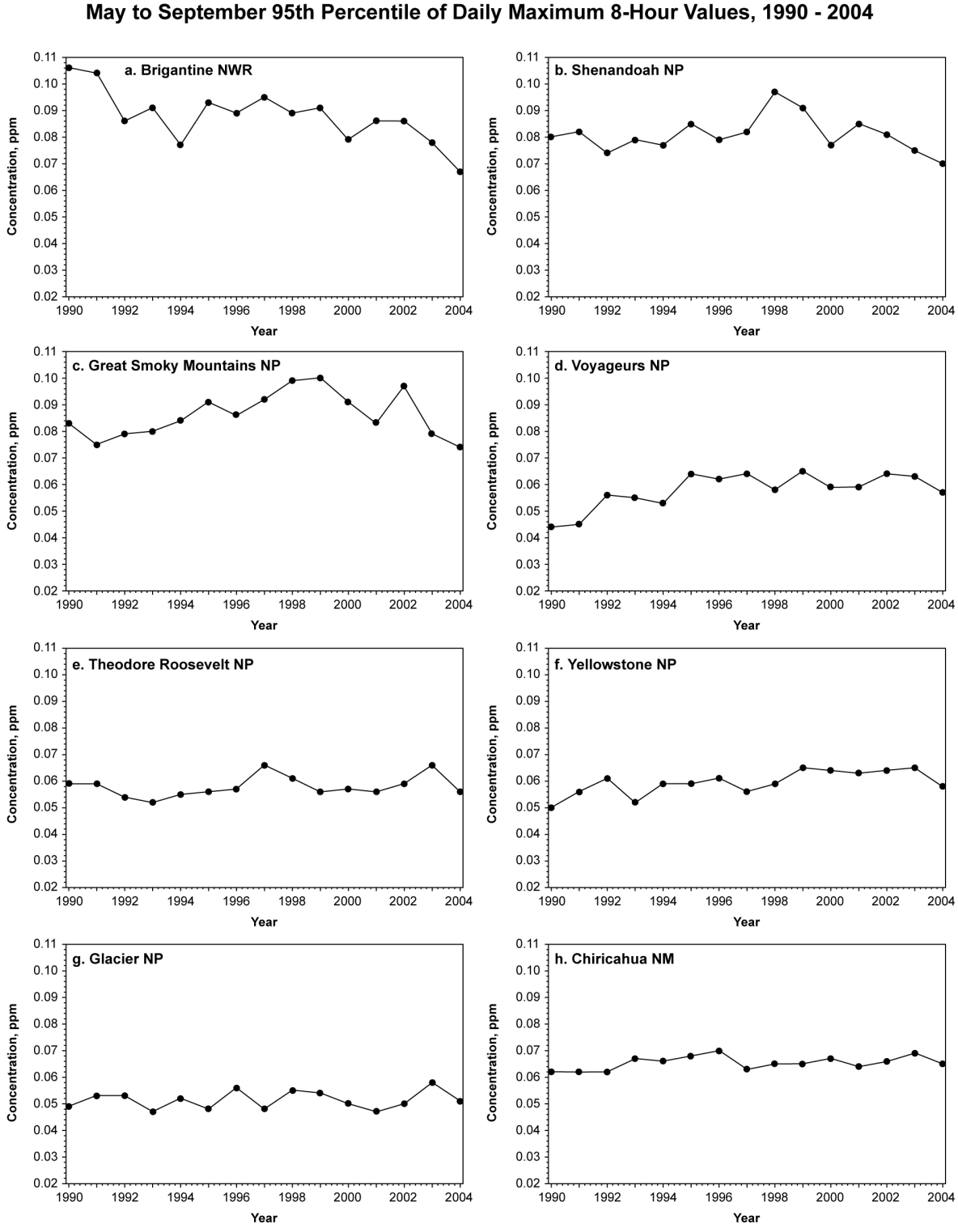


Figure 3-20a-h. Year-to-year variability in 95th percentile of daily maximum 8-h O_3 concentrations at selected national park (NP), national wildlife refuge (NWR), and national monument (NM) sites.

Source: Fitz-Simons et al. (2005).

Some species (e.g., O_3 and organic nitrates) are closely related photochemically and are highly correlated. Others (e.g., O_3 and H_2O_2) show a more complex correlation pattern. Further details are given in Annex AX3 in Section AX3.7.

Relationships between primary and secondary components are illustrated by considering data for O_3 and $PM_{2.5}$. Ozone and $PM_{2.5}$ concentrations observed at a monitoring site in Fort Meade, MD are plotted as binned means for different intervals in Figure 3-21, based on data collected between July 1999 and July 2001. As can be seen from the figure, $PM_{2.5}$ tends to be negatively correlated with O_3 to the left of the inflection point (at about 30 ppbv O_3) and tends to be positively correlated with O_3 to the right of the inflection point. Data to the left of the minimum in $PM_{2.5}$ were collected mainly during the cooler months of the year, while data to the right of the minimum were collected during the warmer months. This situation arises because $PM_{2.5}$ contains a large secondary component during the summer and has a larger primary component during winter. During the winter, O_3 comes mainly from the free troposphere, above the planetary boundary layer and, thus, may be considered a tracer for relatively clean air, and it is titrated by NO in the polluted boundary layer. Unfortunately, data for $PM_{2.5}$ and O_3 are collected concurrently at relatively few U.S. sites throughout an entire year. So these results, while highly instructive, are not readily extrapolated to areas where appreciable photochemical activity occurs throughout the year. Ito et al. (2005) examined the relation between PM_{10} and O_3 on a seasonal basis in several urban areas (cf., Figure 7-24). Although PM_{10} contains proportionately more primary material than does $PM_{2.5}$, relations similar to those shown in Figure 3-21 are found, reflecting the dominant contribution of $PM_{2.5}$ to PM_{10}.

Other Oxidants

Measurements of gas phase peroxides in the atmosphere were reviewed by Lee et al. (2000). Ground level measurements of H_2O_2 taken during the 1970s indicated values of 180 ppb in Riverside, CA and 10 to 20 ppb during smog episodes in Claremont and Riverside, with values approaching 100 ppb in forest fire plumes. However, later surface measurements always found much lower values. For example, in measurements made in Los Angeles and nearby areas in the 1980s, peak values were always less than about 2 ppb and in a methods intercomparison study in Research Triangle Park, NC in June 1986, concentrations were <2.5 ppbv. Higher

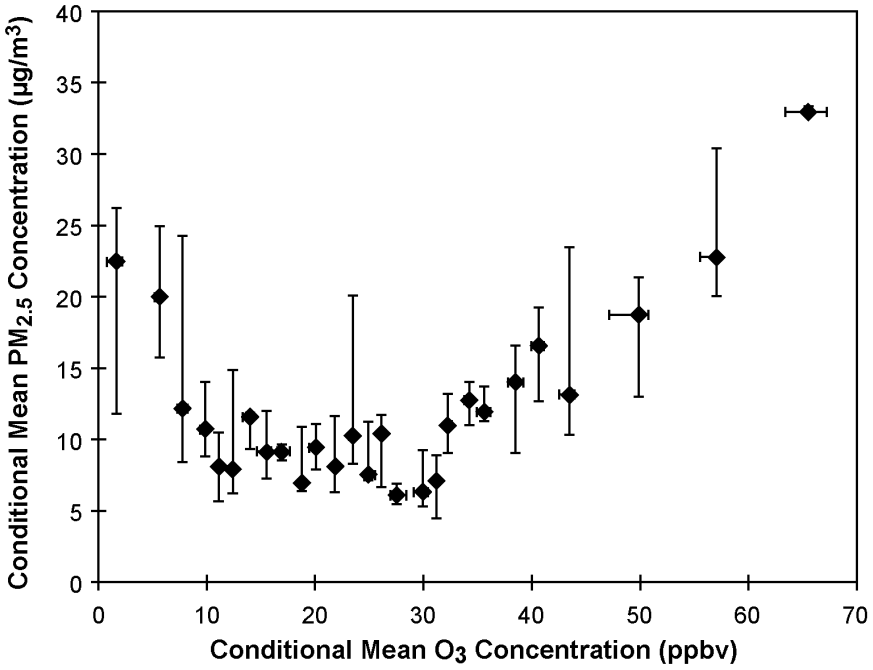


Figure 3-21. Binned mean $PM_{2.5}$ concentrations versus binned mean O_3 concentrations observed at Fort Meade, MD from July 1999 to July 2001.

Source: Chen (2002).

values ranging up to 5 ppb were found in a few other studies in Kinterbish, Alabama and Meadview, Arizona. Several of these studies found strong diurnal variations (typically about a factor of three) with maximum values in the mid-afternoon and minimum values in the early morning. Mean concentrations of organic hydroperoxides at the surface at Niwot Ridge, CO in the summer of 1988 and State Park, GA during the summer of 1991 were all less than a few ppb.

Aircraft measurements of hydroperoxide (H_2O_2, CH_3OOH and $HOCH_2OOH$) concentrations were made as part of the Southern Oxidants Study intensive campaign in Nashville, TN in July 1995 (Weinstein-Lloyd et al., 1998). The median concentration of total hydroperoxides in the boundary layer between 1100 and 1400 CDT was about 5 ppbv, with more than 50% contribution from organic hydroperoxides. Median O_3 was about 70 ppbv at the same time. The concentrations of the hydroperoxides depended strongly on wind direction with values about 40% lower when winds originated from the N/NW as opposed to the S/SW suggesting that local source areas were important.

Peroxyacetylnitrate (PAN) is produced during the photochemical oxidation of a wide range of VOCs in the presence of NO_x. It is removed by thermal decomposition and also by uptake to vegetation (Sparks et al., 2003; Teklemariam and Sparks, 2004). PAN is the dominant member of the broader family of peroxyacylnitrates (PANs) which includes as other significant atmospheric components peroxypropionyl nitrate (PPN) of anthropogenic origin, and peroxymethacrylic nitrate (MPAN) produced from oxidation of isoprene. Measurements and models show that PAN in the United States includes major contributions from both anthropogenic and biogenic VOC precursors (Horowitz et al., 1998; Roberts et al., 1998). Measurements in Nashville during the 1999 summertime Southern Oxidants Study (SOS) showed PPN and MPAN amounting to 14% and 25% of PAN respectively (Roberts et al., 2002). Measurements during the TexAQS 2000 study in Houston indicated PAN concentrations of up to 6.5 ppbv (Roberts et al., 2003). PAN measurements in southern California during the SCOS97-NARSTO study indicated peak concentrations of 5-10 ppbv, which can be contrasted to values of 60 to 70 ppbv measured back in 1960 (Grosjean, 2003). Vertical profiles measured from aircraft over the U.S. and off the Pacific coasts show PAN concentrations above the boundary layer of only a few hundred pptv, although there are significant enhancements associated with long-range transport of pollution plumes including from Asia (Kotchenruther et al., 2001a; Roberts et al., 2004). Decomposition of this anthropogenic PAN as it subsides over North America can lead to significant O_3 production, enhancing the O_3 background (Kotchenruther et al., 2001b; Hudman et al., 2004).

Oxidants are also present in airborne cloud droplets, rain drops and particulate matter. Measurements of hydroperoxides, summarized by Reeves (2003), are available mainly for hydrometeors, but are sparse for ambient particles. Venkatachari et al. (2005a) sampled the concentrations of total reactive oxygen species (ROS) in particles using a cascade impactor in Rubidoux, CA during July 2003. Although the species constituting ROS were not identified, the results were reported in terms of equivalent H_2O_2 concentrations. Unlike O_3 and gas phase H_2O_2 which show strong diurnal variability (i.e., about a factor of three variation between afternoon maximum and early morning minimum), the diurnal variation of particle phase ROS was found to be much weaker (i.e., less than about 20%) at least for the time between 8 a.m. and midnight. Because the ROS were measured in the fine aerosol size fraction, which has a lifetime with respect to deposition of much greater than a day, little loss is expected but their concentrations

might also be expected to increase because of nighttime chemistry, perhaps involving NO_3 radicals. The ROS concentration, about 7×10^{-9} M/m^3 (expressed as equivalent H_2O_2), was at most 1% that of O_3 (6.2 to 38×10^{-7} M/m^3 or 15 to 90 ppb), with highest values at night. In a companion study conducted in Queens, NY during January and early February 2004, Venkatachari et al. (2005b) found much lower concentrations of ROS of about 1.2×10^{-9} M/m^3. However, O_3 levels were also substantially lower, but ROS concentrations were still less than 1% those of O_3. It is of interest to note that gas phase OH concentrations measured at the same time ranged from about 7.5×10^4/cm^3 to about 1.8×10^6/cm^3, implying the presence of significant photochemical activity even in New York City during winter.

Co-occurrence of Ozone with Other Pollutants

The characterization of co-occurrence patterns under ambient conditions is important for relating human health and vegetation effects under ambient conditions to controlled research results as described in Annex AX3.8. Several attempts have been made to characterize gaseous air pollutant mixtures. The previous 1996 O_3 AQCD discussed various patterns of pollutant mixtures of SO_2, NO_2, and O_3. Pollutant combinations can occur at or above a threshold concentration at either the same or different times.

The 1996 O_3 AQCD noted that studies of the joint occurrence of gaseous NO_2/O_3 and SO_2/O_3 reached two conclusions: (1) hourly simultaneous and daily simultaneous-only co-occurrences are fairly rare (when both pollutants were present at an hourly average concentration $\geq$0.05 ppm) and (2) when co-occurrences are present, complex-sequential and sequential-only co-occurrence patterns predominate. Year-to-year variability was found to be insignificant.

Using 2001 hourly data for O_3 and NO_2 and for O_3 and SO_2, the co-occurrence patterns for the data are similar to those of previous studies. As shown in Figure 3-22, fewer than 10 co-occurrences of O_3 and NO_2 were found for most of the collocated monitoring sites. Likewise, Figure 3-23 shows that fewer than 10 co-occurrences of O_3 and SO_2 were found for most of the collocated monitoring sites analyzed.

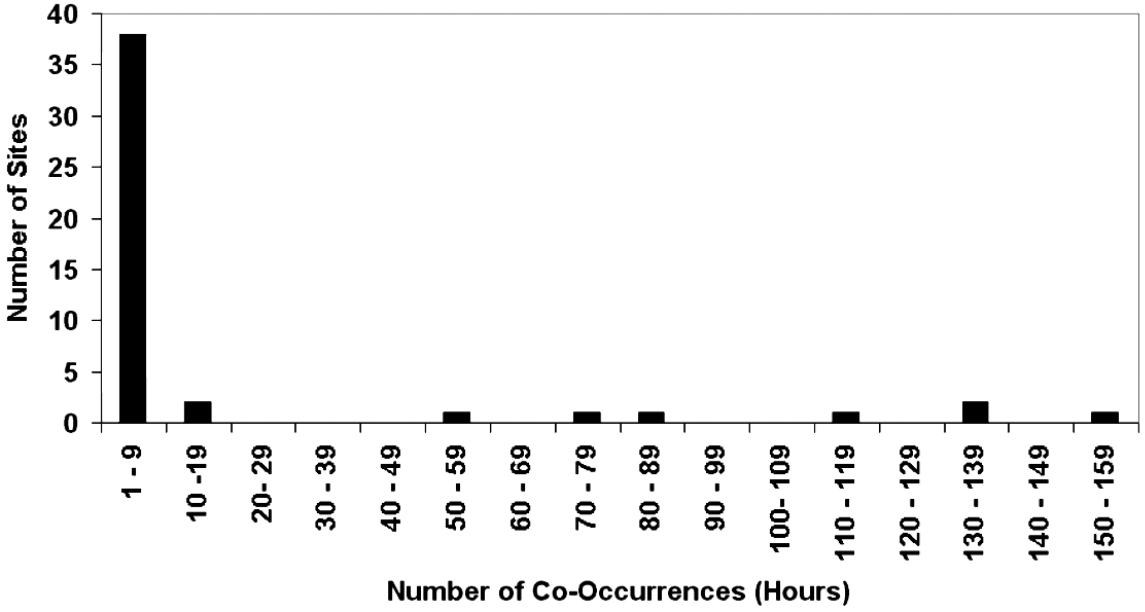


Figure 3-22. The co-occurrence pattern for O_3 and nitrogen dioxide using 2001 data from the AQS. There is co-occurrence when hourly average concentrations of O_3 and another pollutant are both ≥0.05 ppm.

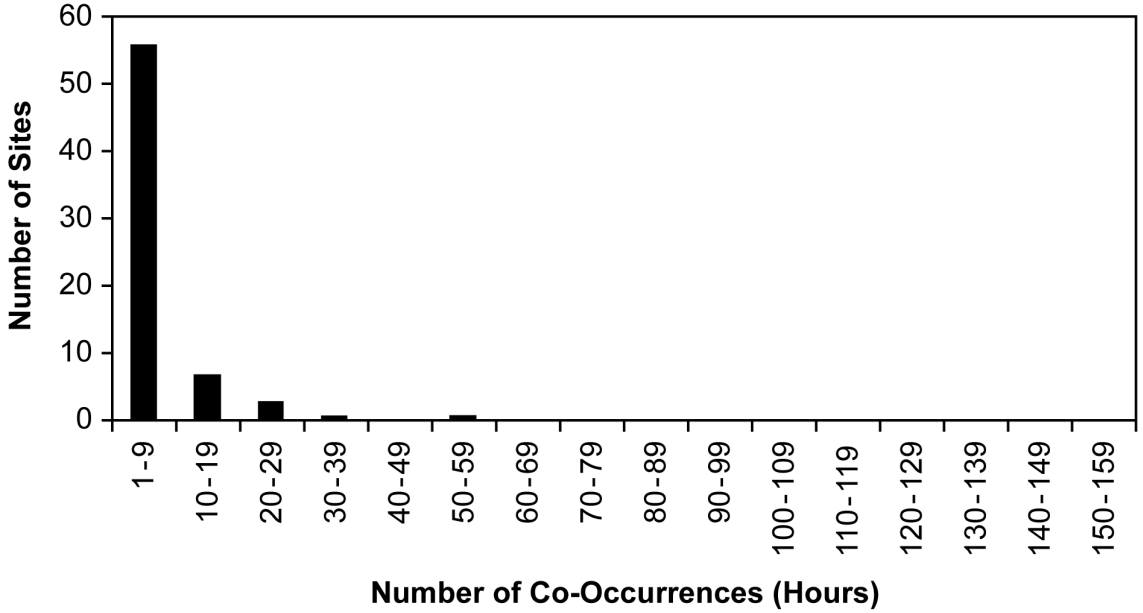


Figure 3-23. The co-occurrence pattern for O_3 and sulfur dioxide using 2001 data from AQS. There is co-occurrence when hourly average concentrations of O_3 and another pollutant are both ≥0.05 ppm.

Since 1999, monitoring stations across the United States have routinely measured 24-h average concentrations for $PM_{2.5}$. Daily co-occurrence of $PM_{2.5}$ and O_3 over a 24-h period was also characterized. Because $PM_{2.5}$ data are mostly summarized as 24-h average concentrations in the AQS database, a daily co-occurrence of O_3 and $PM_{2.5}$ was subjectively defined as an hourly average O_3 concentration $\geq$0.05 ppm and a $PM_{2.5}$ 24-h concentration $\geq$40 $\mu g/m^3$ (corresponding to the EPA Air Quality Index, Level of Concern for $PM_{2.5}$) occurring during the same 24-h period. Using 2001 data from the AQS database, the daily co-occurrence of $PM_{2.5}$ and O_3 was infrequent (Figure 3-24). Only limited data are available on the co-occurrence of O_3 and other pollutants (e.g., acid precipitation and acidic cloudwater). In most cases, routine monitoring data are not available from which to draw general conclusions.

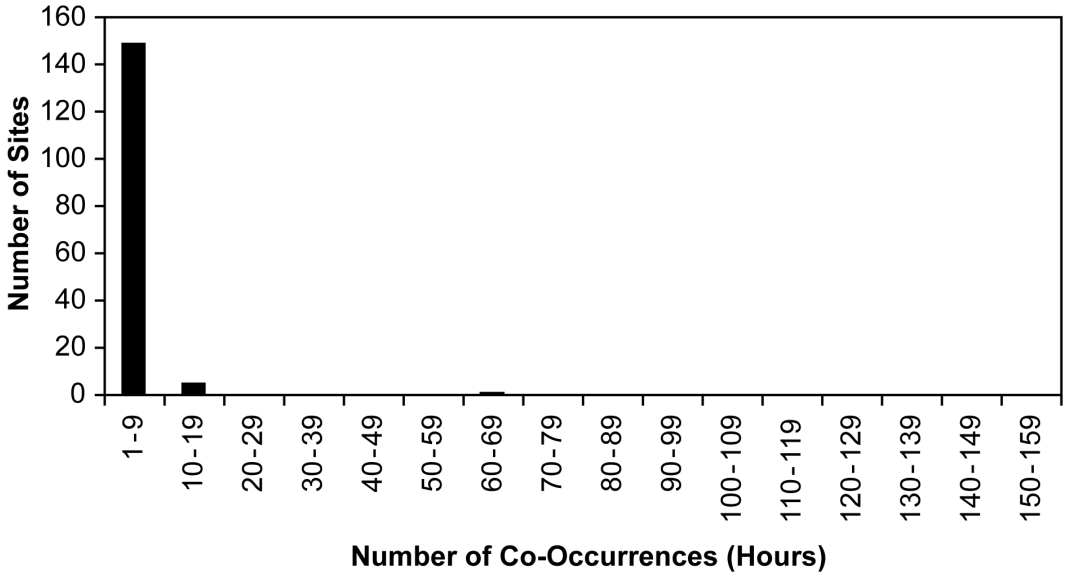


Figure 3-24. The co-occurrence pattern for O_3 and $PM_{2.5}$ using 2001 data from AQS.

3.7 POLICY RELEVANT BACKGROUND OZONE CONCENTRATIONS

Background O_3 concentrations used for NAAQS-setting purposes are referred to as Policy Relevant Background (PRB) O_3 concentrations. Policy Relevant Background concentrations are those concentrations that would occur in the United States in the absence of anthropogenic emissions in continental North America (defined here as the United States, Canada, and Mexico). Policy Relevant Background concentrations include contributions from natural sources

everywhere in the world and from anthropogenic sources outside these three countries. For the purposes of informing decisions about O_3 NAAQS, EPA assesses risks to human health and environmental effects from O_3 levels in excess of PRB concentrations. Issues concerning the methodology for estimating PRB O_3 concentrations are described in detail in Annex AX3, Section AX3.9.

Contributions to PRB O_3 include photochemical actions involving natural emissions of VOCs, NO_x, and CO as well as the long-range transport of O_3 and its precursors from outside North America and the stratospheric-tropospheric exchange (STE) of O_3. Processes involved in STE are described in detail in Annex AX2.3. Natural sources of O_3 precursors include biogenic emissions, wildfires, and lightning. Biogenic emissions from agricultural activities are not considered in the formation of PRB O_3.

Springtime maxima are observed at relatively remote (Annex AX3 and Figures 3-25a,b) national park sites, located mainly in the western United States and at a number of other relatively unpolluted monitoring sites throughout the Northern Hemisphere. The major issues concerning the calculation of PRB O_3 center on the capability of the current generation of global-scale, three-dimensional, CTMs to simulate the causes of high O_3 concentrations observed at monitoring sites in relatively unpolluted areas of the United States from late winter through spring (i.e., February through June). The issues raised do not affect interpretations of the causes of summertime O_3 episodes as strongly. Summertime O_3 episodes are mainly associated with slow- moving high-pressure systems characterized by limited mixing between the planetary boundary layer and the free troposphere, as noted in Annex AX2, Section AX2.3.

A large number of case studies document the occurrence of STE mainly during winter and spring in mid- and high-latitudes in Europe, Asia, and North America. These studies were based on aircraft, satellite, and ground-based measurements. Considerable uncertainty exists in the magnitude of the exchange; however, these studies have found that STE occurs throughout the year, but with a distinct preference for the transport of O_3 directly to the middle and lower troposphere during late winter and spring. Transport to the upper troposphere occurs throughout the year.

Springtime maxima in tropospheric O_3 observed at high latitudes are also associated with the winter buildup of O_3 precursors and thermally labile reservoir species, such as PAN and other reactive nitrogen species. These pollutants originate from all continents in the Northern

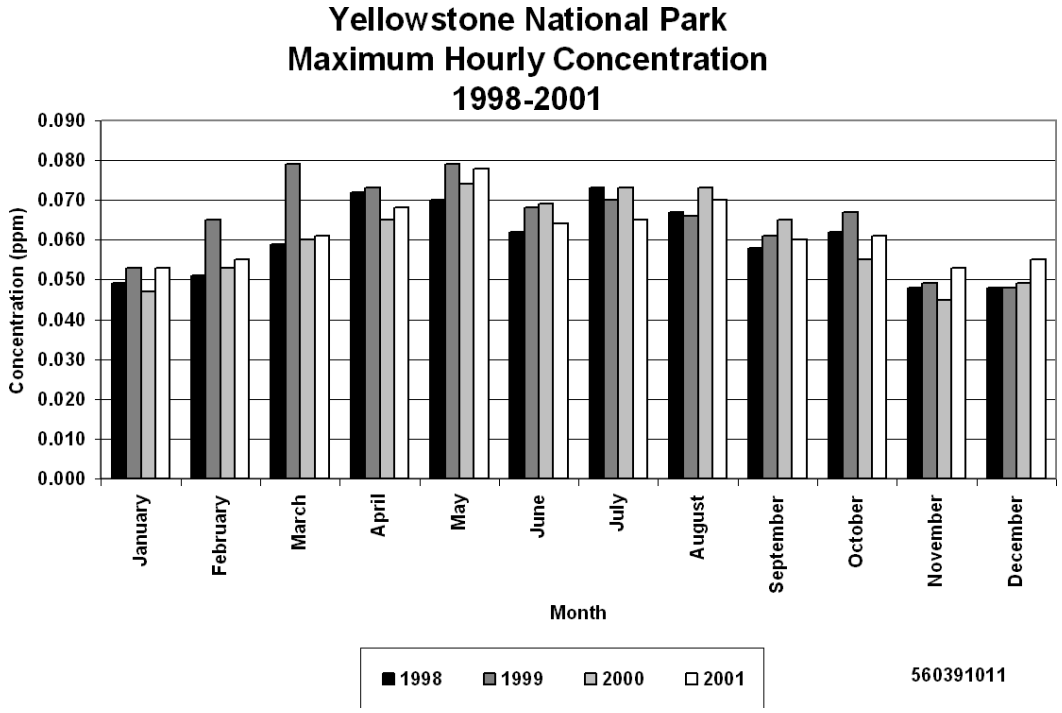


Figure 3-25a. Monthly maximum hourly average O_3 concentrations at Yellowstone National Park (WY) in 1998, 1999, 2000, and 2001.

Source: U.S. Environmental Protection Agency (2003a).

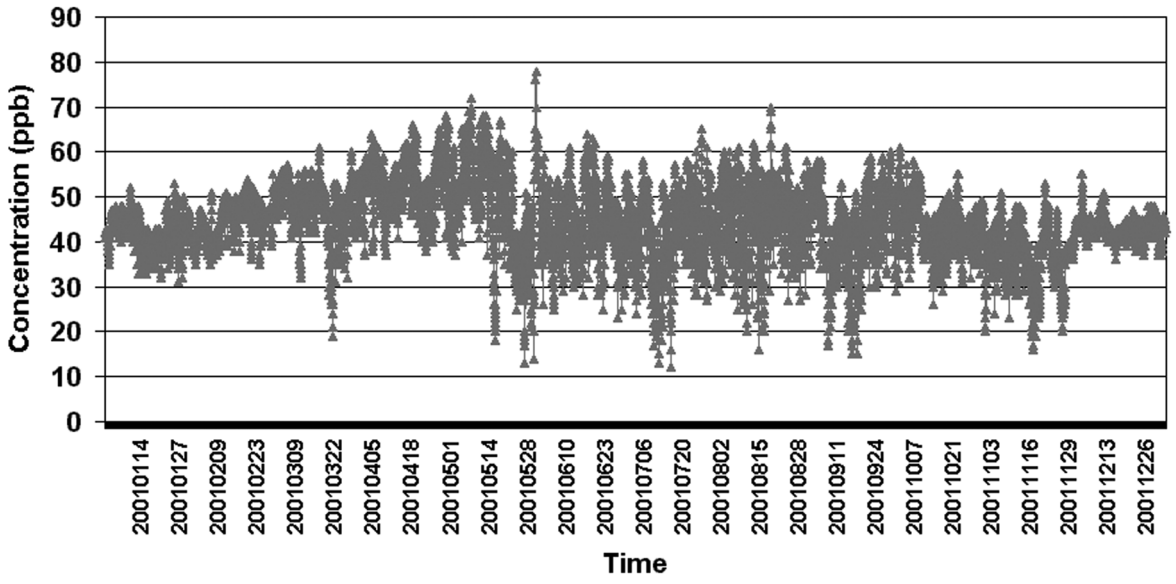


Figure 3-25b. Hourly average O_3 concentrations at Yellowstone National Park (WY) for the period January to December 2001.

Source: U.S. Environmental Protection Agency (2003a).

Hemisphere. Ozone precursor concentrations reach a maximum in late March; and as sunlight returns to the Arctic, photochemical reactions generate tropospheric O_3 (Section AX3.9.1). The contribution of Asian sources to the U.S. levels is also largest during spring, reflecting the efficient lifting of Asian pollution ahead of cold fronts originating in Siberia and transport by strong westerly winds across the Pacific (e.g., Hudman et al., 2004). The longer lifetime of O_3 during spring also contributes to springtime maxima (Wang et al., 1998).

Estimates of PRB concentrations cannot be obtained solely by examining measurements of O_3 obtained at RRMS in the United States (Annex AX3, Section AX3.2.3) because of the long-range transport from anthropogenic source regions within North America. It should also be noted that it is impossible to determine sources of O_3 without ancillary data that could be used as tracers of sources or to calculate photochemical production and loss rates. The current definition of PRB implies that only CTMs can be used to estimate the range of PRB values. On the synoptic and larger spatial scales at least, all evidence indicates that global CTMs are adequate tools to investigate the factors controlling tropospheric O_3; and three-dimensional CTMs, as typified by Fiore et al. (2003) appear to offer the best methodology for estimating PRB concentrations that cannot be measured directly (Annex AX3, Section AX3.9.2), at least for averaging periods of longer than one hour.

Previous estimates of background O_3 concentrations, based on different concepts of background, are given in Table 3-2. Results from global three-dimensional CTMs, where the background is estimated by zeroing anthropogenic emissions in North America (Table 3-8) are on the low end of the 25 to 45 ppbv range. Lefohn et al. (2001) have argued that frequent occurrences of O_3 concentrations above 50 to 60 ppbv at remote northern U.S. sites in spring are mainly stratospheric in origin. Fiore et al. (2003) used a global CTM to determine the origin of the high-O_3 events reported by Lefohn et al. (2001), and to conduct a more general quantitative analysis of background O_3 as a function of season, altitude, and local O_3 concentration.

Figure 3-26 shows a comparison between observations obtained at CASTNet sites and model results of Fiore et al. (2003). They classified the CASTNet monitoring sites into low-lying sites (generally <1.5 km) and elevated sites (>1.5 km). All elevated sites are in the West. Results were then aggregated to construct the cumulative probability distributions shown in Figure 3-26 for the 58 low-altitude sites and the 12 high-altitude sites as well as for the three seasons. The calculated mean background at the surface sites in spring is 27 ppbv, compared to

Table 3-2. Previous Estimates of Background O_3 in Surface Air Over the United States

Study	Method	Time Period	Region	Background Estimate (ppbv)
Trainer et al. (1993)	y-intercept of O_3 vs. NO_y-NO_x regression line[a]	Summer 1988	Eastern United States	30-40[b]
Hirsch et al. (1996)	y-intercept of O_3 vs. NO_y-NO_x regression line	May-Sep 1990-1994	Harvard Forest[c]	25 (Sept) – 40 (May)[d]
Altshuller and Lefohn (1996)	y-intercept of O_3 vs. NO_y regression line, and observations at remote/rural sites	Apr-Oct 1988-1993	Continental United States	25-45 (inland)[e] 25-35 (coastal)
Liang et al. (1998)	Sensitivity simulation in a 3-D model with anthropogenic NO_x emissions in the continental U.S. set to zero	Full year	Continental United States	20-30 (East)[f] 20-40 (West) (spring maximum)
Lin et al. (2000)	Median O_3 values for the lowest 25th percentiles of CO and NO_y concentrations	1990-1998	Harvard Forest	35 (fall) – 45 (spring)[g]
Fiore et al. (2002)	O_3 produced outside of the North American boundary layer in a global 3-D model	Summer 1995	Continental United States	15-30 (East)[h] 25-35 (West)

[a] NO_y is the chemical family including NO_x and its oxidation products; NO_y-NO_x denotes the chemically processed component of NO_y.
[b] 1300-1700 local time (LT) in flatland and valley sites; all daytime measurements at elevated sites.
[c] rural site in central Massachusetts.
[d] 1100-1700 EST hourly means.
[e] seasonal 7-h (0900-1559) daylight average.
[f] 1300-1600 LT monthly mean.
[g] daily max 8-h averages.
[h] 1300-1700 average.

Source: Fiore et al. (2003).

23 ppbv in summer and fall. At these sites, the background is highest for O_3 concentrations near the center of the distribution, and it declines as total surface O_3 concentrations increase, for reasons summarized below and discussed by Fiore et al. (2002). The observed O_3 concentration thus serves a surrogate for meteorological variability (i.e., stagnant versus ventilated conditions), such that the background O_3 is smaller on days when total O_3 is highest. At the elevated sites, the calculated mean background is 36 ppbv in spring versus 30 ppbv in the summer and fall. Background concentrations in the fall resemble those in summer but show less variability and do not exceed 40 ppbv anywhere in this analysis.

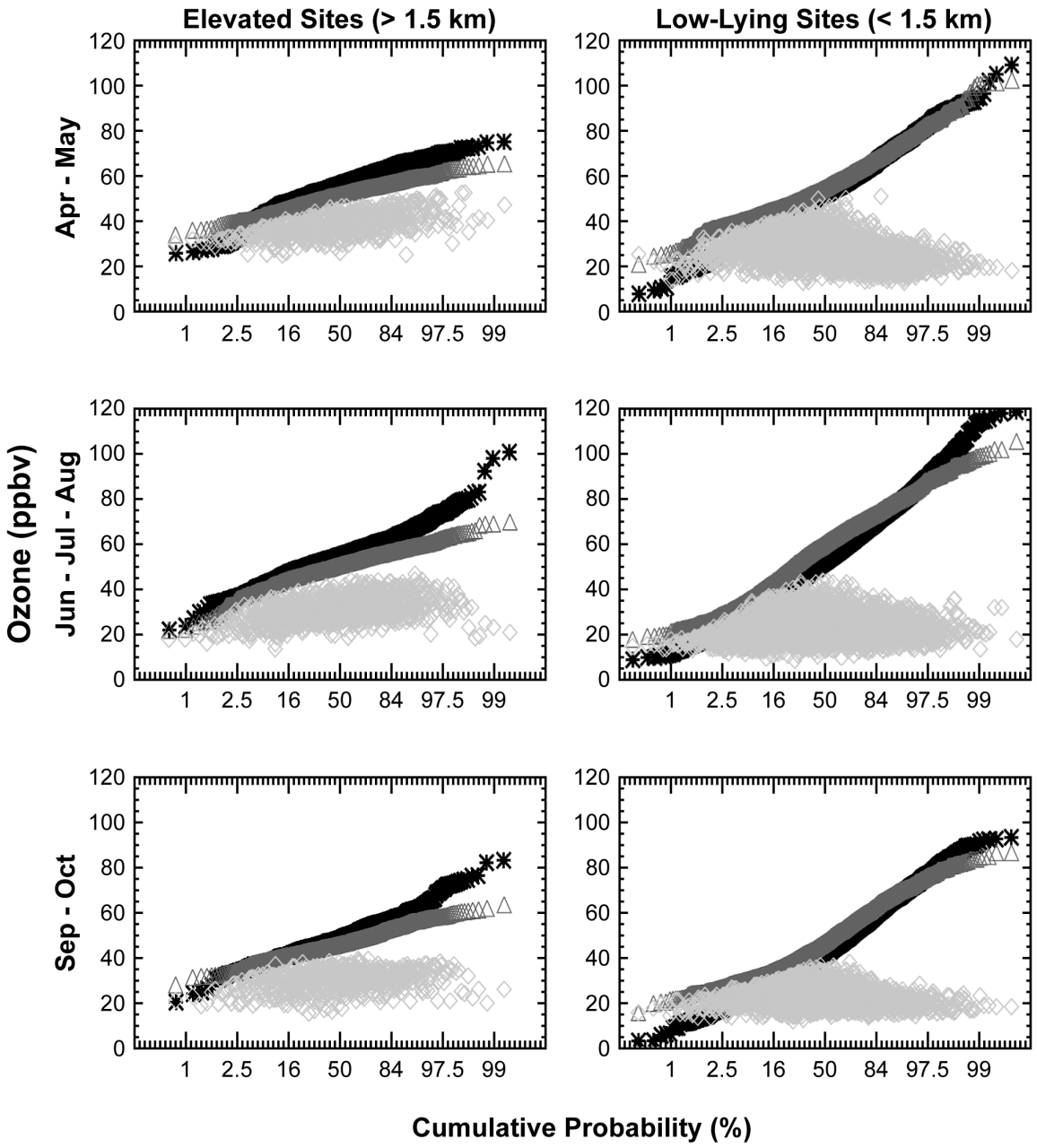


Figure 3-26. Estimates of background contribution to surface afternoon (13 to 17 LT) O_3 concentrations in the United States as a function of local O_3 concentration, site altitude, and season. The figure shows cumulative probability distributions of O_3 concentrations for the observations (asterisks) and the model (triangles). The corresponding distribution of background O_3 concentrations is shown as grey diamonds.

Source: Fiore et al. (2003).

Major conclusions from the Fiore et al. (2003) study (discussed in detail in Annex AX3, Sections AX3.9.3 and AX3.9.4) are:

- PRB O_3 concentrations in U.S. surface air from 1300 to 1700 local time are generally 15 to 35 ppbv. They decline from spring to summer and are generally <25 ppbv under the conditions conducive to high-O_3 episodes.
- PRB O_3 concentrations can be represented as a function of season, altitude, and total surface O_3 concentration, as illustrated in Figure 3-26.
- High PRB concentrations (40 to 50 ppbv) occur occasionally at high-elevation sites (>1.5 km) in spring due to the free-tropospheric influence, including a 4- to 12-ppbv contribution from hemispheric pollution (O_3 produced from anthropogenic emissions outside North America). These sites cannot be viewed as representative of low-elevation surface sites (Cooper and Moody, 2000), where the background is lower when O_3 >60 ppbv.
- The stratospheric contribution to surface O_3 is of minor importance, typically well <20 ppbv. While stratospheric intrusions might occasionally elevate surface O_3 at high-altitude sites, these events are rare.

Appropriate background concentrations should thus be allowed to vary as a function of season, altitude, and total O_3 level. The diamonds in Figure 3-26 can be applied for this purpose. In particular, the depletion of the background during high-O_3 events should be taken into account (i.e., background O_3 is depleted by reactions in the atmosphere and by deposition to the surface but is not replenished at a significant rate in the stable, polluted boundary layer). This depletion is shown in the right-hand panels of Figure 3-26 for the highest O_3 values. Note that the model is generally able to reproduce the overall frequency distributions in Figure 3-26. Typically, models produce distributions flatter than are observed. Underpredictions, especially at the upper end of the frequency distribution during the warmer months, are likely related to sub-grid-scale processes that the model cannot resolve explicitly. The highest observed O_3 concentrations in all three seasons and at all altitudes are associated with regional pollution (i.e., North American anthropogenic emissions), rather than stratospheric influence.

Chemistry transport models should be evaluated with observations given earlier in Chapter 3, in Annex AX3, and to simulate the processes causing the intra-day variability in O_3 concentrations shown in Figure 3-27 in addition to those summarized in Chapter 2. The diurnal patterns shown in Figure 3-27 do not fit the smooth pattern shown in Figure 3-15 and indicate

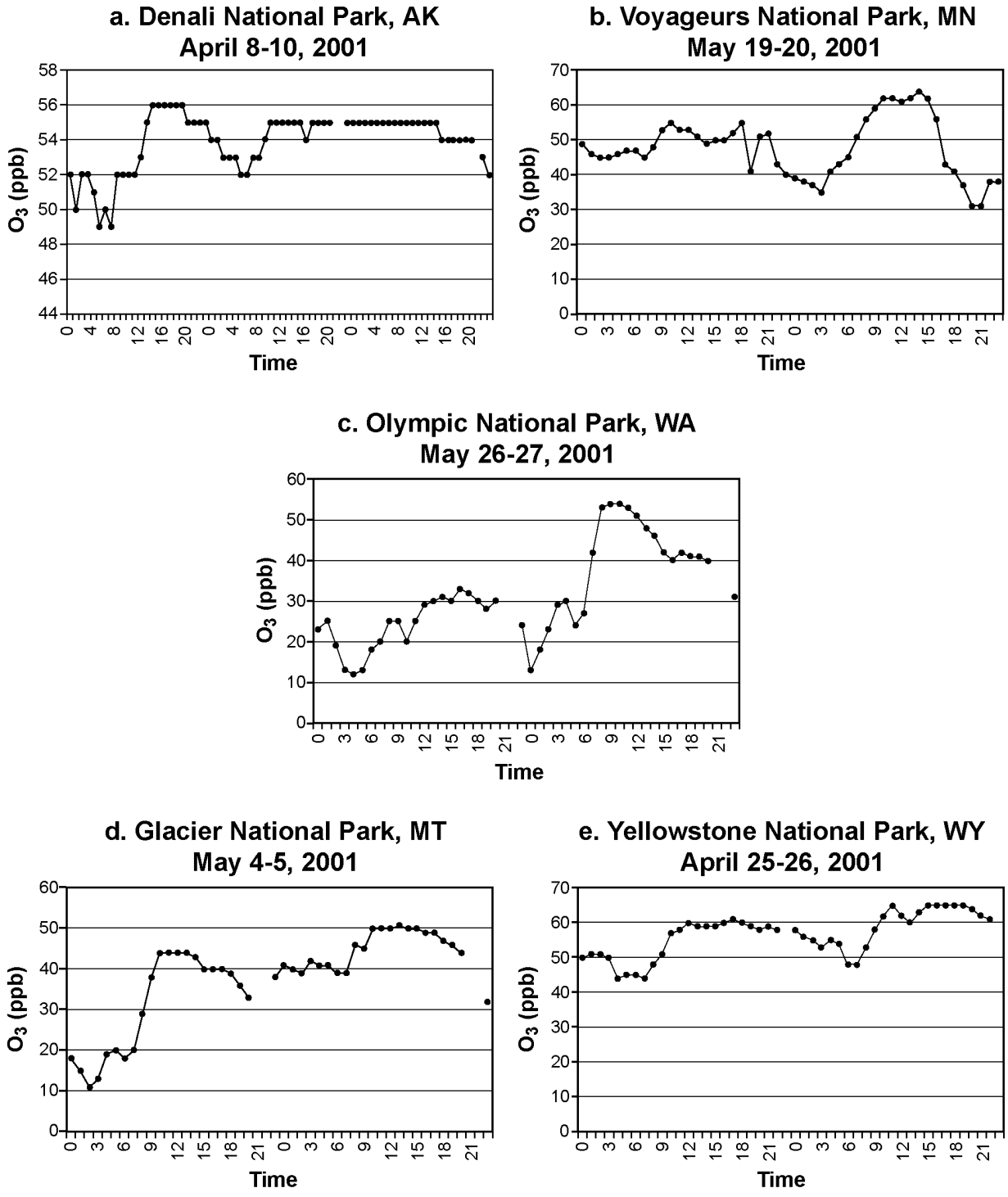


Figure 3-27. Time-series of hourly average O_3 concentrations observed at five national parks: Denali (AK), Voyageur (MN), Olympic (WA), Glacier (MT), and Yellowstone (WY).

processes capable of producing rapid rises in O_3 at times when substantial photochemical activity is not present and may indicate stratospheric effects. Higher resolution models capable of spatially and temporally resolving stratospheric intrusions and capable of resolving O_3 variability on hourly timescales have not been applied to this problem. Ebel et al. (1991) have demonstrated that regional-scale CTMs could be used to study individual stratospheric intrusions. As an example of the utility of different types of models, Zanis et al. (2003) were able to forecast, observe, and model a stratospheric intrusion (maximum penetration depth was to slightly >2 km altitude) that occurred from June 20 to 21, 2001, over a large swath of central Europe. Roelofs et al. (2003) compared results from six global tropospheric CTMs with lidar observations obtained during that event and concluded that the models qualitatively captured the features of this intrusion. It was also found that the coarser resolution models overestimated transport to lower altitudes. The use of higher resolution models, perhaps nested inside the coarser resolution models, may have helped solve this problem. They would also better address issues related to temporal (i.e., 1-h versus 8-h averages) and spatial (i.e., populated versus remote areas) scales needed by policymakers.

Although many of the features of the day-to-day variability of O_3 at RRMS in the United States are simulated reasonably well by Fiore et al. (2003), uncertainties in the calculation of the temporal variability of O_3 originating from different sources on shorter time scales must be recognized. The uncertainties stem in part from an underestimate in the seasonal variability in the STE of O_3 (Fusco and Logan, 2003), the geographical variability of this exchange, and the variability in the exchange between the free troposphere and the planetary boundary layer in the model.

Ideally, the predictions resulting from an ensemble of models should be compared with each other and with observations, so that the range of uncertainty inherent in the model predictions can be evaluated.

3.8 OZONE EXPOSURE IN VARIOUS MICROENVIRONMENTS

Humans are exposed to O_3 and related photochemical oxidants through the exchange boundary, the skin and the openings into the body such as the mouth, the nostrils, and punctures and lesions in the skin (U.S. Environmental Protection Agency, 1992; Federal Register, 1986).

Inhalation exposure to O_3 and related photochemical oxidants is determined by pollutant concentrations measured in the breathing zone that is not affected by exhaled air as the individual moves through time and space. A discussion of the basic terminology associated with exposure appears in AX3.

Quantification of Exposure

Ambient O_3 concentrations vary with time of day (peaking during the latter portion of the day) and season and among locations. Consequently, exposure to O_3 will change as a function of time of day and as an individual moves among locations. A hypothetical exposure is demonstrated in Figure 3-28. The actual dose received also changes during the day and is dependent on the O_3 concentration in the breathing zone and the individual's breathing rate, which is, in turn, dependent on the individual's level of exertion.

When measuring or modeling exposure to O_3 and related photochemical oxidants consideration should be given to the diurnal weekly (weekday-weekend) and seasonal variability. Peak concentrations lasting for several hours typically occur toward the latter portion of the day during the summer months. Regional O_3 episodes often co-occur with high concentrations of airborne fine particles, making it difficult to assess O_3 dynamics and exposure patterns. Also, while there are few indoor O_3 sources, O_3 will react with materials and other pollutants in the indoor environment in an analogous fashion to that occurring in the ambient atmosphere, potentially exposing subjects to other more toxic pollutants (Nazaroff and Weschler, 2004; Lee and Hogsett, 1999; Wainman et al., 2000; Weschler and Shields, 1997). (See discussion on O_3 chemistry and indoor sources and concentrations later in this chapter.).

Personal Exposure and Ambient Concentrations

The two approaches for measuring personal exposure are (a) the direct approach, using a personal exposure monitor (PEM) consisting of a passive sampler worn around the breathing zone, and (b) the indirect approach, which measures or estimates the O_3 concentrations through the use of models or biomarkers. Both approaches are associated with measurement error.

Although it is difficult to develop passive monitors for personal exposure measurements because of problems in identifying chemical or trapping reagents that can react with O_3, several modified passive samplers have been developed for use in personal O_3 exposure measurements

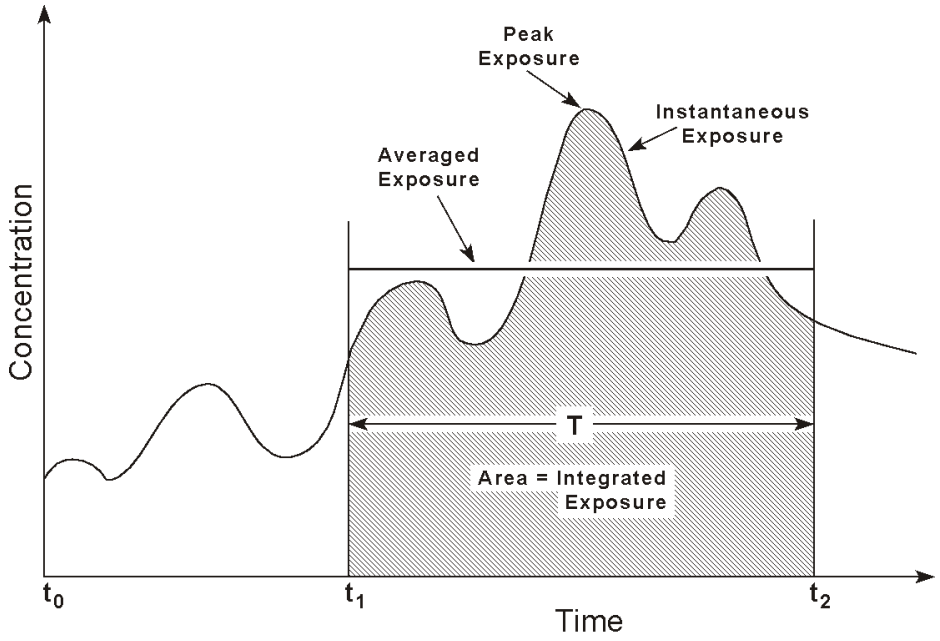


Figure 3-28. Hypothetical exposure time profile: pollutant exposure as a function of time showing how the average exposure, integrated exposure, and peak exposure relate to the instantaneous exposure. ($t_2 - t_1 = T$)

Source: U.S. Environmental Protection Agency (2004a).

(Bernard et al., 1999; Koutrakis et al., 1993; Avol et al., 1998b; Geyh et al., 1997, 1999). Some personal exposure measurements using passive samplers show O_3 exposures below those O_3 concentrations measured at outdoor stationary sites (Delfino et al., 1996; Avol et al., 1998b; Sarnat et al., 2000; Geyh et al., 2000; Brauer and Brook, 1997). However, other studies have found strong correlations between O_3 measured at stationary sites and personal monitored concentrations (Liard et al., 1999; Bauer and Brook, 1997; Linn et al., 1996; Lee et al., 2004; Avol et al., 1998b; O'Neill et al., 2003) when the time spent outdoors, age, gender, and occupation of the subjects were considered.

The indirect approach determines and measures the concentrations in all of the locations or "microenvironments". The concept of microenvironments is important in the understanding of human exposure modeling. Often identified with a perfectly mixed compartment, microenvironments are more recently viewed as a controlled volume, indoors or outdoors, that can be characterized using a set of either mechanistic or phenomenological governing equations. This allows for a nonhomogeneous environment, including sources and sinks within the

microenvironment. Microenvironments include indoor residences, other indoor locations, outdoors near roadways, other outdoor locations, and areas within vehicles.

Microenvironmental Concentration and Ozone Exposure Models

Outdoor concentrations of O_3 are estimated either through emissions-based mechanistic modeling, or through ambient-data-based modeling. Emissions-based models determine the spatiotemporal fields of the O_3 concentrations using precursor emissions and meteorological conditions as inputs. (They are described in Annex AX2.). The ambient-data-based models determine spatial or spatiotemporal distributions of O_3 through the use of interpolation schemes. The kriging approach provides standard procedures for generating an interpolated O_3 spatial distribution for a given period of time (Georgopoulos et al., 1997a,b). The Spatio-Temporal Random Field (STRF) approach has been used to interpolate monitoring data in both space and time (Christakos and Vyas, 1998a,b). The STRF approach can analyze information on temporal trends which cannot be directly incorporated by kriging.

Several approaches are available for modeling microenvironmental concentrations: empirical, mass balance, and detailed computational fluid dynamics (CFD) models. Empirical relationships provide the basis for future, "prognostic" population exposure models. Mass balance modeling is the most common approach used to model pollutant concentrations in enclosed microenvironments. Mass balance modeling ranges from very simple formulations, assuming ideal (homogeneous) mixing and only linear physicochemical transformations with sources and sinks, to models that account for complex multiphase chemical and physical interactions and nonidealities in mixing. Mass balance models take into account the effects of ventilation, filtration, heterogeneous removal, and direct emission as well as photolytic, thermal, and chemical reactions. The simplest form of the model is represented by the following differential equation:

$$\frac{dC_{IN}}{dt} = vC_{OUT} + \frac{S}{V} - vC_{IN}$$

where dC_{IN} is the indoor pollutant concentration (mass/volume), dt is time in hours, v is the air exchange rate, C_{OUT} is the outdoor pollutant concentration (mass/volume), V is the volume of the microenvironment, and S is the indoor source emission rate. When the model was used to

estimate indoor O_3 concentrations, indoor concentrations were found to be 33% of outdoor O_3 concentrations (Freijer and Bloemen, 2000). A more in-depth discussion of the mass balance model has been reported in Nazaroff and Cass (1986). The pNEM/O_3 model, discussed later in this chapter, includes a sophisticated mass balance model for indoor and vehicle microenvironments (Johnson, 2003). CFD models take into account the complex, multiphase processes that affect indoor concentrations of interacting gas phase pollutants, such as the interactions of O_3 with indoor sinks and sources (surfaces, gas releases) and with entrained gas (Sarwar et al., 2001, 2002; Sørensen and Weschler, 2002).

Exposure modeling is often used in evaluating exposure to large populations over time. The use of models is complicated by the fact that O_3 is a secondary pollutant with complex nonlinear and multiscale dynamics in space and time. Ozone is formed in the atmosphere through a series of chemical reactions involving precursor VOCs and NO_x. Therefore, O_3 exposures may be affected by: (1) emission levels and spatiotemporal patterns of VOCs and NO_x; (2) ambient atmospheric as well as indoor microenvironmental transport, removal and mixing processes; and (3) chemical transformations that take place over a multitude of spatial scales. The transformations are dependent on the presence of co-occurring pollutants and the nature of surfaces interacting with the pollutants.

Exposure models may be classified as (1) potential exposure models, typically the maximum outdoor concentrations versus "actual" exposure, including locally modified microenvironmental outdoor and indoor exposures; (2) population versus "specific individual"-based exposure models; (3) deterministic versus probabilistic models; and (4) observation versus mechanistic air quality model-driven estimates of spatially and temporally varying O_3 concentrations.

There are several steps involved in defining exposure models. The steps are based on frameworks described in the literature over the last 20 years and the structure of various existing inhalation exposure models (NEM/pNEM, MENTOR/SHEDS, REHEX, TRIM.Expo also known as APEX, AIRPEX, AIRQUIS). The steps include (1) estimation/ determination of the background or ambient levels of O_3; (2) estimation/determination of levels and temporal profiles of O_3 in various microenvironments; (3) characterization of relevant attributes of individuals or populations under study (age, gender, weight, occupation, other physiological characteristics); (4) development of activity event or exposure event sequences; (5) determination of appropriate inhalation rates during the exposure events; (6) determination of dose; (7) determination of

event-specific exposure and intake dose distributions for selected time periods; and (8) extrapolation of population sample (or cohort) exposures and doses to the entire populations of interest. Figure 3-29 provides a conceptual overview of a current exposure model. A more detailed overview of an exposure model can be found in Annex AX3.

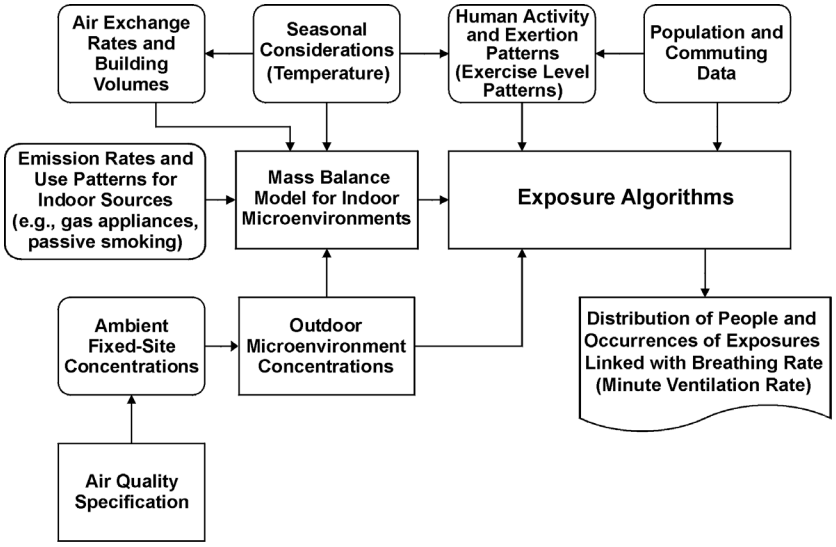


Figure 3-29. Conceptual overview of an exposure model. Model inputs (e.g., activity patterns, ambient monitoring data, air exchange rates) are in round-corner boxes and model calculations are shown in rectangles.

Source: Johnson et al. (1999).

To estimate the actual O_3 dose delivered to the lung, information on the concentration, minute ventilation rate, activity level, and the morphology of the respiratory tract are needed. Limited data have been compiled for ventilation rates for different age groups, both healthy and compromised individuals, at varies levels of activity (Klepeis et al., 1996, 2001; Avol et al., 1998b; Adams, 1993). Based on the available information, the highest level of outdoor activity occurs during the spring and summer months, during the mid- to late afternoon and early evening—the times when O_3 concentrations are highest. Children are likely more susceptible to the effects of O_3 than other groups. School-age children spend more time outdoors engaged in high-level activities than do other groups and breath more air in than adults relative to body

surface area, breathing frequency, and heart rate. Asthmatic children spend the same amount of time outdoors as other more healthy children but the time spent engaged in high levels of activity are less.

Estimates of activity level have been compiled based on questionnaire data. The National Human Activity Pattern Survey (NHAPS), a probability-based telephone survey, was conducted in the early 1990s. The survey concluded that outdoor work-related activities were highest during the springtime and were more frequent during the morning and early afternoon. Exercise/sports-related activities were highest from noon to 3 p.m. during the summer months. During the spring months, exercise/sports-related activities were highest from mid- to late afternoon (Klepeis et al., 1996, 2001). A pilot study by Gonzales et al. (2003) evaluated the use of retrospective questionnaires for reconstructing past time-activity and location pattern information. Ozone concentration estimates using ambient stationary monitors and estimates derived from diaries and questionnaires differed slightly. However, both estimates were greater than O_3 personal exposure measurements.

Existing comprehensive inhalation exposure models (NEM and pNEM) (Johnson, 2003), (MENTOR/SHEDS) Burke et al., 2001; McCurdy et al., 2000), and the Air Pollutants Exposure model (TRIM.Expo) treat human activity patterns as sequences of exposure events in which each event is defined by a geographic location and microenvironment and then assigned activity diary records from the CHAD (Consolidated Human Activities Database; www.epa.gov/chadnet1) (Glen et al., 1997; McCurdy, 2000; McCurdy et al., 2000). There are now about 22,600 person-days of sequential daily activity pattern data in CHAD representing all ages and both genders. The data for each subject consist of one or more days of sequential activities, in which each activity is defined by start time, duration, activity type (140 categories), and microenvironment classification (110 categories). Activities vary from 1 min to 1 h in duration. Activities longer than 1 h are subdivided into clock-hour durations to facilitate exposure modeling. A distribution of values for the ratio of oxygen uptake rate to body mass (referred to as metabolic equivalents or METs) is provided for each activity type listed. A table listing the activity patterns included in CHAD appears in AX3.

pNEM divides the population of interest into representative cohorts based on the combinations of demographic characteristics (age, gender, and employment), home/work district, and residential cooking fuel. TRIM.Expo and MENTOR/SHEDS generate a population demographic file containing a user-defined number of person-records for each census tract of the

population based on proportions of characteristic variables (age, gender, employment, and housing) obtained for the population of interest, and then assigns the matching activity information from CHAD to each individual record of the population based on the characteristic variables.

The TRIM.Expo model is capable of simulating individual movement through time and space to provide estimates of exposure to a given pollutant in various microenvironments (e.g., indoor, outdoor, and in-vehicle microenvironments). One of the key strengths of the TRIM.Expo model is its ability to estimate hourly exposures and doses for all simulated individuals in a sampled population. However, TRIM.Expo is limited in that uncertainties in the predicted distributions (e.g., age, activity data, commuting patterns, personal activities) have not been addressed.

MENTOR/SHEDS is capable of simulating individuals exposures in various microenvironments (outdoors, residence, office, school, store, restaurant, bar, and vehicles) using spatial concentration data for each census tract. The indoor and in-vehicle pollutant concentrations are calculated using specific equations for the microenvironment and ambient pollutant concentration relationship. Randomly selected characteristics for a fixed number of individual are selected to match demographics within the census tract for age, gender, employment status, and housing type. Smoking prevalence statistics by gender and age is randomly selected for each individual in the simulation. Diaries for activity patterns are matched for the simulated individual by demographic characteristics (Burke et al., 2001).

An important source of uncertainty in existing exposure modeling involves the creation of multiday, seasonal, or year-long exposure activity sequences based on 1- to 3-day activity data for any given individual from CHAD. Currently, appropriate longitudinal data are not available and the existing models use various rules to derive longer-term activity sequences utilizing 24-h activity data from CHAD.

Of the above models, only NEM/pNEM have been used extensively in O_3 exposure modeling. The pNEM probabilistic model builds on the earlier NEM deterministic exposure model. The model takes into consideration the temporal and spatial distribution of people and O_3 in the area of consideration, variations in O_3 concentrations in the microenvironment, and the effects of exercise-increased ventilation on O_3 uptake. There are three versions of the pNEM/O_3 model: (1) general population (Johnson et al., 1996a), (2) outdoor workers (Johnson et al., 1996b), and (3) outdoor children (Johnson et al., 1996c, 1997). The pNEM models have been

applied to nine urban areas and a summer camp. The models used activity data from the Cincinnati Activity Diary Study (CADS) along with time-activity data from several other studies. Data from stationary monitoring sites were used to estimate outdoor O_3 exposure. Indoor O_3 decay was assumed to be proportional to the indoor O_3 concentration. An algorithm assigned the EVR associated with each exposure event. The EVR for the outdoor children model was generated using a module based on heart rate data by Spier et al. (1992) and Linn et al. (1992).

Characterization of Exposure

The use of ambient air monitoring stations is the most common surrogate for assigning exposure in epidemiological studies. Since the primary source of O_3 exposure is the ambient air, monitoring concentration data would provide the exposure outdoors while exercising, a potential important exposure to evaluate in epidemiological studies. Monitored concentrations are useful for a relative assignment of exposure with time if the concentration were uniform across the region; the time-activity pattern were the same across the population; and the housing characteristics, such as ventilation rates and the O_3 sinks contributing to its indoor decay rates, were constant for the study area. Since these factors vary by population and location there will be errors in the magnitude of the total exposure and in the relative total exposure assignment based solely on ambient monitoring data.

Personal O_3 exposure measurements have been made for potentially susceptible populations (children, outdoor workers, the elderly, and individuals with chronic obstructive pulmonary disease). Children and outdoor workers have somewhat higher exposures than other individuals because they spend more time outdoors engaged in moderate and heavy exertion. Children are also more active outside and, therefore, have a higher minute ventilation rate than most adults (Klepeis et al., 1996, 2001). Available exposure studies suggest trends in exposure magnitude for some populations, however, additional exposure studies are needed to generalize differences in exposure between the general population and potentially susceptible populations. Table 3-3 summaries the findings of available exposure studies.

Table 3-3. Personal Exposure Concentrations

Location, Population, Sample Duration	n	Personal Exposure Mean[a] (range) (ppb)	Reference
San Diego, CA, Asthmatics ages 9-18 years, 12 hour	12	12 ± 12 (0-84) 10 weekend 12 weekday	Delfino et al. (1996)
Vancouver, Canada, Adult Workers, Daily	585		Brauer and Brook (1997)
High indoor time		(ND-9)	
Moderate indoor time		(ND-12)	
Only outdoor		(2-44)	
Southern California, Subjects 10-38 years	24		Liu et al. (1997)
Spring		13.6 ± 2.5 (- to 80)	
Fall		10.5 ± 2.5 (- to 50)	
Montpellier, France, Adults, Hourly	16	34.3 ± 17.6 (6.5-88)	Bernard et al. (1999)
Winter		15.4 ± 7.7 (6.5-40)	
Summer		44.1 ± 18.2(11-88)	
Souther California, Children 6-12 years,	169		Geyh et al. (2000)
≥ 6 days		6.2 ± 4.7 (0.5-41)	
Upland - winter		19 ± 18 (0.5-63)	
- summer		5.7 ± 4.2 (0.5-31)	
Mountain - winter		25 ± 24 (0.5-72)	
- summer			
Baltimore, MD, Technician, Hourly[b]	1		Chang et al. (2000)
Winter		3.5 ± 7.5 (ND-49)	
Summer		15 ± 18 (ND-76)	
Baltimore, MD, Adults 75 ± 7 years, Daily	20		Sarnat et al. (2000)
Winter		3.5 ± 3.0 (ND-9.9)	
Summer		0. ± 1.8 (ND-2.8)	

[a]ND = not detected.

[b]Measurements made following scripted activities for 15 days.

Ozone concentrations in various microenvironments under a variety of environmental conditions have been reported in the literature. In the absence of an indoor O_3 source, concentrations of O_3 indoors are lower than that found in the ambient air. Ozone concentrations in microenvironments were found to be primarily controlled by ambient O_3 concentrations and the AER: they increase with increasing AER. To a lesser extent, O_3 concentrations in microenvironments are influenced by the ambient temperature, time of day, indoor characteristics (e.g., presence of carpeting), and the presence of other pollutants in the microenvironment. Table 3-4 describes the findings of the available studies.

Factors Affecting Ozone Concentrations

Ozone and other photochemical oxidants are formed in the ambient air from the reaction of sunlight with vehicle emissions, gasoline fumes, solvent vapors, and power plant and industrial emissions (See Chapter 2 for a discussion of O_3 atmospheric chemistry). Ozone enters the indoor environment primarily through infiltration from outdoors through building components, such as windows, doors, and ventilation systems. There are also a few indoor sources of O_3 (photocopiers, facsimile machines, laser printers, and electrostatic air cleaners and precipitators) (Weschler, 2000). Generally O_3 emissions from office equipment and air cleaners are low except under improper maintenance conditions. Reported O_3 emissions from office equipment range from 1300 to 7900 μg/h (Leovic et al., 1996, 1998). Most air cleaners (particulate ionizers) emitted no or only a small amount (56 to 2757 μg/h) of O_3 during operation (Niu et al., 2001). Emissions from O_3 generators can range from tens to thousands of micrograms per hour (Weschler, 2000; U.S. Environmental Protection Agency, 1996).

Other photochemical oxidants (peroxyacyl nitrates; PAN and PPN) have no known direct emission sources indoors. PAN may be formed in the indoor environment from the reaction of the $OH\cdot$ or NO_3 with acetaldehyde to form the acetyl radical, CH_3CO (Grosjean et al., 1996). The acetyl radical then reacts with oxygen to for an acetylperoxy radical which reacts with NO_2 to form PAN. Peroxyacyl nitrates primarily occur in the indoor environment from infiltration through the building envelop and through openings in the building envelopment.

The concentration of O_3 in indoor environments is dependent on the outdoor O_3 concentration, the AER or outdoor infiltration, indoor circulation rate, and O_3 removal processes through contact with indoor surfaces and reactions with other indoor pollutants. Since O_3 concentrations are generally higher during the warmer months, indoor concentrations will likely be highest during that time period. (See earlier discussion on ambient concentrations of O_3.).

Air exchange rates vary depending on temperature differences, wind effects, geographical region, type of heating/mechanical ventilation system, and building type (Weschler and Shields, 2000; Colome et al., 1994). The balance of the flow of air in and out of a microenvironment is greatest in a residential building when a window or door is open (Johnson et al., 2004; Howard-Reed et al., 2002). The opening of windows or doors is dependent on the building occupancy, season, housing density, the presence of air conditioning, and wind speed (Johnson and Long, 2004). When windows and doors are closed, the dominant mechanism controlling AERs is

Table 3-4. Indoor/Outdoor Ozone Concentrations in Various Microenvironments

Location and Ventilation Conditions	Indoor/Outdoor Concentrations	Comments	Reference
New England States (9) Fall	20 ppb/40 ppb	Schools represented a variety of environmental conditions - varying ambient O_3 concentrations, sources, geographic locations, population density, traffic patterns, building types. Average O_3 concentrations were low in the morning and peaked during the early afternoon. O_3 concentrations averaged for all schools monitored.	NESCAUM (2002)
Mexico City, School Windows/Doors Open (27) Windows/Doors Closed Cleaner Off (41) Windows/Doors Closed Cleaner On (47)	0 to 247 ppb/ 64 to 361 ppb	Study conducted over 4 d period during winter months. Two-minute averaged measurements were taken both inside and outside of the school every 30 min from 10 a.m. to 4 p.m. Estimated air exchange rates were 1.1, 2.1, and 2.5 h^{-1} for low, medium, and high flow rates. Ozone concentrations decreased with increasing relative humidity.	Gold et al. (1996)
Mexico City Homes Schools	5 ppb/27ppb (7 d) 7 ppb/37 ppb (14 d) 22 ppb/56 to 733 ppb	Ozone monitoring occurred between September and July. Study included 3 schools and 145 homes. Most of the homes were large and did not have air conditioning. Ninety-two percent of the homes had carpeting, 13% used air filters, and 84% used humidifiers. Thirty-five percent opened windows frequently, 43% sometimes, and 22% never between 10 a.m. and 4 p.m. Ozone was monitored at the schools sites from 8 a.m. to 1 p.m. daily for 14 consecutive days. Homes were monitored for continuous 24-h periods for 7 and 14 consecutive days.	Romieu et al. (1998)
Boston, MA, Homes (9) Winter - continuously Summer - continuously	 0 to 20.4 ppb/4.4 to 24.5 ppb 0 to 34.2 ppb/8.2 to 51.8 ppb	Study examined the potential for O_3 to react with VOCs to form acid aerosols. Carbonyls were formed. No clear trend of O_3 with AERs. The average AER was 0.9 h^{-1} during the winter and 2.6 h^{-1} during the summer. Four residences in winter and nine in summer with over 24 h samples collected.	Reiss et al. (1995)
Los Angeles, Homes (239)	13 ppb/37 ppb	Four hundred and eighty-one samples collected inside and immediately outside of home from February to December. Concentrations based on 24-h average O_3 concentrations indoors and outdoors. Low outdoor concentrations resulted in low indoor concentrations. However, high outdoor concentrations resulted in a range of indoor concentrations.	Avol et al. (1998a)

Table 3-4 (cont'd). Indoor/Outdoor Ozone Concentrations in Various Microenvironments

Location and Ventilation Conditions	Indoor/Outdoor Concentrations	Comments	Reference
Burbank, CA Telephone Switching Station	0.2/21.1 ppb	Major source of O_3 was transport from outdoors. From early spring to late fall O_3 concentrations peaked during the early afternoon and approach zero at sunset. AER ranged from 1.0 to 1.9 h^{-1}.	Weschler et al. (1994)
Munich Germany		Indoor concentrations were dependent on the type of ventilation.	Jakobi and Fabian (1997)
Office	0.4/0.9 ppb		
Gymnasium	0.49/0.92 ppb		
Classroom	0.54/0.77 ppb		
Residence			
Bedroom	0.47/1.0 ppb		
Livingroom	0.74/1.0 ppb		
Montpellier, France, Homes (110)	15.5/32.0 ppb	Ozone measurements were made over 5-d periods in and outside of 21 homes during the summer and winter months. The winter I/O ratio was 0.31 compared to 0.46 during the summer months.	Bernard et al. (1999)
Southern CA, Homes		Ozone measurements were taken at 119 homes (57 in Upland and 62 in towns located in the mountains) during April and May. Concentrations were based on average monthly outdoor concentrations and average weekly indoor concentrations. Indoor based on the home location, number of bedrooms, and the presence of an air conditioner.	Geyh et al. (2000) Lee et al. (2002)
Upland	11.8/48.2 ppb		
Mountains	2.8/35.7 ppb		
Krakow, Poland, Museums		Ozone continuously monitored at five museums and cultural centers. Monitoring conducted during the summer months for 21 to 46 h or 28 to 33 days at each of the sites. The indoor concentration was found to be dependent on the ventilation rate, i.e., when the ventilation rate was high the indoor O_3 concentrations approached that of ambient O_3. Rooms sequestered from the outdoor air or where air was predominantly recycled through charcoal filters the O_3 levels indoors were greatly reduced.	Salmon et al. (2000)
Cloth Hall	3.2/25.7-27.4 ppb		
Matejko	8.5/20.0 ppb		
Wawel Castle	2.5/14.7 ppb		
National	1.5/11.0 ppb		
Buildings, Greece		There was no heating/air conditioning system in the building at Thessaloniki. Windows were kept closed during the entire monitoring period. Complete air exchange took place every 3 h. The air conditioning system in continuous use at the Athens site recirculated the air. Complete air exchange was estimated to be 1 h. Monitoring lasted for 30 days at each site but only the 7 most representative days were used.	Drakou et al. (1995)
Thessalonki	9.39/15.48 ppb		
Athens	8.14/21.66 ppb		

Table 3-4 (cont'd). Indoor/Outdoor Ozone Concentrations in Various Microenvironments

Location and Ventilation Conditions	Indoor/Outdoor Concentrations	Comments	Reference
Patrol cars, NC	11.7/28.3 ppb	Patrol cars were monitored Mon. through Thurs. between the hours of 3 p.m. to midnight on 25 occasions during the months of Aug., Sept., and Oct. Outdoor O_3 concentrations were taken from ambient monitoring station. Air inside the patrol car was recirculated cool air.	Riediker et al. (2003)
University of CA Photocopy room	<20 to 40 ppb/—	Room volume was 40 m^3. Ozone concentrations increased proportionately with increasing use of photocopier.	Black et al. (2000)
Home/office O_3 generators	14 to 200 ppb/—	Room volume was 27 m^3. Doors and windows were closed. Heating/air conditioning and mechanical ventilation systems were off. Ozone generator was operated for 90 min. High O_3 concentrations noted when O_3 generator used at high setting. AER was 0.3 h^{-1}.	Steiber et al. (1995)

infiltration through unintentional openings in the building envelope. Williams et al. (2003a, 2003b) reported AERs of 0.001 to 4.87 h^{-1} in 37 homes in Research Triangle Park, NC. Chan et al. (2005) compared air leakage measurements for 70,000 houses. Older and smaller houses had higher normalized leakage areas than newer and larger houses. Meng et al. (2004) also attributed higher AERs to the age of the housing stock. AERs for homes in Houston, TX and Elizabeth, NJ were averaged for all four seasons, the highest AER, 1.22 h^{-1}, was noted for homes in Elizabeth, NJ where the homes were older. Evaluations of AERs for residential structures was reported by Murray and Burmaster (1995) and includes AERs for 2,844 residential structures in four different climatic regions by season (winter, spring, summer, and fall). The AER for all seasons across all regions was 0.76 h^{-1} (arithmetic mean) (Region 1: IN, MN, MT, NH, NY1, VT, WI; Region 2: CO, CT, IL, NJ, NY2, OH, PA, WA; Region 3: CA3, MD, OR, WA; Region 4: AZ, CA4, FL, TX). The AERs were generally higher during the warm seasons, when ambient O_3 concentrations are highest. Data for the warmest region during the summer months may not be representative of all homes because measurements were made in southern California where windows are open and air conditioning is not used.

Average mean (median) AERs of 2.45 (2.24), 1.35 (1.09), and 2.22 (1.79) h^{-1} were reported by Lagus Applied Technology, Inc. (1995) for schools, offices, and retail establishments in California. Mean AERs for schools, offices, and retail establishments in Oregon and Washington were 0.32, 0.31, and 1.12 h^{-1} (Turk et al., 1989)—considerably less than that reported by Lagus Applied Technology. Park et al. (1998) reported mean AERs ranging from 1.0 to 47.5 h^{-1} for stationary vehicles under varying ventilating conditions. Where available, AERs for other studies are included in Table 3-10.

The most important removal process for O_3 in the indoor environment is deposition on and reaction with indoor surfaces. The rate of deposition is material-specific. The removal rate will depend on the indoor dimensions, surface coverings, and furnishings. Smaller rooms generally have larger surface-to-volume ratio (A/V) and remove O_3 faster than larger rooms. Fleecy materials, such as carpets, have larger surface-to-volume ratios and remove O_3 faster than smooth surfaces (Weschler, 2000). However, the rate of O_3 reaction with carpet diminishes with cumulative O_3 exposure (Morrison and Nazaroff, 2000, 2002). Weschler (2000) compiled the O_3 removal rates for a variety of microenvironments. Generally, the removal rates ranged between 3.0 and 4.3 k_d $(A/V)/h^{-1}$. The highest removal rate, 7.6 k_d $(A/V)/h^{-1}$, was noted for a clean room (Weschler et al, 1989).

Ozone chemical reactions in the indoor environment are analogous to those reactions occurring in the ambient air (See discussion on atmospheric chemistry in Chapter 2). Ozone reacts with unsaturated VOCs in the indoor environment, primarily terpenes or terpene-related compounds from cleaning products, air fresheners, and wood products. The reactions are dependent on the O_3 indoor concentration, the indoor temperature and, in most cases, the air exchange rate/ventilation rate. Some of the reaction products may more negatively impact human health and artifacts in the indoor environment than their precursors (Wolkoff et al., 1999; Wilkins et al., 2001; Weschler et al., 1992; Weschler and Shields, 1997; Rohr et al., 2002; Nøjgaard et al., 2005). Primary reaction products are Criegee biradicals, nitrate radicals, and peroxyacetyl radicals. Secondary reaction products are hydroxy, alkyl, alkylperoxy, hydroperoxy, and alkoxy radicals. Reactions with alkenes can produce aldehydes, ketones, and organic acids (Weschler and Shields, 2000; Weschler et al., 1992).

Hydroxyl radicals formed from the reaction of O_3 with VOCs, nitric oxide and hydroperoxy, and other intermediate products can react with various nitrogen compounds, sulfur dioxide, carbon monoxide and other compounds to produce significantly more toxic compounds (Sarwar et al., 2002; Orzechowska and Paulson, 2002; Fick et al., 2003, 2004; Van den Bergh et al., 2000; Fan et al., 2003; Wilkins et al., 2001; Clausen et al., 2001; Rohr et al., 2002, 2003; Poupard et al., 2005; Blondeau et al., 2005). The reaction between O_3 and terpenes also has been shown to increase the concentration of indoor particles (Weschler and Shields, 1999, 2003; Weschler, 2004; Clausen et al., 2001; Fan et al, 2003; Wainman et al., 2000), possibly from further reactions of the hydroxy radical with terpenes (Sarwar et al., 2002).

Decomposition and formation of PAN in the indoor environment are influenced by NO_2 and NO. Decomposition of PAN is expected to be a relatively fast process when indoor O_3 levels are low and when motor vehicle emissions are large or there is an indoor source of NO_x (Weschler and Shields, 1997).

Factors Affecting the Relationship between Ambient Concentrations and Personal Exposures to O_3

Ambient O_3 concentrations vary with the time of day, season of the year, and among locations. Personal exposure to O_3 is influenced by the microenvironmental concentration and the amount of time spent in each microenvironment. Because the majority of the population spends on average nearly 90% of their time in an indoor microenvironment, the majority of the

O_3 exposure will occur in the indoor environment. Since there are few indoor sources of O_3, O_3 ambient concentration may be the most important factor that affects average population exposure in the indoor environment.

Indoor O_3 concentrations also are affected by several other factors and mechanisms. Studies have shown that in addition to the ambient O_3 concentrations, indoor O_3 concentrations are influenced by the air exchange rate or outdoor infiltration, increasing with increasing air exchange. Once indoors, the O_3 concentration is affected by the indoor circulation rate and O_3 removal through contact with indoor surfaces and reactions with other indoor pollutants.

In some instances, ambient O_3 monitors are located in areas outside the breathing zone. Studies on the effect of elevation on O_3 concentrations found that concentrations increased with increasing elevation (Väkevä et al., 1999; Johnson, 1997). Also, since O_3 monitors are frequently located on rooftops in urban settings, the concentrations measured there may overestimate the exposure to individuals outdoors in streets and parks, locations where people exercise and their maximum O_3 exposure is more likely to occur.

In epidemiologic studies investigating acute and chronic health outcomes using ambient monitoring data from stationary monitoring sites, O_3 exposure assessment was affected by the distance between home and the monitoring site, gender, time-activity patterns (e.g., percentage of time spent outdoors, type of outdoor activity, time of day during outdoor activity), and indoor air exchange rates (e.g., ventilation conditions, home characteristics) (Geyh et al., 2000; Lee et al., 2002, 2004; Liu et al., 1995, 1997; Chang et al., 2000; Chan et al., 2005; O'Neill et al., 2003; Brauer and Brook, 1997; O'Neill et al., 2003). People that work outdoors tend to be exposed to higher levels of O_3 (Brauer and Brook, 1997; O'Neill et al., 2003). Geyh et al. (2000) observed higher indoor and personal O_3 concentrations in a southern California community with 2% air-conditioned homes compared to a community with 93% air-conditioned homes during the summer (high O_3) months, but showed no difference in O_3 levels during the winter (low O_3) months. Lee et al. (2004) observed that personal O_3 exposure was positively correlated with outdoor time ($r = 0.19$, $p < 0.01$) and negatively correlated with indoor time ($r = -0.17$, $p < 0.01$). Additional factors that affected indoor O_3 levels were air conditioning, window fans, and window opening. The O_3 exposure assessment study by Liu et al. (1995) found that after adjusting for time spent in various indoor and outdoor microenvironments (e.g., car with windows open, car with windows closed, school, work, home, outdoors near home, outdoors

other than near home), mean 12-hour ambient O_3 concentrations explained 32% of the variance in personal exposure in the summer.

In a southern California study by Avol et al. (1998b), boys were found to spend more time outdoors and be more physically active than girls. Another southern California study found that boys were outdoors 30 minutes longer than girls, and had higher personal O_3 exposure during both high and low O_3 months (Geyh et al., 2000).

The announcement of smog alerts or air quality indices may influence personal exposures to O_3 by causing individuals to alter behaviors (avoidance behavior). Neidell (2004), in his evaluation of the effect of pollution on childhood asthma, examined the relationship between the issuance of smog alerts or air quality indices for several counties in California and hospital admissions for asthma in children under age 18 years (not including newborns). Smog alerts are issued in California on days when O_3 concentrations exceed 200 ppb. There was a significant reduction in the number of asthma-related hospital admissions in children ages 1 to 12 years on smog alert days, indicating that avoidance behavior might be present on days of high O_3 concentrations. Changes in population behavior as a function of concentration complicate the estimation of health effects from population-based studies; thus, it may be desirable to include sensitivity analyses that eliminate high O_3 days, particularly in areas where avoidance behavior is expected.

Potential Sources of Error Resulting from the Use of Ambient Ozone Concentrations in Epidemiological Analyses

There is no clear consensus among exposure analysts as to how well stationary monitor measurements of ambient O_3 concentrations represent a surrogate for personal O_3 exposure. The approaches available for assessing exposure in air pollution epidemiology studies, the microenvironmental (indirect) approach and the personal sampling (direct) approach (Navidi et al., 1999; Ott, 1982, 1985), are associated with measurement error. To determine personal exposure using the microenvironmental approach, the concentrations of the various microenvironments are multiplied by the time spent in each microenvironment. Both the concentration and time component contribute to the measurement error. There is no time component to the measurement error in the personal sampling approach, however, the estimation of exposure using personal monitoring devices contributes to measurement error, especially in the case of O_3. Passive badges are commonly used for monitoring O_3 integrated personal

exposure. Their sensitivity to wind velocity, badge placement, and interference with other copollutants may result in measurement error.

Results from the error analysis models developed by Navidi et al. (1999) indicated that neither the microenvironmental nor personal sampling approach gave reliable health effect estimates when measurement errors were uncorrected. The nondifferential measurement error biased the effect estimates toward zero under the model assumptions. However, if the measurement error was correlated with the health response, a bias away from the null could result. The use of central ambient monitors to estimate exposure also biased the estimates toward the null. Since most people spend the majority of their time indoors, where O_3 levels tend to be much lower than outdoor ambient levels, using ambient concentrations to determine exposure generally overestimates true personal O_3 exposure, resulting in effect estimates biased toward the null.

Several studies have examined the relationship between measured ambient O_3 concentrations from fixed monitoring sites and personal O_3 exposure (Avol et al., 1998a; Brauer and Brook, 1995, 1997; Chang et al., 2000; Delfino et al., 1996; Lee et al., 2004; Liard et al., 1999; Linn et al., 1996; Liu et al., 1995, 1997; O'Neill et al., 2003; Sarnat et al., 2001). In a Baltimore, MD study of older adults, individuals with COPD, and children, 24-h average ambient O_3 concentrations from a monitoring site were not found to be significantly associated with personal O_3 exposure (Sarnat et al., 2001). The mixed regression effect estimates were $\beta = 0.01$ ($t = 1.21$) and $\beta = 0.00$ ($t = 0.03$), for summer and winter, respectively. Chang et al. (2000) compared one-hour personal and ambient O_3 measurements in older adults in various microenvironments using activity data from the National Human Activity Pattern Survey study (Klepeis, 1999). There was no correlation between personal and ambient O_3 concentrations in the indoor residence ($r = 0.09$ and $r = 0.05$, for summer and winter, respectively), although a moderate correlation was found in other indoor environments such as restaurants, hospitals, and shopping malls ($r = 0.34$ in summer, $r = 0.46$ in winter). In comparison, the correlation in outdoor environments (near and away from roads) was moderate to high ($0.68 \leq r \leq 0.91$) and statistically significant. Slopes for the relationship between personal and ambient O_3 concentrations were not reported in this study.

Brauer and Brook (1995, 1997) observed that the daily averaged personal O_3 measurements and ambient concentrations were well-correlated after stratifying groups by time spent outdoors. Clinic workers ($n = 25$; 24-hour samples), teenage camp counselors ($n = 25$; 24- hour samples),

and farm workers (n = 15; 6-14 h work shift samples) spent 0 to 25%, 7.5 to 45%, and 100% of their monitored time outdoors, respectively. The personal to ambient O_3 concentration ratios were significantly different for the clinic workers (0.28) and farm workers (0.96). Ambient O_3 concentrations and time spent outdoors explained more of the variability in the personal O_3 measurements for outdoor farm workers compared to the clinical workers. However, the Spearman correlation coefficients were comparable, 0.60 and 0.64 for the clinic workers and farm workers, respectively, indicating that the variability of nonambient O_3 exposures was similar in the two groups. A study by O'Neill et al. (2003) examined 107 pairs of ambient and personal O_3 measurements from 39 outdoor workers in Mexico City using a longitudinal analysis method. Two to seven personal measurements were collected on each of the 26 monitoring days, which were averaged then compared with the ambient concentrations. They estimated that a 1 ppb increase in ambient O_3 concentration was associated with a 0.56 ppb (95% CI: 0.43, 0.69) increase in personal O_3 concentration. In a Paris, France study by Liard et al. (1999), adults (n = 55) and children (n = 39) wore passive O_3 monitors for 4 consecutive days during three periods. For each period, all adults wore the O_3 monitors over the same 4 days. Likewise, all children wore monitors over the same 4 days for each of the three periods, but on different days from the adults. The ambient O_3 concentrations from the stationary monitoring sites did not explain a high percentage of the variance of personal O_3 exposure (nonsignificant [value not stated] in adults and 21% in children). However, when personal measurements from all subjects were aggregated for each of the six periods, the 4-day mean personal O_3 exposure was found to be highly correlated with the corresponding mean ambient concentration ($r = 0.83$, $p < 0.05$). Similarly, a study of Los Angeles school children by Linn et al. (1996) found that daily 24-h average ambient O_3 concentrations from a central site were well-correlated ($r = 0.61$) with daily averaged personal O_3 exposures.

The low correlation observed between personal O_3 exposures and ambient O_3 concentrations in the study by Sarnat et al. (2001) suggests that O_3 concentrations measured at central ambient monitors do not explain the variance of individual personal exposures. However, daily averaged personal exposures from the aggregate population have been found to be correlated with monitored ambient O_3 concentrations, which is of greater relevance in time-series studies. Although there are correlations between aggregate personal and monitored ambient O_3 concentrations, the absolute personal concentrations may be considerably lower than the monitored ambient O_3 concentrations.

In summary, results indicate that the relationship between ambient O_3 concentrations and personal exposure will vary depending on factors such as O_3 concentrations, time spent in the various microenvironments, and activity levels, creating potential measurement errors. The expectations based on statistical modeling considerations are that these exposure measurement errors or uncertainties will reduce the statistical power of the O_3 health effects analysis, making it difficult to detect a true underlying association between the correct exposure metric and the health outcome studied. However, until more data on O_3 exposure become available, the use of monitored ambient O_3 concentrations as a surrogate for exposures is not expected to change the principal conclusions from O_3 epidemiologic studies using community average health and pollution data.

Exposure to Related Photochemical Oxidants

A variety of related photochemical oxidants produced outdoors, such as PAN and peroxypropionyl nitrate (PPN), can infiltrate into indoor environments. These compounds are thermally unstable and decompose to peroxacetyl radicals and NO_2. Exposure to related photochemical oxidants has not been measured, nor are these compounds routinely monitored at stationary monitoring sites. Available monitored concentrations of related photochemical oxidants may be found in Annex AX3.

3.9 SUMMARY OF KEY POINTS

The median of the daily maximum 8-h O_3 concentration averaged over May to September is about 0.049 ppm from 2000 to 2004. The daily maximum 1-h O_3 concentrations could have been much higher in large urban areas or in areas downwind of large urban areas. For example, in Houston, TX, the daily maximum 1-h O_3 concentrations have approached 0.20 ppm during this period.

Daily maximum 8-h average O_3 concentrations are lower than the maximum 1-h O_3 concentrations, but they are highly correlated. Within individual MSAs, O_3 concentrations tend to be well correlated across monitoring sites. However, there can be substantial variations in O_3 concentrations. Ozone in city centers tends to be lower than in regions either upwind or downwind because of titration by NO emitted by motor vehicles.

Ozone concentrations tend to peak in early- to mid-afternoon in areas where there is strong photochemical activity and later in the day in areas where transport is more important in determining the O_3 abundance. Summertime maxima in O_3 concentrations occur in areas in the United States where there is substantial photochemical activity involving O_3 precursors emitted from human activities. Monthly maxima can occur anytime from June through August. However, springtime maxima are observed in national parks, mainly in the western United States and at a number of other relatively unpolluted monitoring sites throughout the Northern Hemisphere. For example, the highest O_3 concentrations at Yellowstone NP tend to occur during April and May. Generally, monthly minima O_3 concentrations tend to occur from November through February at polluted sites and during the fall at relatively remote sites.

Nationwide, daily maximum 8-h O_3 concentrations have decreased at the upper end of the distribution from 1990 to 2004. However, the daily maximum 8-h O_3 concentrations toward the center of the distribution have not reflected these changes. Trends have not been consistent at national park sites; with downward trends observed at some sites and upward or no trends observed at others. At some sites, trends reversed direction in going from the 98th to the 95th percentile values.

Sufficient data are not available for other atmospheric oxidants (e.g., H_2O_2, PAN) and oxidation products (e.g., HNO_3, H_2SO_4) to relate concentrations of O_3 to these species for use in time series studies. Data for these species are only obtained as part of specialized field studies. In general, secondary species, such as HNO_3, H_2SO_4, H_2O_2, and PAN, are expected to be at least moderately correlated with O_3. On the other hand, primary species are expected to be more highly correlated with each other than with secondary species, provided that the primary species originate from common sources. Concentrations of other oxidants are much lower than for O_3 and range from $\leq$1% for oxidants in particles to several percent for gas phase species. The relationship of O_3 to $PM_{2.5}$ is complex, because PM is not a distinct chemical species but is a mix of primary and secondary species. $PM_{2.5}$ concentrations were positively correlated with O_3 during summer, but negatively correlated with O_3 during winter at Ft. Meade, MD. PM_{10} concentrations show similar relations with O_3.

Co-occurrences of O_3 (defined when both pollutants are present at an hourly average concentration of $\geq$0.05 ppm) with NO_2 and SO_2 are rare. For example, there were fewer than 10 co-occurrences with either NO_2 or SO_2 in 2001. The number of co-occurrences for O_3 and $PM_{2.5}$ (defined as an hourly average O_3 concentration $\geq$0.05 ppm and a 24-h average $PM_{2.5}$

concentration ≥40 μg/m^3 occurring during the same 24-h period) also tended to be infrequent (<10 times) at most sites, but there were up to 20 such co-occurrences at a few sites.

Policy relevant background O_3 concentrations are used for assessing risks to human health associated with O_3 produced from anthropogenic sources in continental North America. Because of the nature of the definition of PRB concentrations, they cannot be directly derived from monitored concentrations, instead they must be derived from modeled estimates. Current model estimates indicate that ambient air PRB concentrations in the United States are generally 0.015 ppm to 0.035 ppm. They decline from spring to summer and are generally <0.025 ppm under conditions conducive to high O_3 episodes. However, PRB concentrations can be higher, especially at elevated sites during spring, due to enhanced contributions from hemispheric pollution and stratospheric exchange.

Ozone exposure changes as a function of time of day, season, and microenvironment. Ambient O_3 concentrations are generally higher during warmer seasons and during the weekday, peaking during the later portion of the day. Ozone concentrations in indoor microenvironments are generally lower than those concentrations encountered in the ambient air. There are few indoor sources of O_3. Ozone occurs in indoor microenvironments primarily through infiltration through the building envelop and through windows, doors, and ventilation systems. The indoor O_3 concentration is dependent on the outdoor concentration, the AER, indoor circulation rate, and removal processes. Consequently, measured and modeled exposures should take into consideration O_3 diurnal weekly and seasonal variability and varying microenvironmental concentrations.

Once indoors, O_3 reacts with indoor surfaces, including surface coverings and furnishings. Ozone also will react with VOCs in indoor environments, primarily terpenes or terpene-related compounds. Ozone reactions with pollutants indoors are analogous to those reactions occurring in the ambient air, potentially exposing subjects to compounds significantly more toxic than O_3. The reaction products include Criegee biradicals, nitrate radicals, peroxyacetyl radicals, and hydroxy, alkyl, alkyperoxy, hydroperoxy, and alkoxy radicals. The hydroxy radical will react with various nitrogen compounds, sulfur dioxide, carbon monoxide, and other compounds. The formation of submicron particles has been attributed to the reaction of O_3 and the hydroxy radical with terpene and terpene-related compounds.

The available approaches for measuring personal O_3 exposure include the direct approach, using a PEM, and the indirect approach, which measures or models exposure in the

microenvironments the individual encounters. Both approaches are associated with measurement errors.

There are difficulties in identifying chemical trapping agents for PEMs that can react with O_3, and PEMs are sensitive to wind velocity, badge placement, and interference with other copollutants. Some studies using PEMs have shown personal O_3 exposures below those concentrations measured at stationary monitoring sites, while other studies have found strong correlations between O_3 measured at stationary monitoring sites and personal monitored concentrations.

The use of measured O_3 concentrations from stationary ambient monitoring sites as surrogates for personal exposure may be affected by the O_3 ambient concentration, percentage of time spent outdoors, and type of outdoor activity. Epidemiologic studies investigating health outcomes using data from stationary monitoring sites found O_3 exposure to be affected by the distance between the subjects' location and the stationary monitor, individual activity patterns, and the O_3 concentration in the microenvironment.

The use of exposure models to evaluate O_3 exposure to large populations over time is complicated by the fact that O_3 is a secondary pollutant with complex nonlinear and multiscale dynamics in space and time. The existing comprehensive inhalation exposure models (NEM, pNEM, MENTOR/SHEDS, TRIM.Expo) treat human activity patterns as sequences of exposure events. Estimates of activity levels are assigned from CHAD, the Consolidated Human Activities Database.

Ambient O_3 concentrations are estimated using emissions-based mechanistic models or ambient-data-based models. Models for estimating microenvironmental concentrations include the empirical, mass balance, and detailed CFD models. Mass balance modeling is the most common modeling approach to estimating concentrations in enclosed microenvironments. The pNEM/O_3 population exposure model, the model used more extensively in O_3 exposure modeling, includes a sophisticated mass balance model for indoor and vehicle microenvironments. There are three versions of the pNEM/O_3 model: the general population, outdoor workers, and outdoor children.

Results from O_3 exposure studies indicate that the relationship between ambient O_3 concentrations and personal exposure/dose will vary depending on O_3 concentrations and time spent in the various microenvironments, particularly the time spent outdoors where O_3 concentrations tend to be higher, and the personal activity level. Consequently, the O_3

exposure/dose may differ from the concentrations measured at stationary monitoring sites. However, until more data on O_3 exposure become available, the use of monitored ambient O_3 concentrations as a surrogate for exposures is not expected to change the principal conclusions from O_3 epidemiologic studies using community average health and pollution data.

REFERENCES

Adams, W. C. (1993) Measurement of breathing rate and volume in routinely performed daily activities [final report]. Sacramento, CA: California Environmental Protection Agency, Air Resources Board; contract no. A033-205.

Altshuller, A. P.; Lefohn, A. S. (1996) Background ozone in the planetary boundary layer over the United States. J. Air Waste Manage. Assoc. 46: 134-141.

Avol, E. L.; Navidi, W. C.; Colome, S. D. (1998a) Modeling ozone levels in and around southern California homes. Environ. Sci. Technol. 32: 463-468.

Avol, E. L.; Navidi, W. C.; Rappaport, E. B.; Peters, J. M. (1998b) Acute effects of ambient ozone on asthmatic, wheezy, and healthy children. Cambridge, MA: Health Effects Institute; research report no. 82.

Bernard, N. L.; Gerber, M. J.; Astre, C. M.; Saintot, M. J. (1999) Ozone measurement with passive samplers: validation and use for ozone pollution assessment in Montpellier, France. Environ. Sci. Technol. 33: 217-222.

Black, D. R.; Harley, R. A.; Hering, S. V.; Stolzenburg, M. R. (2000) A new, portable, real-time monitor. Environ. Sci. Technol. 34: 3031-3040.

Blondeau, P.; Iordache, V.; Poupard, O.; Genin, D.; Allard, F. (2005) Relationship between outdoor and indoor air quality in eight French schools. Indoor Air 15: 2-12.

Brauer, M.; Brook, J. R. (1995) Personal and fixed-site ozone measurements with a passive sampler. J. Air Waste Manage. Assoc. 45: 529-537.

Brauer, M.; Brook, J. R. (1997) Ozone personal exposures and health effects for selected groups residing in the Fraser Valley. In: Steyn, D. G.; Bottenheim, J. W., eds. The Lower Fraser Valley Oxidants/Pacific '93 Field Study. Atmos. Environ. 31: 2113-2121.

Burke, J. M.; Zufall, M. J.; Özkaynak, H. (2001) A population exposure model for particulate matter: case study results for $PM_{2.5}$ in Philadelphia, PA. J. Exposure Anal. Environ. Epidemiol. 11: 470-489.

Chan, C.-C.; Wu, T.-H. (2005) Effects of ambient ozone exposure on mail carriers' peak expiratory flow rates. Environ. Health Perspect. 113: 735-738.

Chang, L.-T.; Koutrakis, P.; Catalano, P. J.; Suh, H. H. (2000) Hourly personal exposures to fine particles and gaseous pollutants—results from Baltimore, Maryland. J. Air Waste Manage. Assoc. 50: 1223-1235.

Chen, L.-W. A. (2002) Urban fine particulate matter: chemical composition and possible origins (dissertation). College Park, MD: University of Maryland, Department of Chemical Physics. Available from: University Microfilms, Ann Arbor, MI; AADAA-I3078297.

Christakos, G.; Vyas, V. M. (1998a) A composite space/time approach to studying ozone distribution over eastern United States. Atmos. Environ. 32: 2845-2857.

Christakos, G.; Vyas, V. M. (1998b) A novel method for studying population health impacts of spatiotemporal ozone distribution. Soc. Sci. Med. 47: 1051-1066.

Clausen, P. A.; Wilkins, C. K.; Wolkoff, P.; Nielsen, G. D. (2001) Chemical and biological evaluation of a reaction mixture of R-(+)-limonene/ozone: formation of strong airway irritants. Environ. Int. 26: 511-522.

Code of Federal Regulations. (2000) Appendix D to part 58—Network design for state and local air monitoring stations (SLAMS), national air monitoring stations (NAMS), and photochemical assessment monitoring stations (PAMS). C. F. R. 40: pt. 58, app. D.

Colome, S. D.; Wilson, A. L.; Tian, Y. (1994) California residential indoor air quality study. Volume 2. Carbon monoxide and air exchange rate: an univariate and multivariate analysis. Chicago, IL: Gas Research Institute; report no. GRI-93/0224.3.

Cooper, O. R.; Moody, J. L. (2000) Meteorological controls on ozone at an elevated eastern United States regional background monitoring site. J. Geophys. Res. [Atmos.] 105: 6855-6869.

Delfino, R. J.; Coate, B. D.; Zeiger, R. S.; Seltzer, J. M.; Street, D. H.; Koutrakis, P. (1996) Daily asthma severity in relation to personal ozone exposure and outdoor fungal spores. Am. J. Respir. Crit. Care Med. 154: 633-641.

Drakou, G.; Zerefos, C.; Ziomas, I. (1995) A preliminary study on the relationship between outdoor and indoor air pollution levels. Fresenius' Environ. Bull. 4: 689-694.

Ebel, A.; Hass, H.; Jakobs, J. H.; Laube, M.; Memmesheimer, M.; Oberreuter, A. (1991) Simulation of ozone intrusion caused by a tropopause fold and cut-off low. Atmos. Environ. Part A 25: 2131-2144.

Fan, Z.; Lioy, P.; Weschler, C.; Fiedler, N.; Kipen, H.; Zhang, J. (2003) Ozone-initiated reactions with mixtures of volatile organic compounds under simulated indoor conditions. Environ. Sci. Technol. 37: 1811-1821.

Federal Register. (1986) Guidelines for estimating exposures. F. R. (September 24) 51: 34,042-34,054.

Fick, J.; Pommer, L.; Nilsson, C.; Andersson, B. (2003) Effect of OH radicals, relative humidity, and time on the composition of the products formed in the ozonolysis of α-pinene. Atmos. Environ. 37: 4087-4096.

Fick, J.; Nilsson, C.; Andersson, B. (2004) Formation of oxidation products in a ventilation system. Atmos. Environ. 38: 5895-5899.

Fiore, A. M.; Jacob, D. J.; Bey, I.; Yantosca, R. M.; Field, B. D.; Fusco, A. C.; Wilkinson, J. G. (2002) Background ozone over the United States in summer: origin, trend, and contribution to pollution episodes. J. Geophys. Res. (Atmos.) 107(D15): 10.1029/2001JD000982.

Fiore, A.; Jacob, D. J.; Liu, H.; Yantosca, R. M.; Fairlie, T. D.; Li, Q. (2003) Variability in surface ozone background over the United States: implications for air quality policy. J. Geophys. Res. (Atmos.) 108(D24): 10.1029/2003JD003855.

Fitz-Simons, T.; McCluney, L.; Rizzo, M. (2005) Analysis of 2004 ozone data for the ozone NAAQS review [memorandum to Dr. Joseph Pinto]. Research Triangle Park, NC: U.S. Environmental Protection Agency; August 22.

Freijer, J. I.; Bloemen, H. J. T. (2000) Modeling relationships between indoor and outdoor air quality. J. Air Waste Manage. Assoc. 50: 292-300.

Fusco, A. C.; Logan, J. A. (2003) Analysis of 1970-1995 trends in tropospheric ozone at Northern Hemisphere midlatitudes with the GEOS-CHEM model. J. Geophys. Res. (Atmos.) 108: 10.1029/2002JD002742.

Garratt, J. R. (1992) The atmospheric boundary layer. Cambridge, United Kingdom: Cambridge University Press. (Houghton, J. T.; Rycroft, M. J.; Dessler, A. J., eds. Cambridge atmospheric and space science series).

Georgopoulos, P. G.; Arunachalam, S.; Wang, S. (1997a) Alternative metrics for assessing the relative effectiveness of NO_x and VOC emission reductions in controlling ground-level ozone. J. Air Waste Manage. Assoc. 47: 838-850.

Georgopoulos, P. G.; Walia, A.; Roy, A.; Lioy, P. J. (1997b) Integrated exposure and dose modeling and analysis system. 1. Formulation and testing of microenvironmental and pharmacokinetic components. Environ. Sci. Technol. 31: 17-27.

Geyh, A. S.; Wolfson, J. M.; Koutrakis, P.; Mulik, J. D.; Avol, E. L. (1997) Development and evaluation of a small active ozone sampler. Environ. Sci. Technol. 31: 2326-2330.

Geyh, A. S.; Roberts, P. T.; Lurmann, F. W.; Schoell, B. M.; Avol, E. L. (1999) Initial field evaluation of the Harvard active ozone sampler for personal ozone monitoring. J. Exposure Anal. Environ. Epidemiol. 9: 143-149.

Geyh, A. S.; Xue, J.; Özkaynak, H.; Spengler, J. D. (2000) The Harvard Southern California chronic ozone exposure study: assessing ozone exposure of grade-school-age children in two southern California communities. Environ. Health Perspect. 108: 265-270.

Glen, G.; Lakkadi, Y.; Tippett, J. A.; del Valle-Torres, M. (1997) Development of NERL/CHAD: the National Exposure Research Laboratory consolidated human activity database. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Research and Development; contract no. 68-D5-0049.

Gold, D. R.; Allen, G.; Damokosh, A.; Serrano, P.; Hayes, C.; Castillejos, M. (1996) Comparison of outdoor and classroom ozone exposures for school children in Mexico City. J. Air Waste Manage. Assoc. 46: 335-342.

Gonzales, M.; Ngo, L.; Hammond, S. K.; Tager, I. (2003) Validation of a questionnaire and microenvironmental model for estimating past exposures to ozone. Int. J. Environ. Health Res. 13: 249-260.

Grosjean, D. (2003) Ambient PAN and PPN in southern California from 1960 to the SCOS97-NARSTO. Atmos. Environ. 37(suppl. 2): S221-S238.

Grosjean, E.; Grosjean, D.; Fraser, M. P.; Cass, G. R. (1996) Air quality model evaluation data for organics: 3. Peroxyacetyl nitrate and peroxypropionyl nitrate in Los Angeles air Environ. Sci. Technol. 30: 2704-2714.

Heuss, J. M.; Kahlbaum, D. F.; Wolff, G. T. (2003) Weekday/weekend ozone differences: what can we learn from them? J. Air Waste Manage. Assoc. 53: 772-788.

Hirsch, A. I.; Munger, J. W.; Jacob, D. J.; Horowitz, L. W.; Goldstein, A. H. (1996) Seasonal variation of the ozone production efficiency per unit NOx at Harvard Forest, Massachusetts. J. Geophys. Res. [Atmos.] 101: 12,659-12,666.

Horowitz, L. W.; Liang, J.; Gardner, G. M.; Jacob, D. J . (1998) Export of reactive nitrogen from North America during summertime: sensitivity to hydrocarbon chemistry. J. Geophys. Res. [Atmos.] 103(D11): 13451-13476.

Horváth, L.; Nagy, Z.; Weidinger, T.; Artz, R.; Luke, W. T.; Valigura, R.; Pinto, J. P.; Womack, J. (1995) Measurement of fluxes of trace gases (O_3, NO_x, SO_2, CO_2, HNO_3), particulate sulfate and nitrate, water vapour over short vegetation by gradient and eddy correlation techniques in Hungary. EGS XX. General Assembly; April; Hamburg, Germany. Ann. Geophys. 13(suppl. 2): C490.

Horváth, L.; Pinto, J.; Weidinger, T. (2003) Estimate of the dry deposition of atmospheric nitrogen and sulfur species to spruce forest. Idõjárás (Q. J. Hung. Meteorol. Serv.) 107: 249-255.

Howard-Reed, C.; Wallace, L. A.; Ott, W. R. (2002) The effect of opening windows on air change rates in two homes. J. Air Waste Manage Assoc. 52: 147-159.

Hudman, R. C.; Jacob, D. J.; Cooper, O. C.; Evans, M. J.; Heald, C. L.; Park, R. J.; Fehsenfeld, F.; Flocke, F.; Holloway, J.; Hubler, G.; Kita, K.; Koike, M.; Kondo, Y.; Neuman, A.; Nowak, J.; Oltmans, S.; Parrish, D.; Roberts, J. M.; Ryerson, T. (2004) Ozone production in transpacific Asian pollution plumes and implications for ozone air quality in California. J. Geophys. Res. (Atmos.) 109(D23): 10.1029/2004JD004974.

Ito, K.; De Leon, S. F.; Lippmann, M. (2005) Associations between ozone and daily mortality, analysis and meta-analysis. Epidemiology 16: 446-457.

Jaffe, D.; Price, H.; Parrish, D.; Goldstein, A.; Harris, J. (2003) Increasing background ozone during spring on the west coast of North America. Geophys. Res. Lett. 30: 10.1029/2003GL017024.

Jakobi, G.; Fabian, P. (1997) Indoor/outdoor concentrations of ozone and peroxyacetyl nitrate (PAN). Int. J. Biometeorol. 40: 162-165.

Johnson, T. R. (1995) Recent advances in the estimation of population exposure to mobile source pollutants. J. Exposure Anal. Environ. Epidemiol. 5: 551-571.

Johnson, T. (1997) A pilot study in Los Angeles to measure personal ozone exposures during scripted activities. Washington, DC: American Petroleum Institute, Health and Environmental Sciences Department; API publication no. DR 218.

Johnson, T. (2003) A guide to selected algorithms, distributions, and databases used in exposure models developed by the Office of Air Quality Planning and Standards. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Research and Development; EPA grant no. CR827033. Available: http://www.epa.gov/ttn/fera/data/human/report052202.pdf [9 April, 2004].

Johnson, T.; Long, T. (2004) Determining the frequency of open windows in residences: a pilot study in Durham, North Carolina during varying temperature conditions. J. Exposure Anal. Environ. Epidemiol.: 10.1038/sj.jea.7500409.

Johnson, T.; Capel, J.; McCoy, M. (1996a) Estimation of ozone exposures experienced by urban residents using a probabilistic version of NEM and 1990 population data. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards; contract no. 68-DO-0062.

Johnson, T.; Capel, J.; McCoy, M.; Mozier, J. W. (1996b) Estimation of ozone exposures experienced by outdoor workers in nine urban areas using a probabilistic version of NEM. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards; contract no. 63-D-30094, work assignment nos. 0-1 and 1-4.

Johnson, T.; Capel, J.; Mozier, J.; McCoy, M. (1996c) Estimation of ozone exposures experienced by outdoor children in nine urban areas using a probabilistic version of NEM. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards; contract no. 63-D-30094.

Johnson, T.; Mozier, J.; Capel, J. (1997) Supplement to "Estimation of ozone exposures experienced by outdoor children in nine urban areas using a probabilistic version of NEM (April 1996)". Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards.

Johnson, T.; Mihlan, G.; LaPointe, J.; Fletcher, K.; Capel, J. (1999) Estimation of carbon monoxide exposures and associated carboxyhemoglobin levels in Denver residents using pNEM/CO (version 2.0) [draft]. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards; March 15.

Johnson, T.; Myers, J.; Kelly, T.; Wisbith, A.; Ollison, W. (2004) A pilot study using scripted ventilation conditions to identify key factors affecting indoor pollutant concentrations and air exchange rate in a residence. J. Exposure Anal. Environ. Epidemiol. 14: 1-22.

Klepeis, N. E. (1999) An introduction to the indirect exposure assessment approach: modeling human exposure using microenvironmental measurements and the recent National Human Activity Pattern Survey. Environ. Health Perspect. Suppl. 107(2): 365-374.

Klepeis, N. E.; Tsang, A. M.; Behar, J. V. (1996) Analysis of the national human activity pattern survey (NHAPS) respondents from a standpoint of exposure assessment. Washington, DC: U.S. Environmental Protection Agency, Office of Research and Development; report no. EPA/600/R-96/074.

Klepeis, N. E.; Nelson, W. C.; Ott. W. R.; Robinson, J. P. Tsang, A. M.; Switzer, P.; Behar, J. V.; Hern, S. C.; Engelmann, W. H. (2001) The National Human Activity Pattern Survey (NHAPS): a resource for assessing exposure to environmental pollutants. J. Exposure Anal. Environ. Epidemiol. 11: 231-252.

Kotchenruther, R. A.; Jaffe, D. A.; Beine, H. J.; et al. (2001a) Observations of ozone and related species in the northeast Pacific during the PHOBEA campaigns. 2. Airborne observations. J. Geophys. Res. (Atmos.) 106(D7): 7463-7483.

Kotchenruther, R. A.; Jaffe, D. A.; Jaegle, L. (2001b) Ozone photochemistry and the role of peroxyacetyl nitrate in the springtime northeastern Pacific troposphere: results from the photochemical ozone budget of the eastern north Pacific atmosphere (PHOBEA) campaign. J. Geophys. Res. (Atmos.) 106(D22): 28731-28742.

Koutrakis, P.; Sioutas, C.; Ferguson, S. T.; Wolfson, J. M.; Mulik, J. D.; Burton, R. M. (1993) Development and evaluation of a glass honeycomb denuder/filter pack system to collect atmospheric gases and particles. Environ. Sci. Technol. 27: 2497-2501.

Lagus Applied Technology, Inc. (1995) Air change rates in non-residential buildings in California. Sacramento, CA: California Energy Commission; contract no. 400-91-034; July.

Lee, E. H.; Hogsett, W. E. (1999) Role of concentrations and time of day in developing ozone exposure indices for a secondary standard. J. Air Waste Manage. Assoc. 49: 669-681.

Lee, M.; Heikes, B. G.; O'Sullivan, D. W. (2000) Hydrogen peroxide and organic hydroperoxide in the troposphere. Atmos. Environ. 34: 3475-3494.

Lee, K.; Xue, J.; Geyh, A. S.; Ozkaynak, H.; Leaderer, B. P.; Weschler, C. J.; Spengler, J. D. (2002) Nitrous acid, nitrogen dioxide, and ozone concentrations in residential environments. Environ. Health Perspect. 110: 145-150.

Lee, K.; Parkhurst, W. J.; Xue, J.; Özkaynak, H.; Neuberg, D.; Spengler, J. D. (2004) Outdoor/indoor/personal ozone exposures of children in Nashville, Tennessee. J. Air Waste Manage. Assoc. 54: 352-359.

Lefohn, A. S.; Runeckles, V. C. (1987) Establishing standards to protect vegetation - ozone exposure/dose considerations. Atmos. Environ. 21: 561-568.

Lefohn, A. S.; Oltmans, S. J.; Dann, T.; Singh, H. B. (2001) Present-day variability of background ozone in the lower troposphere. J. Geophys. Res. [Atmos.] 106: 9945-9958.

Lehman, J.; Swinton, K.; Bortnick, S.; Hamilton, C.; Baldridge, E.; Ender, B.; Cox, B. (2004) Spatio-temporal characterization of tropospheric ozone across the eastern United States. Atmos. Environ. 38: 4357-4369.

Leovic, K. W.; Sheldon, L. S.; Whitaker, D. A.; Hetes, R. G.; Calcagni, J. A.; Baskir, J. N. (1996) Measurement of indoor air emissions from dry-process photocopy machines. J. Air Waste Manage. Assoc. 46: 821-829.

Leovic, K.; Whitaker, D.; Northeim, C.; Sheldon, L. (1998) Evaluation of a test method for measuring indoor air emissions from dry-process photocopiers. J. Air Waste Manage. Assoc. 48: 915-923.

Levinson, D. H., ed. (2005) State of the climate in 2004. Bull. Am. Meteorol. Soc. 86: S1-S86.

Levinson, D. H.; Waple, A. M., eds. (2004) State of the climate in 2003. Bull. Am. Meteorol. Soc. 85: S1-S72.

Liang, J.; Horowitz, L. W.; Jacob, D. J.; Wang, Y.; Fiore, A. M.; Logan, J. A.; Gardner, G. M.; Munger, J. W. (1998) Seasonal budgets of reactive nitrogen species and ozone over the United States, and export fluxes to the global atmosphere. J. Geophys. Res. (Atmos.) 103: 13,435-13,450.

Liard, R.; Zureik, M.; Le Moullec, Y.; Soussan, D.; Glorian, M.; Grimfeld, A.; Neukirch, F. (1999) Use of personal passive samplers for measurement of NO_2, NO, and O_3 levels in panel studies. Environ. Res. 81: 339-348.

Lin, C.-Y.; Jacob, D. J.; Munger, J. W.; Fiore, A. M. (2000) Increasing background ozone in surface air over the United States. Geophys. Res. Lett. 27: 3465-3468.

Linn, W. S.; Shamoo, D. A.; Hackney, J. D. (1992) Documentation of activity patterns in 'high-risk' groups exposed to ozone in the Los Angeles area. In: Tropospheric ozone and the environment II: effects, modeling and control: papers from an Air & Waste Management Association international specialty conference; November, 1991; Atlanta, GA. Pittsburgh, PA: Air & Waste Management Association; pp. 701-712. (A&WMA publication TR-20).

Linn, W. S.; Shamoo, D. A.; Anderson, K. R.; Peng, R.-C.; Avol, E. L.; Hackney, J. D.; Gong, H., Jr. (1996) Short-term air pollution exposures and responses in Los Angeles area schoolchildren. J. Exposure Anal. Environ. Epidemiol. 6: 449-472.

Liu, L.-J. S.; Koutrakis, P.; Leech, J.; Broder, I. (1995) Assessment of ozone exposures in the greater metropolitan Toronto area. J. Air Waste Manage. Assoc. 45: 223-234.

Liu, L.-J. S.; Delfino, R.; Koutrakis, P. (1997) Ozone exposure assessment in a southern California community. Environ. Health Perspect. 105: 58-65.

McCurdy, T. (2000) Conceptual basis for multi-route intake dose modeling using an energy expenditure approach. J. Exposure Anal. Environ. Epidemiol. 10: 86-97.

McCurdy, T.; Glen, G.; Smith, L.; Lakkadi, Y. (2000) The National Exposure Research Laboratory's Consolidated Human Activity Database. J. Exposure Anal. Environ. Epidemiol. 10: 566-578.

Meng, Q. Y.; Turpin, B. J.; Korn, L.; Weisel, C. P.; Morandi, M.; Colome, S.; Zhang, J.; Stock, T.; Spektor, D.; Winer, A.; Zhang, L.; Lee, J. H.; Giovanetti, R.; Cui, W.; Kwon, J.; Alimokhtari, S.; Shendell, D.; Jones, J.; Farrar, C.; Maberti, S. (2005) Influence of ambient (outdoor) sources on residential indoor and personal $PM_{2.5}$ concentrations: analyses of RIOPA data. J. Exposure Anal. Environ. Epidemiol. 15: 17-28.

Morrison, G. C.; Nazaroff, W. W. (2000) The rate of ozone uptake on carpets: experimental studies. Environ. Sci. Technol. 34: 4963-4968.
Morrison, G. C.; Nazaroff, W. W. (2002) Ozone interactions with carpet: secondary emissions of aldehydes. Environ. Sci. Technol. 36: 2185-2192.
Murray, D. M.; Burmaster, D. E. (1995) Residential air exchange rates in the United States: empirical and estimated parametric distributions by season and climatic region. Risk Anal. 15: 459-465.
Navidi, W.; Thomas, D.; Langholz, B.; Stram, D. (1999) Statistical methods for epidemiologic studies of the health effects of air pollution. Cambridge, MA: Health Effects Institute; research report no. 86.
Nazaroff, W. W.; Cass, G. R. (1986) Mathematical modeling of chemically reactive pollutants in indoor air. Environ. Sci. Technol. 20: 924-934.
Nazaroff, W. W.; Weschler, C. J. (2004) Cleaning products and air fresheners: exposure to primary and secondary air pollutants. Atmos. Environ. 38: 2841-2865.
Neidell, M. J. (2004) Air pollution, health, and socio-economic status: the effect of outdoor air quality on childhood asthma. J. Health Econ. 23: 1209-1236.
Niu, J.; Tung, T. C. W.; Burnett, J. (2001) Ozone emission rate testing and ranking method using environmental chamber. Atmos. Environ. 35: 2143-2151.
Nøjgaard, C.; Söndergaard, S. B.; Madsen, J. L.; Möller, S. (2005) [Does the administration of dipyridamole affect the ventilatory capacity of patients with chronic obstructive lung disease?] Ugeskr. Laeg. 167: 1750-1753.
Northeast States for Coordinated Air Use Management (NESCAUM). (2002) Indoor/outdoor school air monitoring project. Boston, MA. Available: http://www.nescaum.org/pdf/schoolmonitoring.pdf [29 October, 2003].
Oltmans, S. J.; Lefohn, A. S.; Scheel, H. E.; Harris, J. M.; Levy, H., II; Galbally, I. E.; Brunke, E.-G.; Meyer, C. P.; Lathrop, J. A.; Johnson, B. J.; Shadwick, D. S.; Cuevas, E.; Schmidlin, F. J.; Tarasick, D. W.; Claude, H.; Kerr, J. B.; Uchino, O.; Mohnen, V. (1998) Trends of ozone in the troposphere. Geophys. Res. Lett. 25: 139-142.
O'Neill, M. S.; Ramirez-Aguilar, M.; Meneses-Gonzalez, F.; Hernández-Avila, M.; Geyh, A. S.; Sienra-Monge, J. J.; Romieu, I. (2003) Ozone exposure among Mexico City outdoor workers. J. Air Waste Manage. Assoc. 53: 339-346.
Orzechowska, G. E.; Paulson, S. E. (2002) Production of OH radicals from the reactions of C_4-C_6 internal alkenes and styrenes with ozone in the gas phase. Atmos. Environ. 36: 571-581.
Ott, W. R. (1982) Concepts of human exposure to air pollution. Environ. Int. 7: 179-196.
Ott, W. R. (1985) Total human exposure: an emerging science focuses on humans as receptors of environmental pollution. Environ. Sci. Technol. 19: 880-886.
Park, J.-H.; Spengler, J. D.; Yoon, D.-W.; Dumyahn, T.; Lee, K.; Ozkaynak, H. (1998) Measurement of air exchange rate of stationary vehicles and estimation of in-vehicle exposure. J. Exposure Anal. Environ. Epidemiol. 8: 65-78.
Pinto, J. P.; Lefohn, A. S.; Shadwick, D. S. (2004) Spatial variability of $PM_{2.5}$ in urban areas in the United States. J. Air Waste Manage. Assoc. 54: 440-449.
Poupard, O.; Blondeau, P.; Iordache, V.; Allard, F. (2005) Statistical analysis of parameters influencing the relationship between outdoor and indoor air quality in schools. Atmos. Environ. 39: 2071-2080.
Reeves, C. E.; Penkett, S. A. (2003) Measurements of peroxides and what they tell us. Chem. Rev. 103: 5199-5218.
Reiss, R.; Ryan, P. B.; Koutrakis, P.; Tibbetts, S. J. (1995) Ozone reactive chemistry on interior latex paint. Environ. Sci. Technol. 29: 1906-1912.
Riediker, M.; Williams, R.; Devlin, R.; Griggs, T.; Bromberg, P. (2003) Exposure to particulate matter, volatile organic compounds, and other air pollutants inside patrol cars. Environ. Sci. Technol. 37: 2084-2093.
Roberts, J. M.; Williams, J.; Baumann, K.; Buhr, M. P.; Goldan, P. D.; Holloway, J.; Hübler, G.; Kuster, W. C.; McKeen, S. A.; Ryerson, T. B.; Trainer, M.; Williams, E. J.; Fehsenfeld, F. C.; Bertman, S. B.; Nouaime, G.; Seaver, C.; Grodzinsky, G.; Rodgers, M.; Young, V. L. (1998) Measurements of PAN, PPN, and MPAN made during the 1994 and 1995 Nashville Intensives of the Southern Oxidant Study: implications for regional ozone production from biogenic hydrocarbons. J. Geophys. Res. [Atmos.] 103: 22,473-22,490.
Roberts, J. M.; Flocke, F.; Stroud, C. A.; Hereid, D.; Williams, E.; Fehsenfeld, F.; Brune, W.; Martinez, M.; Harder, H. (2002) Ground-based measurements of peroxycarboxylic nitric anhydrides (PANs) during the 1999 Southern Oxidants Study Nashville intensive. J. Geophys. Res. [Atmos.] 107(D21): 10.1029/2001JD000947.
Roberts, J. M.; Jobson, B. T.; Kuster, W.; Goldan, P.; Murphy, P.; Williams, E.; Frost, G.; Riemer, D.; Apel, E.; Stroud, C.; Wiedinmyer, C.; Fehsenfeld, F. (2003) An examination of the chemistry of peroxycarboxylic nitric anhydrides and related volatile organic compounds during Texas Air Quality Study 2000 using ground-based measurements. J. Geophys. Res. [Atmos.] 108(D16): 10.1029/2003JD003383.

Roberts, J. M.; Flocke, F.; Chen, G.; de Gouw, J.; Holloway, J. S.; Hübler, G.; Neuman, J. A.; Nicks, D. K., Jr.; Nowak, J. B.; Parrish, D. D.; Ryerson, T. B.; Sueper, D. T.; Warneke, C.; Fehsenfeld, F. C. (2004) Measurement of peroxycarboxylic nitric anhydrides (PANs) during the ITCT 2K2 aircraft intensive experiment. J. Geophys. Res. 109(D23S21): 10.1029/2004JD004960.

Roelofs, G. J.; Scheeren, H. A.; Heland, J.; Ziereis, H.; Lelieveld, J. (2003) A model study of ozone in the eastern Mediterranean free troposphere during MINOS (August 2001). Atmos. Chem. Phys. 3: 1199-1210.

Rohr, A. C.; Wilkins, C. K.; Clausen, P. A.; Hammer, M.; Nielsen, G. D.; Wolkoff, P.; Spengler, J. D. (2002) Upper airway and pulmonary effects of oxidation products of (+)-α-pinene, *d*-limonene, and isoprene in BALB/*c* mice. Inhalation Toxicol. 14: 663-684.

Rohr, A. C.; Shore, S. A.; Spengler, J. D. (2003) Repeated exposure to isoprene oxidation products causes enhanced respiratory tract effects in multiple murine strains. Inhalation Toxicol. 15: 1191-1207.

Romieu, I.; Lugo, M. C.; Colome, S.; Garcia A. M.; Avila, M. H.; Geyh, A.; Velasco, S. R.; Rendon, E. P. (1998) Evaluation of indoor ozone concentration and predictors of indoor-outdoor ratio in Mexico City. J. Air Waste Manage. Assoc. 48: 327-335.

Salmon, L. G.; Cass, G. R.; Bruckman, K.; Haber, J. (2000) Ozone exposure inside museums in the historic central district of Krakow, Poland. Atmos. Environ. 34: 3823-3832.

Sarnat, J. A.; Koutrakis, P.; Suh, H. H. (2000) Assessing the relationship between personal particulate and gaseous exposures of senior citizens living in Baltimore, MD. J. Air Waste Manage. Assoc. 50: 1184-1198.

Sarnat, J. A.; Schwartz, J.; Catalano, P. J.; Suh, H. H. (2001) Gaseous pollutants in particulate matter epidemiology: confounders or surrogates? Environ. Health Perspect. 109: 1053-1061.

Sarwar, M.; Corsi, R.; Kimura, Y.; Allen, D.; Weschler, C. (2001) Hydroxyl radicals in indoor environments. In: Proceedings of the Air & Waste Management Association's 94th Annual Conference & Exhibition; June; Orlando, FL. Pittsburgh, PA: Air & Waste Management Association.

Sarwar, G.; Corsi, R.; Kumura, Y.; Allen, D.; Weschler, C. J. (2002) Hydroxyl radicals in indoor environments. Atmos. Environ. 36: 3973-3988.

Sørensen, D. N.; Weschler, C. J. (2002) Modeling-gas phase reactions in indoor environments using computational fluid dynamics. Atmos. Environ. 36: 9-18.

Sparks, J. P.; Roberts, J. M.; Monson, R. K. (2003) The uptake of gaseous organic nitrogen by leaves: a significant global nitrogen transfer process. Geophys. Res. Lett. 30(23): 10.1029/2003GL018578.

Spier, C. E.; Little, D. E.; Trim, S. C.; Johnson, T. R.; Linn, W. S.; Hackney, J. D. (1992) Activity patterns in elementary and high school students exposed to oxidant pollution. J. Exposure Anal. Environ. Epidemiol. 2: 277-293.

Steiber, R. S. (1995) Ozone generators in indoor air settings. Research Triangle Park, NC: U.S. Environmental Protection Agency, National Risk Management Research Laboratory; report no. EPA-600/R-95-154. Available from: NTIS, Springfield, VA; PB96-100201.

Teklemariam, T. A.; Sparks, J. P. (2004) Gaseous fluxes of peroxyacetyl nitrate (PAN) into plant leaves. Ecol. Soc. Am. Ann. Meeting Abst. 89: 501-502.

Trainer, M.; Parrish, D. D.; Buhr, M. P.; Norton, R. B.; Fehsenfeld, F. C.; Anlauf, K. G.; Bottenheim, J. W.; Tang, Y. Z.; Wiebe, H. A.; Roberts, J. M.; Tanner, R. L.; Newman, L.; Bowersox, V. C.; Meagher, J. F.; Olszyna, K. J.; Rodgers, M. O.; Wang, T.; Berresheim, H.; Demerjian, K. L.; Roychowdhury, U. K. (1993) Correlation of ozone with NO_y in photochemically aged air. J. Geophys. Res. [Atmos.] 98: 2917-2925.

Turk, B. H.; Grimsrud, D. T.; Brown, J. T.; Geisling-Sobotka, K. L.; Harrison, J.; Prill, R. J. (1989) Commercial building ventilation rates and particle concentrations. ASHRAE Trans. 95(part 1): 422-433.

U.S. Environmental Protection Agency. (1986) Air quality criteria for ozone and other photochemical oxidants. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report nos. EPA-600/8-84-020aF-eF. 5v. Available from: NTIS, Springfield, VA; PB87-142949.

U.S. Environmental Protection Agency. (1992) Guidelines for exposure assessment. Washington, DC: Risk Assessment Forum, USEPA 600Z-92/001.

U.S. Environmental Protection Agency. (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: http://cfpub2.epa.gov/ncea/.

U.S. Environmental Protection Agency. (1998) Guideline on ozone monitoring site selection. Research Triangle Park, NC: U.S. Environmental Protection Agency, National Center for Environmental Assessment; EPA-454/R-98-002.

U.S. Environmental Protection Agency. (2003a) Technology Transfer Network: Air Quality System (AQS). Washington, DC: Office of Air and Radiation. Available: http://www.epa.gov/ttn/airs/airsaqs/ [24 August, 2005].

U.S. Environmental Protection Agency. (2004a) Air quality criteria for particulate matter. Research Triangle Park, NC: National Center for Environmental Assessment; report no. EPA/600/P-99/002aF-bF. 2v. Available: http://cfpub.epa.gov/ncea/ [9 November, 2004].

U.S. Environmental Protection Agency. (2004b) The ozone report: measuring progress through 2003. Research Triangle Park, NC: Office of Air Quality Planning and Standards; report no. EPA-454/K04-001. Available: http://www.epa.gov/air/airtrends/pdfs/2003ozonereport.pdf [12 May, 2005].

Väkevä, M.; Hämeri, K.; Kulmala, M.; Lahdes, R.; Ruuskanen, J.; Laitinen, T. (1999) Street level versus rooftop concentrations of submicron aerosol particles and gaseous pollutants in an urban street canyon. Atmos. Environ. 33: 1385-1397.

Van den Bergh, V.; Vanhees, I.; De Boer, R.; Compernolle, F.; Vinckier, C. (2000) Identification of the oxidation products of the reaction between α-pinene and hydroxyl radicals by gas and high-performance liquid chromatography with mass spectrometric detection. J. Chromatogr. A 896: 135-148.

Venkatachari, P.; Hopke, P. K.; Grover, B. D.; Eatough, D. J. (2005) Measurement of particle-bound reactive oxygen species in Rubidoux aerosols. J. Atmos. Chem. 50: 49-58.

Venkatachari, P.; Hopke, P .K.; Brune, W. H.; Ren, X.; Lesher, R.; Mao, J.; Mitchell, M. (2005) Characterization of reactive oxygen species trends in Flushing, New York. Atmos. Environ.: in press.

Volz-Thomas, A.; Geiss, H.; Hofzumahaus, A.; Becker, K.-H. (2003) Introduction to special section: photochemistry experiment in BERLIOZ. J. Geophys. Res. [Atmos.] 108(D4): 10.1029/JD002029.

Wainman, T.; Zhang, J.; Weschler, C. J.; Lioy, P. J. (2000) Ozone and limonene in indoor air: a source of submicron particle exposure. Environ. Health Perspect. 108: 1139-1145.

Wang, Y.; Logan, J. A.; Jacob, D. J. (1998) Global simulation of tropospheric O_3-NO_x-hydrocarbon chemistry 2. Model evaluation and global ozone budget. J. Geophys. Res. (Atmos.) 103: 10,727-10,755.

Weinstein-Lloyd; et al. (1998) Measurements of peroxides and related species during the 1995 summer intensive of the Southern oxidants study in Nashville, Tennessee. J. Geophys. Res. (Atmos.) 103: 22361-22373.

Weschler, C. J. (2000) Ozone in indoor environments: concentration and chemistry. Indoor Air 10: 269-288.

Weschler, C. J. (2004) Chemical reactions among indoor pollutants: what we've learned in the new millennium. Indoor Air 14(suppl. 7): 184-194.

Weschler, C. J.; Shields, H. C. (1997) Potential reactions among indoor pollutants. Atmos. Environ. 31: 3487-3495.

Weschler, C. J.; Shields, H. C. (1999) Indoor ozone/terpene reactions as a source of indoor particles. Atmos. Environ. 33: 2301-2312.

Weschler, C. J.; Shields, H. C. (2000) The influence of ventilation on reactions among indoor pollutants: modeling and experimental observations. Indoor Air. 10: 92-100.

Weschler, C. J.; Shields, H. C. (2003) Experiments probing the influence of air exchange rates on secondary organic aerosols derived from indoor chemistry. Atmos. Environ. 37: 5621-5631.

Weschler, C. J.; Shields, H. C.; Naik, D. V. (1989) Indoor ozone exposures. JAPCA 39: 1562-1568.

Weschler, C. J.; Hodgson, A. T.; Wooley, J. D. (1992) Indoor chemistry: ozone, volatile organic compounds, and carpets. Environ. Sci. Technol. 26: 2371-2377.

Weschler, C. J.; Shields, H. C.; Naik, D. V. (1994) Indoor chemistry involving O3, NO, and NO2 as evidenced by 14 months of measurements at a site in southern California. Environ. Sci. Technol. 28: 2120-2132.

Wilkins, C. K.; Clausen, P. A.; Wolkoff, P.; Larsen, S. T.; Hammer, M.; Larsen, K.; Hansen, V.; Nielsen, G. D. (2001) Formation of strong irritants in mixtures of isoprene/ozone and isoprene/ozone/nitrogen dioxide. Environ. Health Perspect. 109: 937-941.

Williams, R.; Suggs, J.; Rea, A.; Leovic, K.; Vette, A.; Croghan, C.; Sheldon, L.; Rodes, C.; Thornburg, , J.; Ejire, A.; Herbst, M.; Sanders, W., Jr. (2003a) The Research Triangle Park particulate matter panel study: PM mass concentration relationships Atmos. Environ. 37: 5349-5363.

Williams, R.; Suggs, J.; Rea, A.; Sheldon, L.; Rodes, C.; Thornburg, J. (2003b) The Research Triangle Park particulate matter panel study: modeling ambient source contribution to personal and residential PM mass concentrations. Atmos. Environ. 37: 5365-5378.

Wolkoff, P.; Clausen, P. A.; Wilkins, C. K.; Hougaard, K. S.; Nielsen, G. D. (1999) Formation of strong airway irritants in a model mixture of (+)-α-pinene/ozone. Atmos. Environ. 33: 693-698.

Zanis, P.; Trickl, T.; Stohl, A.; Wernli, H.; Cooper, O.; Zerefos, C.; Gaeggeler, H.; Schnabel, C.; Tobler, L.; Kubik, P. W.; Priller, A.; Scheel, H. E.; Kanter, H. J.; Cristofanelli, P.; Forster, C.; James, P.; Gerasopoulos, E.; Delcloo, A.; Papayannis, A.; Claude, H. (2003) Forecast, observation and modelling of a deep stratospheric intrusion event over Europe. Atmos. Chem. Phys. 3: 763-777.

4. DOSIMETRY, SPECIES HOMOLOGY, SENSITIVITY, AND ANIMAL-TO-HUMAN EXTRAPOLATION

4.1 INTRODUCTION

The dosimetry of ozone (O_3) in humans has been examined in a series of studies published in the past decade. These studies further characterize the dose of O_3 delivered to various sites in the respiratory tract (RT). Ozone, classified as a reactive gas, interacts with surfactant, antioxidants, and other compounds in the epithelial lining fluid (ELF). Researchers have attempted to obtain a greater understanding of how these complex interactions affect O_3 uptake and O_3-induced injury. New work has also been completed evaluating species differences in responses to O_3 exposures, which allow more accurate quantitative extrapolation from animals to humans.

This chapter is not intended to be a complete overview of O_3 dosimetry and animal-to-human comparisons, but rather, it is an update of the dosimetry/extrapolation chapter from the last O_3 criteria document (U.S. Environmental Protection Agency, 1996), or 1996 O_3 AQCD, and other reviews of the earlier published literature. The framework for presenting this chapter is first a discussion in Section 4.2 of general concepts of the dosimetry of O_3 in the RT. Bolus-response studies are then presented in Section 4.2.1 followed by general uptake studies in Section 4.2.2. Dosimetry modeling is presented in Section 4.2.3 followed by the summary and conclusions for the dosimetry material in Section 4.2.4. The chapter continues in Section 4.3 with a discussion of species comparisons and ends with a discussion of animal-to-human extrapolation. More detailed discussions of the studies are presented in the supporting material to this chapter (Annex AX4). The toxicological effects of O_3 in laboratory animals and in vitro test systems are discussed in Chapter 5 and direct effects of O_3 in humans are discussed in Chapter 6. The historical O_3 literature is very briefly summarized in this chapter, providing a very concise overview of previous work. The reader is referred to the 1996 O_3 AQCD for more detailed discussion of the literature prior to the early 1990s.

4.2 DOSIMETRY OF OZONE IN THE RESPIRATORY TRACT

Ozone dosimetry refers to the measurement or estimation of the amount of O_3 or its reaction products reaching and persisting at specific sites in the RT following an exposure. The compound most directly responsible for toxic effects may be the inhaled gas O_3 or one of its chemical reaction products. Complete identification of the actual toxic agents and their integration into dosimetry is a complex issue that has not been resolved. Dosimetric studies attempt to quantify the amount of O_3 retained in the lung (i.e., not exhaled) or the dose of O_3 or its active metabolites delivered to target cells or tissues (i.e., dose per cell or tissue surface area). For comparison, epidemiologic studies may simply consider exposure concentration while clinical studies may consider the total amount of O_3 inhaled (product of exposure concentration, duration, and minute ventilation). Hence, dosimetric studies seek to accurately quantify dose to target lung regions or tissues, whereas epidemiologic and clinical studies typically consider exposures.

Understanding dosimetry as it relates to O_3-induced injury is complex due to the fact that O_3 interacts primarily with the ELF which contains surfactant and antioxidants. In the upper airways ELF is thick and highly protective against oxidant injury. Figure 4-1 illustrates the structure of the lower airways with progression from the large airways to the alveolus. In lower airways ELF is thinner, has lower levels of antioxidants, and thus, allows more cellular injury. Adding to the complexity is the fact that O_3 can react with molecules in the ELF to create even more reactive metabolites, which can then diffuse within the lung or be transported out of the lung to generate systemic effects.

A considerable number of dosimetric studies were summarized in the 1996 O_3 AQCD. These studies provided estimates of absorbed O_3 in the RT as a whole or in regions such as the upper airways (URT) or lower airways (LRT), defined as being proximal or distal to the tracheal entrance, respectively. Estimates were obtained for both humans and animals via direct measurement and mathematical modeling. The mathematical models also estimated O_3 doses to specific target sites such as the proximal alveolar region (PAR; first generation distal to the terminal bronchioles) and the centriacinar region (CAR; junction of conducting airways and gas exchange region).

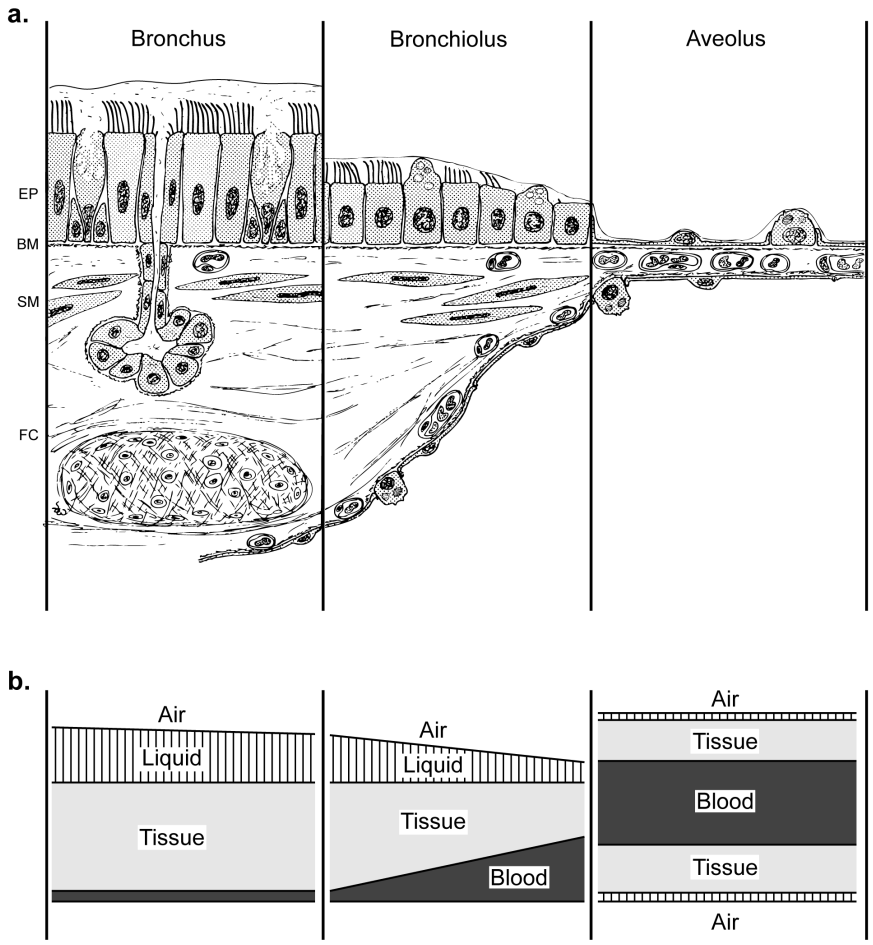


Figure 4-1. Structure of lower airways with progression from the large airways to the alveolus. Panel (a) illustrates basic airway anatomy. Structures are epithelial cells, EP; basement membrane, BM; smooth muscle cells, SM; and fibrocartilaginous coat, FC. Panel (b) illustrates the relative amounts of liquid, tissue, and blood with distal progression. In the bronchi there is a thick liquid lining over a relatively thick layer of tissues. Even highly soluble materials moving from the air into the liquid layer have minimal systemic access via the blood. With distal progress, the protective liquid lining diminishes allowing increased access of compounds crossing the air-liquid interface to the tissues and the blood.

Source: Panel (a) reproduced with permission (Weibel, E. R. [1980] Design and structure of the human lung. In: Fishman, A. P., ed. Pulmonary Diseases and Disorders. New York, NY: McGraw-Hill; p. 231).

In general, the consensus of experimental and modeling studies summarized in the 1996 O_3 AQCD supported the following conclusions: (1) for the URT, animal and human studies suggested that O_3 uptake is greater in the nose than the mouth but the effect of flow on uptake was equivocal; (2) for the LRT, predicted tissue doses (O_3 flux to liquid-tissue interface) were very low in the trachea, increased to a maximum in the terminal bronchioles or first airway generation in the pulmonary region, and rapidly decreased with distal progression; (3) increasing tidal volume (V_T) increases O_3 uptake, whereas, increasing flow or breathing frequency (f_B) decreases O_3 uptake; (3) increasing flow shifts O_3 uptake to the smaller peripheral airways, i.e., toward the CAR; and (4) similarly, the effect of exercise is to significantly increase the pulmonary region total dose (mass of O_3) and the CAR dose (mass per unit surface area).

Some cross-species *in vivo* comparisons were described in the 1996 O_3 AQCD. For instance, comparing bronchoalveolar lavage (BAL) cells from rats and humans, it was estimated that a 0.4 ppm O_3 exposure in exercising humans gave 4 to 5 times the O_3 dose (retained) relative to rats exposed at rest to the same concentration. *In vitro* dosimetry studies in the 1996 O_3 AQCD using isolated lung preparations showed that uptake efficiency is chemical-reaction dependent, indicating the importance of reaction product formation. These reaction products, created mainly by the ozonolysis of polyunsaturated fatty acids, included hydrogen peroxide, aldehydes, and hydroxyhydroperoxides, which are mediators of O_3 toxicity. Other products are created by the reaction of O_3 with other ELF constituents, all of which must be considered in understanding the dosimetry of O_3.

The next two sections (4.2.1 and 4.2.2) review the available new experimental studies on O_3 dosimetry, all of which were conducted by Ultman and colleagues. Table AX4-1 in Annex AX4 summarizes theses studies.

4.2.1 Bolus-Response Studies

The bolus-response method has been used by the Ultman group as an approach to explore the distribution of O_3 absorption in the airways of humans. This non-invasive method consists of an injection of a known volume and concentration of O_3 during inspiration. Ozone uptake is the amount of O_3 absorbed during a breath relative to the amount contained in the inhaled bolus. Figure 4-2 illustrates the uptake of a series of O_3 boli as a function of volumetric penetration (V_P), i.e., the volume between the center of mass of an inhaled bolus and the end of

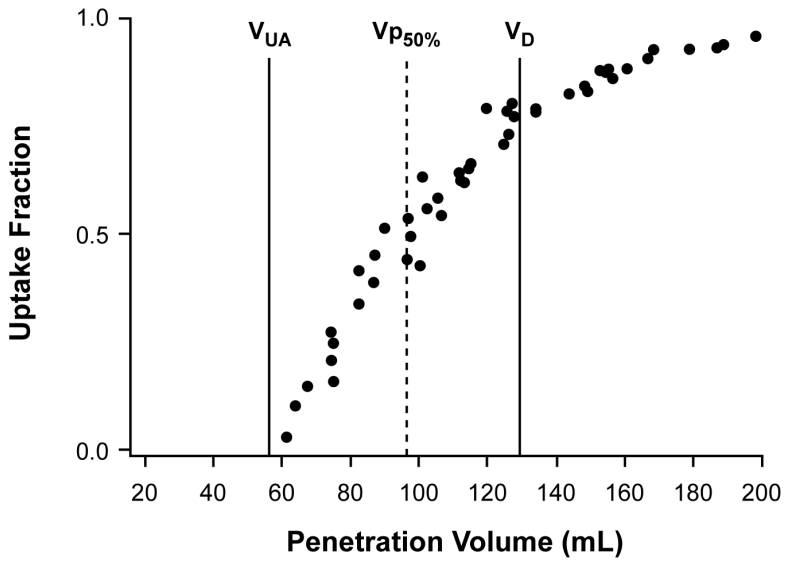


Figure 4-2. Ozone uptake fraction as a function of volumetric penetration (V_P) in a representative subject. Each point represents the O_3 uptake of a bolus inspired by the subject. The volumes, V_{UA} and V_D, are the volume of the upper airways and anatomical dead space, respectively, and $VP_{50\%}$ is the V_P at which 50% of the inspired bolus was absorbed. In 47 healthy subjects (24 M, 23 F), Ultman et al. (2004) found that $VP_{50\%}$ was well correlated with V_D ($r = 0.57$, $p < 0.001$) and better correlated with the volume of the conducting airways, i.e., V_D minus V_{UA}, ($r = 0.65$, $p = 0.001$).

Source: Adapted from Ultman et al. (2004).

inspiration. The inspired O_3 boli (for which the uptake fractions are illustrated in Figure 4-2) were 20 ml of 2 ppm O_3. Kabel et al. (1994) have previously shown that varying the O_3 concentration of inspired boli between 0.4 and 4 ppm does not affect the distribution of uptake as a function of V_P.

The O_3 bolus-technique was used by Bush et al. (1996a) to ascertain differences in lung anatomy and gender that can alter the exposure-dose cascade. Forced vital capacity (FVC), total lung capacity (TLC) and anatomic dead space (V_D) were determined for ten male and ten female subjects, who then inhaled to a 20 ml bolus of 3 ppm O_3 injected into the airstream. In all subjects, dosimetry differences could be explained by differences in V_D. In a subsequent study, Ultman et al. (2004) showed that the volume at which 50% of an inspired O_3 bolus is absorbed

was better associated with the volume of the conducting airways than V_D (see Figure 4-2). Bush et al. (1996a) pointed out that the applicability of their results may be limited because of their assumptions that the intrinsic mass transfer parameter (K_a) was independent of location in the RT and that there was no mucous resistance. They further suggested that the dependence of K_a on flowrate and V_D be restricted to flowrates $\leq$1000 mL/s until studies at higher rates have been performed.

Nodelman and Ultman (1999) demonstrated that the uptake distributions of O_3 boli were sensitive to the mode of breathing and to the airflow rate. As flowrates increased from 150 to 1000 mL/s, O_3 penetrated deeper into the lung and penetration was further increased by oral relative to nasal breathing. The authors suggest that the switch from nasal to oral breathing coupled with increases in respiratory flow as occurs during exercise causes a shift in the O_3 dose distribution, allowing O_3 to penetrate deeper into the lung, increasing the potential for damage to bronchiolar and alveolar tissues.

More recently, Ultman et al. (2004) measured O_3 uptake using the bolus technique in 60 young heathy nonsmoking adults (32 M, 28 F). Bolus were inspired at a rate of 1 mL/s, equivalent to a moderate exercise rate with a minute ventilation of 30 L/min. Figure 4-2 illustrates the O_3 uptake fraction as a function of V_P in a representative subject. Anatomic dead space was measured in 47 of the subjects (24 M, 23 F). In these subjects, the volume at which 50% of an inhaled bolus was absorbed ($VP_{50\%}$) was correlated with V_D ($r = 0.57$, $p < 0.001$) and the volume of the conducting airways, i.e., V_D minus the volume of the upper airways, ($r = 0.65$, $p = 0.001$). Both $VP_{50\%}$ and V_D were significantly greater in males than females, although the volume of the upper airways was not. These findings suggest that in females the smaller airways, and associated larger surface-to-volume ratio, enhance local O_3 uptake and cause reduced penetration of O_3 into the distal lung. It is not clear from these findings, however, if the actual anatomical location of $VP_{50\%}$ differed between males and females.

A few studies have measured the effect of a continuous pollutant exposure on O_3 bolus uptake. Asplund et al. (1996) randomly exposed young healthy adults (8 M, 3 F) for 2 h [presumably at rest] to 0.0 (air), 0.12, or 0.36 ppm O_3 on 3 separate occasions separated by at least 1-wk. Ozone bolus uptake was measured preexposure and subsequently at 30 minute intervals during the exposure. Ozone uptake over the V_P range of 70 to 120 ml increased after the air exposure, decreased slightly after the 0.12 ppm O_3 exposure, and decreased

more substantially following the 0.36 ppm O_3 exposure. Relative to uptake during the air exposure, O_3 bolus uptake was significantly decreased by 30 minutes of the 0.12 and 0.36 ppm O_3 exposures and remained significantly decreased for the duration of these exposures.

Using a similar protocol, Rigas et al. (1997) randomly exposed young healthy adults (6 M, 6 F) for 2 h at rest to filtered air, 0.36 ppm NO_2, 0.75 ppm NO_2, 0.36 ppm SO_2, or 0.36 ppm O_3. Ozone bolus uptake (V_P range of 70 to 120 ml) was measured preexposure and every 30 minute during the exposures. The results of an *F* test indicated that exposure duration (30-, 60-, 90-, 120-min) was not a significant factor, but treatment (NO_2, SO_2, etc.) was ($p < 0.01$). Ozone bolus uptake was increased by 30 minutes during the NO_2 and SO_2 exposures and decreased during the O_3 exposure. The authors suggested that there may be increased production of an O_3-reactive substrate in the ELF due to airway inflammation. During NO_2 and SO_2 exposures the substrate was not depleted by these gases and so could react with the O_3 bolus. During O_3 exposure the substrate was depleted, causing the fractional absorption of the O_3 bolus to decrease.

4.2.2 General Uptake Studies

Ultman and colleagues have recently completed some general uptake studies to determine the ratio of O_3 uptake to the quantity of O_3 inhaled. Uptake efficiency was determined at exposures of 0.2 or 0.4 ppm O_3 while exercising at a minute volume of approximately 20 L/min for 60 minutes or 40 L/min for 30 minutes in both men and women (Rigas et al., 2000). Uptake efficiency ranged from 0.56 to 0.98 and had a statistically significant but weak dependence on concentration, minute volume, and exposure time. Intersubject differences had the largest influence on uptake efficiency, resulting in a variation of approximately 10%. As the quantity of O_3 retained by the RT is equal to uptake efficiency times the quantity of O_3 inhaled, relatively large changes in concentration, minute volume, or exposure time may result in relatively large changes in the amount of O_3 retained by the RT or absorbed locally. The authors concluded that for exposure times <2 h, inhaled dose (product of O_3 concentration, exposure duration, and minute ventilation) is a reasonable predictor of actual uptake as long as there are fixed concentrations of O_3 and fixed levels of exercise. More importantly, similarly exposed individuals vary in the amount of actual dose received.

Santiago et al. (2001) studied the effects of airflow rate (3 to 15 L/min) and O_3 concentration (0.1, 0.2, or 0.4 ppm) on O_3 uptake in nasal cavities of males and females. As would be expected, uptake efficiency in the nose was inversely related to the flowrate and the concentration of O_3 in the inlet air. They computed a gas-phase diffusion resistance of <24% of overall diffusion resistance which suggested to them that simultaneously occurring diffusion and chemical reactions in the mucous layer were the limiting factors in O_3 uptake. Difference in O_3 uptake ranged from 0.63 to 0.97 at flowrates of 3 L/min and 0.25 to 0.50 at 15 L/min. The small effects of flowrate and concentration on uptake efficiency were statistically significant, but intersubject differences accounted for approximately half of the total variation in uptake efficiency. Both these general uptake studies, done at environmentally relevant O_3 concentrations, indicate that inter-individual differences in fractional uptake are extremely important in O_3 dose-response relationships.

In the research mentioned above, Ultman et al. (2004) also completed continuous exposure studies. The same 60 subjects were exposed continuously for 1 h to either clean air or 0.25 ppm ozone while exercising at a target minute ventilation of 30 L/min. This is the first study to assess ventilatory and dosimetric parameters for an entire hour of exposure. Additionally they measured bronchial cross-sectional area available for gas diffusion in addition to other ventilatory parameters. At a fixed minute ventilation of 30 L/min, the uptake fraction of O_3 decreased with increasing f_B (see Figure 4-3) and increased with increasing V_T. The uptake fraction was significantly greater in males (91.4%) than females (87.1%), which is consistent with the larger f_B and smaller V_T of the females than males. There was a small but significant reduction in the breath-by-breath uptake of O_3 from 90.6% on average for the first 15 minutes to 87.3% on average for the last 15 minutes of exposure. Ozone uptake rate correlated with percent changes in individual bronchial cross-sectional area but did not correlate with individual FEV_1 responses. Neither of these parameters correlated with the penetration volume determined in the bolus studies mentioned above. The authors concluded that the intersubject differences in forced respiratory responses were not due to differences in O_3 uptake. However, these data did partially support the hypothesis that changes in cross-sectional area available for gas diffusion are related to overall O_3 retention.

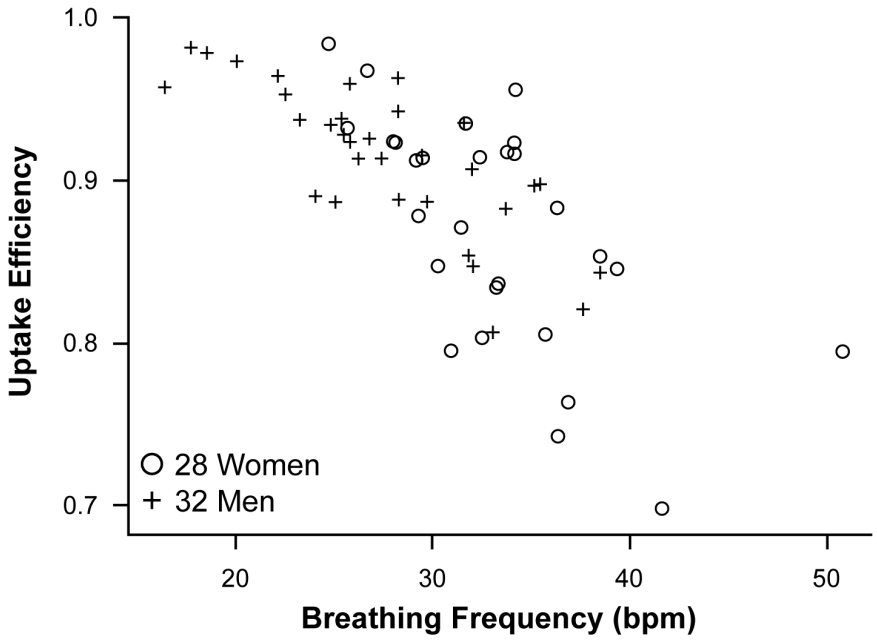


Figure 4-3. Ozone uptake efficiency as a function of breathing frequency at a minute ventilation of 30 L/min. The uptake efficiency was well correlated with breathing frequency ($r = -0.723$, $p < 0.001$) and tidal volume (*not illustrated;* $r = 0.490$, $p < 0.001$).

Source: From Ultman et al. (2004).

4.2.3 Dosimetry Modeling

When all of the animal and human in vivo O_3 uptake efficiency data are compared, there is a good degree of consistency across data sets, which raises the level of confidence with which these data sets can be used to support dosimetric model formulations. Models predict that the net O_3 dose (O_3 flux to air-liquid interface) gradually decreases distally from the trachea toward the end of the TB and then rapidly decreases in the pulmonary region. However, the tissue dose (O_3 flux to liquid-tissue interface) is low in the trachea, increases to a maximum in the terminal bronchioles and the first generation of the pulmonary region, and then decreases rapidly distally into the pulmonary region. The increased V_T and flow, associated with exercise in humans or CO_2-stimulated ventilation increases in rats, shifts O_3 dose further into the periphery of the lung, causing a disproportionate increase in distal lung dose.

Localized damage to lung tissue has been modeled showing variation of O_3 dose among anatomically equivalent ventilatory units as a function of path length from the trachea with shorter paths showing greater dose (Overton and Graham, 1995). More recent data indicate that the primary site of acute cell injury occurs in the conducting airways (Postlethwait et al., 2000). These data must be considered when developing models that attempt to predict site-specific locations of O_3-induced injury. The early models computed relationships between delivered regional dose and response with the assumption that O_3 was the active agent responsible for injury. It is now known that reactive intermediates such as hydrohydroxyperoxides and aldehydes are important agents mediating the response to O_3 (further discussed in Section 5.3.1). Thus, models must consider O_3 reaction/diffusion in the ELF and ELF-derived reactions products.

Table AX4-2 in the annex presents a summary of new theoretical studies of the uptake of O_3 by the RTs (or regions) of humans and laboratory animals that have become available since the 1996 review. They are discussed below.

Overton and Graham (1995) created a rat model combining multiple path anatomic models and one-dimensional convection-dispersion equations which simulates transport and uptake of O_3 in airways and airspaces of the modeled TB region. Predictions from this model realistically detail O_3 transport and uptake of different but morphologically equivalent sites. Using computational fluid dynamics (CFD), Cohen-Hubal et al. (1996) modeled the effect of the mucus layer thickness in the nasal passage of a rat. Predictions of overall uptake were within the range of measured uptake. Predicted regional O_3 flux was correlated with measured cell proliferation for the CFD simulation that incorporated two regions, each with a different mucus thickness. But using bolus-response data described above, Hu et al. (1994) and Bush et al. (2001) estimate a reaction rate constant that is more than 1000 times as large as that used by Cohen-Hubal et al. (1996).

With a RT dosimetry model, Overton et al. (1996) investigated the sensitivity of uptake efficiency, proximal alveolar region (PAR) dose, and PAR dose ratio to TB region volume (V_{TB}) and TB region expansion in humans and rats. The PAR was defined as the first generation distal to terminal bronchioles and the PAR dose ratio was defined as the ratio of a rat's predicted PAR dose to a human's predicted PAR dose. This ratio relates human and rat exposure concentrations so that both species receive the same PAR dose. In rats, the PAR is a region of major damage

from O_3. For each species, three values of V_{TB} were used: a mean value from the literature and the mean ± twice the SD. For both the rat and human simulations, there were several general findings: (1) uptake efficiency and PAR dose both increased with decreasing V_{TB}, e.g., using the highest TB region mass transfer coefficient (k_{TB}), the PAR dose for V_{TB} – 2SD was five times greater than the PAR dose for V_{TB} + 2SD, (2) uptake efficiency and PAR dose both decreased with TB expansion relative to no expansion, 3) PAR dose increased with tidal volume, 4) PAR dose increased with decreasing k_{TB}, and 5) uptake efficiency increased with k_{TB}.

Bush et al. (2001) modified their single-path model (Bush et al., 1996b) so that simulations would coincide with experimental uptake efficiency data for O_3 and Cl_2 during oral and nasal breathing. Relative to their original model, the Bush et al. (2001) model added lung expansion and modified the mass transfer coefficients for both the gas-phase (k_g) and the liquid-phase (k_l). Consistent with Overton et al. (1996), considering expansion of the TB airways reduced uptake efficiency versus no expansion. As very little inhaled O_3 reaches the peripheral lung, it was not surprising that alveolar expansion had minimal affect on uptake efficiency. Ignoring the O_3 reaction rate constant (k_r), the simulations for O_3 and Cl_2 were nearly the same since the gas-phase diffusion coefficients of O_3 and Cl_2 are similar. But for a given V_P the TB airways of the lung, experimental bolus uptake are always less for O_3 than for Cl_2. The authors surmised that the difference between the uptake for these gases could be explained adequately based solely on the diffusive resistance of O_3 in airways surface fluid (modeled by k_r). Qualitatively, model simulations also agreed well with the experimental data of Gerrity et al. (1995).

Age- and gender-specific differences in both regional and systemic uptake in humans was modeled using a physiologically-based pharmacokinetic (PBPK) approach (Sarangapani et al., 2003). The model estimated that regional (URT, TB, pulmonary) extraction efficiency of O_3 is relatively insensitive to age and gender.

A recent attempt was made (Mudway and Kelly, 2004) to model O_3 dose-inflammatory response using a meta-analysis of 23 exposures in published human chamber studies. The O_3 concentrations ranged from 0.08 to 0.6 ppm and the exposure durations ranged from 60 to 396 minutes. The analysis showed linear relationships between O_3 dose and neutrophilia in bronchoalveolar lavage fluid (BALF). Linear relationships were also observed between O_3 dose and protein leakage into BALF, which suggested to the authors that a large-scale study could determine a possible O_3 threshold level for these inflammatory responses. These recent findings

seem consistent with the linear relationship between O_3 dose to pulmonary tissues normalized for body weight and lavage fluid protein in rats, guinea pigs, and rabbits (Miller et al., 1988).

4.2.4 Summary and Conclusions - Dosimetry

Ozone is a highly reactive gas and powerful oxidant with a short half-life. Uptake occurs in mucous membranes of the RT where O_3 reacts with components of the ELF. Uptake efficiency is chemical-reaction dependent and the reaction products (hydrogen peroxide, aldehydes, and hydroxyhydroperoxides) created by ozonolysis of polyunsaturated fatty acids mediate O_3 toxicity. The 1996 O_3 AQCD reported that uptake of O_3 in rat is about 0.50 and in humans at rest is about 0.8 to 0.95. In humans, about 0.07 of the O_3 is removed in the larynx/trachea, about 0.50 in the head, and about 0.43 in the lungs, where the primary site of damage was believed to be the CAR. Increasing flow shifted O_3 uptake distally toward smaller airways of the lung. Studies in humans showed that increasing minute ventilation with exercise (by increasing both breathing frequency and tidal volume) causes only a small decrease in uptake efficiency by the total RT. The nasal passages appeared to absorb more O_3 than the oral passages. Comparing BAL cells, a 0.4 ppm exposure in exercising humans showed 4 to 5 times the retained dose of O_3 relative to rats exposed at rest to the same concentration.

New research on O_3 uptake has been performed in humans but not in laboratory animals. Bolus-response studies demonstrated that a previous continuous exposure to O_3 decreases the absorption of a bolus of O_3, probably due to depletion of compounds able to absorb O_3. Continuous exposure to NO_2 and SO_2 increased absorption of a bolus of O_3. These data are of some relevance to environmental exposures where humans may receive differing concentrations of O_3 depending on time of day. Verifying prior work, the bolus-response method was used to demonstrate that O_3 bolus uptake is sensitive to the mode of breathing and to the airflow rate. As flow is increased from 150 to 1000 mL/s, O_3 boli penetrated deeper into the lung and penetration was further increased by oral versus nasal breathing. This suggests that the switch from nasal to oral breathing coupled with increases in respiratory flow as occurs during exercise causes a shift in regional O_3 dose deeper into the lung, increasing the potential of damage to bronchiolar and alveolar tissues. The finding that O_3 uptake is inversely related to airflow also agrees with earlier animal studies.

New general uptake study data demonstrate that exercising men and women receiving 0.2 or 0.4 ppm O_3 at 20 L/min for 60 minutes or 40 L/min for 30 minutes absorb 0.56 to 0.98. The absorbed fraction or FA is affected only by large changes in concentration, minute volume, and exposure time. This suggests that for exposure times <2 h, inhaled dose is a reasonable predictor of actual uptake as long as there are fixed concentrations of O_3 and fixed levels of exercise. Individuals exposed to similar concentrations vary considerably in the amount of actual dose received. This intersubject variability was also demonstrated in a study of O_3 uptake in nasal cavities of men and women. The FA in the nose was inversely related to the flowrate and the concentration of O_3, suggesting that simultaneously occurring diffusion and chemical reactions in the mucous layer were the limiting factors in O_3 uptake. Both these general uptake studies, done at environmentally relevant O_3 concentrations, indicate that inter-individual differences in fractional uptake, which can range from 0.25 to 0.97, are extremely important in O_3 dose-response relationships.

The consistency of uptake data generated in animal and human studies allow a high level of confidence in their use in dosimetry modeling. Early models predicted that net O_3 dose to ELF and tissue gradually decreases distally from the trachea toward the end of the TB and then rapidly decreases in the pulmonary region. Exercise-induced or CO_2-stimulated increases in V_T and flow, shift O_3 dose further into the periphery of the lung, causing a disproportionate increase in distal lung dose. Localized damage to lung tissue has been modeled showing variation of O_3 dose among anatomically equivalent ventilatory units as a function of path length from the trachea with shorter paths showing greater damage.

New models have produced some refinements of earlier models such as: (1) the use of mucus resistance and thickness in describing O_3 dosimetry and determining the patterns of O_3-induced lesions; (2) the shape of the dose versus generation plot along any path from the trachea to alveoli is independent of path, with the tissue dose decreasing with increasing generation index; (3) simulations sensitive to conducting airway volume but relatively insensitive to characteristics of the respiratory airspace; (4) the importance of TB region expansion; (5) the importance of dose received in the PAR both inter-individual differences and extrapolations based on dose; and (6) revaluation of mass transfer coefficients for conducting airways. Additionally, more recent data indicate that the primary site of acute cell injury occurs

in the conducting airways and that reactive intermediates in the ELF, rather than O_3 itself, are responsible for pulmonary injury. These data must be considered when developing new models.

4.3 SPECIES HOMOLOGY, SENSITIVITY, AND ANIMAL-TO-HUMAN EXTRAPOLATION

Basic similarities exist across human and other animals species with regard to basic anatomy, physiology, biochemistry, cell biology, and disease processes. However, there are obviously some species differences that have the potential to affect both the patterns of O_3 uptake in the respiratory tract as well as responses. For instance, primates are oronasal breathers with a dichotomous branching lung structure, whereas, rodents are obligate nasal breathers with a monopodial branching lung structure (Miller et al., 1993). Even when comparing nasal breathing, differences in the nasal structure between primates and rodents can affect both the site and amount of gaseous uptake in this region (DeSesso, 1993; Morgan et al., 1989). Cellular profiles also differ between species as a function of location in the respiratory tract (Miller et al., 1993; Plopper et al., 1989; Stone et al., 1992).

The homology as it exists creates similarities in acute O_3-induced effects, especially in the respiratory tract and in lung defense mechanisms. Rodents appear to have a slightly higher tachypneic response to O_3, which is clearly concentration-dependent in most species and shows parallel exacerbation when hyperventilation (e.g., exercise or CO_2) is superimposed. What is not known is whether this is evidence of pulmonary irritant sensitivity, perhaps as a prelude to toxicity, or whether tachypnea is a defensive action taken by the respiratory system to minimize distal lung O_3 deposition. Airway or lung resistance in humans is not affected appreciably by acute exposure to O_3, except under conditions of heavy exercise; animals appear to need high-level exposures or special preparations that bypass nasal scrubbing. Dynamic lung compliance (Cdyn) has been shown to have small magnitude decreases in response to O_3 in some studies across species, but it is thought that these changes are of little biological significance for ambient exposures. Spirometric changes, the hallmark of O_3 response in humans, occur in rats, but to a lesser degree. It is unclear, however, the degree to which anesthesia (rat) and the comparability of hyperventilation induced by CO_2 (rat) or exercise (human) may influence this difference in

responsiveness. Collectively, the acute functional response of laboratory animals to O_3 appears quite homologous to that of the human.

Examination of BAL constituents show that the influx of inflammatory cells and protein from the serum is influenced by species, but perhaps to less extent than by ventilation and antioxidant status. Adjustment for these factors can modulate responses to approximate animal responses to those of humans. Unfortunately, these influential factors are rarely measured and, even less often, controlled. Increases in protein levels in BALF with O_3 exposures in guinea pigs are also a factor in the species' susceptibility to the effects of O_3. Species comparisons of acute O_3 exposures to mice, guinea pigs, rats, hamsters, and rabbits found that guinea pigs were the most responsive (to $\geq$0.2 ppm); rabbits were the least responsive (2.0 ppm only); and rats, hamsters, and mice were intermediate (effects at $\geq$1.0 ppm). Rats and humans have subtle species-specific differences in inflammatory responses to O_3 in terms of the timing of PMN influx in the nasal and bronchoalveolar regions.

When humans are exposed to O_3 repeatedly for several consecutive days, lung function decrements subside, and normal spirometric parameters are regained (see Section 6.6). This phenomenon of functional attenuation also has been demonstrated in rats, not only in terms of spirometry, but also in terms of the classic tachypneic ventilatory response. Full or partial attenuation of some BAL parameters also appears to occur in both rats and humans, but exposure scenario appears to play a role; other cellular changes do not attenuate (see Section 6.9.4). Existing epidemiologic studies provide only suggestive evidence that persistent or progressive deterioration in lung function is associated with long-term oxidant-pollutant exposure (See Chapter 7). With chronic, repeated exposures to $\geq$0.12 ppm O_3, however, laboratory animals demonstrate changes in lung structure, function, and biochemistry that are indicative of airway irritation and inflammation with the possible development of chronic lung disease (U.S. Environmental Protection Agency, 1996). Based on the apparent homology of these responses between humans and laboratory animals, animal studies appear to provide a means for assessing such chronic health concerns.

A species' susceptibility to the effects of O_3 exposure may be due, in part, to biochemical differences among species. Evidence for this is provided by differences in activity of SD rat and rhesus monkey CYP moonoxygenases elicited by O_3 exposure (Lee et al., 1998). Additional characterization of species- and region-specific CYP enzymes will create a better understanding

of the differences in response to O_3. This will allow more accurate extrapolation from animal exposures to human exposures and toxic effects.

Antioxidant metabolism varies widely among species, which can greatly influence the effects of O_3 (discussed in greater detail in 5.2.1.3). The guinea pig appears to be the species most susceptible to O_3. Early studies ranked mice > rats > guinea pigs in order of antioxidant responsiveness to O_3 challenge. Guinea pigs have been shown to have lower basal levels of GSH transferase activity, lower activity of GSH peroxidases, and lower levels of vitamin E compared to rats. These lower levels of antioxidants combined with increases in protein levels in BALF (discussed above) with O_3 exposures likely explain, at least in part, the species' susceptibility to the effects of O_3.

Because cytokine and chemokine responses are so important in an animal's defense against O_3 exposure, comparisons of differences in species expression and activity of these inflammatory mediators is necessary. Arsalane et al. (1995) compared guinea pig and human AM recovered in BALF and subsequently exposed in vitro to 0.1 to 1 ppm for 60 minutes. Measurement of inflammatory cytokines showed a peak at 0.4 ppm in both species. Guinea pig AM had an increase in IL-6 and TNFa while human AM had increases in TNFa, IL-1b, IL-6 and IL-8. This exposure also caused an increase in mRNA expression for TNFa, IL-1b, IL-6 and IL-8 in human cells. At 0.1 ppm exposures, only TNFa secretion was increased. These data suggest similar cytokine responses in guinea pigs and humans, both qualitatively and quantitatively.

Species differences in morphological responses to O_3 exposure have been characterized by Dormans et al. (1999), as discussed in previous sections. Dormans et al. (1999) continuously exposed rats, mice, and male guinea pigs to filtered air, 0.2, or 0.4 ppm O_3 for 3, 7, 28, and 56 days. The animals exposed for 28 days were examined at 3, 7, or 28 days PE. Depending on the endpoint studied, the species varied in sensitivity. Greater sensitivity was shown in the mouse as determined by biochemical endpoints, persistence of bronchiolar epithelial hypertrophy, and recovery time. Guinea pigs were more sensitive in terms of the inflammatory response though all three species had increases in the inflammatory response after three days that did not decrease with exposure. These data on inflammation are in general agreement with Hatch et al., (1986), discussed above. In all species, the longest exposure to the highest O_3 concentration caused increased collagen in ductal septa and large lamellar bodies in Type II

cells, but that response also occurred in rats and guinea pigs at 0.2 ppm. No fibrosis was seen at the shorter exposure times and the authors question whether fibrosis occurs in healthy humans after continuous exposure. The authors do not rule out the possibility that some of these differences may be attributable to differences in total inhaled dose or dose actually reaching a target site. Overall, the authors rated mice as most susceptible, followed by guinea pigs and rats.

Comparisons of airway effects in rats, monkeys and ferrets resulting from exposures of 1.0 ppm O_3 for 8 h (Sterner-Kock et al. 2000) demonstrated that monkeys and ferrets had similar inflammatory responses and epithelial necrosis. The response of these two species was more severe than that seen in rats. These data suggest that ferrets are a good animal model for O_3-induced airway effects due to the similarities in pulmonary structure between primates and ferrets. However, the mechanisms of O_3 effects at these high concentrations may differ from those at more realistic levels.

A number of species, including nonhuman primates, dogs, cats, rabbits, and rodents, have been used to study the effects of O_3 exposure on airway bronchoconstriction. A commonly used model of bronchospasm utilizes guinea pigs acutely exposed to high O_3 concentrations (2 to 3 ppm) to induce airway hyperreactivity (AHR). As mentioned earlier, the model is helpful for determining mechanistic aspects of AHR, but is not really relevant for extrapolation to potential airway responses in humans exposed to ambient levels of O_3. Additionally, guinea pigs have been shown to have AHR in other studies that is very similar to asthmatic humans, but the utility of guinea pig data is somewhat limited by their disparity from other animal models.

The rat is a key species used in O_3 toxicological studies, but the rat has both behavioral and physiological mechanisms that can lower core temperature in response to acute exposures, thus limiting extrapolation of rat data to humans. Iwasaki et al. (1998) evaluated cardiovascular and thermoregulatory responses to O_3 at exposure of 0.1, 0.3, and 0.5 ppm O_3 8 hrs/day for 4 consecutive days. A dose-dependent disruption of HR and T_{co} was seen on the first and second days of exposure, which then recovered to control values. Watkinson et al. (2003) exposed rats to 0.5 ppm O_3 and observed this hypothermic response, which included lowered HR, lowered T_{co}, and increased inflammatory components in BALF. The authors suggested that the response is an inherent reflexive pattern that can possibly attenuate O_3 toxicity in rodents. They discuss the cascade of effects created by decreases in T_{co}, which include: (1) lowered metabolic rate, (2) altered enzyme kinetics, (3) altered membrane function, (4) decreased oxygen

consumption and demand, (5) reductions in minute ventilation, which would act to limit the dose of O_3 delivered to the lungs. These effects are concurrent with changes in HR which lead to: (1) decreased CO, (2) lowered BP, and (3) decreased tissue perfusion, all of which may lead to functional deficits. The hypothermic response has not been observed in humans except at very high exposures, which complicates extrapolation of effects in rats to humans.

The importance of animal studies derives from their utilization in determining cause-effect relationships between exposure and health outcome, but the animal data must be integrated with epidemiological studies and controlled human clinical studies. Animal studies can corroborate both clinical and epidemiology studies and further provide important data that is impossible to collect in human studies. Toxic pulmonary and extrapulmonary effects following O_3 exposure have been well-studied in rodents, nonhuman primates, and a few other species; so, extrapolation, both qualitative and quantitative, to human exposures and consequent health effects is possible. Quantitative extrapolation, required to determine what specific exposure is likely to cause an effect in humans, is theoretically founded on the equivalency of mechanisms across species. At the molecular level, O_3 acts on the carbon-carbon double bond in polyunsaturated fatty acids and on sulfhydryl groups in proteins, both of which are found within cell membranes in animals and humans. At higher levels of cellular organization, cells affected in animals (e.g., AMs, Type 1 cells) have similar functions in humans, and organ systems (e.g., respiratory system) have major interspecies similarities. However, interspecies differences do occur and complicate extrapolation.

Quantitative extrapolation, which involves a combination of dosimetry and species sensitivity, still requires more research before it can be fully realized. Knowledge of dosimetric animal-to-human extrapolation is more advanced than that of species-sensitivity, but extrapolation models have not been completely validated, and therefore, significant uncertainties remain. Mathematical modeling of O_3 deposition in the lower respiratory tract (i.e., from the trachea to alveoli) of several animal species and humans shows that the pattern of regional dose is similar, but that absolute values differ. In spite of structural and ventilatory differences between species, the greatest predicted tissue dose is to the CAR. Even though the CAR of rats has very rudimentary respiratory bronchioles, compared to well-developed ones in primates, the CAR of both rats and nonhuman primates respond similarly to O_3.

Experimental measurement of delivered O_3 doses estimate that total respiratory uptake is ~47% in laboratory animals and ~87% in exercising humans, while nasopharyngeal removal is ~17% in rats and ~40% in humans. The previous O_3 AQCD (U.S. Environmental Protection Agency, 1996) provided the first quantitative animal-to-human extrapolation of morphological changes in the proximal alveolar region using rat and monkey studies. The extrapolation predicted that a 9-year-old child would have a 20% or 75% increase in PAR tissue thickness if their sensitivity to O_3 was equal to that of a rat or monkey, respectively. Adults would have 15 or 70% increase, suggesting the potential for chronic effects in humans. In spite of the significant uncertainties, this extrapolation raises concern about the potential for chronic effects in humans

Experiments using 2 h exposures to 0.4 ppm $^{18}O_3$ suggested that exercising (15 min intervals, rest and exercise at 60 L/min) humans received a 4- to 5-fold higher $^{18}O_3$ concentrations in BAL than resting rats (Hatch et al., 1994). That level of exposure increased BAL protein and PMNs in humans, while a concentration of 2.0 ppm in rats was necessary for similar effects. Caveats in the interpretation of $^{18}O_3$ studies include: (1) only a very small portion of the labeled compound is recoverable to assess incorporation; and (2) if species being compared differ in physiocochemical factors controlling mass transfer and downstream O_3 metabolism, it could cause significant differences in the amount of inhaled $^{18}O_3$ that is detected during subsequent tissue analysis. Further, species differences in pulmonary anatomy, ventilation, antioxidants, and susceptibility all influence dose, repair processes, and tolerance to subsequent O_3 exposure. Important differences between exercising humans and resting rats that can affect tissue O_3 dose include: (1) increased ventilation and O_3 delivery with exercise; (2) decreased pulmonary ventilation and body temperature during O_3 exposure in rats; (3) diminished dose received in rats due to their burying their noses in their fur during exposure; and (4) increased concentration of antioxidants in ELF in rats compared to humans. These antioxidants are important for converting O_3 to inactive products before toxicity occurs (Kari et al., 1997; Gunnison and Hatch, 1999; Plopper et al., 1998), though this quenching is not quantitative. These and possibly other differences between rats and humans suggest that a 2 ppm exposure in nonexercising rats approximates a 0.4 ppm exposure in exercising humans. Further comparisons of exercising human exposure to 0.1 ppm for 6 hours (Devlin et al., 1991)

and resting rat exposure to 0.3 ppm show inflammatory and permeability changes in humans but not rats.

4.3.1 Summary and Conclusions: Species Homology, Sensitivity, and Animal-to-Human Extrapolation

Comparisons of acute exposures in rats and humans suggest that, though both species have similar qualitative responses to O_3 exposure, there are interspecies mechanistic disparities that necessitate careful comparisons of dose-response relationships. There is no perfect nonhuman species with which to model O_3 toxicity. All have limitations that must be considered when attempting to extrapolate to human exposures. Awareness of these limitations, even at the level of subtle strain differences within a test species, is extremely important. The currently available data suggest that LOELs in resting rats are approximately 4- to 5-fold higher than for exercising humans for toxicological endpoints including BAL protein and BAL PMNs. Studies comparing species-specific differences in O_3-induced effects showed that guinea pigs were the most susceptible, rabbits the least susceptible, and rodents intermediate in susceptibility. The recent work being done utilizing various mouse strains with differing sensitivities to O_3 will help us to understand the extremely complex inter-individual differences in human sensitivity to O_3.

REFERENCES

Arsalane, K.; Gosset, P.; Vanhee, D.; Voisin, C.; Hamid, Q.; Tonnel, A.-B.; Wallaert, B. (1995) Ozone stimulates synthesis of inflammatory cytokines by alveolar macrophages *in vitro*. Am. J. Respir. Cell Mol. Biol. 13: 60-68.

Asplund, P. T.; Ben-Jebria, A.; Rigas, M. L.; Ultman, J. S. (1996) Longitudinal distribution of ozone absorption in the lung: effect of continuous inhalation exposure. Arch. Environ. Health 51: 431-438.

Bush, M. L.; Asplund, P. T.; Miles, K. A.; Ben-Jebria, A.; Ultman, J. S. (1996a) Longitudinal distribution of O_3 absorption in the lung: gender differences and intersubject variability. J. Appl. Physiol. 81: 1651-1657.

Bush, M. L.; Raybold, T.; Abeles, S.; Hu, S.-C.; Ben-Jebria, A.; Ultman, J. S. (1996b) Longitudinal distribution of ozone absorption in the lung: simulation with a single-path model. Toxicol. Appl. Pharmacol. 140: 219-226.

Bush, M. L.; Zhang, W.; Ben-Jebria, A.; Ultman, J. S. (2001) Longitudinal distribution of ozone and chlorine in the human respiratory tract: simulation of nasal and oral breathing with the single-path diffusion model. Toxicol. Appl. Pharmacol. 173: 137-145.

Cohen-Hubal, E. A.; Kimbell, J. S.; Fedkiw, P. S. (1996) Incorporation of nasal-lining mass-transfer resistance into a CFD model for prediction of ozone dosimetry in the upper respiratory tract. Inhalation Toxicol. 8: 831-857.

DeSesso, J. M. (1993) The relevance to humans of animal models for inhalation studies of cancer in the nose and upper airways. Qual. Assur. (San Diego) 2: 213-231.

Devlin, R. B.; McDonnell, W. F.; Mann, R.; Becker, S.; House, D. E.; Schreinemachers, D.; Koren, H. S. (1991) Exposure of humans to ambient levels of ozone for 6.6 hours causes cellular and biochemical changes in the lung. Am. J. Respir. Cell Mol. Biol. 4: 72-81.

Dormans, J. A. M. A.; Van Bree, L.; Boere, A. J. F.; Marra, M.; Rombout, P. J. A. (1999) Interspecies differences in time course of pulmonary toxicity following repeated exposure to ozone. Inhalation Toxicol. 11: 309-329.

Gerrity, T. R.; Biscardi, F.; Strong, A.; Garlington, A. R.; Brown, J. S.; Bromberg, P. A. (1995) Bronchoscopic determination of ozone uptake in humans. J. Appl. Physiol. 79: 852-860.

Gunnison, A. F.; Hatch, G. E. (1999) O_3-induced inflammation in prepregnant, pregnant, and lactating rats correlates with O_3 dose estimated by ^{18}O. Am. J. Physiol. 276: L332-L340.

Hatch, G. E.; Slade, R.; Stead, A. G.; Graham, J. A. (1986) Species comparison of acute inhalation toxicity of ozone and phosgene. J. Toxicol. Environ. Health 19: 43-53.

Hatch, G. E.; Slade, R.; Harris, L. P.; McDonnell, W. F.; Devlin, R. B.; Koren, H. S.; Costa, D. L.; McKee, J. (1994) Ozone dose and effect in humans and rats: a comparison using oxygen-18 labeling and bronchoalveolar lavage. Am. J. Respir. Crit. Care Med. 150: 676-683.

Hu, S.-C.; Ben-Jebria, A.; Ultman, J. S. (1994) Longitudinal distribution of ozone absorption in the lung: effects of respiratory flow. J. Appl. Physiol. 77: 574-583.

Iwasaki, T.; Takahashi, M.; Saito, H.; Arito, H. (1998) Adaptation of extrapulmonary responses to ozone exposure in conscious rats. Ind. Health 36: 57-60.

Kabel, J. R.; Ben-Jebria, A.; Ultman, J. S. (1994) Longitudinal distribution of ozone absorption in the lung: comparison of nasal and oral quiet breathing. J. Appl. Physiol. 77: 2584-2592.

Kari, F.; Hatch, G.; Slade, R.; Crissman, K.; Simeonova, P. P.; Luster, M. (1997) Dietary restriction mitigates ozone-induced lung inflammation in rats: a role for endogenous antioxidants. Am. J. Respir. Cell Mol. Biol. 17: 740-747.

Lee, C.; Watt, K. C.; Chang, A. M.; Plopper, C. G.; Buckpitt, A. R.; Pinkerton, K. E. (1998) Site-selective differences in cytochrome P450 isoform activities: comparison of expression in rat and rhesus monkey lung and induction in rats. Drug Metab. Dispos. 26: 396-400.

Miller, F. J.; Overton, J. H.; Gerrity, T. R.; Graham, R. C. (1988) Interspecies dosimetry of reactive gases. In: Mohr, U.; Dungworth, D.; McClellan, R.; Kimmerle, G.; Stöber, W.; Lewkowski, J., eds. Inhalation toxicology: the design and interpretation of inhalation studies and their use in risk assessment. New York, NY: Springer-Verlag; pp. 139-155.

Miller, F. J.; Overton, J. H.; Kimbell, J. S.; Russell, M. L. (1993) Regional respiratory tract absorption of inhaled reactive gases. In: Gardner, D. E.; Crapo, J. D.; McClellan, R. O., eds. Toxicology of the lung. 2nd ed. New York, NY: Raven Press; pp. 485-525. (Target organ toxicology series).

Morgan, K. T.; Monticello, T. M.; Patra, A. L.; Fleishman, A. (1989) Preparation of rat nasal airway casts and their application to studies of nasal airflow. In: Crapo, J. D.; Smolko, E. D.; Miller, F. J.; Graham, J. A.; Hayes, A. W., eds. Extrapolation of dosimetric relationships for inhaled particles and gases. New York, NY: Academic Press, Inc.; pp. 45-58.

Mudway, I. S.; Kelly, F. J. (2004) An investigation of inhaled ozone dose and the magnitude of airway inflammation in healthy adults. Am. J. Respir. Crit. Care Med. 169: 1089-1095.

Nodelman, V.; Ultman, J. S. (1999) Longitudinal distribution of chlorine absorption in human airways: a comparison to ozone absorption. J. Appl. Physiol. 87: 2073-2080.

Overton, J. H.; Graham, R. C. (1995) Simulation of the uptake of a reactive gas in a rat respiratory tract model with an asymmetric tracheobronchial region patterned on complete conducting airway cast data. Comput. Biomed. Res. 28: 171-190.

Overton, J. H.; Graham, R. C.; Menache, M. G.; Mercer, R. R.; Miller, F. J. (1996) Influence of tracheobronchial region expansion and volume on reactive gas uptake and interspecies dose extrapolations. Inhalation Toxicol. 8: 723-745.

Plopper, C. G.; St. George, J.; Mariassy, A.; Nishio, S.; Heidsiek, J.; Weir, A.; Tyler, N.; Wilson, D.; Cranz, D.; Hyde, D. (1989) Species differences in airway cell distribution and morphology. In: Crapo, J. D.; Smolko, E. D.; Miller, F. J.; Graham, J. A.; Hayes, A. W., eds. Extrapolation of dosimetric relationships for inhaled particles and gases. New York, NY: Academic Press, Inc.; pp. 19-34.

Plopper, C. G.; Hatch, G. E.; Wong, V.; Duan, X.; Weir, A. J.; Tarkington, B. K.; Devlin, R. B.; Becker, S.; Buckpitt, A. R. (1998) Relationship of inhaled ozone concentration to acute tracheobronchial epithelial injury, site-specific ozone dose and glutathione depletion in rhesus monkeys. Am. J. Respir. Cell Mol. Biol. 19: 387-399.

Postlethwait, E. M.; Joad, J. P.; Hyde, D. M.; Schelegle, E. S.; Bric, J. M.; Weir, A. J.; Putney, L. F.; Wong, V. J.; Velsor, L. W.; Plopper, C. G. (2000) Three-dimensional mapping of ozone-induced acute cytotoxicity in tracheobronchial airways of isolated perfused rat lung. Am. J. Respir. Cell Mol. Biol. 22: 191-199.

Rigas, M. L.; Ben-Jebria, A.; Ultman, J. S. (1997) Longitudinal distribution of ozone absorption in the lung: effects of nitrogen dioxide, sulfur dioxide, and ozone exposures. Arch. Environ. Health 52: 173-178.

Rigas, M. L.; Catlin, S. N.; Ben-Jebria, A.; Ultman, J. S. (2000) Ozone uptake in the intact human respiratory tract: relationship between inhaled dose and actual dose. J. Appl. Physiol. 88: 2015-2022.

Santiago, L. Y.; Hann, M. C.; Ben-Jebria, A.; Ultman, J. S. (2001) Ozone adsorption in the human nose during unidirectional airflow. J. Appl. Physiol. 91: 725-732.

Sarangapani, R.; Gentry, P. R.; Covington, T. R.; Teeguarden, J. G.; Clewell, H. J., III. (2003) Evaluation of the potential impact of age- and gender-specific lung morphology and ventilation rate on the dosimetry of vapors. Inhalation Toxicol. 15: 987-1016.

Sterner-Kock, A.; Kock, M.; Braun, R.; Hyde, D. M. (2000) Ozone-induced epithelial injury in the ferret is similar to nonhuman primates. Am. J. Respir. Crit. Care Med. 162: 1152-1156.

Stone, K. C.; Mercer, R. R.; Gehr, P.; Stockstill, B.; Crapo, J. D. (1992) Allometric relationships of cell numbers and size in the mammalian lung. Am. J. Respir. Cell Mol. Biol. 6: 235-243.

U.S. Environmental Protection Agency. (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available online at: www.epa.gov/ncea/ozone.htm.

Ultman, J. S.; Ben-Jebria, A.; Arnold, S. F. (2004) Uptake distribution of ozone in human lungs: intersubject variability in physiologic response. Boston, MA: Health Effects Institute. Available: http://www.healtheffects.org/Pubs/Ultman.pdf [29 July, 2005].

Watkinson, W. P.; Campen, M. J.; Wichers, L. B.; Nolan, J. P.; Costa, D. L. (2003) Cardiac and thermoregulatory responses to inhaled pollutants in healthy and compromised rodents: modulation via interaction with environmental factors. Environ. Res. 92: 35-47.

Weibel, E. R. (1980) Design and structure of the human lung. In: Fishman, A. P., ed. Pulmonary diseases and disorders. New York, NY: McGraw-Hill; p. 231.

5. TOXICOLOGICAL EFFECTS OF OZONE AND RELATED PHOTOCHEMICAL OXIDANTS IN LABORATORY ANIMALS AND IN VITRO TEST SYSTEMS

5.1 INTRODUCTION

A wide range of effects of ozone (O_3) has been demonstrated in laboratory animals. The major research findings are that environmentally relevant levels of O_3 cause lung inflammation; decreases in host defenses against infectious lung disease; acute changes in lung function, structure, and metabolism; chronic lung disease, some elements of which are irreversible; and systemic effects on target organs (e.g., brain, heart, liver, immune system) distant from the lung. The research also has served to expand the understanding of mechanisms of O_3 toxicity and the relationships between concentration and duration of exposure.

The framework for presenting the health effects of O_3 in animals begins with a presentation of respiratory tract effects, followed by systemic effects, and then interactions of O_3 with other common co-occurring pollutants. The information discussed in this chapter is founded on a very wide body of literature on studies in laboratory animals and on in vitro test systems of animal cell lines and organ systems that may mimic responses in intact animals. The direct effects of O_3 in humans are discussed in the following chapter (Chapter 6).

This chapter is not intended to be a compendium of all that is known about O_3; rather, it is an update of the toxicology chapter from the last O_3 criteria document (U.S. Environmental Protection Agency, 1996), or 1996 O_3 CD, and other reviews of the earlier published literature. The historical O_3 literature is very briefly summarized in an opening paragraph of each section or subsection. This paragraph is intended as a very concise overview of previous work, and the reader is referred to the 1996 O_3 CD for more detailed discussion of the literature prior to the early 1990's. Each section then continues with brief discussions of the key new studies (or somewhat older studies that were not included in the previous CD). Longer discussions of new studies are included where warranted. Sections are ended with comparisons of data from the previous CD with new data and basic conclusions are drawn. Summaries of new studies and results are provided in tables in Annex AX5.

Except for nitrogen dioxide (NO_2), the subject of another criteria document (U.S. Environmental Protection Agency, 1993), there is very little relevant information on other photochemical oxidants in the published literature. What is known about the effects of these other oxidants is also summarized briefly in this chapter.

5.2 RESPIRATORY TRACT EFFECTS OF OZONE

5.2.1 Biochemical Effects

Biochemically detected effects of O_3 are integrally involved in effects on both structure and function (respiratory and nonrespiratory) of the respiratory tract. Changes in xenobiotic metabolism, antioxidant metabolism and oxygen consumption, lipids and arachidonic acid metabolism, and collagen metabolism are all observed with O_3 exposure, though the mechanisms and associations are not fully understood.

5.2.1.1 Cellular Targets of O_3 Interaction

Ozone has the potential to interact with a wide range of different cellular components that include polyunsaturated fatty acids (PUFAs); some protein amino acid residues; and some low-molecular-weight compounds that include glutathione (GSH), urate, vitamins C and E, and free amino acids. Early work demonstrated that O_3, being a highly reactive compound, does not penetrate much beyond the epithelial lining fluid (ELF). Reaction/diffusion analyses suggest that O_3, at environmentally-relevant concentrations, diffuses no more than 0.1 to 0.2 μm into the ELF. Ozone-induced cell damage most likely results from its reactions with PUFAs to form stable but less reactive ozonide, aldehyde, and hydroperoxide reaction products. These reaction products (Crigee ozonides and hydroxyhydroperoxides) may act as signal transduction molecules involved in signaling of cellular responses such as inflammation, and thus mediate O_3 toxicity. These reactions are summarized in Figure 5-1 and studies published since the 1996 AQCD are listed in Table AX5-1.

Frampton et al. (1999) demonstrated the ozonation of PUFA to form nonanal and hexanal in rat BAL after exposures to 0.22 ppm O_3 for 4 h with exercise. Increases in nonanal were not accompanied by significant changes in lung function, in epithelial permeability, or in airway inflammation. Hexanal levels did not increase significantly and levels of both aldehydes

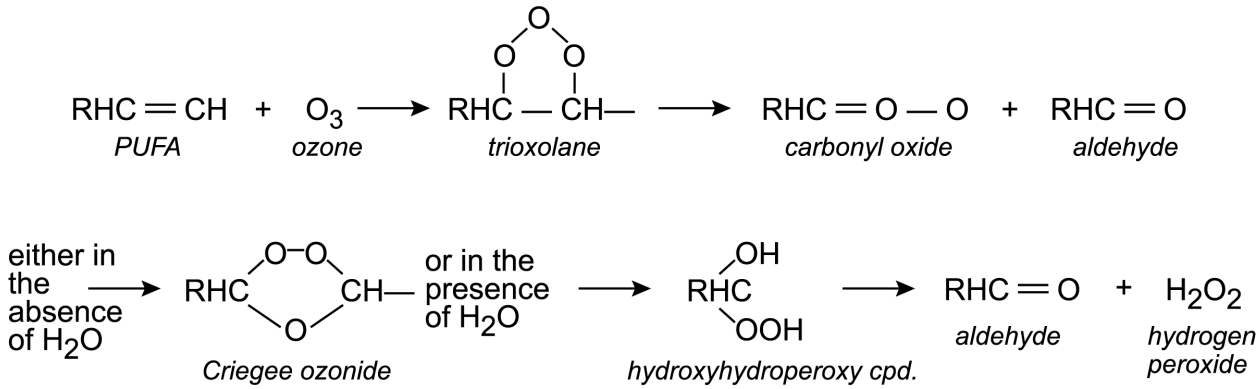


Figure 5-1. Major secondary products of ozone interaction with epithelial lining fluid and lung cells.

returned to baseline by 18 h PE. Pryor et al. (1996) exposed rats to 0.5 to 10 ppm O_3 both with and without 5% CO_2 to measure the amount of aldehyde generated in BAL, and also the rate of disappearance of aldehydes from the ELF following the O_3 exposure. Ozone exposure with CO_2 increased the tidal volume and the yield of aldehydes with a maximal aldehyde yield at 2.5 ppm for 1 h. Absolute yields were impossible to ascertain in this system because deposition of O_3 is unknown and aldehyde recovery is not complete due to loss of aldehyde by volatization and by diffusion into underlying tissue. The data showed that at 0.5 ppm O_3 with 5% CO_2, levels of hexanal and nonanal increased at 30 minutes, decreased slightly from that level at 60 minutes, was maximal at 90 minutes and then dropped to 60 minutes levels at 120 minutes. Levels of heptanal did not change appreciably during this time course. Levels of these aldehydes were dependent on a dynamic relationship between their production and the disappearance from the ELF. The authors stated that O_3 is the limiting reagent in this process because the amount of PUFA far exceeds the amount of O_3 on a molar basis. Because of the limitations of measuring aldehydes in this study paradigm, it is not useful for quantitative dosimetry; however, the authors suggest the study does serve to demonstrate the use of aldehydes as biomarkers of O_3 exposure since nonanol is produced in an O_3-specific pathway.

Postlethwait et al. (1998) utilized three biologically relevant models (isolated epithelial lining fluid, intact lung, and liposome suspensions) to determine the O_3-induced production of heptanal, nonanal and hexanal in an attempt to estimate formation of lipid-derived bioactive

compounds. Exposures used were 0.25 to 1.0 ppm for 30 to 60 minutes. Data suggest that PUFAs directly react with O_3 and the amount of bioactive lipids produced is inversely related to ascorbic acid availability. The authors caution that there are limitations to the use of measurements of these reactions products in determining O_3 dose-response relationships due to the heterogenous nature of O_3 reactions in the epithelial lining fluid. Connor et al. (2004) have recently examined the reactive absorption of O_3 (0.3 to 1.1 ppm for 1 to 2 h) within ELF using interfacial films composed of dipalmitoylglycero-3-phosphocholine (DPPC) and rat lung lavage fluid. The films reduced O_3 reactive absorption by antioxidants. Further experiments using a human lung fibroblast cell line exposed to O_3 demonstrated that ascorbic acid (AA) produced cell injury that high levels of O_3 and AA were needed to induce cell injury, and the DPPC films reduced the amount of cell injury. From these data the authors suggest that O_3 reactions with ELF substrates cause cell injury that films of active, saturated phospholipids reduce the local dose of O_3-derived reaction products, and that these interfacial phospholipids modulate the distribution of inhaled O_3 and the extent of site-specific cell injury.

Recent studies have examined the formation of ozonation products such as 4-hydroxynonenal (HNE), a toxic aldehyde that reacts with cysteine, histamine, and lysine amino acid residues and creates protein adducts. Hamilton et al. (1998) demonstrated (see Chapter 6) using human AM exposed to 0.4 ppm O_3 for 1 h that exposure caused apoptosis, an increase in a 32-kDa protein adduct, and an increase in ferritin and a 72-kDa heat shock protein. By exposing AM to HNE in vitro, all of these effects are replicated, which the authors interpret to mean that creation of protein adducts and apoptotic cell death are cellular toxic effect of acute O_3 exposure and that it is mediated, at least in part by HNE.

These recent reports combined with observations reported in the previous O_3 CD (US Environmental Protection Agency, 1996) suggest that interactions of O_3 with cellular components and ELF generate toxic ozonation products and mediate toxic effects through these products.

5.2.1.2 Monooxygenases

Both short- and long-term exposures to O_3 have been shown to enhance lung xenobiotic metabolism, possibly as a result of changes in the number and function of bronchiolar epithelial Clara cells and alveolar epithelial Type 2 cells. Studies of the effects of O_3 on lung

monooxygenases are listed in Table AX5-2. Early studies showed that exposure to O_3 increased CYP 2B1 (the major CYP isoform in rat lung) content and activity in rat lung. Ozone exposures also caused hypertrophy and hyperplasia of CYP 2B1-immunoreactive Clara cells. Comparisons of rat and rhesus monkey CYP isoforms demonstrated species-specific and region-specific (e.g., trachea, parenchyma) differences in the activities of P450 isoforms (Lee et al., 1998)

Watt et al. (1998) found that 1 ppm O_3 in both acute (8h, 1 ppm) and chronic (90 days, 1 ppm) exposures in rat increased CYP 2E1 in a region-specific manner. Paige et al. (2000a) showed that a long term exposure (0.8 ppm, 8h/day for 90 days) increased the activity of CYP 2B in distal lung but not trachea or intrapulmonary airways. Studies have focused on P450 gene expression to examine possible genetic mechanisms that may explain differential O_3-sensitivity (Mango et al., 1998). Mice (129 strain) deficient in Clara cell secretory protein (CCSP-/-), which are oxidant-sensitive, were exposed to 1 ppm O_3 for 2 hours. The CCSP null mice demonstrated increases in IL-6 and metallothionein (Mt) mRNA that preceded decreases in Clara cell CYP 2F2 mRNA (normally expressed at high levels in mouse lung) levels. In 129 strain wildtype (WT) mice, RNA levels changed similarly, to a lesser degree. These data suggest a protective role against oxidant damage for CCSP, and further, that genetic susceptibility to oxidant stress may be mediated, in part, by the gene coding for CCSP.

5.2.1.3 Antioxidants, Antioxidant Metabolism, and Mitochondrial Oxygen Consumption

Ozone also undergoes reactions with ascorbic acid (AA), reduced glutathione (GSH), and uric acid (UA), all antioxidants present in ELF (see Figure 5-2, A). In vivo experiments have shown that reactions with O_3 occur preferentially with antioxidants compared to proteins and lipids also present in ELF. This is a protective interaction, but even with environmentally relevant exposures to O_3, the reactivity of O_3 is not quantitatively quenched. Antioxidants offer some protection from O_3 exposure but often are not maintained at concentrations sufficient to fully protect the lung. Thus, O_3-induced cell injury occurs in both the lower and upper respiratory tract. Early work has shown that acute (1 week) exposures to <1 ppm O_3 increase antioxidant metabolism, including levels of cytosolic enzymes glucose-6-phosphate dehydrogenase (G6PD), 6-phosphogluconate dehydrogenase (6PGD), glutathione reductase (GR), and glutathione peroxidase (GSHPx). Re-exposure after a recovery period causes increases equivalent to first-time exposures, thus previous exposure appears to not be protective.

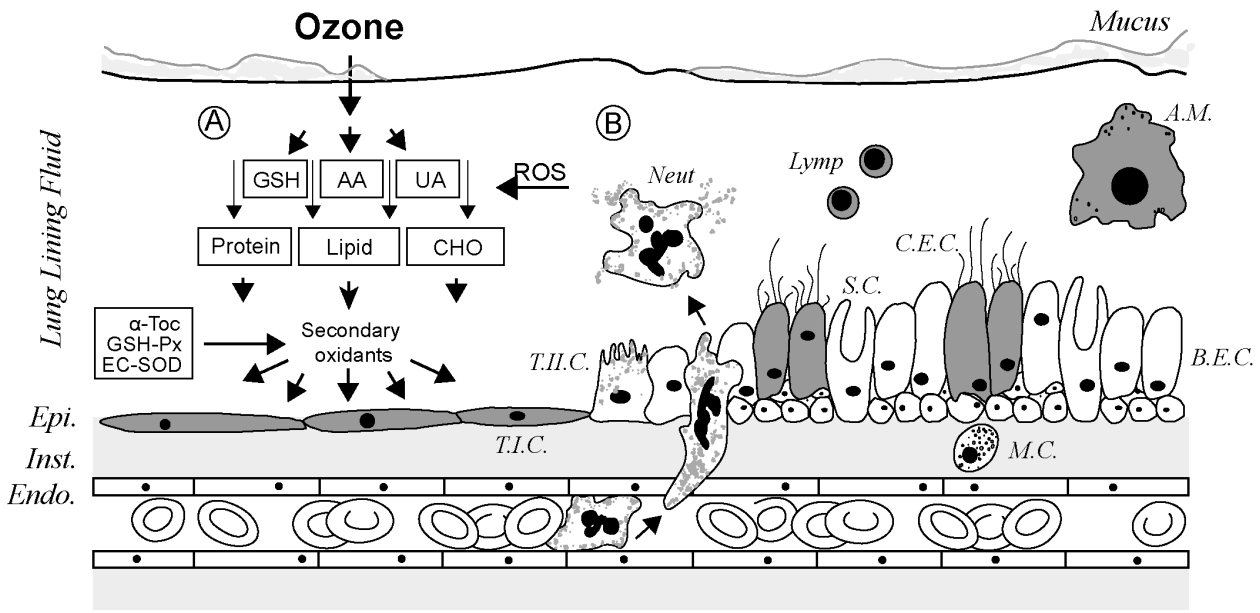


Figure 5-2. (Reprinted from Molecular Aspects of Medicine, I.S. Mudway and F.J. Kelly, Ozone and the Lung: a sensitive issue, page 36, (2000), with permission from Elsevier).

Increases in enzyme activity appear to increase as a function of age, suggesting that O_3 exposure can cause greater lung injury in the older animal. This has been attributed to differences in dose reaching lung target sites, differing base levels of antioxidants and antioxidant enzymes, and differences in cellular sensitivity. Species differences exist in antioxidant metabolism, with guinea pigs being very sensitive to O_3 due to their diminished increases in antioxidants and antioxidant enzymes. Chronic exposures of rats to urban patterns of O_3 (daily peaks of 0.25 ppm) caused increases in GSHPx and GR, but not superoxide dismutase (SOD). The enzyme changes could be accounted for by differences in the steady-state cell population or in cellular antioxidant capacity. More recent studies examining antioxidants and O_3 exposure are listed in Table AX5-3.

Ozone induced both site- and cell-specific changes in copper-zinc (Cu-Zn) and manganese (Mn) SOD in rats exposed to 1.0 ppm O_3 for up to 3 months (Weller et al., 1997). Cu-Zn SOD labeling was decreased in epithelial cells in airways and parenchyma. Mn SOD labeling was increased in both AM and epithelial type II cells of the centriacinar region (CAR), which the

authors suggest may allow these cells to tolerate further O_3 exposure. This work is in agreement with earlier work suggesting a role of SOD in protection of cells against oxidative stress.

Freed et al. (1999) evaluated the role of antioxidants in O_3-induced oxidant stress in dogs (exposed to 0.2 ppm in a 6 h exposure) by inhibiting the antioxidant transport using probenecid (an anion-transport inhibitor). Blocking antioxidant transport caused heterogeneously distributed increases in peripheral airway resistance and reactivity, supporting the hypothesis that in the lung periphery, endogenous antioxidants moderate the effects of O_3 and that this exposure is a subthreshold stimulus for producing effects on peripheral airway resistance and reactivity in dogs. The authors further found that treatment with probenecid also inhibited O_3-induced neutrophilic inflammation, providing evidence for a dissociation between airway function and inflammation. This suggests that O_3-induced inflammation and airway hyperreactivity (AHR) are independent phenomena operating through multiple mechanistic pathways.

Mudway and Kelly (1998) modeled the interactions of O_3 with ELF antioxidants using a continually mixed, interfacial exposure set up with O_3 concentrations of ranging from 0.1 to 1.5 ppm for durations ranging from 30 to 720 min. Uric acid was ranked the most O_3-reactive, AA the second most reactive, and GSH the least reactive. Thus, they concluded that GSH is not an important substrate for O_3, while UA appeared to be the most important reactive substrate, which confers protection from O_3 by removing it from inhaled air and limiting the amount that reaches the distal lung. By providing a substrate for O_3 reactions in the ELF, UA effectively reduces the diffusive resistance of O_3 (see Bush et al., 2001) in the TB airways and thus may serve to limit the amount of O_3 reaching the distal lung. The authors acknowledge limitations in extrapolating these data to in vivo O_3 exposures due to the absence of surfactant lipids and airway mucus in the model.

5.2.1.4 Lipid Metabolism and Content of the Lung

One of the major postulated molecular mechanisms of action of O_3 is peroxidation of mono- and polyunsaturated fatty acids and unsaturated neutral lipids in the lung. Because all of these lipids appear both in cell membranes and as secretions in the ELF, it is difficult to ascertain which lipid pool contributes to the formation of lipid ozonation products. As mentioned, O_3 can penetrate only about 0.1 to 0.2 μm into the ELF, so it is unlikely that O_3 reacts directly with

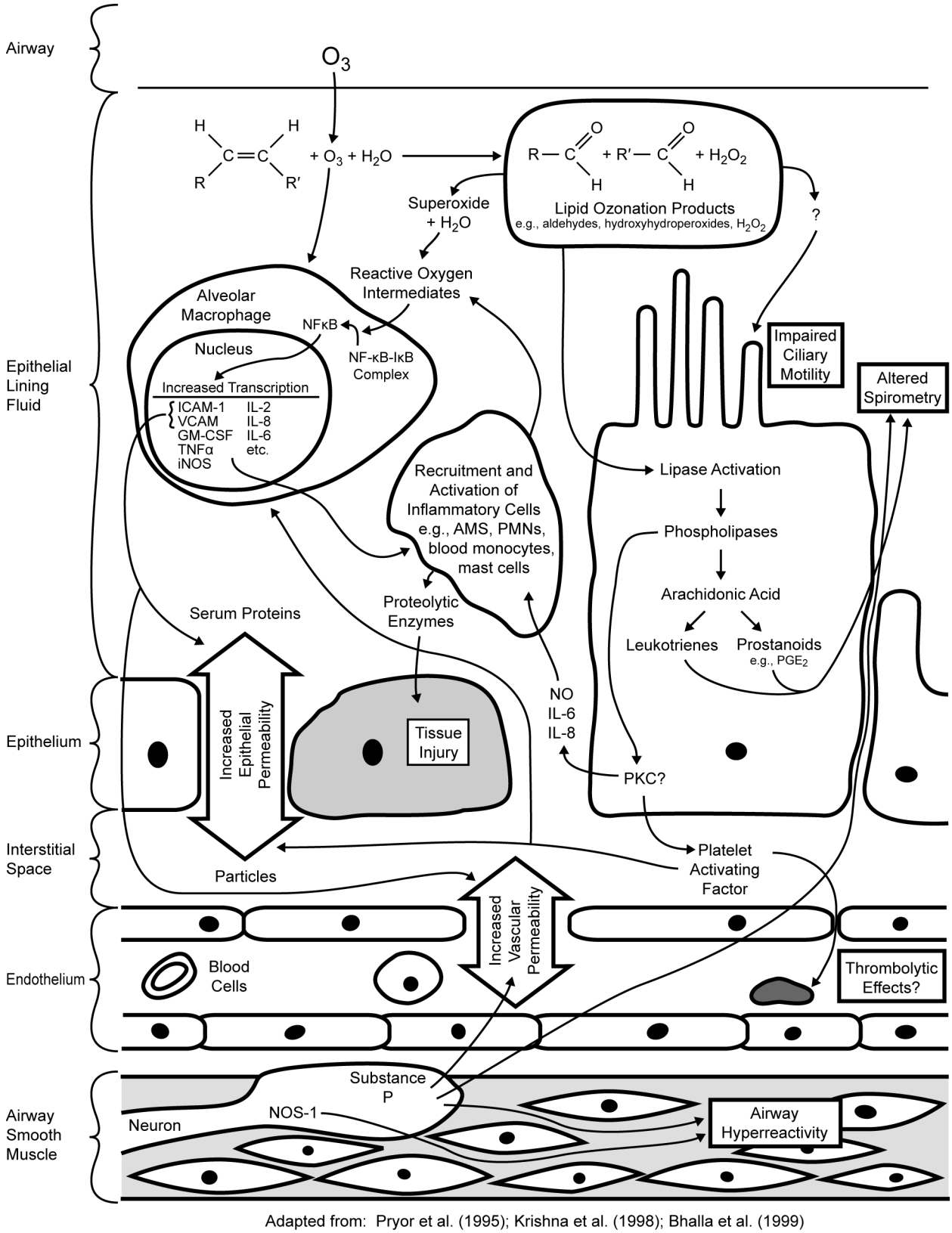
epithelial cell membranes, except in regions of distal lung where ELF is very thin or absent. The inflammatory cascade (shown in Figure 5-3) initiated by O_3 generates a mix of secondary reactants (e.g., aldehydes) which then are likely to oxidize lipids and proteins in cell membranes.

In both acute and short-term studies, a variety of lung lipid changes occur, including an increase in arachidonic acid. Metabolism of arachidonic acid produces a variety of biologically active mediators that can, in turn, affect host defenses, lung function, the immune system, and other functions. The protein A component of surfactant is also a primary target of O_3 interaction. During the first few days of O_3 exposure, the changes in lung lipid biosynthesis can be accounted for by the alveolar epithelial proliferative repair. With longer exposures (e.g., 0.12 ppm for 90 days) an increase in PUFAs and a decrease in cholesterol-esters are seen, indicative of long-term alterations of surfactant lipid composition.

Several new studies listed in Table AX5-4 examined the effects of O_3 exposure on phospholipids in lung tissue. Ozonation of PUFAs has been shown to generate other aldehydes such as nonanal and hexanal in rat (Pryor et al., 1996; Frampton et al., 1999). These aldehydes are short-lived and found to not affect lung function (Frampton et al., 1999). These observations suggest that levels of these aldehydes are dependent on a dynamic relationship between their production and their disappearance from the ELF.

Pryor et al. (1995) proposed a cascade mechanism whereby ozonation products cause activation of specific lipases, which then trigger the activation of second messenger pathways (e.g., phospholipase A_2 or phospholipase C). This group (Kafoury et al., 1999) showed that exposure of cultured human bronchial epithelial cells to the lipid ozonation product 1 -palmitoyl-2-(9-oxononanoyl)-sn-glycero-3-phosphocholine elicited release of platelet-activating factor (PAF) and prostaglandin E2, but not IL-6. The lipid ozonation product 1-hydroxy-1-hydroperoxynonane caused release of PAF and IL-6 in these cells, but not prostaglandin E2. These results suggest to the authors that O_3-induced production of lipid ozonation products causes release of proinflammatory mediators that then generate an early inflammatory response.

Very new work (Ballinger et al., 2005) has shown that ozone-induced membrane oxidation is augmented by antioxidants present in ELF. They utilized a red cell membrane model exposed to 0.8 ppm O_3 for 30 min. The monolayer of cells was intermittently covered by an aqueous film consisting of rat BALF or BALF plus added antioxidants. AA and GSH induced dose-dependent



Adapted from: Pryor et al. (1995); Krishna et al. (1998); Bhalla et al. (1999)

Figure 5-3. Mechanisms of ozone toxicity.

oxidative damage to the cell membrane proteins and lipids via secondary oxidant formation. The authors concluded that early in O_3 exposure, ELF antioxidants are high enough to drive reactive absorption of O_3 into the ELF and to concurrently quench secondary reaction products, thus limiting cell injury. With continued exposure, antioxidants levels decrease such that unreacted O_3 and cytotoxic products can diffuse to the cell membranes, causing injury. Limitations of this in vitro study are the possible differences in chemical species and mechanisms compared to in vivo systems.

Uhlson et al. (2002) reacted O_3 with calf lung surfactant which resulted in the production of 1-palmitoyl-2-(9'-oxo-nonanoyl)-glycerophosphocholine (16:0a/9-al-GPCho). The biological activity of this oxidized phospholipid included: (1) decreased macrophage viability, (2) induction of apoptosis in pulmonary epithelial-like A549 cells, (3) and release of IL-8 from A549 cells. Exposure levels of 0.125 ppm O_3 for 2–4 h in this system were capable of generating biologically active phospholipids that were capable of mediating toxic effects of O_3.

In addition to PUFA, cholesterol, the most abundant neutral lipid present in ELF, is also a target of O_3. Pulfer and Murphy (2004) demonstrated the ozonolysis of cholesterol in an in vitro system using BALF isolated from rats that had been exposed to 2.0 ppm O_3 for 4 h. Production of 5-hydroperoxy-*B*-homo-6-oxa-cholestan-3,7a-diol, 5β,6β-epoxycholesterol, and 3β-hydroxy-5oxo-5,6-seco-cholestan-6-al was shown. Additionally, both 5β,6β-epoxycholesterol and its most abundant metabolite, cholestan-6-oxo-3β,5α-diol, were demonstrated to be cytotoxic to 16-HBE cells and to inhibit cholesterol synthesis. Studies (Pulfer et al., 2005) in C57BL/6J mice exposed to 0.5, 1.0, 2.0 or 3.0 ppm O_3 for 3 h demonstrated that these oxysterols were produced in vivo also. The authors suggest that this may be an additional mechanism of O_3 toxicity. Though these oxysterol reaction products have not been fully characterized, they may be involved in O_3-induced inflammation by disrupting cellular membranes or altering signaling between cells. Similar oxysterols have been implicated in the inflammatory cascade associated with atherosclerosis.

Thus, new work has attempted to elucidate the mechanisms by which reactions of O_3 with lipids create phospholipids that then mediate downstream toxic effects. It is uncertain whether these described changes in lipid content and/or metabolism lead to significant changes in surface tension or compliance properties of the lung.

5.2.1.5 Ozone Interactions with Proteins and Effects on Protein Synthesis

Epithelial lining fluid contains proteins arising from airway secretions and from blood. Ozone can react with four amino acid residues (cysteine, histidine, methionine, and tryptophan) and can cause oxidation of functional groups on proteins, including aldehydes, alcohol, amines and sulfhydryls. A number of enzymes have been shown to be inhibited by O_3 including cholinesterase, α1-antiproteinase, and prostaglandin synthetase. Additionally, O_3 decreases the inhibitory activity of α1-proteinase inhibitor, which is implicated in development of emphysema. Surfactant protein A (SP-A) is a target for O_3 toxicity by modulation of SP-A self association, vesicle aggregation, phospholipid secretion, and stimulation of AM superoxide anion generation (see Section 5.2.2.3). Further, O_3 is thought to interfere in SP-A's homeostatic role in surfactant release from alveolar Type 2 cell lamellar bodies and its subsequent uptake by Type 2 cells and AMs.

Lung collagen, collagen synthesis, and prolyl hydroxylase activity associated with fibrogenesis have been shown to increase in rodents with O_3 exposure of $\geq$0.45 ppm. Some studies have shown that this increase persists after exposure stops and that there is an influence of exposure pattern on the response. The increased collagen has been correlated with structural changes in the lung. Rats exposed to an urban pattern of O_3 with daily peaks of 0.25 ppm for 38 weeks displayed extracellular matrix thickening. Increased levels of collagen in CAR were demonstrated in female rats exposed to 0.5 to 1.0 ppm O_3 for 6 h/day for 20 months and in monkeys exposed to 0.61 ppm for 1 year. Both increased age and health status (e.g., emphysemic) were implicated in the increased collagen formation in response to O_3 exposure.

A time-course study (van Bree et al., 2001 Table AX5-5) evaluating the lung injury and changes in collagen content in rats exposed acutely or subchronically to 0.4 ppm O_3 demonstrated CAR thickening of septa which progressed from 7 through 56 days of exposure. Though collagen content decreased with PE recovery, the structural fibrotic changes in ductular septa and respiratory bronchioles persisted, suggesting that subchronic O_3 exposures in rats creates a progression of structural lung injury that can evolve to a more chronic form, which included fibrosis. The biological relevance and adverse health effects of altered protein synthesis and collagen accumulation are uncertain.

5.2.1.6 Differential Gene Expression

Gohil et al. (2003) examined differential gene expression in C57BL/6 mice exposed to 1 ppm O_3 for three consecutive nights for 8 hours (see Table AX5-6). Ozone exposure induced changes in expression of 260 genes (80% repressed and 20% induced). Differentially expressed genes included those involved in progression of the cell cycle such as *S*-adenosyl methionine decarboxylase 3 (SAMD), ribonucleotide reductase (RR), and clusterin. Increased transcription of these genes suggests O_3-induced activation of the cell cycle with subsequent cellular proliferation. This is in accord with the finding of increased epithelial proliferation with acute O_3 exposure as discussed in studies in Sections 5.2.4.1 and 5.2.5.1. Several NF-κB-induced genes were upregulated, included serum amyloid protein, topoisomerase IIα, monocyte chemoattractant protein, platelet-derived growth factor, and inhibitor of apoptosis. Upregulation of these genes suggests to the authors that they may account for O_3-induced proliferation of nonciliated cells and Clara cells. Downregulation of transcripts for isoforms of myosins and actins were also observed, which may explain, in part, a mechanism of O_3-induced vascular permeability. Several members of the CYP family were downregulated, including 2a4,and 2e1, and 2f2, as were aryl-hydrocarbon receptor and several glutathione transferases. Metallothionein 1 and 2 and lactotransferrin were upregulated, indicative of their function as antioxidants and anti-inflammatory agents. Ozone-induced suppression of immune function is suggested by downregulation of transcripts encoding major histocompatibility complex genes, lymphocyte-specific proteins, and immunoglobulins. Section 5.2.2.3 discusses the effects of O_3 exposure on the immune system.

Quinlan et al. (1994) have reviewed the regulation of antioxidant enzymes in lung after oxidant injury. A comparison of alterations in gene expression in rat following O_3 or hyperoxia exposure, both of which induce reactive oxygen species and injury to vascular endothelial cells and cells of the alveoli, show that both ~1 ppm O_3 and 85-95% O_2 increase expression of CuZnSOD, glutathione peroxidase, and catalase. Studies in mice (Johnston et al, 1998) also demonstrate that changes in gene expression indicative of inflammation and epithelial injury that occur with hyperoxia in mice (95% O_2) compare to similar injury that occurs following O_3 exposure.

5.2.1.7 Summary and Conclusions - Biochemical Effects

Ozone has been shown to interact with a wide range of different cellular components including PUFAs, amino acid residues, and some low-molecular-weight compounds (GSH, urate, vitamins C and E). As O_3 does not penetrate much beyond the ELF, damage likely results from its PUFA ozonation products (mostly hydroxyhydroperoxides) involvement in signaling of cellular responses such as inflammation. New work has shown that ozonation of PUFA also forms the aldehydes nonanal, heptanal, and hexanal, the production of which is dependent on AA availability. Saturated phospholipids are thought to reduce the local dose and limit site-specific cell injury from O_3 exposure. Another ozonation product HNE creates protein adducts that have been linked to apoptosis and heat shock proteins in vitro.

Both short- and long-term exposures to O_3 have been shown to enhance lung xenobiotic metabolism, possibly as a result of changes in the number and function of bronchiolar epithelial Clara cells and alveolar epithelial Type 2 cells. This modulation is both species- and region-specific and includes the isoforms CYP 2B1, CYP 2E1. CCSP is also involved in inflammatory responses to O_3 exposure. Mice strains with differing sensitivities to O_3 show that responses in protein, LDH and inflammatory cell influx are due to CCSP levels and changes in lung epithelial permeability.

Reactions of O_3 with AA, GSH, and UA (all antioxidants present in ELF) are a protective mechanism. But even with environmentally relevant exposures, the reactivity of O_3 is not quantitatively quenched and cell injury occurs in both the lower and upper respiratory tract. Early work has shown that short-term exposures to <1 ppm O_3 increase antioxidant metabolism. Re-exposure after a recovery period causes increases equivalent to first-time exposures, suggesting that previous exposure is not protective. Increases in enzyme activity appear to increase as a function of age, suggesting that O_3 exposure can cause greater lung injury in the older animal. Long-term urban patterns of exposure to O_3 (daily peaks of 0.25 ppm) caused increases in GSHPx and GR, but not SOD. Recent work has suggested that endogenous antioxidants moderate the effects of O_3 and that this exposure is a subthreshold stimulus for producing effects on peripheral airway resistance and reactivity, thus indicating a dissociation between airway function and inflammation.

In both acute and short-term studies, a variety of lung lipid changes occur with O_3 exposure, including an increase in AA. With longer exposures (e.g., 0.12 ppm for 90 days),

an increase in PUFAs and a decrease in cholesterol-esters are seen, indicative of long-term alterations of surfactant lipid composition. Whether these changes in lipid content and/or metabolism lead to significant changes in surface tension or compliance properties of the lung remains unknown. New studies evaluating O_3-induced alterations in lipid metabolism have not been completed.

Collagen, a structural protein involved in fibrosis, increases with O_3 exposure, and some studies have shown that this increase persists after exposure stops. Urban patterns of exposure (daily peaks of 0.25 ppm for 38 weeks) created extracellular matrix thickening. Increases in centriacinar collagen were demonstrated in female rats exposed to 0.5 to 1.0 ppm O_3 for 6 h/day for 20 months and in monkeys exposed to 0.61 ppm for 1 year. New work examining the time course of lung injury and changes in collagen content in rats exposed acutely or subchronically to 0.4 ppm O_3 showed centriacinar thickening of septa. Collagen content decreased with PE recovery but not the structural fibrotic changes in ductular septa and respiratory bronchioles, which suggests that subchronic O_3 exposures in rats creates a progression of structural lung injury that can evolve to a more chronic form, which includes fibrosis.

5.2.2 Lung Host Defenses

Defense mechanisms, including the mucociliary clearance system, AMs, and humoral- and cell-mediated immune system, exist in the lung to protect it from infectious and neoplastic disease and inhaled particles. Summaries of key new animal studies examining the effects of O_3 on lung host defenses are presented in Table AX5-7 of Annex AX5. Acute human exposures to O_3 result in similar effects on AMs (see Chapter 6).

5.2.2.1 Clearance

Early studies of the effect of O_3 on the mucociliary escalator showed morphological damage to ciliated epithelial cells of the tracheobronchial tree at doses of <1 ppm. Functionally, O_3 slowed particle clearance in rats at doses of 0.8 ppm for 4 h and in rabbits at 0.6 ppm for 2 h exposures. Acute exposures at 0.5 ppm O_3 in sheep caused increased basal secretion of glycoproteins, while longer exposures reduced tracheal glycoprotein secretions, both of which can alter the effectiveness of the mucociliary escalator. Early postnatal exposures of sheep to 1 ppm O_3 caused retardation of normal morphologic development of the tracheal

epithelium, decreased epithelial mucosa density, decreased tracheal mucous velocity, and delayed development of carbohydrate composition. Conversely, alveolar clearance in rabbits after acute exposure (0.1 ppm, 2 h/day, for 1 to 4 days) is increased. Longer exposures showed no effect and increased O_3 (1.2 ppm) slowed clearance. This pattern of clearance occurs in rats also. A study using rat tracheal explants exposed to O_3 for 10 min (Churg et al., 1996) showed that uptake of TiO_2 and asbestos was enhanced at 0.01 and 0.1 ppm, respectively. The authors attribute the increased uptake as a direct effect of O_3, suggesting mediation by H_2O_2 or hydroxyl radical. Studies of the clearance of the radiolabled chelate ^{99m}Tc diethylenetriamine pentaacetic acid (Tc-DTPA) have shown that clearance is significantly increased following a 3 h exposure to 0.8 ppm O_3 in SD rats (Pearson and Bhalla,1997). Examination of regional clearance of ^{99m}Tc-DTPA in dogs following a 6 h isolated sublobar exposure to 0.4 ppm O_3 or air showed that O_3 decreased the clearance halftime by 50% at 1 day following exposure (Foster and Freed, 1999). Clearance was still elevated at 7 d PE but had recovered by 14 d. So, a single local exposure to O_3 increases transepithelial clearance but without any influence on contralateral segments, i.e., only for epithelia directly exposed to O_3.

Alveolar clearance is slower than tracheobronchial clearance and involves particle movement through interstitial pathways to the lymphatic system or movement of particle-laden AMs to the bottom of the mucociliary escalator. Exposures of rabbits to 0.1 ppm accelerated clearance while 1.2 ppm slowed clearance. A chronic exposure has been shown to slow clearance. New evaluations of the effects of O_3 on alveolar clearance have not been performed.

5.2.2.2 Alveolar Macrophages

A primary function of AMs is to clear the lung of infectious and noninfectious particles by phagocytosis, detoxification, and removal. Further, AMs secrete cellular mediators that recruit and activate inflammatory cells in the lungs (see Figure 5-3). Ozone has been shown to inhibit phagocytosis at 0.1 ppm for 2 h in rabbits. This inhibition returns to control levels if exposures are repeated for several days. The production of superoxide anion radicals and the activity AM lysosomal enzymes (both involved in bactericidal activity) are inhibited by 3 h exposures to 0.4 and 0.25 ppm O_3 in rodents and rabbits, respectively. Production of IFNγ was decreased in rabbit AM by 1 ppm O_3 for 3 h.

New studies have shown that O_3 affects AM chemotaxis, cell adhesion, and surface expression of cell adhesion molecules (Bhalla, 1996). AM from SD rats exposed to 0.8 ppm O_3 for 3 h showed greater mobility and greater adhesion than air exposed controls. This increased mobility and adhesion were attenuated by CD16b and ICAM-1 antibodies, suggesting these adhesion molecules modulate O_3-induced inflammation. Antibodies to TNFα and IL1α also mitigated AM adherence, suggesting further that the inflammatory response to O_3 is mediated by these cytokines (Pearson and Bhalla, 1997). Cohen et al. (1996) showed that 1 ppm O_3 for 4 h reduces binding of INF γ to AM in WEHI-3 cells, and additionally reduces phagocytic activity, production of reactive oxygen intermediates, and elevation of intracellular Ca^{++}.

Cohen et al. (2001, 2002) exposed male F-344 rats to either 0.1 or 0.3 ppm O_3 for 4 h/day, 5 days/week or either 1 or 3 weeks. In this study, superoxide anion production was increased at 1 week. Hydrogen peroxide production was reduced at both exposure concentrations and durations and was further reduced with INFγ stimulation, suggesting that one effect of O_3 is compromised killing of bacteria by AM due to the reduction in hydrogen peroxide production.

Ozone treatment (2 ppm O_3, 3 h in female SD rats) caused a time-dependent increase in NO levels in both AM and type II epithelial cells that was correlated with increased expression of iNOS mRNA and protein (Laskin et al., 1998). Inhibition of NF-κB, caused a dose-dependent inhibition of NO and iNOS production. Additionally, O_3 caused a time-dependent increase in NF-κB binding activity in the nucleus of both cell types. The authors hypothesize that O_3 exposure causes the cytokines TNFα and IL-1βα to bind to surface receptors and initiate intracellular signaling pathways in AM leading to activation of NF-κB, its entry into the nucleus, and its binding to the regulatory sequences of genes such as iNOS to allow their transcription. Additional studies (Laskin et al., 2002) using AM isolated from C57Bl6x129 mice with a targeted disruption of the gene for iNOS showed no toxicity to 0.8 ppm O_3 for 3 h, as measured by BALF protein levels and nitrotyrosine staining of the lung. Additionally, mice overexpressing human Cu, Zn superoxide dismutase (SOD) and mice with a targeted disruption of p50 NF-κB were also resistant to O_3 toxicity. WT mice exposed to O_3 showed an increase in expression of STAT-1, a protein that binds to the regulatory region of iNOS. Taken together, these results suggest to the authors that a number of proteins including NF-κB, phosphoinoside 3-kinase, and STAT-1 that bind to and regulate expression of iNOS are modulated by O_3 exposure. The same iNOS knockout mice strain exposed to 0.8 ppm O_3 for 3 h (Fakhrzadeh

et al., 2002) showed no increase in AM superoxide anion and prostaglandin. These data provide further evidence the NO and its reactive oxidative product peroxynitrite are important in O_3-induced lung injury. Further discussions of the role of nitric oxide synthase/reactive nitrogen and cytokines/chemokines in O_3-induced inflammation are provided in Section 5.2.3.

5.2.2.3 Immune System

Other than by natural protection (e.g., opsonizing antibody, nonspecific phagocytosis by AM), the immune system defends the lung by mounting three major waves of response: natural killer (NK) cells (nonspecific lymphocytes that kill viruses, bacteria, and tumor cells), followed by cytotoxic T-lymphocytes (T_{CTL}- lymphocytes that lyse specifically recognized microbial and tumor-cell targets), followed by antigen-specific antibodies. These T-cell types are involved with other immunologically active cells (e.g., B-cells and AM), which in a complex manner, interact in immunological defense. To date, only a few of these mechanisms have been investigated in the context of their role in O_3 susceptibility. The effects of O_3 on the immune system are complex and depend on the exposure parameters and observation periods. T-cell-dependent functions appear to be more affected than B-cell-dependent functions. Generally, there is an early immunosuppressive effect that can, with continued exposure, either return to normal or actually enhance immunity. Changes in immune cell population occur with O_3 exposure including T:B-cell ratios in the MLN. Natural killer (NK) cell activity increases with 1 week exposures of 0.2 to 0.4 ppm O_3 but decreases with exposures to 0.82 ppm. Ozone exposure has also shown to be responsible for enhancement of allergic sensitization at levels of 0.5 to 0.8 ppm for 3 days. Studies of the effects of O_3 on the immune system are summarized in Table AX5-7.

Garssen et al. (1997) have studied the effects of O_3 on non-IgE-mediated pulmonary hyper-immune reactions induced by picryl chloride (PCI). BALB/c mice sensitized with PCI, both actively and passively (by adoptive transfer of lymphoid cells from pre-sensitized mice), were then challenged with picryl sulfonic acid (PSA). The mice were exposed to12 h of 0.4, 0.8, or 1.6 mg/m^3 O_3 during one night, at 4 days or 7 days after skin sensitization (which was either just before or just after PSA challenge, i.e., during the induction or effector phase). Nonsensitized mice showed no changes in tracheal reactivity to carbacol with O_3 exposure. Sensitized mice were hyperreactive to carbachol 48 h after PSA challenge, whereas sensitized

mice exposed to all concentrations of O_3 showed no significant tracheal hyperreactivity to carbachol. The sensitized mice also demonstrated a suppressed inflammatory reaction (PMN) with 1.6 mg O_3 exposure. Ozone exposure following PSA challenge also caused a suppression of tracheal hyperresponsiveness. In a separate experiment wherein mice were exposed to O_3 before sensitization and then lymphoid cells from these mice were injected into nonexposed mice, the recipients also demonstrated an inhibition of the induction of hyperreactivity. These results are opposite to the effect on type I (IgE-mediated) allergic reactions, which the authors suggest is due to activation of Th-2 cell-dependent reactions that are possibly potentiated by O_3 or to a direct effect by O_3 on Th-1 cells or other cells that are crucial for the tracheal hyperreactivity and inflammation seen in this mouse model.

Kleeberger et al. (2000, 2001a) have demonstrated a potential interaction between the innate and acquired immune system with O_3 exposure. Using O_3-susceptible (C57BL/6J) and O_3-resistant (C3H/HeJ) mice, they identified a candidate gene on chromosome 4, Toll-like receptor 4 (*Tlr4*). Ozone exposure (0.3 ppm for 24 to 72 h) of C3H/HeJ and C3H/HeOuJ mice, the latter differing from the O_3-resistant strain by a polymorphism in the coding region of *Tlr4*, demonstrated greater protein concentrations in the OuJ strain. The two strains exhibited differential expression of *Tlr4* mRNA with O_3 exposure. Thus, a quantitative trait locus on chromosome 4 appears to be responsible for a significant portion of the genetic variance in O_3-induced lung hyperpermeability. In these mouse strains lavageable protein concentration was lowered by inhibition of inducible nitric oxide synthase (iNOS) and by targeted disruption of *Nos2*. Comparisons of C3H/HeJ and C3H/HeOuJ O_3 exposures demonstrated reduced *Nos2* and *Tlr4* mRNA levels in the O_3-resistant C3H/HeJ mice. These data are consistent with the hypothesis that O_3 -induced lung hyperpermeability is mediated by iNOS. These studies suggest a role for TLR4 in the host response to O_3 similar to the role it has demonstrated in lipopolysaccharide (LPS) sensitivity (Schwartz 2002; Wells et al. 2003). TLR4 signaling is thought to be critical to linking the innate and acquired immune system through antigen presenting cells and Th1/Th2 differentiation.

Ozone exposure has been shown to affect Ig responses both in vitro and in mice. Becker et al. (1991) demonstrated changes in IgG production in cultured human lymphocytes with O_3 exposures of 1.0, 0.5, and 0.1 ppm for 2 h. Subsequent to O_3 exposure, cells were stimulated with pokeweed mitogen (PWM, a T-cell-dependent stimulus) or Staphylococcus aureus Cowan 1

strain (SAC, a T-cell-independent stimulus). Both B and T cells were affected by O_3. T cells also demonstrated an increase in IL-6 and a decrease in IL-2, which suggested to the authors that O_3 may have direct effects on IgG producing cells and concurrently an effect that is mediated by altered production of T cell immunoregulatory molecules. Responses to repeated O_3 (0.08 - 0.25 ppm) and OVA (1%) exposures were compared in "IgE-high responder" (BALB/c) and "IgE-low responder" (C57BL/6) mice (Neuhaus-Steinmetz et al., 2000). Ozone appeared to shift the immune response toward a Th2-like pattern in the two mouse strains with differing potentials for developing allergic reactions.

Another study (Depuydt et al., 2002) demonstrated that O_3 (0.1 ppm for 2 h) increases allergen-induced airway inflammation in previously sensitized mice but has no effect on the sensitization process itself. This study uses OVA-pulsed dendritic cells instead of systemic adjuvant, which the authors consider a more relevant model of sensitization as it clearly separates the immune response from the challenge and does not obscure regulatory processes as does i.p. injections of OVA. They further suggest that dendritic cells, the principal antigen-presenting cells in the airway, are an important component of O_3-induced eosinophilic airway inflammation.

Surfactant protein A and D (SP-A and SP-D) were shown to create an inflammatory feedback loop with perturbations in lung immune defenses (reviewed in Hawgood and Poulain, 2001). Earlier studies suggested that SP-A is a target for O_3 toxicity by causing inhibition of SP-A self-association and SP-A-mediated lipid vesicle aggregation. Further, O_3 reduced the ability of SP-A to inhibit phospholipid secretion by alveolar type II cells and reduced the capacity of SP-A to induce superoxide anion production and enhance phagocytosis of herpes simplex virus. Bridges et al. (2000) reported that both SP-A and SP-D directly protect surfactant phospholipids and macrophages from oxidative damage by blocking accumulation of TBARS and conjugated dienes.

Eight human variants of SP-A in CHO cells exposed to O_3 (1ppm for 4 h) showed decreased ability to stimulate cytokine (TNF-α and IL-8) production in THP-1 cells, a macrophage-like cell line (Wang et al., 2002). Each variant had a unique time- and dose-dependent pattern of stimulation of cytokine production with O_3 exposure which the authors attribute to possible differences in susceptibility to O_3 oxidation. Targeted disruption of mouse SP-A and SP-D (Hawgood et al, 2002) caused increases in BAL phospholipid,

macrophage, and protein through 24 weeks of age. Further, the deficient mice developed patchy lung inflammation and air space enlargement consistent with emphysema. Future experiments using these null mice will help to establish the role of SP-A and SP-D in pulmonary host defense to O_3 exposure.

5.2.2.4 Interactions with Infectious Microorganisms

Ozone-induced dysfunction of host defense systems results in enhanced susceptibility to bacterial lung infections. Acute exposures of 0.08 ppm (3 h) O_3 can overcome the ability of mice to resist infection (by decreasing lung bactericidal activity) with Streptococcal bacteria, resulting in mortality. Changes in antibacterial defenses are dependent on exposure regimens, species and strain of test animal, species of bacteria, and age of animal, with young mice more susceptible to the effects of O_3. The effect of O_3 exposure on antibacterial host defenses appears to be concentration- and time-dependent. Early studies using the mouse "infectivity model," consisting of exposure to clean air or O_3 followed by exposure to an aerosolized microorganism, showed that the difference in mortality between O_3-exposed groups and controls is concentration-related. Chronic exposures (weeks, months) of 0.1 ppm do not cause greater effects on infectivity than short exposures, due to defense parameters becoming reestablished with prolonged exposures.

More recent studies of O_3-induced modulation of cell-mediated immune responses showed effects on the onset and persistence of infection. Cohen et al. (2001, 2002) exposed male F-344 rats subchronically to either 0.1 or 0.3 ppm O_3 for 4 h/day 15 days/week, for 1 or 3 weeks. Subsequent exposure with viable Listeria monocytogenes demonstrated no observed effect on cumulative mortality but did show a concentration-related effect on morbidity onset and persistence. These data suggest that O_3 may cause a possible imbalance between Th-1 and Th-2 cells, which can subsequently lead to suppression of the resistance to intracellular pathogens.

Effects of O_3 on viral infections are dependent on the temporal relationship between O_3 exposure and viral infection. Only high concentrations (1.0 ppm O_3, 3 h/day, 5 days, mice) increased viral-induced mortality. No detrimental effects were seen with a 120-day exposure to 0.5 ppm O_3 on acute lung injury from influenza virus administered immediately before O_3 exposure started. But there were O_3-enhanced postifluenzal alveolitis and lung parenchymal changes. As O_3 does not affect lung influenza viral titers, it apparently does not impact antiviral

clearance mechanisms. In general, the evidence suggests that O_3 can enhance both bacterial and viral lung infections, but the key mechanisms have not yet been identified. New studies on the interactions of O_3 and viral infections have not been published.

5.2.2.5 Summary and Conclusions - Lung Host Defenses

New data on lung host defenses support earlier work which suggests that mucociliary clearance is affected in most test species at just under 1 ppm, with lower levels (~0.1 ppm) increasing clearance and somewhat higher levels decreasing clearance. These data also propose mechanisms whereby O_3 affects clearance, which include uptake being a direct effect of O_3, but modulated by ROS and hydroxyl radicals.

Alveolar macrophage function is disrupted by O_3 as shown by a number of studies demonstrating inhibition of phagocytosis at concentrations ranging from 0.1 to 1.2 ppm. This inhibition returns to control levels if exposures are repeated for several days. Two new studies corroborate earlier findings of increases in AM number in that same exposure range. In this environmentally relevant exposure range, new studies support older findings of decreased resistance to microbial pathogens as shown by the endpoints examining superoxide radical formation, altered chemotaxis/motility, decreased INFγ levels, decreased lysosomal activity, increased PGE levels, and increased NO mRNA and protein.

New research evaluating the effects of O_3 on immune function advances previous work that has shown that exposures can enhance or suppress immune responsiveness depending on the species studied, concentration of O_3, route of exposure of allergen, and timing of exposure. Continuous exposure to O_3 impairs immune responses for the first several days of exposure, followed by an adaptation to O_3 that allows a return of normal immune responses. Most species show little effect of O_3 exposures prior to immunization, but a suppression of responses to antigen in O_3 exposures post-immunization. The use of mouse strains with genetically determined sensitivity or resistance to O_3 indicated a possible interaction between the innate and acquired immune system, and further, that O_3 may shift the immune response towards a Th-2-like pattern. Work has also focused the deleterious effects of O_3 exposure on SP-A and SP-D and their immunomodulatory function in protecting against oxidative stress.

Several new studies evaluating the effects of O_3 exposures on infectious microorganisms are in concurrence with previous studies which showed, in general, increased mortality and

morbidity, decreased clearance, increased bacterial growth, and increased severity of infection at exposure levels of 0.1 to 1 ppm O_3 for 1 week.

5.2.3 Inflammation and Lung Permeability Changes

The normal lung has an effective barrier function that controls bidirectional flow of fluids and cells between the air and blood compartments. Ozone disrupts this function, resulting in two well-characterized effects of O_3 exposure, lung inflammation and increased permeability, which are distinct events controlled by independent mechanisms. Ozone initiates inflammation of lung tissue by reactions with antioxidants and lipids in ELF (discussed in 5.2.1, see Figure 5-2). Secondary reaction products generated in this process then cause changes in cell membranes, disruption of the lung barrier leading to leakage of serum proteins, influx of polymorphonuclear leukocytes (PMNs), release of bioactive mediators, and movement of compounds from the airspaces into the blood. This increased permeability allows accumulation of co-occurring pollutants into the lung tissue. The framework for presenting this stereotypical response to O_3 consists of discussions covering: 1) the time course of these changes; 2) concentration × time (C × T) relationships; 3) susceptibility factors; 4) mediators of inflammation; and 5) nitric oxide and reactive nitrogen.

Rats appear to be more resistant to O_3-induced inflammation than humans (see Chapter 4). With comparable exposure protocols, both species have similar observed inflammatory and permeability changes, i.e., controlled human exposure studies discussed in Chapter 6 indicate that the majority of acute responses in humans are similar to those observed in animals.

Ozone also increases the permeability from the air to the blood compartment. Ozone (0.8 ppm; 2 h) caused a 2-fold increase of the transport of labeled DTPA from the rat tracheal lumen to the blood. This coincided with a 2-fold increase in the number of endocytic vesicles in epithelial cells that contained intraluminally instilled HRP as a tracer. These studies also suggest an uneven disruption of tight junctions and alternate transport through endocytotic mechanisms. In studies aimed at detecting the effects of O_3 exposure on regional permeability, O_3 increased the transmucosal transport of DTPA and BSA more in the trachea and bronchoalveolar zone than in the nose. These changes in barrier integrity may allow increased entry of antigens and other bioactive compounds (e.g., bronchoconstrictors) into lung tissue. Data from analyses at regular

intervals PE indicate that maximal increases in BALF protein, albumin and number of PMNs occur 8 to 18 h (depending on the study) after an acute exposure ceases.

Increases in permeability and inflammation have been observed at levels as low as 0.1 ppm O_3 for 2 h/day for 6 days in rabbit and 0.12 ppm in mice (24 h exposure) and rats (6 h exposure). After acute exposures, the influence of the time of exposure increases as the concentration of O_3 increases. The exact role of inflammation in causation of lung disease is not known, nor is the relationship between inflammation and changes in lung function. Table AX5-8 in Annex AX5 summarizes new key studies describing the potential for O_3 exposure effects on lung permeability and inflammation.

5.2.3.1 Time Course of Inflammation and Lung Permeability Changes

The maximal increase in BALF protein, albumin, and PMN occurs in most species 8 to 18 h after the cessation of acute exposures of 0.5 to 1.0 ppm. A study of OVA-sensitized male Dunkin-Hartley guinea pigs exposed to 1.0 ppm O_3 for 3 h showed that levels of PMN significantly increased at 3 h PE, but BAL protein levels did not, suggesting a lack of correlation between the two endpoints (Sun et al., 1997). Increased PMN without a concordant increase in BAL protein levels were found when the guinea pigs were exposed to 1.0 ppm O_3 for 1 h and evaluated 24-h PE. The first group also had an increase in AHR, but not the second group, which suggests a dissociation between PMN levels and AHR.

Earlier work demonstrated that O_3 exposures of 0.8 to 1 ppm in rat and guinea pig transiently increase the permeability from the air to the blood compartment. This permeability is greatest in trachea and bronchoalveolar zone, and may allow increased entry of antigens and other bioactive compounds (e.g., bronchoconstrictors) into lung tissues. The time course of the influx of PMNs into the lung and the BALF fluid levels of macrophage inflammatory protein-2 (MIP-2) were found to be roughly similar to that for proteins (Bhalla and Gupta, 2000). Adherence of neutrophils to pulmonary vascular endothelium is maximal within 2 h after exposure and returns to control levels by 12 h PE (Lavnikova et al., 1998). Cheek et al. (1995) cultured monolayers of rat alveolar type II cells and exposed them to 0.1 or 0.5 ppm O_3 for 0.5 h to evaluate the effects of O_3 on permeability. Permeability increased dose-dependently and the higher exposures elicited greater numbers of injured epithelial cells. Exposure to 0.1 ppm O_3 was thought to expedite the restoration of epithelial barrier functions, while in higher exposures,

neutrophils exacerbated the O_3-induced injury. Vesely et al. (1999a) have demonstrated that neutrophils contribute to the repair process in O_3-injured airway epithelium and they may play a role in removal of O_3-injured cells.

Exposures of 3 to 7 days have been found to cause increases in BALF protein and PMNs that typically peak after a few days (depending upon species tested and exposures) and return towards control even with continuing exposure. Van Bree et al. (2002) observed lower BALF levels of protein, fibronectin, IL-6 and inflammatory cells in rats exposed for 5 days to 0.4 ppm O_3 than in rats exposed for 1 day, suggesting adaptation to O_3 exposure. Postexposure challenge with single O_3 exposures at different time points showed recovery of susceptibility to O_3. McKinney et al. (1998) observed differences in IL-6 levels due to repetitive exposures and demonstrated a role of IL-6 in the adaptive response induced by repeated O_3 exposures of 0.5 ppm for 4 h.

5.2.3.2 Concentration and Time of Exposure

The relative influence of concentration and duration of exposure (i.e., C × T) has been investigated extensively in rats, using BALF protein as an endpoint. Earlier work utilizing concentrations of 0.1 to 2 ppm O_3 and durations of 1 to 8 h has shown that the interaction between C and T is complex. At these levels of exposure, concentration generally dominates the response. C × T studies using the endpoints of changes in lung protein or cell type showed that acute damage is a function of cumulative dose. The impact of T is C-dependent (at higher Cs, the impact of T is greater); at the lowest C and T values, this dependence appears to be lost. The controlled human exposure data described in Chapter 6 concur with most animal data, showing that concentration of O_3 is the most important factor determining O_3 responses, and that duration of exposure and ventilation rate are secondary factors.

New studies evaluating C × T relationships in animal models have not been completed. However, a full understanding of C × T relationships in ambient exposures must include the recognition that 'real world' exposures are cyclic in nature, due to the daily and seasonal variations in O_3 levels. The concentration of O_3, the duration of the exposure, and duration between exposures are all relevant to the type and level of O_3-induced injury.

5.2.3.3 Susceptibility Factors

Factors that have been studied for potential impact on the effects of O_3 exposure include age, gender, nutritional status, exposure to co-pollutants, exercise, and genetic variability. A full characterization of the effects of age on O_3 responses has not been completed. Data available indicate that effects of age on O_3 responses are endpoint-dependent, with young mice, rats, and rabbits having greater prostaglandin levels with exposure and senescent rats having greater IL-6 and N-acetyly-β-D-glucosaminidase levels with exposure.

A study (Johnston et al., 2000a) compared gene expression of chemokines and cytokine in newborn and 8-week-old C57Bl/6J mice exposed to 1.0 or 2.5 ppm for 4, 20, or 24 h. The newborn mice displayed increased levels of Mt mRNA only, while the 8-week-old mice had increases in MIP-1α, MIP-2, IL-6, and Mt mRNA. Comparisons were made with mice of the same age groups with exposures to endotoxin (10 ng/mouse for 10 min). Both age groups displayed similar cytokine/chemokine profiles with endotoxin exposure. This suggested to the authors that the responses to endotoxin, which does not cause epithelial injury, and the responses to O_3, which does, demonstrate that differences in inflammatory control between newborn and adult mice is secondary to epithelial injury.

Pregnancy and lactation increased the susceptibility of rats to acute O_3, but no clear effects of gender have been identified. The effects of vitamin C deficiency on O_3 responses are unclear. Ascorbate-deficient guinea pigs exposed to O_3 demonstrated only minimal effects on injury and inflammation (Kodavanti et al., 1995). Utilizing a diet-restricted (20% of the freely-fed diet) rat model, Elsayed (2001) demonstrated higher survivability on exposure to higher O_3 (0.8 ppm continuously for 3 days) compared to freely-fed rats. Pre-exposure to sidestream cigarette smoke had been found to cause increased lung injury (Yu et al., 2002). In vitro studies on the macrophages from smoke + O_3- exposed animals responded by a greater release of TNF-α following LPS stimulation when compared to macrophages exposed to air, smoke or O_3 (0.5 ppm, 24 h) alone.

Lines of evidence illustrate that genetic background is an extremely important determinant of susceptibility to O_3. Earlier studies using inflammation-prone (susceptible) C57BL/6J (B6) and inflammation-resistant C3H/HeJ (C3) mouse strains and high doses of O_3 (2 ppm for 3 h) identified *Inf*-2 as a locus controlling susceptibility. Further studies in these two strains of mice using more relevant exposures (0.3 ppm for 72 h) identified that the acute and subacute

exposures are controlled by two distinct genes, referred to as *Inf*-1 and *Inf*-2, respectively (Tankersley and Kleeberger, 1994). Exposures to 0.3 ppm O_3 for 48 or 72 h, when repeated fourteen days after the initial exposures, caused a smaller increase in BALF protein and number of macrophages, lymphocytes and epithelial cells in both strains, but PMN number was greater in both strains compared to initial exposure (Paquette et al., 1994). Kleeberger et al. (1997) also identified another potential susceptibility gene, tumor necrosis factor (*Tnf,* which codes for the pro-inflammatory cytokine TNF-α) on a qualitative trait locus on mouse chromosome 17. By neutralizing the function of TNF-α with a specific antibody, they were able to confer protection against O_3 (0.3 ppm, 48 h) injury in susceptible mice. The group then demonstrated a role for TNF receptor 1 and 2 (TNFR1 and TNFR2, respectively) signaling in subacute (0.3 ppm for 48 h) O_3-induced pulmonary epithelial injury and inflammation (Cho et al., 2001). TNFR1 and TNFR2 knockouts were less sensitive to subacute O_3 exposure than WT C57BL/6J mice.

An integrated and more comprehensive effort to identify the genetic basis for the susceptibility to O_3-induced lung injury was reported by Savov et al. (2004). In this report, acute lung injury to high dose of O_3 (2 ppm for 3 h) was assessed and integrated with physiological, biochemical, and genetic observations using 9 inbred mouse strains. This work indicated the presence of genetic loci on chromosomes 1, 7, and 15 associated with phenotypic characteristics for resistance to acute O_3-induced lung injury. They identified C3H/HeJ and A/J as consistently O_3-resistant, C57BL/6J and 129/SvIm as consistently O_3-vulnerable, and CAST/Ei, BTBR, DBA/2J, FVB/NJ, and BALB/cJ as intermediate in response to O_3.

Ozone-induced changes in CCSP (called CC16 by this group) expression were evaluated in five inbred mouse strains: C57BL/6J and CBA both considered sensitive to acute O_3-induced inflammation, C3H/HeJ and AKR/J both considered resistant, and SJL/J considered intermediate (Broeckaert et al., 2003). Two exposures paradigms were used, 1.8 ppm O_3 for 3 h or 0.11 ppm O_3, 24/h day for up to 3 days, and BALF and serum was assayed immediately after exposure or at 6 h PE. Both exposure levels caused a transient increase in CC16 in serum that correlated with BALF changes in protein, LDH, and inflammatory cells. There was an inverse relationship between preexposure levels of CC16 in BALF and epithelial damage based on serum CC16 levels and BALF markers of inflammation. There was also an inverse relationship between preexposure levels of albumin in BALF and lung epithelium damage. Based on these results, the authors conclude that a major determinant of susceptibility to O_3 is basal lung

epithelial permeability. As all of the mouse strains had similar levels of preexposure CC16 mRNA, they explored the possible role of CC16 isozymes in differences among strains. The CC16 monomer a 7kD protein exists in two isoforms with differing p*I* values, CC16a (4.9) and CC16b (5.2). To evaluate the role of CC16 isoform profiles in permeability differences between C57BL/6J and C3H/HeJ, this group evaluated the CC16 protein profiles in BALF of the strains before and after O_3 exposure following two-dimensional protein electrophoresis analysis. C57BL/6J mice had lower levels of CC16a (the more acidic form) than C3H/HeJ. But both the strains had similar levels of CC16b. Based on these observations, Broeckaert et al (2003) conclude that greater epithelial permeability observed in C57BL/6J may be due to difference in the expression of CC16a and possibly other antioxidant/inflammatory proteins.

Wattiez et al. (2003) examined BALF protein from C57BL/6J (O_3-sensitive) and C3H/HeJ (O_3-resistant) mice exposed to filtered air using a two-dimensional polyacrylamide gel approach to analyze the protein profiles. C3H/HeJ mice expressed 1.3 times more Clara cell protein16 (CC16) than C57BL/6J mice and, further, expressed more of the acidic isoform of CC16. Strain-specific differential expression of isoforms of the antioxidant protein 2 (AOP2), the isoelectric point 5.7 isoform in C3H/HeJ and isoelectric point 6.0 isoform in C57BL/6J were observed. These data suggest a protective role for CCSP against oxidative damage, and further, that genetic susceptibility to oxidant stress may be moderated, in part, by the gene coding for CCSP. Taken together, these mouse studies of genetic susceptibility are useful for understanding underlying mechanisms leading to O_3-induced effects. However, at this point, corresponding human polymorphisms have not yet been identified which associate with differing human sensitivities to O_3.

5.2.3.4 Mediators of Inflammatory Response and Injury

Ozone reacts with lipids in the ELF or epithelial cell membranes, creating ozonation products which then stimulate airway epithelial cells, AMs, and PMNs to release a host of pro-inflammatory mediators including cytokines, chemokines, reactive oxygen species, eicosanoids, and platelet activating factor (see Figure 5-3). While neutrophils in the lung characterize an inflammatory response to O_3, the release of chemotactic mediators by inflammatory cells indicates their state of activation and their role in continued inflammation and injury. At O_3 exposures of $\geq$1 ppm, these mediators recruit PMN, and increase expression of

MIP-2 mRNA or BALF levels of MIP-2 (Driscoll et al., 1993; Haddad et al., 1995; Bhalla and Gupta, 2000). The increased mRNA expression was associated with an increased neutrophilia in the lung. Zhao et al. (1998) showed that 0.6 ppm O_3 exposure for 2 h in mice and rats causes an increase in monocyte chemotactic protein-1 (MCP-1).

Fibronectin, an extracellular matrix glycoprotein, is thought to have a role in lung inflammation and inflammatory disorders, and has shown to be increased with exposure to 1 ppm O_3 for 14 days. Gupta et al. (1998) observed an increase in both fibronectin protein and mRNA expression in the lung of rats exposed to 0.8 ppm O_3 for 3 h. A mechanistic role of fibronectin in O_3-induced inflammation and injury was suggested on the basis of comparability of temporal changes in BALF protein, fibronectin and alkaline phosphatase activity with exposures of 1 ppm for 3 h (Bhalla et al., 1999). Studies have reported an effect of O_3 on other cytokines and inflammatory mediators. An increase occurred for cytokine-induced neutrophil chemoattractant (CINC) and NF-κB expression in vivo (Koto et al., 1997), for IL-8 in vivo and in vitro (Chang et al., 1998), TNFα, fibronectin, IL-1 and CINC release by macrophages ex vivo (Pendino et al., 1994; Ishii et al., 1997), and NF-κB and TNFα (Nichols et al., 2001; see 6.9.2). An increase in lung CINC mRNA occurred within 2 h after the end of a 3 h exposure of rats to 1 ppm O_3. The CINC mRNA expression was associated with neutrophilia at 24 h PE. Exposure of guinea pig AMs recovered in BALF and exposed in vitro to 0.4 ppm O_3 for 1 h produced a significant increase in IL-6 and TNFα (Arsalane et al., 1995). An exposure of human AMs to an identical O_3 concentration increased TNFα, IL-1β, IL-6 and IL-8 and their mRNAs. Ozone exposure (0.3 to 2.5 ppm, 1-48 h) of mice caused an increase in IL-6, MIP-1α, MIP-2, eotaxin and Mt abundance (Johnston et al., 1999a). The IL-6 and Mt increase was enhanced in mice deficient in CCSP, suggesting a protective role of Clara cells and their secretions (Mango et al., 1998). CCSP deficiency, also increased sensitivity of mice to O_3, as determined by an increase in abundance of MIP-1α and MIP-2 following a 4 h exposure to 1.0 ppm O_3 (Johnston et al., 1999b).

Mast cells, which are located below the epithelium, release proinflammatory mediators and have been shown to contribute to O_3-induced epithelial damage. Greater increases in lavageable macrophages, epithelial cells and PMNs were observed in mast cell-sufficient mice than in mast cell-deficient mice exposed to 0.26 ppm O_3 for 8 h per day, 5 days per week (Kleeberger et al., 2001b). Increases in inflammatory cells were also observed in mast cell-deficient mice repleted

of mast cells, however O_3-induced permeability changes were similar between genotypic groups exposed to 0.26 ppm. When a RBL-2H3 mast cell line was exposed to 0.1 to 1.0 ppm O_3 for 1 h, spontaneous release of serotonin and modest generation of prostaglandin D2 occurred only under conditions that caused cytotoxicity (Peden and Dailey, 1995). Additionally, O_3 inhibited IgE- and A23187-induced degranulation. Mast cells recovered from O_3-exposed peripheral airways of ascaris sensitive dogs released significantly less histamine and PGD2 following *in vitro* challenge with ascaris antigen or calcium ionophore (Spannhake, 1996). Ozone (0.4 ppm, 5 weeks) exposure also promoted eosinophil recruitment in the nose and airways in response to instillation of OVA or OVA-pulsed dendritic cells and aggravated allergy like symptoms in guinea pigs (Iijima et al., 2001).

The role of PMNs and cellular mediators in lung injury and epithelial permeability has been investigated using antibodies and inhibitors of known specificity to block inflammatory cell functions and cytokine activity. Treatment of rats with cyclophosphamide prior to O_3 exposure (0.8 ppm, 48 h) resulted in a decreased recovery of PMNs in the BALF and attenuated permeability induced by O_3 (Bassett et al., 2001).

Pretreatment of animals with antiserum against rat neutrophils abrogated PMN accumulation in the lung, but did not alter permeability increase produced by O_3. Studies utilizing antibodies to selected pro- or anti-inflammatory cytokines suggest a role of TNFα, IL-10, and IL-1β in O_3-induced changes in permeability, inflammation and cytokine release (Ishii et al., 1997; Reinhart et al., 1999; Bhalla et al., 2002) in exposures of ~1 ppm for 3-6 h. An attenuation of O_3-induced increases in permeability and inflammation was also observed in mice treated, either before or after exposure, with UK-74505, a platelet-activating factor (PAF) receptor antagonist (Longphre et al., 1999). These results were interpreted to indicate that O_3-induced epithelial and inflammatory changes are mediated in part by activation of PAF receptors.

Ozone exposure stimulates macrophage motility towards a chemotactic gradient, and macrophages isolated from rats exposed to 0.8 ppm O_3 for 3 h adhered to epithelial cells (ARL-14) in culture to a greater extent than macrophages from air-exposed controls (Bhalla, 1996). Both macrophage motility and chemotaxis were attenuated by antibodies to cell adhesion molecules CD-11b and ICAM-1, suggesting a role for cell adhesion molecules in O_3-induced cellular interactions. This may also explain the increased tissue localization and reduced

recovery of macrophages in BALF (Pearson and Bhalla, 1997) following O_3 exposure (0.8 ppm, 3 h). Studies investigating the mechanisms of PMN recruitment in the lung have explored the role of cell adhesion molecules that mediate PMN-endothelial interactions. An exposure of female rats to O_3 (1 ppm, 2 h) had an attenuating effect on CD-18 expression on AMs and vascular PMNs, but the expression of CD62L, a member of selection family, on vascular PMNs was not affected (Hoffer et al., 1999). In monkeys, O_3-induced (0.8 ppm, 8 h) inflammation was blocked by treatment with a monoclonal antibody to CD18, suggesting dependence of PMN recruitment on this adhesion molecule (Hyde et al., 1999). Treatment of monkeys with CD18 antibody also reduced tracheal expression of the β6 integrin (Miller et al., 2001) suggesting that lung epithelial cell expression of this adhesion molecule is associated with sites of neutrophil recruitment. A single 3 h exposure of rats to 1 ppm O_3 caused an elevation in concentration of ICAM-1, but not CD-18, in the BALF (Bhalla and Gupta, 2000). Takahashi et al. (1995a) found an increase in tissue expression of ICAM-1 in mice exposed to 2 ppm O_3 for 3 h, noting a temporal correlation of inflammatory activity and ICAM-1 expression which varied in different regions of the lung. A comparable pattern of time-related changes in total protein, fibronectin and alkaline phosphatase activity in the BALF of rats exposed to 0.8 ppm O_3 for 3 h was also noted by Bhalla et al. (1999). Together, these studies support the role of extracellular matrix protein and cell adhesion molecules in the induction of lung inflammation and injury.

5.2.3.5 The Role of Nitric Oxide Synthase and Reactive Nitrogen in Inflammation

Nitric oxide (NO) is a messenger molecule involved in many biological processes, including inflammation (see Figure 5-3). Cells in the respiratory tract (including mast cells, neutrophils, epithelial cells, neurons, and macrophages) produce three differing forms of nitric oxide synthase (NOS), the enzyme that catalyzes the formation of NO. NOS-1(neuronal) and NOS-3 (endothelial) are constitutively expressed, whereas NOS-2 (also referred to as iNOS) is inducible, commonly by pro-inflammatory cytokines. Macrophages isolated from O_3-exposed (0.8 ppm for 3 h) mice produced increased amounts of NO, superoxide anion, and PGE2, but production of these mediators by macrophages from NOS knockout mice was not elevated (Fakhrzadeh et al., 2002). Additionally, mice deficient in NOS or mice treated with N^G-monomethyl-L-arginine, an inhibitor of total NOS, were protected from O_3-induced permeability, inflammation, and injury, suggesting a role of NO in the production of O_3 effects

(Kleeberger et al., 2001a; Fakhrzadeh et al., 2002). These results contrast with a study showing that O_3 exposure (of 1 ppm for 8 h/night for 3 nights) produced greater injury, as determined by measurement of MIP-2, matrix metalloproteinases, total protein, cell content and tyrosine nitration of whole lung protein, in iNOS knockout mice than in wild type mice (Kenyon et al., 2002). This group suggests that protein nitration is related to inflammation and is not dependent on iNOS-derived NO. They point out the possible experimental differences, such as O_3 concentration, for inconsistency between their results and those of Kleeberger et al. (2001a).

Rats pretreated with ebselen, a potent anti-inflammatory, immunomodulator, and NO/peroxynitrite scavenger, then exposed to 2 ppm O_3 for 4 h had decreased numbers of neutrophils, lowered albumin levels, and inhibited nitration of tyrosine residues in BALF 18 h PE, though macrophage iNOS expression was not changed (Ishii et al., 2000a). These results suggest an iNOS-independent mechanism for O_3-induced inflammation. Jang et al. (2002) showed dose-dependent increases in nitrate (indicative of in vivo NO generation) with O_3 exposure (0.12, 0.5, 1, or 2 ppm for 3 h). Functional studies of enhanced pause (P_{enh}) demonstrated increases with O_3 exposure which were also dose-dependent. Western blot analysis of lung tissue showed increases in NOS-1, but not in NOS -3 or iNOS isoforms. These results suggest that in mice NOS-1 may induce airway responsiveness by a neutrophilic airway inflammation. The literature regarding the effects of O_3 exposure on NOS activity is complex and conflicting. Similarly, the issue of protein nitration as it relates to cell injury due to O_3 exposure is somewhat controversial.

5.2.3.6 Summary and Conclusions - Inflammation and Permeability Changes

Figure 5-3 depicts many of the inflammatory and permeability changes that occur with O_3 exposure. Additionally, the figure demonstrates links between inflammatory/permeability responses and altered spirometric responses (discussed in Section 5.2.5), ciliary motility (discussed in Section 5.2.2.1), airway hyperreactivity (discussed in Section 5.2.5.3), and possible thrombolytic effects (Section 5.3.3). Airway mucosa in the normal lung serves as an effective barrier that controls bidirectional flow of fluids and cells between the air and blood compartments. Ozone disrupts this function, resulting in a cascade of effects which includes an increase in serum proteins, bioactive mediators, and PMNs in the interstitium and air spaces of the lung. Damaged epithelial cells release cytokines, which function to recruit and activate AMs

and PMNs. PMN recruitment into the lung is maximal at several hours PE. PMN recruitment is followed by blood monocytes which enter the lung and enlarge to become AMs. The AMs persist for days to weeks, phagocytizing injured cells. Activated PMNs and AMs continue the cascade of effects by further releasing inflammatory mediators, which serve to amplify the initial effects of O_3. Generally, the initiation of inflammation is an important component of the defense process; however, its persistence and/or repeated occurrence can result in adverse health effects. Activation of this inflammatory cascade takes several hours. Chemical mediators released early in the cascade contribute to effects on pulmonary function. Events later in the cascade, by which time O_3-induced alterations pulmonary function have attenuated, are related to sustained inflammation. Further, mechanistic separation of inflammation, permeability, and airway hyperreactivity (AHR) is suggested by the temporal disparities of their increases.

The O_3-induced disruption of the tight junctions between epithelial cells also increases the permeability between the air and blood compartments. This disruption, occurring with exposures of 0.8 ppm for 2 h, is greater in the trachea and bronchoalveolar zone than in the nose and allows entry of particles, including bioactive compounds, into the lung tissue.

For environmentally-relevant exposures to O_3, concentration of exposure dominates the response. Studies evaluating C × T relationships have not been published recently. Other factors that have been studied for potential impact on the effects of O_3 include age, gender, nutritional status, genetic variability, exercise and exposure to co-pollutants. The effects of age on lung inflammation are not well known. After an acute exposure to 0.8 or 1 ppm, young mice, rats, and rabbits had greater changes in prostaglandins in BALF, but there were no age-dependent effects on BALF protein or cell number. Comparisons of male and female animals, and vitamin C or ascorbate deficiency did not reveal significant differences in the effects of O_3, but exercise during exposure increased susceptibility.

Important new work has revealed that susceptibility to O_3 is, in part, genetically determined. Mouse strains with differing sensitivities to O_3 have identified genes on separate loci controlling various aspects of inflammation, providing additional evidence for the mechanistic separation of responses to O_3. The research is summarized in Figure 5-4. Kleeberger's group has identified Inf-1, which modulates acute inflammatory responses; Inf-2 which modulates responses to subacute exposures; and TNF-α and TNF receptors, which are involved in inflammatory responses. Other research groups have identified loci linked to other

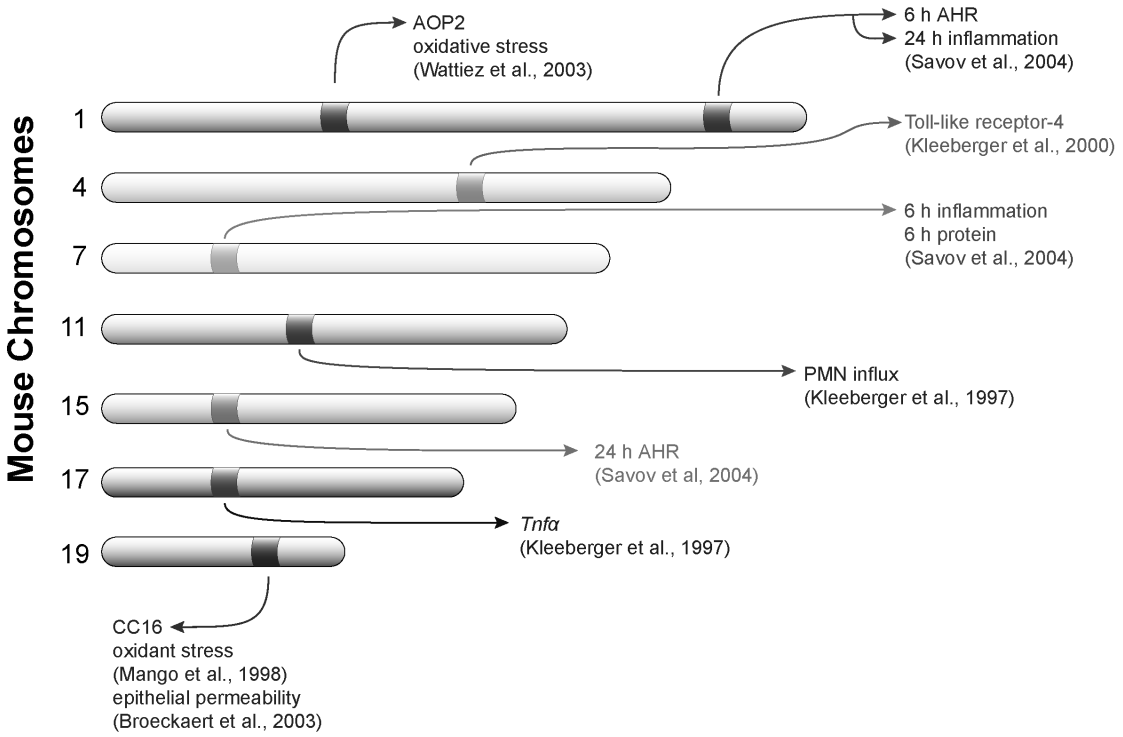


Figure 5-4. Mouse chromosomes on which genes or gene loci have been identified that modulate responses to O_3.

endpoints. This line of research provides a ground work for understanding the underlying mechanisms of O_3-induced injury, and can shed light on genes responsible for human susceptibility to O_3.

Recent studies have placed a major focus on mediators released from inflammatory cells to understand the mechanisms of O_3-induced inflammation and injury. Cytokines and chemokines have been shown to be released as a result of stimulation or injury of macrophages, epithelial cells and PMNs. Exposure of guinea pig AMs recovered in BALF and exposed *in vitro* to 0.4 ppm O_3 produced a significant increase in IL-6 and TNFα. An exposure of human AMs to an identical O_3 concentration increased TNFα, IL-1β, IL-6 and IL-8. The expression of MIP-2 mRNA or BALF levels of MIP-2 increased in mice and rats exposed to O_3 concentrations $\geq$1 ppm. An increase after O_3 exposure has also been reported for other cytokines and inflammatory mediators, including CINC and fibronectin. The CINC mRNA expression

was associated with neutrophilia at 24 hrs PE. Ozone exposure of mice also caused an increase in IL-6, MIP-1α and eotaxin in mice. Further understanding of the role of mediators has come from studies utilizing antibodies and inhibitors of known specificity. In these studies treatment of rats with an anti IL-6 receptor antibody prior to a nighttime exposure to O_3 abolished O_3-induced cellular adaptive response following a subsequent exposure. Studies utilizing antibodies to selected pro- or anti-inflammatory cytokines suggest a role of TNFα, interleukin-10 (IL-10) and IL-1β in O_3-induced changes in permeability, inflammation and cytokine release.

Studies investigating the mechanisms of PMN recruitment in the lung have explored the role of cell adhesion molecules that mediate PMN-endothelial cell interactions. An increase in tissue expression of ICAM-1 occurred in mice exposed to 0.8 ppm O_3. A comparable pattern of time-related changes in total protein, fibronectin and alkaline phosphatase activity in the BALF was observed in rats exposed to 1 ppm O_3. In monkeys, the O_3-induced inflammation was blocked by treatment with a monocolonal antibody to CD18, suggesting dependence of PMN recruitment on this adhesion molecule. Together, these studies support the role of extracellular matrix protein and cell adhesion molecules in lung inflammation and injury.

Ozone exposure also affects macrophage functions, and consequently their role in lung inflammation. Macrophages isolated from O_3-exposed mice produced increased amounts of NO, superoxide anion and PGE_2, but production of these mediators by macrophages from NOS knockout mice was not elevated. Additionally, mice deficient in NOS or mice treated with N^G-monomethyl-L-arginine, an inhibitor of total NOS, were protected from O_3-induced permeability, inflammation and injury. These findings suggest a role of NO in the production of O_3 effects.

5.2.4 Morphological Effects

Most mammalian species show generally similar morphological responses to <1 ppm O_3, which differ only by region, cell type, exposure parameters, and length of time between exposure and examination. Constant low exposures to O_3 create an early bronchoalveolar exudation, which declines with continued exposure and drops in the PE period. Epithelial hyperplasia also starts early, increases in magnitude for several weeks, plateaus with continuing exposure, and declines slowly during PE. Interstitial fibrosis has a later onset, continues to increase throughout

the exposure, and can continue to increase after the exposure ends. Nonhuman primates respond more than rats at this concentration, due to differences in antioxidants, the CAR (predicted to receive the highest dose of O_3), the presence of respiratory bronchioles, acinar volume, and differences in the nasal cavity's ability to "scrub" the O_3. Ciliated epithelial cells of the airway, Type 1 epithelial cells of the gas-exchange region, and ciliated cells in the nasal cavity are the cells most affected by O_3. Ozone-damaged ciliated cells are replaced by nonciliated cells (which are unable to provide clearance function) and Type 1 cells are replaced by Type 2 cells, which are thicker and produce more lipids. Inflammation also occurs, especially in the CAR, wherein the tissue is thickened as collagen accumulates. At exposures of 0.25 ppm O_3 (8 h/day, 18 mo) in monkeys, the distal airway is remodeled as bronchiolar epithelium replaces the cells present in alveolar ducts. In both rodents and monkeys, it appears that the natural seasonal patterns of O_3 exposure alters morphology more than continuous exposures, thus long-term animal studies with uninterrupted exposures may underestimate morphological effects.

5.2.4.1 Acute and Subchronic Exposure Effects

Morphological effects of key acute and subchronic exposure studies are summarized in Table AX5-9. Harkema et al. (1997a) reviewed toxicological studies of the nasal epithelial response to short-term O_3. New information regarding the effects of O_3 in this region include demonstrations that the topical anti-inflammatory corticosteriod fluticasone propionate prevents inflammation and mucous cell metaplasia in rats after cumulative O_3 exposure (0.5 ppm O_3, 8 h/day, for 3 or 5 days) (Hotchkiss et al., 1998). Exposure to bacterial endotoxin, a common ambient air toxicant, can potentiate mucous cell metaplasia in the nasal transitional epithelium of rats caused by a previous 3 day 0.5 ppm O_3 exposure (Fanucchi et al., 1998). Male F344/N Hsd rats were intranasally instilled with endotoxin after exposure to filtered air (FA) or 0.5 ppm O_3, (8 h/d for 3 d). Mucous cell metaplasia was not found in the air/endotoxin group, but was found in the O_3/saline group and was most severe in the O_3/endotoxin group. A similar synergistic effect was demonstrated by Wagner et al. (2001a,b) with exposure of Fischer rats to O_3 for 8 h per day for 3 days and endotoxin. Ozone alone created epithelial lesions in the nasal transitional epithelium, while endotoxin alone caused lesions in the epithelium of the nose and conducting airways. The enhanced O_3-induced mucous cell metaplasia was related to neutrophilic inflammation.

Pre-metaplastic responses, such as mucin mRNA upregulation, neutrophilic inflammation, and epithelial proliferation, were shown to be responsible for O_3-induced mucous cell metaplasia in the transitional epithelium of rats (Cho et al., 1999a, 2000). Male F344/N rats exposed to O_3 (0.5 ppm, 8 h/d for 1, 2, or 3 d) demonstrated a rapid increase in an airway-specific mucin gene mRNA after exposure to O_3, both before and during the onset of mucous cell metaplasia. Neutrophilic inflammation coincided with epithelial DNA synthesis and upregulation of rMuc-5AC, but was resolved before the development of epithelial metaplasia. The mucous cell metaplasia was neutrophil-dependent, whereas O_3-induced epithelial cell proliferation and mucin gene upregulation were neutrophil-independent.

Dormans et al. (1999) compared the extent and time course of fibrotic changes in mice, rats, and guinea pigs exposed to 0.2 and 0.4 ppm O_3 for 3, 7, 28, and 56 days. They found a concentration-related centriacinar inflammation in all three species, with a maximum after 3 days of exposure and total recovery within 3 days after exposure. Repair of O_3-induced damage by removal of injured epithelial cells is enhanced by the influx of neutrophils (Hyde et al., 1999; Veseley et al., 1999b; Miller et al., 2001; see Section 5.2.3). A study examining the kinetics of early cellular responses to O_3 utilized bromodeoxyuridine to label s-phase cells (Hotchkiss et al., 1997). Labeling indices for rat nasal transitional epithelial cell DNA were greatest 20 to 24 h after O_3 (0.5 ppm for 8 h) exposure, and remained greater than control at 36 h PE.

Very few published studies have explicitly explored susceptibility factors such as species, gender, age, antioxidant defense, acute and chronic airway disease, and exercise. Most typical laboratory species studied have qualitatively similar effects associated with O_3 exposure. Dormans et al. (1999) compared morphological, histological, and biochemical effects in the rat, mouse, and guinea pig following O_3 exposure and recovery in clean air. Wistar RIV:Tox male rats, NIH male mice, and Hartley Crl:(HA)BR male guinea pigs were continuously exposed to FA, 0.2, or 0.4 ppm for 3, 7, 28, and 56 days. Recovery from 28 days of exposure was studied at intervals of 3, 7, and 28 days PE. The mouse was the most sensitive as shown by a concentration and exposure-time dependent persistence of bronchiolar epithelial hypertrophy, elevated lung enzymes, and slow recovery from exposure. Exposure to the high dose for 56 d in both rats and guinea pigs caused increased amounts of collagen in ductal septa and large lamellar bodies in

Type II cells. The inflammatory response was greater in the guinea pig. Overall, the authors rated mice as most susceptible, followed by guinea pigs and rats.

Ferrets, monkeys, and rats were exposed to O_3 (1.0 ppm, 8 h) to compare airway effects Sterner-Kock et al. (2000). The ferrets and monkeys had similar epithelial necrosis and inflammation that was more severe than that found in rats. Because ferrets have a similar pulmonary structure as humans (e.g., well-developed respiratory bronchioles and submucosal glands), the authors concluded that the ferret would be a better model than rodents for O_3-induced airway effects. Age susceptibility is dependent on the endpoint examined (see Chapter 4 for discussions of age-related differences in O_3 dosimetry). One study (Dormans et al., 1996) demonstrated that O_3-induced centriacinar lesions are larger in younger rats than in older rats with exposures to 0.4 ppm for 1 to 7 days.

New studies have examined O_3-induced morphological effects in compromised laboratory animals. Rats with endotoxin-induced rhinitis were more susceptible to mucous cell metaplasia in the nasal transitional epithelium caused by a 3 day exposure to 0.5 ppm O_3 (Cho et al.,1999b). Wagner et al. (2002) reported a similar O_3-induced enhancement of inflammatory and epithelial responses associated with allergic rhinitis. Brown Norway rats were exposed to 0.5 ppm O_3, 8 h/day for 1 day or 3 consecutive days and then immediately challenged intranasally with either saline or ovalbumin (OVA). Multiple exposures to O_3 caused greater increases in mucosubstances produced in the nose by allergen challenge.

Recent research has focused on the concept of O_3 susceptible and nonsusceptible sites within the respiratory tract, including in situ antioxidant status and metabolic activity. Plopper et al. (1998) examined whether the variability of acute epithelial injury to short-term O_3 exposure within the tracheobronchial tree is related to local tissue doses of O_3 or to local concentrations of reduced glutathione (GSH). Adult male rhesus monkeys exposed to O_3 (0.4 or 1.0 ppm for 2 h) demonstrated significant cellular injury at all sites, but the most damage, along with increased inflammatory cells, occurred in the proximal respiratory bronchiole.
A significant reduction in GSH was found in the proximal bronchus at 0.4 ppm O_3 and in the respiratory bronchiole at 1.0 ppm O_3. A significant decrease in the percent of macrophages, along with significant increases in the percent of neutrophils and eosinophils, and a doubling of total lavage protein, were found after exposure to 1.0 ppm O_3 only. The authors concluded that

the variability of local O_3 dose in the respiratory tract was related to inhaled O_3 concentration and was closely associated with local GSH depletion and with the degree of epithelial injury.

Plopper and colleagues (e.g., Watt et al.,1998; Paige et al., 2000a) explored the site-specific relationship between epithelial effects of O_3 exposure and the metabolism of bioactivated compounds within the respiratory tract of rats. The distribution of CYP2E1-dependent activity, measured with a selective substrate (p-nitrocatechol), was found to be highest in the distal bronchioles and minor daughter airways, and lower in the lobar bronchi and major daughter airways. Short-term O_3 exposure (1 ppm for 8 h) increased CYP2E1 activity in the lobar bronchi/major daughter airways only; however, long-term O_3 exposure (1 ppm for 90 days) decreased CYP2E1 activity in the major and minor airways, further complicating the interpretation of O_3 effects based on concentration and duration of exposure and recovery. Rats treated i.p. with 1-nitronaphthalene, a pulmonary toxicant requiring metabolic activation, and exposed to 0.8 ppm O_3, 8h/day for 90 days showed greater histopathologic and morphometric effects in the CAR of the lung (Paige et al., 2000b). Despite reported tolerance to oxidant stress after long-term O_3 exposure, there was increased severity of ciliated cell toxicity.

5.2.4.2 Summary of Acute and Subchronic Morphological Effects

Short-term exposures to O_3 cause similar alterations in lung structure in a variety of laboratory animal species at concentrations of 0.15 ppm in rats and lower concentrations in primates. Cells in the CAR are the primary targets of O_3, but ciliated epithelial cells in the nasal cavity and airways and Type 1 epithelial cells in the gas exchange region are also targets. New work has shown that a topical anti-inflammatory corticosteroid can prevent these effects in nasal epithelia, while exposure to bacterial endotoxin can potentiate the effects. Ozone-induced fibrotic changes in the CAR are maximal at 3 d of exposure and recover 3 d PE with exposures of 0.2 ppm in rodents. New studies of susceptibility factors demonstrated that ferrets and monkeys have similar inflammatory and necrotic responses to 1 ppm O_3, which differs from lesser injury seen in rats. Rats with induced allergic rhinitis are more susceptible to 0.5 ppm than are controls. Important new work has demonstrated variability of local O_3 dose and subsequent injury in the RT due to depletion of GSH. The proximal respiratory bronchiole receives the most acute epithelial injury from exposures $\leq$1 ppm, while metabolic effects were greatest in the distal bronchioles and minor daughter airways.

5.2.4.3 Subchronic and Chronic Exposure Effects

Summaries of new studies of morphological effects of subchronic and chronic exposures are listed in Table AX5-10 in Annex AX5. In general, as the duration of exposure lengthens, there is not a concomitant linear increase in the intensity of effect of a given endpoint. Rather, as exposure proceeds past 1 week to 1 year, Type 1 cell necrosis and inflammatory responses generally decrease to near control values, and hyperplastic and fibrotic changes remain elevated. After long-term exposure ended, some indicies of fibrosis persisted and in some cases became more severe during PE periods in clean air.

Effects of O_3 on the upper respiratory tract of F344 rats exposed to O_3 (0.12, 0.5, or 1.0 ppm for 20 months) included marked mucous cell metaplasia in the rats exposed to 0.5 and 1.0 pm O_3, but not at 0.12 ppm O_3 (Harkema et al., 1997a) . In a follow-up study, hyperplasia was found in the nasal epithelium of rats exposed to 0.25 and 0.5 ppm, 8h/day, 7 days/week, for 13 weeks (Harkema et al., 1999). The mucous cell metaplasia, and associated intraepithelial mucosubstances, induced by 0.5 ppm O_3 persisted for 13 weeks after exposure. An acute (8 h) exposure to 0.5 ppm O_3 13 weeks after the chronic exposure induced an additional increase of mucosubstances in the nasal epithelium of rats but not in rats chronically exposed to 0 or 0.25 ppm O_3. The persistent nature of the O_3-induced mucous cell metaplasia in rats reported in this study suggests that O_3 exposure may have the potential to induce similar long-lasting alterations in the airways of humans.

No significant changes in nasal tissue were seen in rats continuously exposed for 49 days to the ambient air of Mexico City, Mexico (Moss et al., 2001). A rat study using 6-month exposures to ambient air of Sao Paulo, with a disparate pollutant composition than that of Mexico City, demonstrated development of secretory hyperplasia in rats (Lemos et al., 1994). However, without information on differences in ambient pollution composition in the two cities, the studies cannot be compared. Because of the persistent nature of these changes in the controlled studies with rats, and the fact that the upper airways of humans are probably more sensitive, like the monkey, the authors suggested that long-term exposure to ambient levels of O_3 could induce significant nasal epithelial lesions that may compromise the upper respiratory tract defense mechanisms of exposed human populations.

Rats exposed to 0.5 ppm O_3 for 1 month exhibited Bcl-2 in protein extracts of nasal epithelium (Tesfaigzi et al., 1998). Further, after 3 and 6 months of exposure, the number of

metaplastic mucous cells in the transitional epithelium was indirectly related to the percentage of cells that were Bcl-2 positive . Cells from rats exposed to FA did not express any Bcl-2. This study suggests that apoptosis regulators like Bcl-2 may play a role in the development and resolution of mucous cell metaplasia in the nasal airway.

A spectrum of lesions was reported (Herbert et al., 1996) in the nasal cavity and centriacinar lung of male and female mice exposed to 0.5 or 1.0 ppm of O_3 for 2 years, which persisted with continued exposure for 30 months. These lesions included bone loss in the maxilloturbinates, mucosal inflammation, mucous cell metaplasia in the nasal transitional epithelium and increased interstitial and epithelial thickening in the proximal alveolar region. In the CAR, there were increased numbers of nonciliated cells. However, changes in other endpoints including lung function and lung biochemistry were not evident. The investigators' interpretation of the entire study is that rodents exposed to the two higher O_3 concentrations had some structural hallmarks of chronic airway disease in humans.

A chronic study using a simulated, seasonal O_3-exposure pattern was reported by Plopper and colleagues (Evans et al., 2003; Schelegle et al., 2003a; Chen et al., 2003; Plopper and Fanucchi, 2000). Infant rhesus monkeys (30 days old) were exposed to FA, house dust mite allergen aerosol (HDMA), or O_3 + HDMA. The 0.5 ppm O_3 exposures were 8 h/day for 5 days, every 14 days for a total of 11 O_3 episodes. Half of the monkeys were sensitized to house dust mite allergen (*Dermatophagoides farinae*) at 14 and 28 days of age. The sensitized monkeys were exposed to HDMA for 2 h/day on Days 3-5 of the FA or O_3 exposures. The lungs were removed during the last FA exposure and the right and left cranial and right middle lobes were separately inflation fixed. Microdisection and morphometric analyses were performed on the conducting airways to the level of the most proximal respiratory bronchiole. Repeated exposures to O_3 or O_3 + HDMA over a 6-month period resulted in an atypical development of the basement membrane zone of airways in nonsensitized developing monkeys. Remodeling in the distal conducting airways was found in the sensitized monkeys as a result of the damage and repair processes occurring with repeated exposure (Evans et al., 2003; Schelegle et al., 2003a). Lung function changes in these monkeys (Schelegle et al., 2003b), and associated adaptation of the respiratory motor responses (Chen et al., 2003), are described in Section 5.2.5.2. Collectively, these findings provide a pathophysiologic basis for changes in airway function

described in children growing up in polluted metropolitan areas (e.g., Tager, 1999) (see Chapter 7).

Necropsy of the left caudal lobe of these infant monkeys showed accumulation of eosinophils and mucous cells within the combined epithelial and interstitial compartments in the conducting airways and in the terminal/respiratory bronchioles (Schelegle et al., 2003a) . House dust mite sensitization and HDMA challenge alone, or combined with O_3 exposure, resulted in significantly greater eosinophil accumulation in the conducting airways when compared to FA and O_3 only exposures. A significant accumulation of eosinophils was found in the terminal/respiratory bronchioles of the sensitized monkeys challenged with HDMA when compared to monkeys exposed to FA, O_3, and HDMA + O_3. The mean mass of mucous cells increased in the fifth generation conducting airways of sensitized animals challenged with HDMA alone and when combined with O_3 exposure, and in the terminal bronchioles of sensitized animals exposed to HDMA + O_3. The tracheal basement membrane of HDMA-sensitized monkeys exposed to HDMA or to HDMA + O_3 was significantly increased over controls; however, there were no significant changes in the airway diameter of proximal and mid-level airways. Exposures of sensitized young monkeys to HDMA alone, or to O_3 alone, resulted in eosinophilia of the mid-level conducting airways and the terminal/respiratory bronchioles, but without alterations in airway structure or function. The authors interpreted these findings to indicate that the combination of cyclic O_3 exposure and HDMA challenge in HDMA-sensitized infant monkeys act synergistically to produce an allergic-reactive airway phenotype characterized by significant eosinophilia of midlevel conducting airways, transmigration of eosinophils into the lumen, and an altered structural development of conducting airways that is associated with increased airway resistance and nonspecific airway reactivity (see Section 5.2.5).

Examination of development of the tracheal basement membrane zone (BMZ) in these monkeys (Evans et al., 2003) showed that with exposures to either O_3 or HDMA + O_3, BMZ development was affected. Abnormalities in the BMZ included: (1) irregular and thin collagen throughout the BMZ; (2) perlecan depeleted or severely reduced; (3) FGFR-1 immunoreactivity reduced; (4) FGF-2 immunoreactivity absent in perlecan-deficient BMZ, but present in the lateral intercellular space (LIS), in basal cells, and in attenuated fibroblasts; (5) syndecan-4 immunoreactivity increased in basal cells. The authors interpret these data as suggesting that O_3

targets cells are associated with synthesis of epithelial BMZ perlecan. The absence of FGF-2, normally stored in the BMZ, could affect downstream signaling in airway epithelium and could be responsible for the abnormal development of the airway seen in this study, and thus be an important mechanism modulating O_3-induced injury. Midlevel bronchi and bronchioles from these monkeys (Larson et al., 2004) demonstrated decrements in the density of epithelial nerves in the axial path between the sixth and seventh airway generations in exposures to O_3. Combined O_3 + HDMA exposures exacerbated this reduction. They attribute this loss of nerve plexuses to neural regression or stunted nerve development, the latter corroborated by the Evans et al. (2003) finding of decreased growth factors following O_3 exposure. Additionally, they found streaks or clusters of cells immunoreactive for protein gene product 9.5 (PGP 9.5, a pan-neuronal marker) and negative for calcitonin gene-related peptide. The functional significance of this is unknown but suggests to the authors a possible injury-repair process induced by O_3.

Remodeling of the distal airways and CAR is one of the most disturbing aspects of the morphological changes occurring after subchronic and chronic exposure to O_3. Recently, bronchiolization was reported in rats exposed to 0.4 ppm O_3 for only 56 days (van Bree et al., 2001). They also found collagen formation progressively increased with increasing O_3 exposure and persisted into PE recovery. In addition to centriacinar remodeling, Pinkerton et al. (1998) reported thickening of tracheal, bronchial, and bronchiolar epithelium after 3 or 20 months exposure to 1 ppm O_3, but not to 0.12 ppm. Although some older literature had reported that chronic exposures to $\leq$1.0 ppm O_3 cause emphysema, none of the more recent literature supports this hypothesis.

5.2.4.4 Summary and Conclusions - Subchronic and Chronic Morphological Effects

The progression of effects during and after a chronic exposure at a range of 0.5 to 1.0 ppm is complex, with inflammation peaking over the first few days of exposure, then dropping, then plateauing, and finally, largely disappearing. Epithelial hyperplasia follows a somewhat similar pattern. Effects of 0.5 ppm O_3 for 20 months on the nasal mucosa include atrophy of nasal turbinates and mucous cell metaplasia, which persisted long after the exposure ceased. Fibrotic changes in the tissue increase very slowly over months of exposure, and, after exposure ceases, the changes sometimes persist or increase. The pattern of exposure in this same concentration range determines effects, with 18 mo of daily exposure causing less morphologic damage than

exposures on alternating months. This is important, given that environmental O_3 exposure is typically seasonal. Plopper and colleagues' long term study of infant rhesus monkeys exposed to simulated, seasonal O_3 (0.5 ppm 8 h/day for 5 days, every 14 days for 11 episodes) demonstrated: (1) remodeling in the distal airways; (2) abnormalities in tracheal basement membrane; (3) eosinophil accumulation in conducting airways; and (4) decrements in airway innervation. These findings advance earlier information regarding possible injury-repair processes occurring with seasonal O_3 exposures.

5.2.5 Effects on Pulmonary Function

5.2.5.1 Acute and Subchronic Exposure Effects on Pulmonary Function

Numerous pulmonary function studies of the effects of acute O_3 exposure (defined here as $\leq$1 week of exposure) in several animal species have been conducted and generally show responses similar to those of humans (e.g., increased breathing frequency, decreased tidal volume, increased resistance, decreased forced vital capacity (FVC) and changes in the expiratory flow-volume curve). These effects are seen at 0.25 to 0.4 ppm O_3 for several h in a number of species. At concentrations of $\geq$1 ppm, breathing mechanics (compliance and resistance) are affected. The breathing pattern returns to normal after O_3 exposure. In rats exposed to 0.35 to 1 ppm O_3 for 2 h/day for 5 days, there was a pattern of attenuation of pulmonary function responses similar to that observed in humans. Concurrently, there was no attenuation of biochemical indicators of lung injury or of morphological changes.

Work demonstrating attenuation of pulmonary functions (see Table AX5-11) was completed by Wiester et al. (1996) who exposed male Fischer 344 rats to 0.5 ppm O_3 for either 6 or 23 h/day over 5 days. Ozone-induced changes in lung volume were attenuated during the 5 exposure days and returned to control levels after 7 days recovery. The responses to repeated O_3 exposure in rats were exacerbated by reduced ambient temperature, presumably as a result of increased metabolic activity.

Researchers have utilized inbred mouse strains with varying ventilatory responses to O_3 to attempt to model susceptible populations. As differences were seen in inflammatory responses to acute O_3 exposures in C57BL/6J and C3H/HeJ mice, comparisons were made of their ventilatory responses also (Tankersley et al., 1993). Following an exposure of 2 ppm O_3 for 3 h,

breathing frequency (f), tidal volume (V_T), and minute ventilation were measured 1 and 24 h in both normocapnia (or air at ~0% CO_2) and hypercapnia (5 or 8% CO_2). They demonstrated that acute O_3 exposures caused altered hypercapnic ventilatory control, which varied between strains. This suggested to the authors that O_3-induced alterations in ventilation are determined, at least in part, by genetic factors. A caveat regarding studies such as this using high exposure concentrations is that events observed at high concentrations may differ from those observed at near-ambient O_3 levels.

Paquette et al. (1994) measured ventilatory responses in C57BL/6J and C3H/HeJ mice given repeated acute exposures of 0.3 ppm for 48 and 72 h. The two strains had differing responses to both normocapnia and hypercapnia. Normocapnic V_E was greater following subacute O_3 exposure in C57BL/6J mice than in C3H/HeJ mice, due to increased *f* and reduced V_T, respectively. This suggests that the increased V_T in C57BL/6J mice may contribute to the increased susceptibility to lung injury due to a greater dose of O_3 reaching the lower lung. Hypercapnic ventilatory responses following subacute O_3 exposures demonstrated reduced V_E (due to decreased V_T) in C57BL/6J only. Evaluations of O_3 dosimetry were performed in these two strains using $^{18}O_3$-labeled ozone (2 ppm for 2-3 h) (Slade et al., 1997). Immediately after exposures of 2 ppm $^{18}O_3$ for 2-3 h, C3H/HeJ mice had 46% less ^{18}O in lungs and 61% less in trachea, than C57BL/6J. Additionally, C3H/HeJ mice had a greater body temperature decrease following O_3 exposure than C57BL/6J mice, suggesting that the differences in susceptibility to O_3 are due to differences the ability to decrease body temperature and, consequently decrease the dose of O_3 to the lung.

Tracheal transepithelial potential has also been shown to differ in eight mouse strains 6 h after exposure to 2 ppm O_3 for 3 h (Takahashi et al., 1995b). AKR/J, C3H/HeJ, and CBA/J were identified as resistant strains and 129/J, A/J, C57BL/6J, C3HeB/FeJ and SJL/J were identified as susceptible strains. The authors noted that strains' responses to this parameter did not show concordance with inflammatory responses, suggesting to the authors that the two phenotypes are not controlled by the same genetic factors.

Savov et al. (2004) characterized ventilatory responses in nine mouse strains exposed to O_3 (2.0 ppm O_3 for 3 h). C57BL/6J was hyporeactive to MCh prior to O_3, but was very responsive to MCh following O_3. Conversely, C3H/HeJ had an intermediate baseline P_{enh} and a small

response to MCh following O_3 exposure. This study corroborates the evidence of no consistent relationship between respiratory P_{enh} and inflammation.

5.2.5.2 Summary and Conclusions - Acute and Subchronic Effects on Pulmonary Function

Early work has demonstrated that during acute exposure of ~0.2 ppm O_3 in rats, the most commonly observed alterations are increased frequency of breathing and decreased tidal volume (i.e., rapid, shallow breathing). Exposures of ~1.0 ppm O_3 affect breathing mechanics (compliance and resistance). Additionally, decreased lung volumes are observed in rats with acute exposures at levels of 0.5 ppm. New work utilizing inbred mouse strains with varying ventilatory responses to O_3 has suggested that: (1) control of the ventilatory response is determined, at least in part, by genetic factors; (2) increased V_T in some strains may contribute to lung injury due to a greater dose of O_3 reaching the lower lung; (3) some strains' ability to reduce body temperature may account for their decreased O_3-induce lung injury; and (4) tracheal transepithelial potential is determined, in part, by genetic factors. Importantly, the genetic loci that appear to be modulating various aspects of pulmonary responses to O_3 differ from each other and from loci controlling inflammatory responses.

Exposures of 2 h/day for 5 days create a pattern of attenuation of pulmonary function in both rats and humans without concurrent attenuation of lung injury and morphological changes, indicating that the attenuation did not result in protection against all the effects of O_3. Chronic O_3 exposure studies evaluating pulmonary function are not available. Earlier work has demonstrated that repeated daily exposure of rats to an episodic profile of O_3 caused small, but significant decrements in lung function that were consistent with early indicators of focal fibrogenesis in the proximal alveolar region, without overt fibrosis.

5.2.5.3 Ozone Effects on Airway Responsiveness

Effects of O_3 on airway reactivity have been observed in a variety of species at an exposure range of 0.5 to 1 ppm. Many of the new studies on pulmonary function in laboratory animals allow a better prediction of the effects of O_3 exposure on the exacerbation of asthma symptoms and the risk of developing asthma in humans. However, it is necessary to understand the factors that determine airway responsiveness across different mammalian species as discussed in Chapter 4.

Traditional studies of airway responsiveness require sedation in both infants and laboratory animals. Laboratory animal studies employ intravenous agonist challenges as well as inhalation challenges, though inhaled agonist challenges are preferred in humans. Exercise testing is not possible with sedation unless exercise is "simulated" by increasing ventilation using elevated F_iCO_2; and the need for artificial ventilation in laboratory animal studies may cause breathing patterns that affect O_3 deposition. Joad et al. (2000) reported that when 1 ppm O_3 for 90 min is administered to isolated rat lung at either 2.4 mL/40 bpm or 1.2 mL/80 bpm, the more rapid breathing pattern elicits less epithelial cell injury than the slower breathing pattern. Though this study design does not really model rapid shallow breathing elicited in the intact animal, it shows greater reduction in injury in the proximal axial airway compared to its adjacent airway branch and terminal bronchiole. The rapid, shallow breathing pattern protects the large conducting airways of rats, but causes a more even distribution of epithelial cell injury to the terminal bronchioles (Schelegle et al., 2001). Postlethwait et al. (2000) demonstrated that the conducting airways are the primary site of acute cytotoxicity from O_3 exposure. Three-dimensional mapping of the airway tree in SD rat isolated lung exposed to 0, 0.25, 0.5, or 1.0 ppm O_3 showed a concentration-dependent increase in injured cells. Injury was evident in proximal and distal conduction airways, lowest in terminal bronchioles, and highest in the small side branches downstream of bifurcations. These exposure levels did not concurrently elicit changes in LDH activity or total protein in BALF, suggesting that the mapping technique is a more sensitive measure of injury and is useful in dosimetry studies.

Whole-body plethysmography of unanesthetized, unrestrained rodents has been used to indirectly measure pulmonary resistance (Shore et al., 2002; Goldsmith et al., 2002; Jang et al., 2002). However, these indices of inspiratory/expiratory pressure differences, including enhanced pause (P_{enh}) may be less sensitive than direct measurements of lung airflow resistance (Murphy, 2002). Changes in airway structure caused by viral infections also must be considered when evaluating laboratory animal studies. Animals with acute viral illness have morphological evidence of inflammatory cell infiltration, bronchiolar wall edema, epithelial hyperplasia, and increased airway mucous plugs that can cause airway narrowing, air trapping, and serious functional changes in the lung (Folkerts et al., 1998).

Exercise-induced bronchoconstriction in humans appears to be mediated by changes in the tonicity of the airway lining fluid (Anderson and Daviskas, 2000). Brannan et al. (1998) suggest

that a test in laboratory animals based on the inhalation of mannitol aerosol (hyperosmolar) might be feasible and provide information similar to that from exercise challenges in cooperative children and adults. Unfortunately, there have been few reports of mannitol or adenosine monophosphate challenges in laboratory animals; most studies have utilized histamine, methacholine, acetylcholine, or carbachol to determine outcome. In active humans with asthma, adenosine monophosphate challenges appear to better reflect ongoing airway inflammation than histamine or methacholine challenges (Polosa and Holgate, 1997; Avital et al, 1995a,b), and might be useful in identifying mechanisms of asthma in laboratory animals and their responsiveness to environmental pollutants.

The increased responsiveness to bronchoconstrictor challenge in asthma is thought to result from a combination of structural and physiological factors that include increased inner-wall thickness, increased smooth-muscle responsiveness, and mucus secretion. These factors also are likely to determine a level of innate airway responsiveness that is genetically influenced. Chapter 6 (Section 6.8) discusses cellular and biochemical changes that have been identified in human asthmatics. These studies suggest that the mechanisms involved in AHR are multifactoral, with general agreement that there is an inconsistent relationship between AHR and markers of inflammation.

A large data base of laboratory animal research has been collected on the role of O_3 in producing an increase in AHR (see Table AX5-12). Exposure levels ($\geq$1 ppm for $\geq$30 min) in many of these studies are not environmentally relevant, but information may be obtained regarding the mechanisms of action of O_3 concerning: O_3 concentration and peak response time, inhaled versus intravenous challenge with nonspecific bronchoconstrictors, neurogenic mediation, neutrophilic inflammation, and interactions with specific biological agents (e.g., antigens and viruses). However, as with other toxicants, high-dose and low-dose mechanisms may differ, so interpretation of results must take this into consideration.

Many species of laboratory animals have been used to study the effects of O_3 on airway bronchoconstriction. Ozone-induced AHR in guinea pigs has been used to model human bronchospasm (van Hoof et al., 1996; 1997a,b; Matsubara et al., 1997a,b; Sun and Chung, 1997; Aizawa et al., 1999a,b; Tsai et al., 1998; Nakano et al., 2000). Because these studies were done at 2 to 3 ppm O_3, these results are not directly relevant for extrapolation to potential airway responses in humans exposed to ambient levels of O_3. Humans with reactive airway disease

(e.g., asthma) appear to be sensitive to ambient levels of O_3 (see Chapters 6 and 7) and the current understanding is that O_3 exacerbates airway responsiveness to specific allergens, presumably by nonspecifically increasing AHR.

Shore et al. (2000, 2002) have shown that O_3-induced AHR is reduced in immature rats and mice. SD rats exposed to 2 ppm O_3 at ages 2, 4, 6, 8, or 12 weeks and A/J mice exposed to 0.3 to 3 ppm for 3 h at age 2, 4, 8, or 12 weeks had similar concentration-related decreases in V_E except at the youngest ages. This smaller decrement in V_E suggested a delivered dose that was much greater in the younger animals. This group (Shore et al., 2003) has also recently shown that obese mice have greater ventilatory responses to O_3. Exposures of 2.0 ppm O_3 for 3 h to lean, WT C57BL/6J and *ob/ob* mice (mice with a genetic defect in the coding for leptin, the satiety hormone) showed that the *ob/ob* mice had enhanced AHR and inflammation compared to the WT mice. These data correlate with epidemiological data showing increased incidence of asthma in overweight children.

Increased AHR to various nonspecific bronchoconstrictive agents (e.g., ACh, methacholine, histamine, carbachol) given by inhalation or intravenous routes has been previously shown in laboratory animals exposed to O_3 concentrations $\leq$1.0 ppm. Dye et al. (1999) showed hyperresponsiveness to methacholine in rats 2 h after exposure to 2 ppm O_3 for 2 h. AHR can be induced by specific antigens as well as O_3. The most commonly used laboratory animal model is the OVA sensitized guinea pig. Animals sensitized with OVA have been shown to have similar responses to nonspecific bronchoconstrictors as control animals.

OVA-sensitized guinea pigs (Sun et al., 1997) and mice (Yamauchi et al., 2002) were used to determine the enhancement of antigen-induced bronchoconstriction by acute, high-level O_3 (1.0 ppm O_3 for 1 h). Male Dunkin-Hartley guinea pigs were sensitized by i.p. injection of OVA and exposed to O_3 alone, OVA aerosol, or O_3 + OVA. Ozone exposure alone increased bronchial responsiveness to ACh at 3 h, but not 24 h, while OVA alone had no effect. Combined exposure to O_3 and OVA (1 ppm for 1 h, then 3 min OVA) increased bronchial responsiveness to ACh 3 h after O_3 exposure. At 24 h following O_3 exposure, AHR increased when OVA challenge was performed at 21 h, suggesting that O_3 pre-exposure can potentiate OVA-induced AHR. Neutrophil counts in the BALF increased at 3 and 24 h after O_3 exposure alone but were not further increased when O_3 exposure was combined with OVA airway challenge; however

protein content of the BALF did increase at 3 and 24 h in the O_3 and OVA groups. Thus, this study also indicates that high-ambient O_3 exposure can augment antigen (OVA)-induced AHR in guinea pigs.

Yamauchi et al. (2002) sensitized male C57BL/6 mice by i.p. injection of OVA and then exposed them to O_3. The sensitized mice had AHR to methacholine. Ozone exposure caused significant decreases in dynamic lung compliance, minute ventilation, and P_aO_2 in OVA-sensitized mice, but not in controls. A marker of inflammation (soluble intercellular adhesion molecule-1 [sICAM-1]) was elevated in the BAL fluid of OVA-sensitized mice, but sICAM-1 levels were not significantly changed by O_3 exposure, indicating that the O_3-induced AHR to methacholine was not caused by O_3-induced inflammation.

Ozone-induced AHR may be temporally associated with inflammatory cells stimulated by cytokines (Koto et al., 1997), mast cells (Igarashi et al., 1998; Noviski et al., 1999), or by oxygen radicals (Takahashi et al., 1993). One study, however, has shown that inflammation is not a prerequisite of AHR (Koto et al., 1997), and it has been suggested that O_3-induced AHR may be epithelium dependent (McGraw et al., 2000). For example, neonatal rats pretreated with capsaicin, which will permanently destroy C-fibers and prevent O_3-induced (1 ppm, 8 h) release of neuropeptides (Vesely et al., 1999a), and then exposed to O_3 when adults, showed a marked increase in airway responsiveness to inhaled aerosolized methacholine (Jimba et al., 1995). Takebayashi et al. (1998) has shown that depletion of tachykinins by capsaicin treatment, or by a specific tachykinin receptor antagonist, can block the induction of AHR by O_3. The seemingly disparate responses in laboratory animals may be due to species- or strain-specific differences in inherent reactivity to bronchoconstrictors, or to inherent differences in susceptibility to O_3-induced inflammation (Zhang et al., 1995; Depuydt et al., 1999; Dye et al., 1999).

Studies that may be potentially relevant to ambient levels of O_3 were conducted in vivo, in an isolated perfused lung model, and in ex vivo lung segments using multihour and repeated multihour exposures with ambient levels of O_3. A study on the relationship between O_3-induced AHR and tracheal epithelial function was conducted in New Zealand white rabbits by Freed et al. (1996). Rabbits exposed to O_3 (0.2 ppm for 7 h) demonstrated significantly decreased tracheal transepithelial potential difference but no changes in lung resistance. Changes in the compartmentalized lung resistance, measured in response to ACh challenge before and after bilateral vagotomy, were not significantly different in air-exposed rabbits; however, bilateral

vagotomy did enhance peripheral lung reactivity in O_3-exposed rabbits. The ACh-induced a 140% increase in lung resistance with O_3 exposure was two times higher than with air exposure, indicating that ambient-level O_3 exposure affects tracheal epithelial function in rabbits and increases central airway reactivity, possibly through vagally-mediated mechanisms.

Pulmonary mechanics and hemodynamics were studied in the New Zealand white rabbit isolated perfused lung model that allowed partitioning of the total pressure gradient into arterial, pre- and post-capillary, and venous components (Delaunois et al., 1998). Exposures to O_3 (0.4 ppm for 4 h) were followed by evaluation of airway responsiveness to ACh, substance P (SP), or histamine immediately or 48 h later. Ozone inhibited pulmonary mechanical reactivity to all three bronchoconstrictors that persisted for 48 h and modified vasoreactivity of the vascular bed, but only at 48 h PE. Arterial segmental pressure, normally insensitive to ACh and SP, was significantly elevated by O_3; precapillary segmental pressure decreased in response to Ach, suggesting that O_3 can induce direct vascular constriction, but the vascular responses are variable and depend on the agonist used and on the species studied.

Airway responsiveness to the same three compounds was evaluated by Segura et al. (1997) in guinea pigs exposed to O_3 (0.15, 0.3, 0.6, or 1.2 ppm for 4 h). Ozone did not cause AHR to ACh or histamine, except at the highest concentration (1.2 ppm O_3) for histamine. However, O_3 did cause AHR to SP at $\geq$0.3 ppm, suggesting that O_3 destroys neutral endopeptidases (responsible for SP inactivation) in airway epithelial cells. Vargas et al. (1998), in a follow-up study, demonstrated that guinea pigs chronically exposed to 0.3 ppm O_3 for 4 h/day became adapted to SP-induced AHR. Ozone caused increased sensitivity to SP after 1, 3, 6, 12, and 24 days of exposure that was associated with airway inflammation; however, after 48 days of exposure, the increased sensitivity to SP was lost.

This study is in accordance with Szarek et al. (1995) who demonstrated that AHR associated with acute O_3 exposures does not persist during long-term exposure to near-ambient-levels of O_3 ($\leq$1 ppm). Fischer 344 rats, exposed to 0.0, 0.12, 0.5, or 1.0 ppm O_3, 6 h/day, 5 days/week for 20 months, demonstrated significantly reduced responses to bethanechol, ACh, and electrical field stimulation in eighth generation airway segments. This suggests that some adaptation had taken place during long-term exposure, possibly increased inner wall thickness.

It is well known that the changes in breathing pattern and lung function caused by O_3 are attenuated with repeated daily exposures for at least 3 to 5 days. But guinea pigs exposed to

0.5 ppm O_3, 8 h/day for 7 days showed enhancement of responsiveness of rapidly adapting airway receptors (Joad et al., 1998). Repeated exposure increased receptor activity to SP, methacholine, and hyperinflation; there were no significant effects on baseline or SP- and methacholine-induced changes in lung compliance and resistance, suggesting that the responsiveness of rapidly adapting receptors was enhanced.

Male and female Hartley guinea pigs exposed to O_3 (0.1 and 0.3 ppm, 4 h/day, 4 days/week for 24 weeks) were evaluated for airway responsiveness following ACh or OVA inhalation challenges (Schlesinger et al., 2002a,b). Ozone exposure did not cause AHR in nonsensitized animals but did exacerbate AHR to both ACh and OVA in sensitized animals that persisted for 4 weeks after exposure. The effects of O_3 on airway responsiveness were gender independent and were concentration-related for the ACh challenges.

Schelegle et al. (2003a) evaluated airway responsiveness in infant rhesus monkeys exposed to a 5 day O_3 episode repeated every 14 days over a 6-month period. Half of the monkeys were sensitized to house dust mite allergen (HDMA; *Dermatophagoides farinae*) at 14 and 28 days of age before exposure to a total of 11 episodes of O_3 (0.5 ppm, 8 h/day for 5 days followed by 9 days of FA), HDMA, or O_3 + HDMA. Baseline R_{aw} was significantly elevated after 10 exposure episodes in the HDMA + O_3 group compared to the FA, HDMA, and O_3 exposure groups. Aerosol challenge with HDMA at the end of the 10th episode did not significantly affect R_{aw}, V_T, f_B, or S_aO_2. Aerosol challenge with histamine was not significantly different after 6 episodes; however, the EC150 R_{aw} for the HDMA + O_3 group was significantly reduced after 10 episodes when compared to the FA, HDMA, and O_3 exposure groups, indicating the development of AHR in this group sometime between episodes 6 and 10. The results are consistent with altered structural development of the conducting airways.

During repeated episodic exposures to O_3, respiratory responses are first altered to a rapid, shallow breathing pattern, which has long been considered protective, especially to the deep lung. This dogma has been discounted recently as discussed above (Schelegle et al., 2001). Alfaro et al. (2004) examined the site-specific deposition of ^{18}O (1 ppm 2 h) at breathing frequencies of 80, 120, 160, or 200 breaths/minute (bpm). At all frequencies, parenchymal areas had a lower content of ^{18}O than trachea and bronchi. As breathing frequency increased from 80 to 160 bpm, the deposition showed a reduction in midlevel trachea and an increase in both mainstream bronchi. At this frequency there was also an increase in deposition in parenchyma

supplied by short (cranial) airway paths, consistent with results seen by Schelegle et al., (2001). At 200 bpm ^{18}O deposition in trachea increased, concurrent with increases in right cranial and caudal bronchi regions. Right cranial parenchymal content decreased at 200 bpm, whereas right caudal parenchymal levels did not change at any breathing frequency. The authors list some limitations of this study, such as the possible effect on regional distribution of ventilation by use of the negative-pressure ventilator, the effect of paralysis on airway geometry, and possible translocation of ^{18}O during the 2 h exposure period. These two studies provide evidence that O_3-induced rapid, shallow breathing creates a more evenly distributed injury pattern, with possibly greater protection from focal injury to the large conducting airways including the trachea and the left mainstem bronchus.

Another study of the adaptive phenomena in SD rats used an exposure paradigm consisting of 5 days of daily 8 h 1 ppm O_3 exposures followed by 9 days of recovery in FA (Schelegle et al., 2003b). This O_3/FA pattern was repeated for 4 cycles and demonstrated that the O_3-induced rapid shallow breathing pattern was followed by adaptation that occurred with each cycle. However, the release of SP from the trachea , the neutrophil content, and cell proliferation became attenuated after the first cycle, suggesting a disconnect from the rapid shallow breathing response. Hypercellularity of the CAR epithelium and thickening of the CAR interstitium, not linked to changes in cell proliferation, were also found. The authors suggest mechanism(s) of injury from repeated O_3 exposures consisting of diminished neutrophilic inflammation/and or release of mitogenic neuropeptides, depressed cell proliferative response, and cumulative distal airway lesion.

Following the initial response of a rapid, shallow breathing pattern, animals eventually adapt with continued episodic exposure despite the continued presence of epithelial damage, altered structural development, and inflammation of the airways. Chen et al. (2003) used a subset of the monkeys from the Schlegele et al. (2003a) study to demonstrate that attenuation of O_3-induced rapid shallow breathing and lung function changes typically seen with repeated O_3 exposure may be caused by the adaptation of the respiratory motor responses. This episodic O_3 exposure appeared to create neuroplasticity of the nucleus tractus solitarius (NTS; a region of the brainstem which controls respiration), including increased nonspecific excitability of the NTS neurons, an increased input resistance, and an increased spiking response to intracellular injections of depolarizing current.

5.2.5.4 Summary and Conclusions - Effects on Airway Responsiveness

Ozone-induced AHR has been reported in a number of laboratory species at an exposure range of 0.5 to 1.0 ppm and in human asthmatics at ambient levels. In asthmatics, O_3 is thought to exacerbate AHR to specific allergens by nonspecifically increasing AHR. New studies have demonstrated that AHR in asthmatics is due in part to chronic inflammation and airway remodeling. Animal studies have shown that O_3 exposure can augment OVA-induced AHR. Importantly, there is a temporal relationship between inflammatory cell influx and O_3-induced AHR, but inflammation is not a prerequisite of AHR. Repeated O_3 exposures enhance AHR, possibly by modulating rapidly adapting airway receptors or by altering the structure of conducting airways.

Currently reported investigations on AHR with repeated O_3 exposure to nonsensitized laboratory animals have shown equivocal results, especially at the most relevant ambient O_3 concentrations of $\leq$0.3 ppm. The few available studies in sensitized laboratory animals are consistent with the O_3-induced exacerbation of AHR reported in atopic humans with asthma (see Chapter 6) but the results are difficult to extrapolate because of interindividual and interspecies differences in responsiveness to bronchoprovocation and possible adaptation of airway responsiveness with long-term, repeated O_3 exposures. Therefore, further studies in laboratory animals are needed to investigate responses to the different challenges in relation to measurements of airway inflammation and the other physiological and structural factors known to contribute to airway responsiveness in human subjects.

Important new information indicates that rapid shallow breathing in response to O_3 causes a more evenly distributed injury pattern rather than protects from injury. New insights into the mechanisms of O_3-induced AHR suggest that: (1) exercise-induced bronchoconstriction may be mediated by changes in tonicity of the bronchial smooth muscles; (2) vagally-mediated mechanisms may affect tracheal epithelial function and increase central airway reactivity; (3) O_3 may induce direct vascular constriction; (4) O_3 may destroy neural endopeptidases in airway epithelial cells, thus preventing the inactivation of SP; and (5) repeated O_3 exposures may diminish neutrophilic inflammation, depress cell proliferation, and cause cumulative distal airway lesions.

5.2.6 Genotoxicity Potential of Ozone

There has been an historical interest in the ability of ground-level pollution to cause cancer, especially lung cancer. This interest has been amplified in recent years by results of an epidemiologic study that suggest association of increased risks of incident lung cancer with elevated long-term ambient concentrations of O_3, PM_{10}, and SO_2 in nonsmoking California males (Beeson et al., 1998; Abbey et al., 1999). However, another larger, nationwide American Cancer Society study (Pope et al., 2002) showed no significant effect of O_3 on mortality risk, but positive associations between warm season (July-September) O_3 concentrations and cardiopulmonary mortality. Studies of children and young adults of southwest metropolitan Mexico City, repeatedly exposed to high levels of O_3, PM, NO_x, aldehydes, metals, and other components in a complex ambient mixture, also report DNA damage in blood leukocytes and nasal epithelial cells (Valverde et al., 1997; Calderón-Garcidueñas et al., 1999) and abnormal nasal biopsies (Calderón-Garcidueñas et al., 2001a). (See Chapter 6 for a discussion of the human studies.)

A number of experimental studies have been done to explore the mutagenic/carcinogenic potential of O_3. In vitro studies are difficult to interpret due to very high exposure levels and culture systems that allowed the potential formation of artifacts. Some recently published in vivo exposure studies (see Table AX5-13) found increased DNA strand breaks in respiratory cells from guinea pigs (Ferng et al., 1997) and mice (Bornholdt et al., 2002) but, again, only on exposure to high doses of O_3 (1 ppm for 72 h and 1 or 2 ppm for 90 min, respectively).

Exposing the A/J mouse strain (known to have a high incidence of spontaneous pulmonary adenomas) to 0.12, 0.50, and 1.0 ppm O_3 for 6 h/day, 5 days/week for up to 9 months, Witschi et al. (1999) did not find O_3 exposure-related differences in lung tumor multiplicity or incidence. Similarly, in a subchronic exposure study ($B6C3F_1$ mice to 0.5 ppm O_3 for 6 h/day, 5 days/week for 12 weeks) Kim et al. (2001) did not find statistically significant increases in the incidence of lung tumors. Significant differences in mean body weight as well as mean absolute and relative weights of several organs (e.g., liver, spleen, kidney, testes, and ovary) were observed between O_3-exposed and air-exposed mice. Histopathologic examination of major organs revealed oviductal carcinomas in 3/10 O_3-exposed female mice.

5.2.6.1 Summary and Conclusions - Genotoxicity Potential of Ozone

The weight of evidence from new experimental studies does not appear to support ambient O_3 as a pulmonary carcinogen in laboratory animal models. These new data are in agreement with a study of carcinogenicity of O_3 from the NTP study (National Toxicology Program, 1994; Boorman et al., 1994), which was negative in male and female rats, ambiguous in male mice, and positive only in female mice at high concentrations of O_3 (i.e., 1.0 ppm). As none of the new experimental studies of genotoxicity provided lifetime exposure durations such as those used in NTP cancer studies, the observation of no effects must be tempered by consideration of the limited duration of the exposure. Overall, then, the new animal studies are inconclusive as are the epidemiologic studies discussed in Chapter 7, which may be due to significant species differences in this health endpoint. Also, O_3 could act as a co-carcinogen functioning to stimulate hyperplasia. In epidemiology studies, exposures typically consist of mixtures of co-pollutants, some of which are known carcinogens (see Section 5.4.3).

5.3 SYSTEMIC EFFECTS OF OZONE EXPOSURE

Ozone indirectly affects organs beyond the respiratory system due to O_3 reaction products entering the bloodstream and being transported to target sites. Extra-pulmonary effects could also be due to the exposure-related production of mediators, metabolic products and cell trafficking. Although systemic effects are of interest and indicate a very broad array of O_3 effects, they are of limited influence and difficult to interpret. By protecting from respiratory tract effects, these systemic effects will likely be protected against also. Systemic effects are only summarized briefly here and in Table AX5-14.

5.3.1 Neurobehavioral Effects

Animal behavior, both motor activity and operant behavior, has been shown to be suppressed by acute O_3 exposures (3 to 6 h) of 0.12 ppm. There is a dose dependent decrease in activity with increasing exposure levels. Additionally, these lowered activity levels tend to attenuate with longer exposure periods. New studies in adult laboratory animals confirm that environmentally relevant O_3 concentrations from 0.2 to 1.0 ppm can decrease motor activity and affect short- and long-term memory, as tested by passive avoidance conditioning in 4 h

exposures in rats (Rivas-Arancibia et al., 1998; Avila-Costa et al., 1999; Dorado-Martinez et al., 2001), or water-maze learning tasks in mice following a 30-day exposure (Sorace et al., 2001). The effects have been attributed to reactive oxygen/nitrogen species and/or ozonation products. The memory deficits could be blocked by administration of vitamin E (Guerrero et al, 1999) or taurine (Rivas-Arancibia et al., 2000). Increased freezing and decreased exploratory behaviors were accompanied by decreased serotonin levels and increased levels of NO, glutamate, dopamine and striatal lipoperoxidation in rats exposed to 1 ppm of O_3 for 4 h (Rivas-Arancibia et al., 2003). The O_3-exposed animals also demonstrated neuronal cytoplasm and dendrite vacuolation and dilation of rough endoplasmic reticulium cisterns, which the authors interpret as a neurodegenerative process resulting from the oxidative stress of acute O_3 exposure. Niño-Cabrera et al. (2002) demonstrated that a 0.7 ppm O_3 exposure for 4 h can induce ultrastructural alterations in the hippocampus and prefrontal cortex in aged rats. These are areas of the brain where degenerative age-related changes in learning and memory functions have been reported (Bimonte et al., 2003).

Paz (1997) reviewed a series of studies that demonstrated significant alterations of electroencephalographic (EEG) patterns during sleep in animals acutely exposed to O_3 (0.35 to 1.0 ppm). Rats and cats both showed loss of paradoxical sleep time after 2 to 8 h of O_3 exposure (Paz and Bazan-Perkins, 1992; Paz and Huitrón-Reséndiz, 1996). Increased total wakefulness, alterations in circadian rhythm, and a permanent 50% loss of paradoxical sleep time were shown in rat pups born to dams exposed to 1.0 ppm O_3 during gestation (Haro and Paz, 1993). Effects on sleep patterns were associated with alterations in brain neurotransmitter levels (Huitrón-Reséndiz et al., 1994; González-Piña and Paz, 1997) thought to be caused by O_3 reaction products or prostaglandins (Koyama and Hayaishi, 1994). The permanent effects in pups caused by high O_3 exposure during gestation were attributed to the diminished antioxidant capability of fetal tissue (Günther et al., 1993).

High, nonambient levels of O_3 (e.g., >1.0 ppm) affect visual and olfactory neural pathways in the rat. For example, Custodio-Ramierez and Paz (1997) reported a significant delay in visual evoked potentials recorded in the visual cortex and the lateral geniculate nucleus of male Wistar rats acutely exposed to high levels of O_3 (1.5, and 3.0 ppm for 4 h). Colin-Barenque et al. (1999), using the same strain, reported cytological and ultrastructural changes in the granule layer of the olfactory bulb after a 4-h exposure to 1 to 1.5 ppm O_3. Although these neural effects

are thought to be caused by O_3 reaction products, especially free radicals, the studies do not add much to an understanding of the underlying mechanisms.

5.3.2 Neuroendocrine Effects

Early studies suggested an interaction of O_3 with the pituitary-thyroid-adrenal axis because thyroidectomy, hypophysectomy, and adrenalectomy protected against the lethal effects of high concentrations of O_3. Concentrations of 0.7 to 1.0 ppm O_3 for a 1 day exposure in male rats caused changes in the parathyroid; thymic atrophy; decreased serum levels of thyroid stimulating hormone, triiodothyronine (T_3), thyroxine (T_4), free T_4, and protein binding; and increased prolactin. In more recent studies, increased toxicity to O_3 was reported in hyperthyroid rats by Huffman et al. (2001) and T_3 supplementation was shown to increase metabolic rate and pulmonary injury in the lungs of O_3-treated animals (Sen et al., 1993).

The mechanisms by which O_3 affects neuroendocrine function are not well understood. Cottet-Emard et al. (1997) examined catecholamine activity in rat sympathetic efferents and brain areas of prime importance to adaptation to environmental stressors. Exposures of 0.5 ppm O_3 for 5 days caused inhibition of norepinephrine turnover in heart (−48% of the control level) but not in lungs and failed to modify the tyrosine hydroxylase activity in superior cervical ganglia and the catecholamine content in the adrenal glands. In the CNS, O_3 inhibited tyrosine hydroxylase activity in noradrenergic brainstem cell groups and decreased catecholamine turnover in the cortex (−49%) and striatum (−18%) but not in the hypothalamus. This suggests that high ambient levels of O_3 can produce marked neural disturbances in structures involved in the integration of chemosensory inputs, arousal, and motor control, effects that may be responsible for some of the behavioral effects seen with O_3 exposure.

5.3.3 Cardiovascular Effects

Studies of the effects on hematological parameters and blood chemistry in rats have shown that erythrocytes are a target of O_3. Exposures to 1.0 ppm O_3 for 3 h have been found to decrease heart rate (HR), mean arterial pressure (MAP), and core temperature (T_{co}) and to induce arrhythmias with some exposures in rats. These effects are more pronounced in adult and awake rats than in younger or sleeping animals. Exposures of 0.2 ppm for 48 h have been shown to

cause bradycardia, while exposures of 0.1 ppm for 3 days have been shown to cause bradyarrhythmia in these animals.

A more recent study of rats exposed to FA for 6 h, followed 2 days later by a 5 h exposure to 0.1 ppm O_3, 5 days later by a 5 h exposure to 0.3 ppm O_3, and 10 days later by a 5 h exposure to 0.5 ppm O_3 used the head-out plethysmograph for continuous measurements (Arito et al., 1997). Each of the O_3 exposures was preceded by a 1 h exposure to FA. Transient rapid shallow breathing with slightly increased HR appeared 1-2 min after the start of O_3 exposures and was attributed to an olfactory response. Persistent rapid shallow breathing with a progressive decrease in HR occurred with a latent period of 12 h. During the last 90-min of exposure, averaged values for relative minute ventilation tended to decrease with the increase in O_3 concentration for young (4-6 mo) but not old (20-22 mo) rats.

Studies utilizing radiotelemetry transmitters in unanesthetized and unrestrained rats, Watkinson et al. (1995; 2001) and Highfill and Watkinson (1996) demonstrated that when HR was reduced during a 5 day 0.5 ppm O_3 exposure, the T_{co} and activity levels also decreased. The decreases in T_{co} and blood pressure reported by in these studies and by Arito et al. (1997) suggest that the changes in ventilation and HR are mediated through physiological and behavioral defense mechanisms in an attempt to minimize the irritant effects of O_3 inhalation. Decreased activity was previously reported in laboratory animals during exposure to O_3 (see above).

Similar cardiovascular and thermoregulatory responses in rats to O_3 were reported by Iwasaki et al. (1998). Repeated exposure to 0.1, 0.3, and 0.5 ppm O_3 8 h/day for 4 consecutive days caused disruption of circadian rhythms of HR and T_{co} on the first and second exposure days that was concentration-dependent. The decreased HR and T_{co} recovered to control values on the third and fourth days of O_3 exposure.

The thermoregulatory response to O_3 was further characterized by Watkinson et al. (2003). Male Fischer-344 rats were exposed to 0.0 ppm for 24 h/day (air), 0.5 ppm for 6 h/day (intermittent) or 0.5 ppm for 23 h/day (continuous) at 3 temperatures, 10 °C (cold), 22 °C (room), or 34 °C (warm). Another protocol examined the effects of O_3 exposure (0.5 ppm) and exercise described as rest, moderate, heavy or CO_2-stimulated ventilation. Both intermittent and continuous O_3 exposure caused decreases in HR and T_{co} and increases in BALF inflammatory markers. Exercise in FA caused increases in HR and T_{co} while exercise in O_3 caused decreases

in those parameters. Carbon dioxide and O_3 induced the greatest deficits in HR and T_{co}. Several factors were suggested that may modulate the hypothermic response, including dose, animal mass, and environmental stress.

Laboratory animals exposed to relatively high O_3 concentrations ($\geq$0.5 ppm) demonstrate tissue edema in the heart and lungs. This may be due to increased circulating levels of atrial natriuretic factor (ANF), which is known to mediate capillary permeability, vasodilation, and blood pressure (Daly et al., 2002). Increased levels of ANF were reported in the heart, lungs, and circulation of rats exposed to 0.5 ppm O_3 for 8 h (Vesely et al., 1994a,b,c).

Earlier work demonstrated O_3-induced release of functionally active PAF from rodent epithelial cells and the presence of PAF receptors on AMs. New work examining lipid metabolism (Section 5.2.1.4) and mediators of inflammatory response and injury (Section 5.2.3.4) confirm these earlier studies that PAF (Kafoury et al., 1999) and PAF receptors (Longphre et al., 1999) are involved in responses to O_3. In addition to the role of PAF in pulmonary inflammation and hyperpermeability, this potent inflammatory mediator may have clotting and thrombolytic effects, though this has not been demonstrated experimentally (see Figure 5-2). This cardiovascular effect may explain, in part, epidemiologic findings of heart attack and stroke (see Chapter 7). The findings of Pulfer and Murphy (2004); Pulfer et al., (2005); Section 5.2.1.4), describing the in vitro and in vivo production of two biologically active oxysterols, are also suggestive of a mechanism whereby O_3 exposure may be implicated in the increased risk of cardiopulmonary disease.

5.3.4 Reproductive and Developmental Effects

Early studies of pre- and postnatal exposure to O_3 were performed at relatively high concentrations. Teratogenic effects were not observed with intermittent exposures of 0.44 to 1.97 ppm O_3 during any part of gestation. Continuous exposure during mid-gestation increased the resorption of embryos while exposures during late gestation delayed some behavioral developments (e.g., righting, eye opening). There were no effects on neonatal mortality up to 1.5 ppm O_3, whereas some transient effects on weight gain were observed at exposures of 0.6 ppm O_3.

More recent studies tend to confirm previous conclusions that prenatal exposures to O_3 concentrations <1.0 ppm do not cause major or widespread somatic or neurobehavioral effects in

the offspring of laboratory animals. These studies generally add some weight toward a negative interpretation of the importance of contributions of low, ambient O_3 to lower birth weights and gross development defects reported in neonates born to women exposed to typical ambient pollution (e.g., Renner, 2002; Chen et al., 2002; Ritz and Yu, 1999). Some postnatal O_3 exposure studies continue to find a few, subtle or borderline somatic and behavioral deficits that will require further research to better assess potential risk to developing humans.

Studies of somatic and neurobehavioral development in female CD-1 mice exposed during pregnancy (days 7 to 17) to O_3 (0, 0.4, 0.8, or 1.2 ppm) failed to show any O_3 effects on reproductive or behavioral performance (Bignami et al., 1994). The study did find significant decreases in body weight gain and delayed eye opening in pups in the 1.2 ppm exposure group. The lack of effect on behavioral performance contrasts with earlier findings, which may be due to the use of different species, differing exposure durations, cross-fostering used in the latter study, different species, and exposure durations during pregnancy. A second study using CD-1 mice exposed in utero from conception through day 17 of pregnancy to 0, 0.2, 0.4, and 0.6 ppm O_3 found no significant deficits in reproductive performance, postnatal somatic and neurobehavioral development, or adult motor activity (Petruzzi et al., 1995). A third study by the same group (Petruzzi et al., 1999), using O_3 exposures (0.3, 0.6, or 0.9 ppm) which continued postnatally until weaning, showed subtle changes in handedness and morphine reactivity. Exposures to 0.6 ppm O_3 caused a reduced preference for the right paw in adulthood. Exposures to 0.9 ppm O_3 altered hot plate avoidance after i.p. treatment with morphine in adulthood.

CD-1 mice exposed to 0.6 ppm O_3 from birth through weaning demonstrated no impairment of navigational performance during acquisition and only subtle changes during reversal (Dell'Omo et al., 1995a). Additionally, there were no O_3-induced effects on reproductive performance, but offspring showed a significant reduction in body weight. Effects on neurobehavioral development with this exposure were minor, with some attenuation of activity responses and impairment of passive avoidance acquisition (Dell'Omo et al. (1995b). The offspring of CD-1 mice continuously exposed from 30 days prior to the formation of breeding pairs until PND 17 to 0.0, 0.3, or 0.6 ppm O_3 showed only small and selective effects on somatic and sensorimotor development (Sorace et al., 2001).

Morphological changes were found in the anterior cerebellar lobe of rat pups born to dams exposed during the entire gestation period to very high (1.0 ppm) O_3 concentrations for 12 h/day.

(Rivas-Manzano and Paz, 1999). Additionally, the dams displayed significantly fewer implantations, increased rate of reabsorptions, a high incidence of spontaneous abortion, and offspring with low birth weight, as noted by previous investigators.

5.3.5 Effects on the Liver, Spleen, and Thymus

Early investigations of the effects of O_3 on liver centered on xenobiotic metabolism, and the prolongation of sleeping time, which was observed at 0.1 ppm O_3. In some species, only adults and especially females were affected. In rats, high (1.0 to 2.0 ppm for 3 h) acute O_3 exposures caused increased production of NO by hepatocytes and enhanced protein synthesis (Laskin et al., 1994; 1996). The O_3-associated effects shown in the liver are thought to be mediated by inflammatory cytokines or other cytotoxic mediators released by activated macrophages in the lungs (Vincent et al., 1996; Laskin et al., 1998; Laskin and Laskin, 2001). Except for the earlier work on xenobiotic metabolism, the responses occurred only after very high acute O_3 exposures.

Examinations of the effects of O_3 on spleen and thymus have shown that O_3 primarily affects T-cell mediated systemic immunity. As with the O_3-associated effects shown in the liver, most of the statistically significant changes occurred after acute exposures to very high O_3 concentrations and relate to systemic oxidative stress. Using more relevant ambient urban O_3 exposure patterns, effects were not found on systemic immune function of rats.

5.3.6 Effects on Cutaneous and Ocular Tissues

Ozone exposure not only affects various organ systems, when inhaled, but also has direct effects on the exposed skin and eyes. The outermost layer of the skin (stratum corneum; SC) may be oxidized, which can lead to compromise of the skin barrier and an epidermal proinflammatory response (Weber et al., 2001; Thiele, 2001). These effects are found only at very high concentrations (>1-5 ppm) and have not been shown at more relevant ambient levels of exposure. The skin possesses a well-developed defense system against oxidative stress, utilizing nonenzymatic (e.g., vitamin C and E, glutathione, uric acid, α-tocopherol) and enzymatic (e.g., superoxide dismutase, catalase, glutathione reductase and peroxidase) antioxidants (Cross et al., 1998). Ocular tissues have similar antioxidant protective function as the skin but are not as well studied (Mucke, 1996; Rose et al., 1998). Effects of ground-level

smog on the eyes have been reported but generally are attributed to related photochemical oxidants like peroxyacetyl nitrate (Vyskocil et al., 1998) or possibly to atmospheric O_3 precursors or reaction products like aldehydes. As in other tissues, O_3 may have disparate high-dose and low-dose mechanisms of effect on skin and eyes, so results must be interpreted in this light.

Hairless mice (SKH-1) exposed to O_3 (0.8 to 10 ppm for 2 h) were used to demonstrate that O_3 depletes the low molecular weight antioxidants (e.g., α-tocopherol, vitamin C, glutathione, uric acid) in the SC at ≥1.0 ppm and causes increased MDA at ≥5 ppm (Weber et al, 1999, 2000, 2001). Valacchi et al. (2000) demonstrated that preexposure to 0.5 O_3 for 2 h followed by low-dose ultraviolet (UV) radiation (0.33 MED) caused depletion of α-tocopherol. This suggests that combined low doses of UV radiation and near-ambient levels of O_3 may cause oxidative stress on the SC. Prolonged exposure to 0.8 ppm O_2 for 6 h also induces cellular stress responses that included the formation of HNE protein adducts, HSP27, and heme-oxygenase-1 in the deeper cellular layers of the skin that continued for up to 18 h after O_3 exposure, followed by repair processes (Valacchi et al., 2003).

The importance of O_3 and UV-induced cellular protein oxidation found in murine skin models to possibly similar environmentally-induced changes in human SC keratins was identified by Thiele et al. (1998, 1999) and Thiele (2001). Using the presence of carbonyl groups in proteins as a marker of reactive oxygen mediated protein oxidation, they reported higher carbonyl levels in the upper SC from the tanned skin of humans and in the skin of healthy human volunteers exposed to model chemical oxidants (e.g., hypochlorite, benzoyl peroxide) that were inversely correlated with vitamin E levels. The environmentally-induced oxidative damage identified in human SC represents an early pathophysiological stage in the development of barrier disruption and inflammation, and possibly has implications for the process of desquamation. The relevance of potentiation of environmental oxidative stress by O_3 exposure of human skin needs further study.

5.3.7 Summary and Conclusions - Systemic Effects of Ozone

Neurobehavioral effects of O_3 at concentrations of 0.2 to 1.0 ppm include decreased motor activity, short- and long-term memory deficits, increased freezing behavior, and decreased exploratory behaviors. These effects have been associated with reactive oxygen/nitrogen

species, ozonation products, altered neurotransmitter levels, morphological changes in several brain regions, and altered EEG patterns during sleep. Neuroendocrine effects of O_3 include morphological and hormonal changes in the pituitary-thyroid-adrenal axis at concentrations of ~0.75 ppm and alterations of visual and olfactory neural pathways at concentrations >1 ppm. Mechanisms underlying these effects are not understood at this time. Cardiovascular effects of O_3 at concentrations of 0.3 to 0.5 ppm include decreased HR, T_{CO}, and BP, which have been termed a hypothermic response. Concentrations of O_3 $\geq$0.5 ppm cause tissue edema (possibly mediated by ANF).

Prenatal exposures to O_3 concentrations <1.0 ppm did not cause noticeable somatic or neurobehavioral effects in offspring, while concentrations of 1.0 to 1.5 ppm caused varying effects on neonatal mortality. Some studies have shown an effect of O_3 on liver xenobiotic enzymes at concentrations as low as 0.1 ppm, while other studies have shown no alterations in metabolic enzymes at even 1 ppm, with the effects appearing to be highly-species specific. Effects on spleen and thymus appear to only occur at high O_3 concentrations (>1.0 ppm), while relevant ambient, urban exposures have no effect on systemic immune function in rats. Effects of O_3 on cutaneous and ocular tissue are only seen at high, nonrelevant concentrations.

5.4 INTERACTIONS OF OZONE WITH OTHER CO-OCCURRING POLLUTANTS

Ozone is part of a complex mixture of air pollutants with a composition and pattern that varies geographically and temporally (by hour of the day, day of the week, and season). Health effects caused by the complex mixture are undoubtedly different (either subtly or significantly) from the additive effects of a few of the hundreds of compounds present. The only disciplinary approach that can evaluate a "real-world" complex mixture is epidemiology (Chapter 7). However, because of the difficulty in evaluation of causative factors and quantitative relationships in epidemiology studies, it is useful to consider animal toxicological studies of mixtures. Such studies can be divided into three categories: (1) ambient air mixtures, (2) laboratory-generated complex mixtures (e.g., gasoline combustion mixtures having ultraviolet-irradiation, other reaction mixtures with O_3 and several other components), and (3) binary mixtures. In most cases, experimental designs in the first two classes did not have

an O_3-only group, making it difficult to impossible to discern the influence of O_3. The more recent mixture studies that are discussed here typically have been with NO_2, sulfuric acid (H_2SO_4), or ammonium sulfate ($[NH_4]_2SO_4$).

Interpreting the mixture studies in terms of real-world risk is difficult because laboratory exposure patterns do not always represent real-world exposure patterns. For example, in the real world, nitrogen dioxide (NO_2) often peaks before O_3 peaks, with a mixture occurring between the peaks, but most laboratory exposures used mixtures only. Also, most studies of O_3 and NO_2 mixtures used ambient levels of O_3 and levels of NO_2 high above ambient. As shall be seen, all interaction possibilities have occurred, depending upon the composition of the mixture, the endpoint examined, and the exposure regimen. In some cases, no interaction was found. Most often, additivity (the effects of the mixture are equal to the sum of the effects of the individual components) or synergism (the effects of the mixture are greater than the sum of the effects of the individual components) was observed. Antagonism (the effects of the mixture are less than the sum of the individual components) was rarely found.

5.4.1 Ozone and Nitrogen Oxides

The most commonly studied copollutant in binary mixtures with O_3 is NO_2. Both early work and more recent studies indicate that, although interaction may occur between these two pollutants, in general, O_3 often masked the effects of the NO_2 or accounted for most of the response, due to the greater toxicity of O_3. Very generally, additivity occurred after acute exposure and synergism occurred with prolonged exposure. Interpreting the mixture studies is challenging because laboratory exposure patterns rarely simulate real-world exposure patterns. In the case of NO_2 and O_3, NO_2 typically peaks before O_3, with a mixture occurring between the peaks, but most laboratory exposures used mixtures only. Also, most studies of O_3 and NO_2 mixtures used ambient levels of O_3 and levels of NO_2 high above ambient. Table AX5-15 lists more recent studies evaluating coexposures to NO_2 and O_3.

Chronic exposures of rats to O_3 (0.8 ppm) and NO_2 (14.4 ppm) for 6 h/day caused development of respiratory insufficiency and severe weight loss. Half of these animals died after 55 to 78 days of exposure due to severe fibrosis (Farman et al., 1997). Increased total lung collagen and elastin were observed, with loss of mature collagen, suggesting breakdown and remodeling of the lung parenchyma. Morphological examination following these coexposures

demonstrates a sequence of events starting with increasing inflammatory and mild fibrotic changes for the first 3 weeks of exposure, stabilized or even reduced changes after 4 to 6 weeks, and severe increases over 7 to 9 weeks of exposure (Farman et al., 1999). This suggests that repair processes occurring during the middle 4 to 6 weeks of exposure become overwhelmed, leading to progressive fibrosis after 7 to 8 weeks of exposure. When the coexposure was extended for 90 days, lesions were noted far into the acinus, but the extent of tissue involvement was the same after 7, 78, and 90 days of exposure. At the end of exposure, high levels of procollagen types I and III mRNA were observed within central acini in the lungs from the combined exposure group but not in lungs from the rats exposed to O_3 or NO_2 alone.

Sprague-Dawley rats exposed to 0.3 ppm O_3 and the combined exposure of O_3 and 1.2 ppm NO_2 for 3 d demonstrated significant DNA single-strand breaks in AMs (Bermúdez et al., 1999). No changes were caused by NO_2-only exposure. The same exposures stimulated the activity of polyADPR synthetase, suggesting a response to lung cellular DNA repair caused by oxidant-induced lung injury (Bermúdez, 2001). The laboratory animal model of progressive pulmonary fibrosis, utilizing long-term, combined O_3 (0.4 to 0.8 ppm) and high-level NO_2 (7 to 14 ppm) exposure, causes an initial acute pulmonary inflammation, followed by adaptation and repair, and eventually causing pulmonary fibrosis after 6 to 13 weeks of exposure (Ishii et al., 2000a; Weller et al., 2000). Unfortunately, this model is not very useful for understanding potential interactive effects of ambient concentrations of O_3 and NO_2.

5.4.2 Ozone and Other Copollutants

Ozone and Formaldehyde

Early studies with combined exposures to O_3 and formaldehyde (HCHO) found evidence of both synergistic and non-interactive effects. Newer work listed in Table AX5-16 includes studies of biochemical and histopathological endpoints in rats exposed to 0.4 ppm O_3 and 3.6 ppm HCHO, alone and combined, for 8 h/day for 3 days (Cassee and Feron, 1994). They demonstrated no interactive effects in the nasal respiratory epithelium, despite the high levels of HCHO when compared to typical ambient levels of 1 to 10 ppb (e.g., Rehle et al., 2001). Mautz (2003) studied changes in breathing pattern and epithelial cell proliferation using exposures of 0.6 ppm O_3 and 10 ppm HCHO alone and in combination for 3 h with exercise at two times resting ventilation. Even with exercise, HCHO does not substantially penetrate to the lower

respiratory tract to interact with O_3 and does not alter breathing patterns to modify local O_3 dose. Parenchymal injury was, therefore, due to O_3 alone. In the nasal transitional epithelium and in the trachea, however, combined exposure produced additive effects due to the increased volume of toxicants during exercise. No other combined pollutant studies have been published in the peer-reviewed literature, although two studies compared the respiratory effects of O_3 to HCHO. Nielsen et al., (1999) compared upper airway sensory irritation caused by HCHO concentrations up to 4 ppm to the lower airway irritation caused by O_3. Using BALB/c mice, they continuously measured f_B, V_T, expiratory flow, T_i, T_e, and respiratory patterns during acute, 30-min exposures. They reported a no effect level of 0.3 ppm for HCHO and 1.0 ppm for O_3.

Thus, O_3 and HCHO do not appear to have additive effects, except during exercise, and that is due to increased volume of gas reaching the tissue. Any possible synergism occurs in the nasal epithelium. HCHO exerts its effects primarily in the upper respiratory tract, whereas the primary site of acute cell injury from O_3 occurs in the conducting airways. EPA is currently completing a toxicological and epidemiological review and risk characterization for formaldehyde.

Ozone and Tobacco Smoke

Early studies of combined exposures of O_3 (1 ppm) and tobacco smoke demonstrated altered airway responsiveness to inhaled bronchoconstrictor challenge and tracheal vascular permeability in guinea pigs. Table AX5-17 lists studies completed since the 1996 AQCD evaluating coexposures of tobacco smoke and O_3.

Wu et al. (1997) reported that inhalation of cigarette smoke evokes a transient bronchoconstrictive effect in anesthetized guinea pigs. Total pulmonary resistance (R_L) and dynamic lung compliance (C_{dyn}) were compared before and after acute exposure to 1.5 ppm O_3 for 1 h. Cigarette smoke alone (7 ml) at a low concentration (33%) induced a mild and reproducible bronchoconstriction that slowly developed and reached its peak after a delay of >1 min. After O_3 exposure, the same cigarette smoke inhalation challenge evoked an intense bronchoconstriction that occurred more rapidly, reaching its peak within 20 s, and was sustained for >2 min. Pretreatment with selective antagonists of neurokinin type 1 and 2 receptors completely blocked the enhanced airway responsiveness suggesting that O_3 exposure induced

AHR to inhaled cigarette smoke, which resulted primarily from the bronchoconstrictive effect of endogenous tachykinins.

The above studies were conducted with undiluted tobacco smoke and high O_3 concentrations. To determine the effects of aged and diluted sidestream cigarette smoke (ADSS) as a surrogate of environmental tobacco smoke (ETS) on O_3-induced lung injury, Yu et al. (2002) exposed male B6C3F1 mice to (1) FA, (2) ADSS, (3) O_3, or (4) ADSS followed by O_3 (ADSS/O_3). Exposure to 30 mg/m^3 ADSS, 6 h/day for 3 days, followed by exposure to 0.5 ppm O_3 for 24 h was associated with a significant increase in the number of cells recovered by BAL compared with exposure to ADSS alone or O_3 alone. Neutrophils, lymphocytes, and total protein levels in BAL were increased following the combined exposure when compared with all other groups. Within the CAR, the percentage of proliferating cells was unchanged from control following exposure to ADSS alone but was significantly elevated following exposure to O_3 and further augmented in a statistically significant manner in mice exposed to ADSS/O_3. Following exposure to O_3 alone or ADSS/O_3, the ability of AMs to release IL-6 under LPS stimulation was significantly decreased, while exposure to ADSS alone or ADSS/O_3 caused a significantly increased release of TNFα from AMs under LPS stimulation. These data suggest that ADSS exposure enhances the sensitivity of animals to O_3-induced lung injury.

Acute exposure to ETS also may make a healthy person more susceptible to sequential O_3 exposure by affecting lung barrier function or the underlying epithelium. Toxicological studies with components of ETS (e.g., nicotine receptor agonists, acrolein, and oxidants) have shown that the vagal bronchopulmonary C-fibers are stimulated by acute exposures that initiate both central and local responses (Bonham et al., 2001; Mutoh et al., 2000). The central responses (e.g., tachypnea, cough, bronchoconstriction, increased mucous secretion) are more protective of the lungs; however, local responses may include increased sensitization of the C-fibers to other irritants, including O_3. Active tobacco smokers should not be similarly affected because they already have significant chronic airway inflammation and increased mucus production. In fact, chronic smokers appear to have diminished lung function responses to O_3 (see Chapter 6).

5.4.3 Complex (Multicomponent) Mixtures Containing Ozone

Ambient pollution in most areas is a complex mix of more than two chemicals. A number of new studies have examined the effects of exposure to multicomponent atmospheres

containing O_3. Some of these studies attempted to simulate photochemical reaction products occurring under actual atmospheric conditions. However, the results of these studies are often difficult to interpret because of chemical interactions between the components, as well as the resultant production of variable amounts of numerous secondary reaction products, and a lack of precise control over the ultimate composition of the exposure environment. In addition, the role of O_3 in the observed biological responses is often obscure. Prior studies using irradiated automobile exhaust mixtures containing total oxidant concentrations (expressed as O_3) in the range of 0.2 to 1.0 ppm have demonstrated pulmonary function changes in several species.

A more recent attempt has been made to examine multicomponent mixtures resulting from the reaction of O_3 with unsaturated hydrocarbons [e.g., isoprene (C_5H_8) and terpene ($C_{10}H_{16}$)], producing HCHO, formic acid, acetone, acrolein, acetic acid, and other oxidation products, many of which are strong airway irritants. Wilkins et al. (2001) evaluated sensory irritation by measuring mean f_B in the mouse bioassay and found a 50% reduction after 30 min of exposure to reaction products of O_3 and isoprene. The mixture at this time period contained <0.2 ppm O_3, so the authors attributed the observed effects to the oxidation products. Clausen et al. (2001), using the same mouse model, evaluated the reaction products of O_3 and limonene. A 33% reduction in mean f_B was produced after 30 min of exposure to the mixture containing <0.3 ppm O_3, again implicating the effects of strong irritant products. Further work needs to be done with these complex reaction mixtures because of their potential impact on the respiratory tract. The results would be particularly important, however, to the reaction of O_3 indoors (see Chapter 3).

Pollutant mixtures containing acid aerosols comprise another type of commonly examined exposure atmosphere (studies summarized in Table AX5-18). Earlier studies that employed simultaneous single, repeated, or continuous exposures of various animal species to mixtures of acid sulfates and O_3 found responses for several endpoints, including tracheobronchial mucociliary clearance, alveolar clearance, pulmonary mechanics, and lung morphology, to be due solely to O_3. Some synergism was noted for bacterial infectivity, response to antigen, and effects on lung protein content and the rate of collagen synthesis.

More recent studies found some differences in airway responses to inhaled acid particle-O_3 mixtures that may have been partly due to airway dosimetry. Various physical and chemical mechanisms may be responsible (see Schlesinger, 1995). For example, physical adsorption or absorption of O_3 or its reaction products on a particle could result in transport to more sensitive

sites, or to sites where O_3, by itself, would not normally be reactive (Madden et al., 2000). Chemical reactions on the surface of particles can form secondary products that are more toxicologically active, or chemical characteristics of the particle may change the residence time or reactivity of oxidation products at the site of deposition. The hypothesis that synergism between O_3 and sulfates is due to decreased pH changing the residence time or reactivity of reactants, such as free radicals, was tested by Chen et al. (1995) and El-Fawal et al. (1995). Male New Zealand white rabbits were exposed for 3 h to 125 $\mu g/m^3$ H_2SO_4, 0.1, 0.3, or 0.6 ppm O_3, and to combinations. Chen et al. (1995) demonstrated that decreased pH following exposure to acid aerosol was correlated with phagocytic activity and capacity of harvested macrophages and that exposure to O_3/ H_2SO_4 removed this relationship. El-Fawal et al. (1995) showed that responsiveness of rabbit harvested bronchial rings to ACh was increased following a 3 h O_3 exposure, but that 0.1 to 0.6 ppm O_3/0.5 to 0.125 mg/m^3 H_2SO_4 combinations resulted in antagonism.

As discussed in Section 5.2.2.1, Churg et al. (1996) demonstrated increased uptake of asbestos or TiO_2 in response to 10 min O_3 (up to 1.0 ppm) pre-exposure suggesting that low concentrations of O_3 may increase the penetration of some types of PM into epithelial cells. Using human epithelial cell cultures, Madden et al. (2000) demonstrated a greater potency for ozonized diesel PM to induce prostaglandin E_2 production. This suggests that 0.1 ppm O_3 for 24 h can modify the biological activity of PM derived from diesel exhaust.

Effects of combined exposures of O_3 and resuspended urban particles on cell proliferation in epithelial cells of the terminal bronchioles and the alveolar ducts were examined by Vincent et al. (1997) and Adamson et al. (1999). Rats exposed to 0.8 ppm O_3 in combination with 5 or 50 mg/m^3 particles for 4 h demonstrated greatly potentiated proliferative effects compared to O_3 exposure alone. These findings using resuspended dusts, although at high concentrations, are consistent with the studies demonstrating interaction between H_2SO_4 aerosols and O_3. Effects of acute coexposure to 0.6 ppm O_3 and fine or ultrafine H_2SO_4 (0.5 to 0.3 mg/m^3) aerosols on lung morphology were examined by Kimmel et al. (1997). They demonstrated that alveolar septal volume was increased in animals co-exposed to O_3 and ultrafine, but not fine, H_2SO_4. Interestingly, cell proliferation was increased only in animals co-exposed to fine H_2SO_4 and O_3, as compared to animals exposed to O_3 alone. Subchronic exposure to acid aerosols (20 to 150 $\mu g/m^3$ H_2SO_4) had no interactive effect on the biochemical and morphometric changes

produced by either intermittent or continuous exposure to 0.12 to 0.2 ppm O_3 for up to 90 days, which suggests that the interactive effects of O_3 and acid aerosol coexposure in the lung disappeared during the long-term exposure (Last and Pinkerton, 1997). Sindhu et al. (1998) observed an increase in rat lung putrescine levels after repeated, combined exposures to O_3 and a nitric acid vapor for 40 weeks.

Other studies have examined interactions between carbon particles and O_3. The interactions of intratracheally instilled carbon particles followed by either a 7-day or 60-day exposure to 0.5 ppm O_3 in rats was evaluated by Creutzenberg et al. (1995). The carbon particles caused diminished phagocytotic capacity and chemotactic migration capability of AMs that was stimulated by the subsequent O_3 exposure. Inflammatory responses following exposures to low- and high-concentration mixtures of O_3 and acidic aerosols (0.2 ppm O_3 + 50 $\mu g/m^3$ carbon + 100 $\mu g/m^3$ H_2SO_4; 0.4 ppm O_3 + 250 $\mu g/m^3$ carbon + 500 $\mu g/m^3$ H_2SO_4, respectively) for 1 or 5 days was examined by Kleinman et al. (1999). The response with the O_3-particle mixture was greater after 5 days (4 h/day) than after day 1. This contrasted with O_3 exposure alone (0.4 ppm), which caused marked inflammation on acute exposure, but no inflammation after 5 consecutive days of exposure.

The effects of a mixture of elemental carbon particles, 0.2 ppm O_3, and 0.5 mg/m^3 ammonium bisulfate on rat lung collagen content and macrophage activity was examined by Kleinman et al. (2000). Decreases in lung collagen, and increases in macrophage respiratory burst and phagocytosis were observed relative to other pollutant combinations. Mautz et al. (2001) used a similar mixture (i.e., elemental carbon particles, 0.16 to 0.59 ppm O_3, ammonium bisulfate 0.5 to 0.22 mg/m^3, but with 0.11 to 0.39 ppm NO_2 also) and exposure regimen as Kleinman et al. (2000). Also observed were decreases in pulmonary macrophage Fc-receptor binding and phagocytosis and increases in acid phosphatase staining. Bronchoalveolar epithelial permeability and cell proliferation were increased. Altered breathing-patterns also were observed, with some adaptations occurring.

Bolarin et al. (1997) exposed rats to 50 or 100 $\mu g/m^3$ carbon particles in combination with ammonium bisulfate and 0.2 ppm O_3. Despite 4 weeks of exposure, they observed no changes in protein concentration in lavage fluid or blood prolyl 4-hydroxylase, an enzyme involved in collagen metabolism. Slight decreases in plasma fibronectin were present in animals exposed to the combined pollutants versus O_3 alone. Thus, the potential for adverse effects in the lungs of

animals challenged with a combined exposure to particles and gaseous pollutants is dependent on numerous factors, including the gaseous co-pollutant, concentration, and time.

In a complex series of studies, Oberdörster and colleagues examined the interaction of several pulmonary oxidative stress pollutants. Elder et al. (2000a,b) reported the results of combined exposure to ultrafine carbon particles (100 $\mu g/m^3$) and O_3 (1 ppm for 6 h) in young and old Fischer 344 rats that were pretreated with aerosolized endotoxin. In old rats, exposure to carbon and O_3 produced an interaction that resulted in a greater influx in neutrophils than that produced by either agent alone. This interaction was not seen in young rats. Oxidant release from lavage fluid cells also was assessed and the combination of endotoxin, carbon particles, and O_3 produced an increase in oxidant release in old rats. This mixture produced the opposite response in the cells recovered from the lungs of the young rats, indicating that the lungs of the aged animals underwent greater oxidative stress in response to a complex pollutant mix of particles, O_3, and a biogenic agent. Johnston et al. (2000a; 2002) reported the results of combined exposure to O_3 (1.0 and 2.5 ppm for 4, 20, or 24 h) and low-dose endotoxin, or to O_3 and endotoxin separately, in newborn and adult C57BL/6J mice. In the first study, adult (8 wk old) mice showed greater sensitivity to O_3 than newborn (36 h old) mice on the basis of mRNAs encoding for various chemokines and cytokines. In contrast, adult and newborn mice responded similarly 2 h after endotoxin exposure (10 ng for 10 min), suggesting that age differences in O_3-generated inflammation is secondary to epithelial cell injury. In the second study, 8 wk old mice exposed to O_3 (1 ppm for 24 h) followed by endotoxin (37.5 EU for 10 min) showed increased responsiveness over either exposure alone, on the basis of increased expression of chemokine and cytokine messages and increased BAL fluid levels of protein and PMNs.

Fanucchi et al. (1998) and Wagner et al. (2001a,b) examined the synergistic effect of coexposure to O_3 and endotoxin on the nasal transitional epithelium of rats that also was mediated, in part, by neutrophils. Fisher 344 rats intranasally instilled with endotoxin and exposed to 0.5 ppm O_3, 8 h per day, for 3 days developed mucous cell metaplasia in the nasal transitional epithelium, an area normally devoid of mucous cells; whereas, intratracheal instillation of endotoxin (20 µg) caused mucous cell metaplasia rapidly in the respiratory epithelium of the conducting airways. A synergistic increase of intraepithelial mucosubstances and morphological evidence of mucous cell metaplasia were found in rat maxilloturbinates upon exposure to both O_3 and endotoxin, compared to each pollutant alone. A similar response was

reported in OVA-sensitized Brown Norway rats exposed to 0.5 ppm O_3, 8 h/day for 3 days (Wagner et al., 2002), indicating that coexposure to O_3 and inflammatory biogenic substances like allergens (e.g., OVA) or bacterial endotoxin can augment epithelial and inflammatory responses in rat nasal passages.

In follow-up studies, Wagner et al. (2003) reported that coexposure of rats to O_3 and endotoxin also enhanced epithelial and neutrophilic inflammatory responses in the pulmonary airways. Fisher 344 rats were intranasally instilled with endotoxin and exposed to 1.0 ppm O_3 for 8 h, which was repeated 24 h later. Three days after the last exposure, BALF was analyzed for inflammatory cells and secreted mucosubstances (mucin 5AC), and lung tissue was processed for morphometric analysis. Endotoxin instillation alone caused a dose-dependent increase in BALF neutrophils that was further increased 2-fold in O_3-exposed rats given 20 μg endotoxin, the highest dose. Mucin glycoprotein 5AC also was increased in the BALF at this dose but not at lower endotoxin doses. Ozone exposure alone did not cause mucus hypersecretion, but it did potentiate mucus secretion in rats given both 2 and 20 μg endotoxin and increased intraepithelial mucosubstances 2-fold, which was further substantiated by significant increases in mucin gene (rMuc5AC) mRNA levels in the conducting airways.

The effect of O_3 modifying the biological potency of PM (diesel PM and carbon black) was examined by Madden et al. (2000) in rats. Reaction of NIST Standard Reference Material # 2975 diesel PM with 0.1 ppm O_3 for 48 hr increased the potency (compared to unexposed or air-exposed diesel PM) to induce neutrophil influx, total protein, and LDH in lung lavage fluid in response to intratracheal instillation. Exposure of the diesel PM to high, nonambient O_3 concentration (1.0 ppm) attenuated the increased potency, suggesting destruction of the bioactive reaction products. Unlike the diesel particles, carbon black particles exposed to 0.1 ppm O_3 did not exhibit an increase in biological potency, which suggested that the reaction of organic components of the diesel PM with O_3 were responsible for the increased potency.

Ulrich et al. (2002) investigated the effect of ambient PM from Ottawa Canada (EHC-93) on O_3-induced inflammation. Male Wistar rats were exposed to 0.8 ppm O_3 for 8 h and allowed to recover before intratracheal instillation of 0.5, 1.5, and 5 mg of EHC-93 in 0.3 ml of saline. The high concentrations of PM used were sufficient to induce pulmonary inflammation, which was not exacerbated by pre-exposure to O_3. Rats from the combined exposure group did have

higher and more persistent lung lavage protein and albumin levels, as well as increased plasma fibrinogen levels when compared to PM exposure alone.

The interaction of PM and O_3 was further examined in a murine model of OVA-induced asthma. Kobzik et al. (2001) investigated whether coexposure to inhaled, concentrated ambient particles (CAPs) from Boston, MA and to O_3 could exacerbate asthma-like symptoms. On days 7 and 14 of life, half of the BALB/c mice used in this study were sensitized by intraperitoneal (ip) injection of OVA and then exposed to OVA aerosol on three successive days to create the asthma phenotype. The other half received the ip OVA but were exposed to a phosphate-buffered saline aerosol (controls). The mice were further subdivided ($n \geq 61$/group) and exposed for 5 h to CAPs, ranging from 63 to 1,569 $\mu g/m^3$, 0.3 ppm O_3, CAPs + O_3, or to FA. Pulmonary resistance and airway responsiveness to an aerosolized MCh challenge were measured after exposures. A small, statistically significant increase in pulmonary resistance and airway responsiveness, respectively, was found in both normal and asthmatic mice immediately after exposure to CAPs alone and to CAPs + O_3 but not to O_3 alone or to FA. By 24 h after exposure, the responses returned to baseline levels. There were no significant increases in airway inflammation after any of the pollutant exposures. In this well-designed study of a small-animal model of asthma, O_3 and CAPs did not appear to be synergistic. In further analysis of the data using specific elemental groupings of the CAPs, the acutely increased pulmonary resistance was found to be associated with the AlSi fraction of PM. Thus, some components of concentrated $PM_{2.5}$ may affect airway caliber in sensitized animals, but the results are difficult to extrapolate to people with asthma.

Animal studies have examined the adverse cardiopulmonary effects of complex mixtures in urban and rural environments of Italy (Gulisano et al., 1997), Spain (Lorz and López, 1997), and Mexico (Vanda et al., 1998; Moss et al., 2001). Some of these studies have taken advantage of the differences in pollutant mixtures of urban and rural environments to report primarily morphological changes in the nasopharynx and lower respiratory tract (Gulisano et al., 1997; Lorz and López, 1997) of lambs and pigeons, respectively, after natural, continuous exposures to ambient pollution. Each study has provided evidence that animals living in urban air pollutants have greater pulmonary changes than those that would occur in a rural and presumably cleaner, environment. However, these studies either did not report ambient O_3 levels, or reported only annual means.

The study by Moss et al. (2001) examined the nasal and lung tissue of rats exposed (23 h/day) to Mexico City air for up to 7 weeks and compared them to controls similarly exposed to FA. No inflammatory or epithelial lesions were found using quantitative morphological techniques; however, the concentrations of pollutants were low. Extrapolation of these results to humans is restricted, however, by uncontrolled exposure conditions, small sample sizes, and other unknown exposure and nutritional factors in the studies in mammals and birds, and the negative studies in rodents. They also bring up the issue of which species of "sentinel" animals is more useful for predicting urban pollutant effects in humans. Thus, in these field studies, it is difficult to assign a specific role to any specific component of the mixture for the significant cardiopulmonary effects reported.

Similar morphological changes (Calderón-Garcidueñas et al., 2000a; 2001) and chest X-ray evidence of mild lung hyperinflation (Calderón-Garcidueñas et al., 2000b) have been reported in children residing in urban and rural areas of Mexico City. (See Chapter 7 for details of these studies.) The ambient air in urban areas, particularly in southwestern Mexico City, is a complex mixture of particles and gases, including high concentrations of O_3 and aldehydes that previously have been shown to cause airway inflammation and epithelial lesions in humans (e.g., Calderón-Garcidueñas et al., 1992, 1994, 1996) and laboratory animals (Morgan et al., 1986; Heck et al., 1990; Harkema et al., 1994, 1997a,b). The described effects demonstrate a persistent, ongoing upper and lower airway inflammatory process and chest X-ray abnormalities in children residing predominantly in highly polluted areas. Again, extrapolation of these results to urban populations of the United States is difficult because of the unique complex mixture of urban air in Mexico City, uncontrolled exposure conditions, and other unknown exposure and nutritional factors.

5.4.4 Summary and Conclusions - Interactions of Ozone with other Co-occurring Pollutants

It is difficult to summarize the role that O_3 plays in exposure responses to binary mixtures, and even harder to determine its role in responses to multicomponent, complex atmospheres. Though the specific mechanisms of action of the individual pollutants within a mixture may be known, the exact bases for toxic interactions have not been elucidated clearly. Certain generic mechanisms that may underlie pollutant interactions: (1) physical, involving adsorption of one

pollutant onto another and subsequent transport to more or less sensitive sites or to sites where one of the components of the mixture normally would not deposit in concentrated amounts (probably not involved in O_3-related interactions); (2) production of secondary products that may be more toxicologically active than the primary materials, demonstrated or suggested in a number of studies as a basis for interaction between O_3 and NO_2 and between O_3 and PM; (3) biological or chemical alterations at target sites that affect response to O_3 or the copollutant, which which has been suggested to underlie interactions with mixtures of O_3 and acid sulfates; 4) O_3- or copollutant-induced physiological change, such as alteration in ventilation pattern, resulting in changes in the penetration or deposition of one pollutant when another is present. This has been implicated in enhanced responses to various O_3-containing mixtures with exercise.

Evaluation of interactions between O_3 and copollutants is a complex procedure. Responses are dependent on a number of host and environmental factors, such that different studies using the same copollutants may show different types or magnitudes of interactions. The occurrence and nature of any interaction is dependent on the endpoint being examined and is also highly related to the specific conditions of each study, such as animal species, health status, exposure method, dose, exposure sequence, and the physicochemical characteristics of the copollutants. Because of this, it is difficult to compare studies, even those examining similar endpoints, that were performed under different exposure conditions. Thus, any description of interactions is really valid only for the specific conditions of the study in question and cannot be generalized to all conditions of exposure to a particular chemical mixture. Furthermore, it is generally not possible to extrapolate the effect of pollutant mixtures from studies on the effects of each component when given separately. In any case, what can be concluded from the database is that interactions of O_3-containing mixtures are generally synergistic (antagonism has been noted in a few studies), depending on the various factors noted above, and that O_3 may produce more significant biological responses as a component of a mixture than when inhaled alone. Furthermore, although most studies have shown that interaction occurs only at higher than ambient concentrations with acute exposure, some have demonstrated interaction at more environmentally relevant levels (e.g., 0.05 to 0.1 ppm O_3 with NO_2) and with repeated exposures.

5.5 EFFECTS OF OTHER PHOTOCHEMICAL OXIDANTS

Peroxyacetyl nitrate (PAN) and peroxypropionyl nitrate (PPN) are the most abundant non-O_3 oxidants in ambient air of industrialized areas, other than the inorganic nitrogenous oxidants such as NO_2, and possibly HNO_3. Ambient levels of PAN and PPN were reported to be decreasing over the 1990's and available air quality data (Grosjean et al., 2001; Grosjean, 2003; Jakobi and Fabian, 1997) indicate that present peak concentrations of PAN and PPN in ambient air from urban areas are in the low ppb range (e.g., <1 to 10 ppb). The levels found in nonurban areas are considerably lower (Gaffney et al., 1993).

Reactions occur in the troposphere between O_3 and hydrocarbons (e.g., d-limonene) to produce epoxides, hydroperoxides, and peroxides. The majority of the measured ambient hydroperoxides produced is hydrogen peroxide (H_2O_2), although a small amount of organic hydroperoxides (ROOH) also may be formed. Friedlander and Yeh (1998) have estimated that atmospheric aerosols can carry as high as 1 mM of H_2O_2 and organic hydroperoxides (e.g., hydroxymethylhydroperoxide) in the associated water. In vitro cell and tissue damage are induced by high concentrations of liquid phase H_2O_2 (50 μM to 1 mM). Morio et al. (2001) (see Table AX5-19) demonstrated that a 2 h exposure of 10 and 20 ppb of inhaled H_2O_2 vapor can penetrate the lower lung where it causes inflammation. It is likely that hygroscopic components of PM transport ambient H_2O_2 into the lower lung and induce tissue injury as well. Exposure of rats to a H_2O_2-fine particle mixture (215 or 429 $\mu g/m^3$ ammonium sulfate) resulted in increased neutrophil influx, and production of inflammatory mediators by AMs (Morio et al., 2001). Hygroscopic secondary organic aerosols generated by the O_3/hydrocarbon reactions and their co-occurrence with H_2O_2 also provides another possible mechanism, yet to be validated, whereby H_2O_2 can be transported into the lower respiratory tract (e.g., Friedlander and Yeh, 1998). Interaction of inhaled O_3 with unsaturated fatty acids on cell membranes and mucus in the airways generates epoxides, hydroperoxides, and secondary ozonation products such as 4-hydroxynonenal (see Section 5.2.1)

Inhalation toxicological information on the effects of the non-O_3 oxidants has been limited to a few studies on PAN, but at concentrations much higher (approximately 100- to 1,000 fold) than levels typically found in ambient air. Such high acute levels cause changes in lung morphology, behavioral modifications, weight loss, and susceptibility to pulmonary infections. Therefore, acute toxicity of PAN is much lower than O_3, and it is unlikely that present ambient

PAN levels would affect pulmonary function responses to O_3 (reviewed in Vyskocil et al., 1998). Cytogenetic studies indicate that PAN is not a potent mutagen, clastogen, or DNA damaging agent in mammalian cells in vivo or in vitro at concentrations several orders of magnitude higher than the generally encountered ambient air levels in most cities (Vyskocil et al., 1998; Kligerman et al., 1995; Heddle et al., 1993). Some studies suggest that PAN may be a weak bacterial mutagen at concentrations much higher than exist in present urban atmospheres (DeMarini et al., 2000; Kleindienst et al., 1990).

An additional level of complexity exists due to the possibility that other ambient oxidants may contribute to effects attributed to O_3. As discussed in Chapter 2, both short-lived radicals and secondary particles containing highly polar compounds are generated in the troposphere by the same photochemical mechanisms that produce O_3. It is plausible that, in addition to the direct effects of O_3, health effects are produced by ambient exposures to these gaseous and particulate secondary compounds. Little is known regarding the composition of these reaction products, and little research has been undertaken evaluating their toxicologic effects. Due to the many oxidizing species present in the atmosphere, interpretation of toxicology data based on O_3 exposures alone have the potential for underestimating health effects of ambient oxidant mixtures.

5.5.1 Summary and Conclusions - Effects of Other Photochemical Oxidants

Concentrations of PAN and PPN (<1 to 10 ppb) in ambient air are unlikely to affect pulmonary function or cause DNA damage. Levels of 10-20 ppm H_2O_2 can penetrate to the lower lung directly or be transported there by PM, where inflammation can result; however, ambient H_2O_2 levels of are typically < ~5 ppb. As toxicology studies of other photochemical oxidants are rare, quantitative scientific evaluations of possible health effects of environmental exposures cannot be completed at this time.

REFERENCES

Abbey, D. E.; Nishino, N.; McDonnell, W. F.; Burchette, R. J.; Knutsen, S. F.; Beeson, W. L.; Yang, J. X. (1999) Long-term inhalable particles and other air pollutants related to mortality in nonsmokers. Am. J. Respir. Crit. Care Med. 159: 373-382.

Adamson, I. Y. R.; Vincent, R.; Bjarnason, S. G. (1999) Cell injury and interstitial inflammation in rat lung after inhalation of ozone and urban particulates. Am. J. Respir. Cell Mol. Biol. 20: 1067-1072.

Aizawa, H.; Shigyo, M.; Nakano, H.; Matsumoto, K.; Inoue, H.; Hara, N. (1999a) Effect of the Chinese herbal medicine, Bakumondo-to, on airway hyperresponsiveness induced by ozone exposure in guinea-pigs. Respirology 4: 349-354.

Aizawa, H.; Shigyo, M.; Matsumoto, K.; Inoue, H.; Koto, H.; Hara, N. (1999b) PACAP reverses airway hyperresponsiveness induced by ozone exposure in guinea pigs. Respiration 66: 538-542.

Alfaro, M. F.; Putney, L.; Tarkington, B. K.; Hatch, G. E.; Hyde, D. M.; Schelegle, E. S. (2004) Effect of rapid shallow breathing on the distribution of ^{18}O-labeled ozone reaction product in the respiratory tract of the rat. Inhalation Toxicol.: in press.

Anderson, S. D.; Daviskas, E. (2000) The mechanism of exercise-induced asthma is ... J. Allergy Clin. Immunol. 106: 453-459.

Arito, H.; Takahashi, M.; Iwasaki, T.; Uchiyama, I. (1997) Age-related changes in ventilatory and heart rate responses to acute ozone exposure in the conscious rat. Ind. Health 35: 78-86.

Arsalane, K.; Gosset, P.; Vanhee, D.; Voisin, C.; Hamid, Q.; Tonnel, A.-B.; Wallaert, B. (1995) Ozone stimulates synthesis of inflammatory cytokines by alveolar macrophages *in vitro*. Am. J. Respir. Cell Mol. Biol. 13: 60-68.

Avila-Costa, M. R.; Colín-Barenque, L.; Fortoul, T. I.; Machado-Salas, J. P.; Espinosa-Villanueva, J.; Rugerio-Vargas, C.; Rivas-Arancibia, S. (1999) Memory deterioration in an oxidative stress model and its correlation with cytological changes on rat hippocampus CA1. Neurosci. Lett. 270: 107-109.

Avital, A.; Springer, C.; Bar-Yishay, E.; Godfrey, S. (1995a) Adenosine, methacholine, and exercise challenges in children with asthma or paediatric chronic obstructive pulmonary disease. Thorax 50: 511-516.

Avital, A.; Picard, E.; Uwyyed, K.; Springer, C. (1995b) Comparison of adenosine 5'-monophosphate and methacoline for the differentiation of asthma from chronic airway diseases with the use of the auscultative method in very young children. J. Pediatr. 127: 438-440.

Ballinger, C. A.; Cueto, R.; Squadrito, G.; Coffin, J. F.; Velsor, L. W.; Pryor, W. A.; Postlethwait, E. M. (2005) Antioxidant-mediated augmentation of ozone-induced membrane oxidation. Free Radical Biol. Med. 38: 515-526.

Bassett, D.; Elbon-Copp, C.; Otterbein, S.; Barraclough-Mitchell, H.; DeLorme, M.; Yang, H. (2001) Inflammatory cell availability affects ozone-induced lung damage. J. Toxicol. Environ. Health A 64: 547-565.

Becker, S.; Quay, J.; Koren, H. S. (1991) Effect of ozone on immunoglobulin production by human B cells in vitro. J. Toxicol. Environ. Health 34: 353-366.

Beeson, W. L.; Abbey, D. E.; Knutsen, S. F. (1998) Long-term concentrations of ambient air pollutants and incident lung cancer in California adults: results from the AHSMOG study. Environ. Health Perspect. 106: 813-823.

Bermúdez, E. (2001) Detection of poly(ADP-ribose) synthetase activity in alveolar macrophages of rats exposed to nitrogen dioxide and ozone. Inhalation Toxicol. 13: 69-84.

Bermúdez, E.; Ferng, S. F.; Castro, C. E.; Mustafa, M. G. (1999) DNA strand breaks caused by exposure to ozone and nitrogen dioxide. Environ. Res. 81: 72-80.

Bhalla, D. K. (1996) Alteration of alveolar macrophage chemotaxis, cell adhesion, and cell adhesion molecules following ozone exposure of rats. J. Cell. Physiol. 169: 429-438.

Bhalla, D. K.; Gupta, S. K. (2000) Lung injury, inflammation, and inflammatory stimuli in rats exposed to ozone. J. Toxicol. Environ. Health 59: 211-228.

Bhalla, D. K.; Gupta, S. K.; Reinhart, P. G. (1999) Alteration of epithelial integrity, alkaline phosphatase activity, and fibronectin expression in lungs of rats exposed to ozone. J. Toxicol. Environ. Health A 56: 329-343.

Bhalla, D. K.; Reinhart, P. G.; Bai, C.; Gupta, S. K. (2002) Amelioration of ozone-induced lung injury by anti-tumor necrosis factor-α. Toxicol. Sci. 69: 400-408.

Bignami, G.; Musi, B.; Dell'Omo, G.; Laviola, G.; Alleva, E. (1994) Limited effects of ozone exposure during pregnancy on physical and neurobehavioral development of CD-1 mice. Toxicol. Appl. Pharmacol. 129: 264-271.

Bimonte, H. A.; Nelson, M. E.; Granholm, A. C. (2003) Age-related deficits as working memory load increases: relationships with growth factors. Neurobiol. Aging 24: 37-48.

Bolarin, D. M.; Bhalla, D. K.; Kleinman, M. T. (1997) Effects of repeated exposures of geriatric rats to ozone and particle-containing atmospheres: an analysis of bronchoalveolar lavage and plasma proteins. Inhalation Toxicol. 9: 423-434.

Bonham, A. C.; Chen, C. Y.; Mutoh, T.; Joad, J. P. (2001) Lung C-fiber CNS reflex: role in the respiratory consequences of extended environmental tobacco smoke exposure ini young guinea pigs. Environ. Health Perspect. 109(suppl. 4): 573-578.

Boorman, G. A.; Hailey, R.; Grumbein, S.; Chou, B. J.; Herbert, R. A.; Goehl, T.; Mellick, P. W.; Roycroft, J. H.; Haseman, J. K.; Sills, R. (1994) Toxicology and carcinogenesis studies of ozone and ozone 4-(*N*-nitrosomethylamino)-1-(3-pyridyl)-1-butanone in Fischer-344/N rats. Toxicol. Pathol. 22: 545-554.

Bornholdt, J.; Dybdahl, M.; Vogel, U.; Hansen, M.; Loft, S.; Wallin, H. (2002) Inhalation of ozone induces DNA strand breaks and inflammation in mice. Mutat. Res. 520: 63-71.

Brannan, J. D.; Koskela, H.; Anderson, S. D.; Chew, N. (1998) Responsiveness to Mannitol in asthmatic subjects with exercise- and hyperventiliation-induced asthma. Am. J. Respir. Crit. Care Med. 158: 1120-1126.

Bridges, J. P.; Davis, H. W.; Demodarasamy, M.; Kuroki, Y.; Howles, G.; Hui, D. Y.; McCormack, F. X. (2000) Pulmonary surfactant proteins A and D are potent endogenous inhibitors of lipid peroxidation and oxidative cellular injury. J. Biol. Chem. 275: 38848-38855.

Broeckaert, F.; Clippe, A.; Wattiez, R.; Falmagne, P.; Bernard, A. (2003) Lung hyperpermeability, Clara-cell secretory potein (CC16), and susceptibility to ozone of five inbred strains of mice. Inhalation Toxicol. 15: 1209-1230.

Bush, M. L.; Zhang, W.; Ben-Jebria, A.; Ultman, J. S. (2001) Longitudinal distribution of ozone and chlorine in the human respiratory tract: simulation of nasal and oral breathing with the single-path diffusion model. Toxicol. Appl. Pharmacol. 173: 137-145.

Calderón-Garcidueñas, L.; Osorno-Velaquez, A.; Bravo-Alvarez, H.; Delgado-Chavez, R.; Barrios-Marquez, R. (1992) Histopathologic changes of the nasal mucosa in Southwest metropolitan Mexico City inhabitants. Am. J. Pathol. 140: 225-232.

Calderón-Garcidueñas, L.; Rodriguez-Alcaraz, A.; García, R.; Sanchez, G.; Barragan, G.; Camacho, R.; Ramirez, L. (1994) Human nasal mucosal changes after exposure to urban pollution. Environ. Health Perspect. 102: 1074-1080.

Calderón-Garcidueñas, L.; Osnaya-Brizuela, N.; Ramírez-Martínez, L.; Villarreal-Calderón, A. (1996) DNA strand breaks in human nasal respiratory epithelium are induced upon exposure to urban pollution. Environ. Health Perspect. 104: 160-168.

Calderón-Garcidueñas, L.; Wen-Wang, L.; Zhang, Y.-J.; Rodriguez-Alcaraz, A.; Osnaya, N.; Villarreal-Calderón, A.; Santella, R. M. (1999) 8-hydroxy-2'-deoxyguanosine, a major mutagenic oxidative DNA lesion, and DNA strand breaks in nasal respiratory epithelium of children exposed to urban pollution. Environ. Health Perspect. 107: 469-474.

Calderón-Garcidueñas, L.; Devlin, R. B.; Miller, F. J. (2000a) Respiratory tract pathology and cytokine imbalance in clinically healthy children chronically and sequentially exposed to air pollutants. Med. Hypotheses 55: 373-378.

Calderón-Garcidueñas, L.; Mora-Tiscareño, A.; Chung, C. J.; Valencia, G.; Fordham, L. A.; García, R.; Osnaya, N.; Romero, L.; Acuña, H.; Villarreal-Calderón, A. (2000b) Exposure to air pollution is associated with lung hyperinflation in healthy children and adolescents in southwest Mexico City: a pilot study. Inhalation Toxicol. 12: 537-561.

Calderón-Garcidueñas, L.; Rodrígues-Alcaraz, A.; Valencia-Salazar, G.; Mora-Tascareño, A.; García, R.; Osnaya, N.; Villarreal-Calderón, A.; Devlin, R. B.; Van Dyke, T. L. (2001) Nasal biopsies of children exposed to air pollutants. Toxicol. Pathol. 29: 558-564.

Cassee, F. R.; Feron, V. J. (1994) Biochemical and histopathological changes in nasal epithelium of rats after 3-day intermittent exposure to formaldehyde and ozone alone or in combination. Toxicol. Lett. 72: 257-268.

Chang, M. M.-J.; Wu, R.; Plopper, C. G.; Hyde, D. M. (1998) IL-8 is one of the major chemokines produced by monkey airway epithelium after ozone-induced injury. Am. J. Physiol. 275: L524-L532.

Cheek, J. M.; McDonald, R. J.; Rapalyea, L.; Tarkington, B. K.; Hyde, D. M. (1995) Neutrophils enhance removal of ozone-injured alveolar epithelial cells in vitro. Am. J. Physiol. 269: L527-L535.

Chen, L. C.; Qu, Q.; Amdur, M. O.; Schlesinger, R. B. (1995) Alteration of pulmonary macrophage intracellular pH following inhalation exposure to sulfuric acid/ozone mixtures. Exp. Lung Res. 21: 113-128.

Chen, L.; Yang, W.; Jennison, B. L.; Goodrich, A.; Omaye, S. T. (2002) Air pollution and birth weight in northern Nevada, 1991-1999. Inhalation Toxicol. 14: 141-157.

Chen, C.-Y.; Bonham, A. C.; Plopper, C. G.; Joad, J. P. (2003) Plasticity in respiratory motor control: selected contribution: neuroplasticity in nucleus tractus solitarius neurons following episodic ozone exposure in infant primates. J. Appl. Physiol. 94: 819-827.

Cho, H. Y.; Hotchkiss, J. A.; Harkema, J. R. (1999a) Inflammatory and epithelial responses during the development of ozone-induced mucous cell metaplasia in the nasal epithelium of rats. Toxicol. Sci. 51: 135-145.

Cho, H. Y.; Hotchkiss, J. A.; Bennett, C. B.; Harkema, J. R. (1999b) Effects of pre-existing rhinitis on ozone-induced mucous cell metaplasia in rat nasal epithelium. Toxicol. Appl. Pharmacol. 158: 92-102.

Cho, H. Y.; Hotchkiss, J. A.; Bennett, C. B.; Harkema, J. R. (2000) Neutrophil-dependent and neutrophil-independent alterations in the nasal epithelium of ozone-exposed rats. Am. J. Respir. Crit. Care Med. 162: 629-636.

Cho, H.-Y.; Zhang, L.-Y.; Kleeberger, S. R. (2001) Ozone-induced lung inflammation and hyperreactivity are mediated via tumor necrosis factor-α receptors. Am. J. Physiol. 280: L537-L546.

Churg, A.; Brauer, M.; Keeling, B. (1996) Ozone enhances the uptake of mineral particles by tracheobronchial epithelial cells in organ culture. Am. J. Respir. Crit. Care Med. 153: 1230-1233.

Clausen, P. A.; Wilkins, C. K.; Wolkoff, P.; Nielsen, G. D. (2001) Chemical and biological evaluation of a reaction mixture of R-(+)-limonene/ozone: formation of strong airway irritants. Environ. Int. 26: 511-522.

Cohen, M. D.; Zelikoff, J. T.; Qu, Q.; Schlesinger, R. B. (1996) Effects of ozone upon macrophage-interferon interactions. Toxicology 114: 243-252.

Cohen, M. D.; Sisco, M.; Li, Y.; Zelikoff, J. T.; Schlesinger, R. B. (2001) Ozone-induced modulation of cell-mediated immune responses in the lungs. Toxicol. Appl. Pharmacol. 171: 71-84.

Cohen, M. D.; Sisco, M.; Baker, K.; Li, Y.; Lawrence, D.; Van Loveren, H.; Zelikoff, J. T.; Schlesinger, R. B. (2002) Effects of inhaled ozone on pulmonary immune cells critical to antibacterial responses in situ. Inhalation Toxicol. 14: 599-619.

Colín-Barenque, L.; Avila-Costa, M. R.; Fortoul, T.; Rugerio-Vargas, C.; Machado-Salas, J. P.; Espinosa-Villanueva, J.; Rivas-Arancibia, S. (1999) Morphologic alteration of the olfactory bulb after acute ozone exposure in rats. Neurosci. Lett. 274: 1-4.

Connor, L. M.; Ballinger, C. A.; Albrecht, T. B.; Postlethwait, E. M. (2004) Interfacial phospholipids inhibit ozone reactive absorption-mediated cytotoxicity in vitro. Am. J. Physiol.: 10.1152/ajplung.00397.2003.

Cottet-Emard, J.-M.; Dalmaz, Y.; Pequignot, J.; Peyrin, L.; Pequignot, J.-M. (1997) Long-term exposure to ozone alters peripheral and central catecholamine activity in rats. Pfluegers Arch. 433: 744-749.

Creutzenberg, O.; Bellmann, B.; Klingebiel, R.; Heinrich, U.; Muhle, H. (1995) Phagocytosis and chemotaxis of rat alveolar macrophages after a combined or separate exposure to ozone and carbon black. Exp. Toxicol. Pathol. 47: 202-206.

Cross, C. E.; Van Der Vliet, A.; Louie, S.; Thiele, J. J.; Halliwell, B. (1998) Oxidative stress and antioxidants at biosurfaces: plants, skin, and respiratory tract surfaces. Environ. Health Perspect. 106(suppl. 5): 1241-1251.

Custodio-Ramírez, V.; Paz, C. (1997) Ozone produces functional deficits in the rat visual pathway. Electroencephalogr. Clin. Neurophysiol. 104: 269-273.

Daly, C.; Fox, K.; Henein, M. (2002) Natriuretic peptides in the diagnosis of heart disease--first amongst equals? Int. J. Cardiol. 84: 107-113.

Delaunois, A.; Segura, P.; Montaño, L. M.; Vargas, M. H.; Ansay, M.; Gustin, P. (1998) Comparison of ozone-induced effects on lung mechanics and hemodynamics in the rabbit. Toxicol. Appl. Pharmacol. 150: 58-67.

Dell'Omo, G.; Fiore, M.; Petruzzi, S.; Alleva, E.; Bignami, G. (1995a) Neurobehavioral development of CD-1 mice after combined gestational and postnatal exposure to ozone. Arch. Toxicol. 69: 608-616.

Dell'Omo, G.; Wolfer, D.; Alleva, E.; Lipp, H.-P. (1995b) Developmental exposure to ozone induces subtle changes in swimming navigation of adult mice. Toxicol. Lett. 81: 91-99.

DeLorme, M. P.; Yang, H.; Elbon-Copp, C.; Gao, X.; Barraclough-Mitchell, H.; Bassett, D. J. P. (2002) Hyperresponsive airways correlate with lung tissue inflammatory cell changes in ozone-exposed rats. J. Toxicol. Environ. Health Part A 65: 1453-1470.

DeMarini, D. M.; Shelton, M. L.; Kohan, M. J.; Hudgens, E. E.; Kleindienst, T. E.; Ball, L. M.; Walsh, D.; de Boer, J. G.; Lewis-Bevan, L.; Rabinowitz, J. R.; Claxton, L. D.; Lewtas, J. (2000) Mutagenicity in lung of Big Blue(R) mice and induction of tandem-base substitutions in *Salmonella* by the air pollutant peroxyacetyl nitrate (PAN): predicted formation of intrastrand cross-links. Mutat. Res. 457: 41-55.

Depuydt, P.; Joos, G. F.; Pauwels, R. A. (1999) Ambient ozone concentrations induce airway hyperresponsiveness in some rat strains. Eur. Respir. J. 14: 125-131.

Depuydt, P. O.; Lambrecht, B. N.; Joos, G. F.; Pauwels, R. A. (2002) Effect of ozone exposure on allergic sensitization and airway inflammation induced by dendritic cells. Clin. Exp. Allergy 32: 391-396.

Dorado-Martínez, C.; Parades-Carbajal, C.; Mascher, D.; Borgonio-Pérez, G.; Rivas-Arancibia, S. (2001) Effects of different ozone doses on memory, motor activity and lipid peroxidation levels, in rats. Int. J. Neurosci. 108: 149-161.

Dormans, J. A. M. A.; Boere, A. J. F.; van Loveren, H.; Rombout, P. J. A.; Marra, M.; van Bree, L. (1996) Age-related toxicity in rat lungs following acute and repeated ozone exposure. Inhalation Toxicol. 8: 903-925.

Dormans, J. A. M. A.; Van Bree, L.; Boere, A. J. F.; Marra, M.; Rombout, P. J. A. (1999) Interspecies differences in time course of pulmonary toxicity following repeated exposure to ozone. Inhalation Toxicol. 11: 309-329.

Driscoll, K. E.; Simpson, L.; Carter, J.; Hassenbein, D.; Leikauf, G. D. (1993) Ozone inhalation stimulates expression of a neutrophil chemotactic protein, macrophage inflammatory protein 2. Toxicol. Appl. Pharmacol. 119: 306-309.

Dye, J. A.; Madden, M. C.; Richards, J. H.; Lehmann, J. R.; Devlin, R. B.; Costa, D. L. (1999) Ozone effects on airway responsiveness, lung injury, and inflammation. Comparative rat strain and in vivo/in vitro investigations. Inhalation Toxicol. 11: 1015-1040.

El-Fawal, H. A. N.; McGovern, T.; Schlesinger, R. B. (1995) Nonspecific bronchial responsiveness assessed in vitro following acute inhalation exposure to ozone and ozone/sulfuric acid mixtures. Exp. Lung Res. 21: 129-139.

Elder, A. C. P.; Gelein, R.; Finkelstein J. N.; Cox, C.; Oberdörster, G. (2000a) Endotoxin priming affects the lung response to ultrafine particles and ozone in young and old rats. In: Phalen, R. F., ed. Inhalation toxicology: proceedings of the third colloquium on particulate air pollution and human health (first special issue); June, 1999; Durham, NC. Inhalation Toxicol. 12(suppl. 1): 85-98.

Elder, A. C. P.; Gelein, R.; Finkelstein, J. N.; Cox, C.; Oberdörster, G. (2000b) Pulmonary inflammatory response to inhaled ultrafine particles is modified by age, ozone exposure, and bacterial toxin. In: Grant, L. D., ed. PM2000: particulate matter and health. Inhalation Toxicol. 12(suppl. 4): 227-246.

Elsayed, N. M. (2001) Diet restriction modulates lung response and survivability of rats exposed to ozone. Toxicology 159: 171-182.

Evans, M. J.; Fanucchi, M. V.; Baker, G. L.; Van Winkle, L. S.; Pantle, L. M.; Nishio, S. J.; Schelegle, E. S.; Gershwhin, L. J.; Miller, L. A.; Hyde, D. M.; Sannes, P. L.; Plopper, C. G. (2003) Atypical development of the tracheal basement membrane zone of infant rhesus monkeys exposed to ozone and allergen. Am. J. Physiol. 285: L931-L939.

Fakhrzadeh, L.; Laskin, J. D.; Laskin, D. L. (2002) Deficiency in inducible nitric oxide synthase protects mice from ozone-induced lung inflammation and tissue injury. Am. J. Respir. Cell Mol. Biol. 26: 413-419.

Fanucchi, M. V.; Hotchkiss, J. A.; Harkema, J. R. (1998) Endotoxin potentiates ozone-induced mucous cell metaplasia in rat nasal epithelium. Toxicol. Appl. Pharmacol. 152: 1-9.

Farman, C. A.; Pinkerton, K. E.; Rajini, P.; Witschi, H.; Last, J. A. (1997) Evolution of lung lesions in rats exposed to mixtures of ozone and nitrogen dioxide. Inhalation Toxicol. 9: 647-677.

Farman, C. A.; Watkins, K.; Van Hoozen, B.; Last, J. A.; Witschi, H.; Pinkerton, K. E. (1999) Centriacinar remodeling and sustained procollagen gene expression after exposure to ozone and nitrogen dioxide. Am. J. Respir. Cell Mol. Biol. 20: 303-311.

Ferng, S.-F.; Castro, C. E.; Afifi, A. A.; Bermúdez, E.; Mustafa, M. G. (1997) Ozone-induced DNA strand breaks in guinea pig tracheobronchial epithelial cells. J. Toxicol. Environ. Health 51: 353-367.

Folkerts, G.; Busse, W. W.; Nijkamp, F. P.; Sorkness, R.; Gern, J. E. (1998) Virus-induced airway hyperresponsiveness and asthma. Am. J. Respir. Crit. Care Med. 157: 1708-1720.

Foster, W. M.; Freed, A. N. (1999) Regional clearance of solute from peripheral airway epithelia: recovery after sublobar exposure to ozone. J. Appl. Physiol. 86: 641-646.

Frampton, M. W.; Pryor, W. A.; Cueto, R.; Cox, C.; Morrow, P. E.; Utell, M. J. (1999) Aldehydes (nonanal and hexanal) in rat and human bronchoalveolar lavage fluid after ozone exposure. Cambridge, MA: Health Effects Institute; research report no. 90. Available: www.healtheffects.org/Pubs/Frampton-C.pdf [2000, February 9].

Freed, A. N.; Chou, C. L.; Fuller, S. D.; Croxton, T. L. (1996) Ozone-induced vagal reflex modulates airways reactivity in rabbits. Respir. Physiol. 105: 95-102.

Freed, A. N.; Cueto, R.; Pryor, W. A. (1999) Antioxidant transport modulates peripheral airway reactivity and inflammation during ozone exposure. J. Appl. Physiol. 87: 1595-1603.

Friedlander, S. K.; Yeh, E. K. (1998) The submicron atmospheric aerosol as a carrier of reactive chemical species: case of peroxides. Appl. Occup. Environ. Hyg. 13: 416-420.

Gaffney, J. S.; Marley, N. A.; Prestbo, E. W. (1993) Measurements of peroxyacetyl nitrate at a remote site in the southwestern United States: tropospheric implications. Environ. Sci. Technol. 27: 1905-1910.

Garssen, J.; Van Bree, L.; Van Der Vliet, H.; Van Loveren, H. (1997) Ozone-induced impairment of pulmonary type IV hypersensitivity and airway hyperresponsiveness in mice. Inhalation Toxicol. 9: 581-599.

Gohil, K.; Cross, C. E.; Last, J. A. (2003) Ozone-induced disruptions of lung transcriptomes. Biochem. Biophys. Res. Commun. 305: 719-728.

Goldsmith, C.-A. W.; Ning, Y.-Y.; Qin, G.; Imrich, A.; Lawrence, J.; Murthy, G. G., K.; Catalano, P. J.; Kobzik, L. (2002) Combined air pollution particle and ozone exposure increases airway responsiveness in mice. Inhalation Toxicol. 14: 325-347.

González-Piña, R.; Paz, C. (1997) Brain monoamine changes in rats after short periods of ozone exposure. Neurochem. Res. 22: 63-66.

Grosjean, D. (2003) Ambient PAN and PPN in southern California from 1960 to the SCOS97-NARSTO. Atmos. Environ. 37(suppl. 2): S221-S238.

Grosjean, E.; Grosjean, D.; Woodhouse, L. F. (2001) Peroxyacetyl nitrate and peroxypropionyl nitrate during SCOS 97-NARSTO. Environ. Sci. Technol. 35: 4007-4014.

Guerrero, A. L.; Dorado-Martínez, C.; Rodriguez, A.; Pedroza-Ríos, K.; Borgonio-Pérez, G.; Rivas-Arancibia, S. (1999) Effects of vitamin E on ozone-induced memory deficits and lipid peroxidation in rats. NeuroReport 10: 1689-1692.

Gulisano, M.; Marceddu, S.; Barbaro, A.; Pacini, A.; Buiatti, E.; Martini, A.; Pacini, P. (1997) Damage to the nasopharyngeal mucosa induced by current levels of urban air pollution: a field study in lambs. Eur. Respir. J. 10: 567-572.

Günther, T.; Höllriegl, V.; Vormann, J. (1993) Perinatal development of iron and antioxidant defence systems. J. Trace Elem. Electrolytes Health Dis. 7: 47-52.

Gupta, S. K.; Reinhart, P. G.; Bhalla, D. K. (1998) Enhancement of fibronectin expression in rat lung by ozone and an inflammatory stimulus. Am. J. Physiol. 275: L330-L335.

Haddad, E.-B.; Liu, S. F.; Salmon, M.; Robichaud, A.; Barnes, P. J.; Chung, K. F. (1995) Expression of inducible nitric oxide synthase mRNA in Brown Norway rats exposed to ozone: effect of dexamethasone. Eur. J. Pharmacol. Environ. Toxicol. Pharmacol. Sect. 293: 287-290.

Hamilton, R. F.; Li, L.; Eschenbacher, W. L.; Szweda, L.; Holian, A. (1998) Potential involvement of 4-hydroxynonenal in the response of human lung cells to ozone. Am. J. Physiol. 274: L8-L16.

Harkema, J. R.; Morgan, K. T.; Gross, E. A.; Catalano, P. J.; Griffith, W. C. (1994) Consequences of prolonged inhalation of ozone on F344/N rats: collaborative studies. Part VII: effects on the nasal mucociliary apparatus. Cambridge, MA: Health Effects Institute; research report no. 65.

Harkema, J. R.; Catalano, P. J.; Hotchkiss, J. A. (1997a) Consequences of prolonged inhalation of ozone on F344/N rats: collaborative studies. Part XII. Atrophy of bone in nasal turbinates. Cambridge, MA: Health Effects Institute; research report no. 65.

Harkema, J. R.; Hotchkiss, J. A.; Griffith, W. C. (1997b) Mucous cell metaplasia in rat nasal epithelium after a 20-month exposure to ozone: a morphometric study of epithelial differentiation. Am. J. Respir. Cell Mol. Biol. 16: 521-530.

Harkema, J. R.; Hotchkiss, J. A.; Barr, E. B.; Bennett, C. B.; Gallup, M.; Lee, J. K.; Basbaum, C. (1999) Long-lasting effects of chronic ozone exposure on rat nasal epithelium. Am. J. Respir. Cell Mol. Biol. 20: 517-529.

Haro, R.; Paz, C. (1993) Effects of ozone exposure during pregnancy on ontogeny of sleep in rats. Neurosci. Lett. 164: 67-70.

Hawgood, S.; Poulain, F. R. (2001) The pulmonary collectins and surfactant metabolism. Annu. Rev. Physiol. 63: 495-519.

Hawgood, S.; Ochs, M.; Jung, A.; Akiyama, J.; Allen, L.; Brown, C.; Edmondson, J.; Levitt, S.; Carlson, E.; Gillespie, A. M.; Villar, A.; Epstein, C. J.; Poulain, F. R. (2002) Sequential targeted deficiency of SP-A and -D leads to progressive alveolar lipoproteinosis and emphysema. Am. J. Physiol. 283: L1002-L1010.

Heck, H. d'A.; Casanova, M.; Starr, T. B. (1990) Formaldehyde toxicity–new understanding. Crit. Rev. Toxicol. 20: 397-426.

Heddle, J. A.; Shepson, P. B.; Gingerich, J. D.; So, K. W. (1993) Mutagenicity of peroxyacetyl nitrate (PAN) in vivo: tests for somatic mutations and chromosomal aberrations. Environ. Mol. Mutagen. 21: 58-66.

Herbert, R. A.; Hailey, J. R.; Grumbein, S.; Chou, B. J.; Sills, R. C.; Haseman, J. K.; Goehl, T.; Miller, R. A.; Roycroft, J. H.; Boorman, G. A. (1996) Two-year and lifetime toxicity and carcinogenicity studies of ozone in B6C3F1 mice. Toxicol. Pathol. 24: 539-548.

Highfill, J. W.; Watkinson, W. P. (1996) Ozone toxicity in the rat. II. Modeling changes due to ambient temperatures and duration. J. Appl. Physiol. 80: 1811-1818.

Hoffer, E.; Baum, Y.; Tabak, A.; Frevert, C. (1999) Adhesion molecules of blood polymorphonuclear leukocytes and alveolar macrophages in rats: modulation by exposure to ozone. Hum. Exp. Toxicol. 18: 547-551.

Hotchkiss, J. A.; Harkema, J. R.; Johnson, N. F. (1997) Kinetics of nasal epithelial cell loss and proliferation in F344 rats following a single exposure to 0.5 ppm ozone. Toxicol. Appl. Pharmacol. 143: 75-82.

Hotchkiss, J. A.; Hilaski, R.; Cho, H.; Regan, K.; Spencer, P.; Slack, K.; Harkema, J. R. (1998) Fluticasone propionate attenuates ozone-induced rhinitis and mucous cell metaplasia in rat nasal airway epithelium. Am. J. Respir. Cell Mol. Biol. 18: 91-99.

Huffman, L. J.; Judy, D. J.; Brumbaugh, K.; Frazer, D. G.; Reynolds, J. S.; McKinney, W. G.; Goldsmith, W. T. (2001) Hyperthyroidism increases the risk of ozone-induced lung toxicity in rats. Toxicol. Appl. Pharmacol. 173: 18-26.

Huitrón-Reséndiz, S.; Custodio-Ramírez, V.; Escalante-Membrillo, C.; González-Piña, R.; Paz, C. (1994) Sleep alterations and brain regional changes of serotonin and its metabolite in rats exposed to ozone. Neurosci. Lett. 177: 119-122.

Hyde, D. M.; Miller, L. A.; McDonald, R. J.; Stovall, M. Y.; Wong, V.; Pinkerton, K. E.; Wegner, C. D.; Rothlein, R.; Plopper, C. G. (1999) Neutrophils enhance clearance of necrotic epithelial cells in ozone-induced lung injury in rhesus monkeys. Am. J. Physiol. 277: L1190-L1198.

Igarashi, A.; Iijima, H.; Tamura, G.; Shirato, K. (1998) Tazanolast inhibits ozone-induced airway hyperresponsiveness in guinea pigs. Am. J. Respir. Crit. Care Med. 157: 1531-1535.

Iijima, M. K.; Kobayashi, T.; Kamada, H.; Shimojo, N. (2001) Exposure to ozone aggravates nasal allergy-like symptoms in guinea pigs. Toxicol. Lett. 123: 77-85.

Ishii, Y.; Yang, H.; Sakamoto, T.; Nomura, A.; Hasegawa, S.; Hirata, F.; Bassett, D. J. P. (1997) Rat alveolar macrophage cytokine production and regulation of neutrophil recruitment following acute ozone exposure. Toxicol. Appl. Pharmacol. 147: 214-223.

Ishii, Y.; Hashimoto, K.; Hirano, K.; Morishima, Y.; Mochizuki, M.; Masuyama, K.; Nomura, A.; Sakamoto, T.; Uchida, Y.; Sagai, M.; Sekizawa, K. (2000) Ebselen decreases ozone-induced pulmonary inflammation in rats. Lung 178: 225-234.

Iwasaki, T.; Takahashi, M.; Saito, H.; Arito, H. (1998) Adaptation of extrapulmonary responses to ozone exposure in conscious rats. Ind. Health 36: 57-60.

Jakobi, G.; Fabian, P. (1997) Indoor/outdoor concentrations of ozone and peroxyacetyl nitrate (PAN). Int. J. Biometeorol. 40: 162-165.

Jang, A.-S.; Choi, I.-S.; Koh, Y.-I.; Park, C.-S.; Lee, J.-S. (2002) The relationship between alveolar epithelial proliferation and airway obstruction after ozone exposure. Allergy 57: 737-740.

Jimba, M.; Skornik, W. A.; Killingsworth, C. R.; Long, N. C.; Brain, J. D.; Shore, S. A. (1995) Role of C fibers in physiological responses to ozone in rats. J. Appl. Physiol. 78: 1757-1763.

Joad, J. P.; Kott, K. S.; Bonham, A. C. (1998) Exposing guinea pigs to ozone for 1 wk enhances responsiveness of rapidly adapting receptors. J. Appl. Physiol. 84: 1190-1197.

Joad, J. P.; Bric, J. M.; Weir, A. J.; Putney, L.; Hyde, D. M.; Postlewait, E. M.; Plopper, C. G. (2000) Effect of respiratory pattern on ozone injury to the airways of isolated rat lungs. Toxicol. Appl. Pharmacol. 169: 26-32.

Johnston, C. J.; Stripp, B. R.; Piedbeouf, B.; Wright, T. W.; Mango, G. W.; Reed, C. K.; Finkelstein, J. N. (1998) Inflammatory and epithelial responses in mouse strains that differ in sensitivity to hyperoxic injury. Exp. Lung Res. 24: 189-202.

Johnston, C. J.; Stripp, B. R.; Reynolds, S. D.; Avissar, N. E.; Reed, C. K.; Finkelstein, J. N. (1999a) Inflammatory and antioxidant gene expression in C57BL/6J mice after lethal and sublethal ozone exposures. Exp. Lung Res. 25: 81-97.

Johnston, C. J.; Finkelstein, J. N.; Oberdörster, G.; Reynolds, S. D.; Stripp, B. R. (1999b) Clara cell secretory protein-deficient mice differ from wild-type mice in inflammatory chemokine expression to oxygen and ozone, but not to endotoxin. Exp. Lung Res. 25: 7-21.

Johnston, C. J.; Oberdörster, G.; Gelein, R.; Finkelstein, J. N. (2000) Newborn mice differ from adult mice in chemokine and cytokine expression to ozone, but not to endotoxin. Inhalation Toxicol. 12: 205-224.

Johnston, C. J.; Oberdörster, G.; Gelein, R.; Finkelstein, J. N. (2002) Endotoxin potentiates ozone-induced pulmonary chemokine and inflammatory responses. Exp. Lung Res. 28: 419-433.

Kafoury, R. M.; Pryor, W. A.; Squadrito, G. L.; Salgo, M. G.; Zou, X.; Friedman, M. (1999) Induction of inflammatory mediators in human airway epithelial cells by lipid ozonation products. Am. J. Respir. Crit. Care Med. 160: 1934-1942.

Kenyon, N. J.; Van Der Vliet, A.; Schock, B. C.; Okamoto, T.; McGrew, G. M.; Last, J. A. (2002) Susceptibility to ozone-induced acute lung injury in iNOS-deficient mice. Am. J. Physiol. 282: L540-L545.

Kim, M. Y.; Son, J. W.; Cho, M. H.; Choi, C. S.; Chae, C. H.; Lee, M. H. (2001) Oviductural carcinoma in B6C3F1 female mice exposed to 0.5 ppm ozone. Vet. Hum. Toxicol. 43: 370-372.

Kimmel, T. A.; Chen, L. C.; Bosland, M. C.; Nadziejko, C. (1997) Influence of acid aerosol droplet size on structural changes in the rat lung caused by acute exposure to sulfuric acid and ozone. Toxicol. Appl. Pharmacol. 144: 348-355.

Kleeberger, S. R.; Levitt, R. C.; Zhang, L.-Y.; Longphre, M.; Harkema, J.; Jedlicka, A.; Eleff, S. M.; DiSilvestre, D.; Holroyd, K. J. (1997) Linkage analysis of susceptibility to ozone-induced lung inflammation in inbred mice. Nat. Genet. 17: 475-478.

Kleeberger, S. R.; Reddy, S.; Zhang, L.-Y.; Jedlicka, A. E. (2000) Genetic susceptibility to ozone-induced lung hyperpermeability: role of toll-like receptor 4. Am. J. Respir. Cell Mol. Biol. 22: 620-627.

Kleeberger, S. R.; Reddy, S. P.; Zhang, L.-Y.; Cho, H.-Y.; Jedlicka, A. E. (2001a) Toll-like receptor 4 mediates ozone-induced murine lung hyperpermeability via inducible nitric oxide synthase. Am. J. Physiol. 280: L326-L333.

Kleeberger, S. R.; Ohtsuka, Y.; Ahang, L.-Y.; Longphre, M. (2001b) Airway responses to chronic ozone exposure are partially mediated through mast cells. J. Appl. Physiol. 90: 713-723.

Kleindienst, T. E.; Shepson, P. B.; Smith, D. F.; Hudgens, E. E.; Nero, C. M.; Cupitt, L. T.; Bufalini, J. J.; Claxton, L. D. (1990) Comparison of mutagenic activities of several peroxyacyl nitrates. Environ. Mol. Mutagen. 16: 70-80.

Kleinman, M. T.; Mautz, W. J.; Bjarnason, S. (1999) Adaptive and non-adaptive responses in rats exposed to ozone, alone and in mixtures, with acidic aerosols. Inhalation Toxicol. 11: 249-264.

Kleinman, M. T.; Bufalino, C.; Rasmussen, R.; Hyde, D.; Bhalla, D. K.; Mautz, W. J. (2000) Toxicity of chemical components of ambient fine particulate matter ($PM_{2.5}$) inhaled by aged rats. J. Appl. Toxicol. 20: 357-364.

Kligerman, A. D.; Mottus, K.; Erexson, G. L. (1995) Cytogenetic analyses of the in vitro and in vivo responses of murine cells to peroxyacetyl nitrate (PAN). Mutat. Res. 341: 199-206.

Kobzik, L.; Goldsmith, C.-A. W.; Ning, Y. Y.; Qin, G.; Morgan, B.; Imrich, A.; Lawrence J.; Murthy, G. G. K.; Catalano, P. J. (2001) Effects of combined ozone and air pollution particle exposure in mice. Boston, MA: Health Effects Institute; research report no. 106. Available: http://www.healtheffects.org/Pubs/Kobzik.pdf [27 January 2003].

Kodavanti, U. P.; Hatch, G. E.; Starcher, B.; Giri, S. N.; Winsett, D.; Costa, D. L. (1995) Ozone-induced pulmonary functional, pathological, and biochemical changes in normal and vitamin C-deficient guinea pigs. Fundam. Appl. Toxicol. 24: 154-164.

Koto, H.; Salmon, M.; Haddad el-B.; Huang, T.-J.; Zagorski, J.; Chung, K. F. (1997) Role of cytokine-induced neutrophil chemoattractant (CINC) in ozone-induced airway inflammation and hyperresponsiveness. Am. J. Respir. Crit. Care Med. 156: 234-239.

Koyama, Y.; Hayaishi, O. (1994) Modulation by prostaglandins of activity of sleep-related neurons in the preoptic/anterior hypothalamic areas in rats. Brain Res. Bull. 33: 367-372.

Krishna, M. T.; Chauhan, A. J.; Frew, A. J.; Holgate, S. T. (1998) Toxicological mechanisms underlying oxidant pollutant-induced airway injury. Rev. Environ. Health 13: 59-71.

Larson, S. D.; Schelegle, E. S.; Walby, W. F.; Gershwin, L. J.; Fanuccihi, M. V.; Evans, M. J.; Joad, J. P.; Tarkington, B. K.; Hyde, D. M.; Plopper, C. G. (2004) Postnatal remodeling of the neural components of the epithelial-mesenchymal trophic unit in the proximal airways of infant rhesus monkeys exposed to ozone and allergen. Toxicol. Appl. Pharmacol. 194: 211-220.

Laskin, D. L.; Laskin, J. D. (2001) Role of macrophages and inflammatory mediators in chemically induced toxicity. Toxicology (Ireland) 160: 111-118.

Laskin, D. L.; Pendino, K. J.; Punjabi, C. J.; del Valle, M. R.; Laskin, J. D. (1994) Pulmonary and hepatic effects of inhaled ozone in rats. Environ. Health Perspect. 102(suppl. 10): 61-64.

Laskin, J. D.; Heck, D. E.; Laskin, D. L. (1996) Nitric oxide production in the lung and liver following inhalation of the pulmonary irritant ozone. In: Snyder, R.; Kocsis, J. J.; Sipes, I. G.; Kalf, G. F.; Jollow, D. J.; Greim, H.; Monks, T. J.; Witmer, C. M., eds. Biological Reactive Intermediates V: Basic Mechanistic Research in Toxicology and Human Risk Assessment: proceedings of the Fifth International Symposium; January 1995; Munich, Germany. Adv. Exp. Med. Biol. 387: 141-146.

Laskin, D. L.; Sunil, V.; Guo, Y.; Heck, D. E.; Laskin, J. D. (1998) Increased nitric oxide synthase in the lung after ozone inhalation is associated with activation of NF-κB. Environ. Health Perspect. 106(suppl. 5): 1175-1178.

Laskin, D. L.; Fakhrzadeh, L.; Heck, D. E.; Gerecke, D.; Laskin, J. D. (2002) Upregulation of phosphoinositide 3-kinase and protein kinase B in alveolar macrophages following ozone inhalation. Role of NF-κB and STAT-1 in ozone-induced nitric oxide production and toxicity. Mol. Cell. Biochem. 234-235: 91-98.

Last, J. A.; Pinkerton, K. E. (1997) Chronic exposure of rats to ozone and sulfuric acid aerosol: biochemical and structural responses. Toxicology 116: 133-146.
Lavnikova, N.; Prokhorova, S.; Lakhotia, A. V.; Gordon, R.; Laskin, D. L. (1998) Distinct inflammatory responses of adherent vascular lung neutrophils to pulmonary irritants. J. Inflammation 48: 56-66
Lee, C.; Watt, K. C.; Chang, A. M.; Plopper, C. G.; Buckpitt, A. R.; Pinkerton, K. E. (1998) Site-selective differences in cytochrome P450 isoform activities: comparison of expression in rat and rhesus monkey lung and induction in rats. Drug Metab. Dispos. 26: 396-400.
Lemos, M.; Lichtenfels, A. J. F. C.; Amaro, E., Jr.; Macchione, M.; Martins, M. A.; King, M.; Böhm, G. M.; Saldiva, P. H. N. (1994) Quantitative pathology of nasal passages in rats exposed to urban levels of air pollution. Environ. Res. 66: 87-95.
Longphre, M.; Zhang, L.-Y.; Harkema, J. R.; Kleeberger, S. R. (1999) Ozone-induced pulmonary inflammation and epithelial proliferation are partially mediated by PAF. J. Appl. Physiol. 86: 341-349.
Lorz, C.; López, J. (1997) Incidence of air pollution in the pulmonary surfactant system of the pigeon (*Columbia livia*). Anat. Rec. 249: 206-212.
Madden, M. C.; Richards, J. H.; Dailey, L. A.; Hatch, G. E.; Ghio, A. J. (2000) Effect of ozone on diesel exhaust particle toxicity in rat lung. Toxicol. Appl. Pharmacol. 168: 140-148.
Mango, G. W.; Johnston, C. J.; Reynolds, S. D.; Finkelstein, J. N.; Plopper, C. G.; Stripp, B. R. (1998) Clara cell secretory protein deficiency increases oxidant stress response in conducting airways. Am. J. Physiol. 275: L348-L356.
Matsubara, S.; Kikkawa, H.; Kaminuma, O.; Ikezawa, K. (1997a) Angiotensin-converting enzyme inhibitors can potentiate ozone-induced airway hyperresponsiveness. Eur. J. Pharmacol. 337: 259-265.
Matsubara, S.; Fushimi, K.; Kaminuma, O.; Kikkawa, H.; Ikezawa, K.; Naito, K. (1997b) Prevention of ozone-induced airway hyperresponsiveness and epithelial injury by phosphodiesterase inhibitors in guinea pigs. Environ. Toxicol. Pharmacol. 3: 201-209.
Mautz, W. J. (2003) Exercising animal models in inhalation toxicology: interactions with ozone and formaldehyde. Environ. Res. 92: 14-26.
Mautz, W. J.; Kleinman, M. T.; Bhalla, D. K.; Phalen, R. F. (2001) Respiratory tract responses to repeated inhalation of an oxidant and acid gas-particle air pollutant mixture. Toxicol. Sci. 61: 331-341.
McGraw, D. W.; Forbes, S. L.; Mak, J. C. W.; Witte, D. P.; Carrigan, P. E.; Leikauf, G. D.; Liggett, S. B. (2000) Transgenic overexpression of beta(2)-adrenergic receptors in airway epithelial cells decreases bronchoconstriction. Am. J. Physiol. 279: L379-L389.
McKinney, W. J.; Jaskot, R. H.; Richards, J. H.; Costa, D. L.; Dreher, K. L. (1998) Cytokine mediation of ozone-induced pulmonary adaptation. Am. J. Respir. Cell. Mol. Biol. 18: 696-705.
Miller, L. A.; Barnett, N. L.; Sheppard, D.; Hyde, D. M. (2001) Expression of the β6 integrin subunit is associated with sites of neutrophil influx in lung epithelium. J. Histochem. Cytochem. 49: 41-48.
Morgan, M. S.; Meyer, P.; Holub, R.; Frank, R. (1986) Overall and regional lung function in dogs exposed acutely to ozone. Environ. Res. 41: 546-557.
Morio, L. A.; Hooper, K. A.; Brittingham, J.; Li, T.-H.; Gordon, R. E.; Turpin, B. J.; Laskin, D. L. (2001) Tissue injury following inhalation of fine particulate matter and hydrogen perioxide is associated with altered production of inflammatory mediators and antioxidants by alveolar macrophages. Toxicol. Appl. Pharmacol. 177: 188-199.
Moss, O. R.; Gross, E. A.; James, R. A.; Janszen, D. B.; Ross, P. W.; Roberts, K. C.; Howard, A. M.; Harkema, J. R.; Calderón-Garcidueñas, L.; Morgan, K. T. (2001) Respiratory tract toxicity in rats exposed to Mexico City air. Cambridge, MA: Health Effects Institute; research report no. 100. Available: http://www.healtheffects.org/pubs-research.htm [15 May, 2003].
Mücke, W. (1996) The environment and the eye. Topics of ophthalmic toxicology. Leban. Med. J. 44: 146-150.
Mudway, I. S.; Kelly, F. J. (1998) Modeling the interactions of ozone with pulmonary epithelial lining fluid antioxidants. Toxicol. Appl. Pharmacol. 148: 91-100.
Mudway, I. S.; Kelly, F. J. (2000) Ozone and the lung: a sensitive issue. Mol. Aspects. Med. 21: 1-48.
Murphy, D. J. (2002) Assessment of respiratory function in safety pharmacology. Fundam. Clin. Pharmacol. 16: 183-196.
Mutoh, T.; Joad, J. P.; Bonham, A. C. (2000) Chronic passive cigarette smoke exposure augments bronchopulmonary C-fibre inputs to nucleus tractus solitarii neurones and reflex output in young guinea-pigs. J. Physiol. (London) 523: 223-233.

Nakano, H.; Aizawa, H.; Matsumoto, K.; Fukuyama, S.; Inoue, H.; Hara, N. (2000) Cyclooxygenase-2 participates in the late phase of airway hyperresponsiveness after ozone exposure in guinea pigs. Eur. J. Pharmacol. 403: 267-275.

National Toxicology Program. (1994) NTP technical report on the toxicology and carcinogenesis studies of ozone (CAS no. 10028-15-6) and ozone/NNK (CAS no. 10028-15-6/64091-91-4) in F344/N rats and B6C3F_1 mice (inhalation studies). Research Triangle Park, NC: U.S. Department of Health and Human Services, National Institutes of Health; publication no. 95-3371. (National Toxicology Program technical report series: no. 440).

Neuhaus-Steinmetz, U.; Uffhausen, F.; Herz, U.; Renz, H. (2000) Priming of allergic immune responses by repeated ozone exposure in mice. Am. J. Respir. Cell Mol. Biol. 23: 228-233.

Nichols, B. G.; Woods, J. S.; Luchtel, D. L.; Corral, J.; Koenig, J. Q. (2001) Effects of ozone exposure on nuclear factor-κB activation and tumor necrosis factor-α expression in human nasal epithelial cells. Toxicol. Sci. 60: 356-362.

Nielsen, G. D.; Hougaard, K. S.; Larsen, S. T.; Hammer, M.; Wolkoff, P.; Clausen, P. A.; Wilkins, C. K.; Alarie, Y. (1999) Acute airway effects of formaldehyde and ozone in BALB/c mice. Hum. Exp. Toxicol. 18: 400-409.

Niño-Cabrera, H. G.; Colín-Barenque, L.; Avila-Costa, M. R.; Espinosa-Villanueva, J.; Fortoul, T. I.; Rivas-Arancibia, S. (2002) Differences between hippocampus and cerebral cortex in aged rats in an oxidative stress model. Int. J. Neurosci. 112: 373-381.

Noviski, N.; Brewer, J. P.; Skornik, W. A.; Galli, S. J.; Drazen, J. M.; Martin, T. R. (1999) Mast cell activation is not required for induction of airway hyperresponsiveness by ozone in mice. J. Appl. Physiol. 86: 202-210.

Paige, R. C.; Royce, F. H.; Plopper, C. G.; Buckpitt, A. R. (2000a) Long-term exposure to ozone increases acute pulmonary centriacinar injury by 1-nitronaphthalene: I. Region-specific enzyme activity. J. Pharmacol. Exp. Ther. 295: 934-941.

Paige, R. C.; Wong, V.; Plopper, C. G. (2000b) Long-term exposure to ozone increases acute pulmonary centriacinar injury by 1-nitronaphthalene: II. Quantitative histopathology. J. Pharmacol. Exp. Ther. 295: 942-950.

Paquette, N. C.; Tankersley, C. G.; Zhang, L.-Y.; Kleeberger, S. R. (1994) Repeated subacute ozone exposure of inbred mice: airway inflammation and ventilation. Exp. Lung Res. 20: 579-594.

Paz, C. (1997) Some consequences of ozone exposure on health. Arch. Med. Res. 28: 163-170.

Paz, C.; Bazan-Perkins, B. (1992) Sleep-wake disorganization in cats exposed to ozone. Neurosci. Lett. 140: 270-272.

Paz, C.; Huitrón-Reséndiz, S. (1996) The effects of ozone exposure on the sleep-wake cycle and serotonin contents in the pons of the rat. Neurosci. Lett. 204: 49-52.

Pearson, A. C.; Bhalla, D. K. (1997) Effects of ozone on macrophage adhesion in vitro and epithelial and inflammatory responses in vivo: the role of cytokines. J. Toxicol. Environ. Health 50: 143-157.

Peden, D. B.; Dailey, L. (1995) Modulation of mast cell functions by in vitro ozone exposure. Am. J. Physiol. 268: L902-L910.

Pendino, K. J.; Shuler, R. L.; Laskin, J. D.; Laskin, D. L. (1994) Enhanced production of interleukin-1, tumor necrosis factor-α, and fibronectin by rat lung phagocytes following inhalation of a pulmonary irritant. Am. J. Respir. Cell Mol. Biol. 11: 279-286.

Petruzzi, S.; Fiore, M.; Dell'Omo, G.; Bignami, G.; Alleva, E. (1995) Medium and long-term behavioral effects in mice of extended gestational exposure to ozone. Neurotoxicol. Teratol. 17: 463-470.

Petruzzi, S.; De Acetis, L.; Chiarotti, F.; Sorace, A.; Alleva, E. (1999) Limited changes in handedness and morphine reactivity in CD-1 mice after pre- and postnatal ozone exposure. Acta Neurobiol. Exp. 59: 115-122.

Pinkerton, K. E.; Weller, B. L.; Menache, M. G.; Plopper, C. G. (1998) Consequences of prolonged inhalation of ozone on F344/N rats: collaborative studies. Part XIII. A comparison of changes in the tracheobronchial epithelium and pulmonary acinus in male rats at 3 and 20 months. Cambridge, MA: Health Effects Institute; research report no. 65.

Plopper, C. G.; Fanucchi, M. V. (2000) Do urban environmental pollutants exacerbate childhood lung diseases? Environ. Health Perspect. 108: A252-A253.

Plopper, C. G.; Hatch, G. E.; Wong, V.; Duan, X.; Weir, A. J.; Tarkington, B. K.; Devlin, R. B.; Becker, S.; Buckpitt, A. R. (1998) Relationship of inhaled ozone concentration to acute tracheobronchial epithelial injury, site-specific ozone dose and glutathione depletion in rhesus monkeys. Am. J. Respir. Cell Mol. Biol. 19: 387-399.

Polosa, R.; Holgate, S. T. (1997) Adenosine bronchoprovocation: a promising marker of allergic inflammation in asthma? Thorax 52: 919-923.

Pope, C. A., III; Burnett, R. T.; Thun, M. J.; Calle, E. E.; Krewski, D.; Ito, K.; Thurston, G. D. (2002) Lung cancer, cardiopulmonary mortality, and long-term exposure to fine particulate air pollution. JAMA J. Am. Med. Assoc. 287: 1132-1141.

Postlethwait, E. M.; Cueto, R.; Velsor, L. W.; Pryor, W. A. (1998) O_3-induced formation of bioactive lipids: estimated surface concentrations and lining layer effects. Am. J. Physiol. 274: L1006-L1016.

Postlethwait, E. M.; Joad, J. P.; Hyde, D. M.; Schelegle, E. S.; Bric, J. M.; Weir, A. J.; Putney, L. F.; Wong, V. J.; Velsor, L. W.; Plopper, C. G. (2000) Three-dimensional mapping of ozone-induced acute cytotoxicity in tracheobronchial airways of isolated perfused rat lung. Am. J. Respir. Cell Mol. Biol. 22: 191-199.

Pryor, W. A.; Squadrito, G. L.; Friedman, M. (1995) A new mechanism for the toxicity of ozone. Toxicol. Lett. 82/83: 287-293.

Pryor, W. A.; Bermúdez, E.; Cueto, R.; Squadrito, G. L. (1996) Detection of aldehydes in bronchoalveolar lavage of rats exposed to ozone. Fundam. Appl. Toxicol. 34: 148-156.

Pulfer, M. K.; Murphy, R. C. (2004) Formation of biologically active oxysterols during ozonolysis of cholesterol present in lung surfactant. J. Biol. Chem. 279: 26331-26338.

Pulfer, M. K.; Taube, C.; Gelfand, E.; Murphy, R. C. (2005) Ozone exposure in vivo and formation of biologically active oxysterols in the lung. J. Pharmacol. Exp. Ther. 312: 256-264.

Quinlan, T.; Spivack, S.; Mossman, B. T. (1994) Regulation of antioxidant enzymes in lung after oxidant injury. Environ. Health Perspect. 102(suppl. 2): 79-87.

Rehle, D.; Leleux, D.; Erdelyi, M.; Tittel, F.; Fraser, M.; Friedfeld, S.; et al. (2001) Ambient formaldehyde detection with a laser spectrometer based on difference-frequency generation in PPLN. Appl. Phys. B: Lasers Opt. 72: 947-952.

Reinhart, P. G.; Gupta, S. K.; Bhalla, D. K. (1999) Attenuation of ozone-induced lung injury by interleukin-10. Toxicol. Lett. 110: 35-42.

Renner, R. (2002) Bad air and birth defects. Environ. Health Perspect. 110: A291.

Ritz, B.; Yu, F. (1999) The effect of ambient carbon monoxide on low birth weight among children born in southern California between 1989 and 1993. Environ. Health Perspect. 107: 17-25.

Rivas-Arancibia, S.; Vazquez-Sandoval, R.; Gonzalez-Kladiano, D.; Schneider-Rivas, S.; Lechuga-Guerrero, A. (1998) Effects of ozone exposure in rats on memory and levels of brain and pulmonary superoxide dismutase. Environ. Res. 76: 33-39.

Rivas-Arancibia, S.; Dorado-Martínez, C.; Borgonio-Pérez, G.; Hiriart-Urdanivia, M.; Verdugo-Diaz, L.; Durán-Vázquez, A.; Colín-Baranque, L.; Avila-Costa, M. R. (2000) Effects of taurine on ozone-induced memory deficits and lipid peroxidation levels in brains of young, mature, and old rats. Environ. Res. 82: 7-17.

Rivas-Arancibia, S.; Dorado-Martínez, C.; Colín-Barenque, L.; Kendrick, K. M.; De la Riva, C.; Guevara-Guzmán, R. (2003) Effect of acute ozone exposure on locomotor behavior and striatal function. Pharmacol. Biochem. Behav. 74: 891-900.

Rivas-Manzano, P.; Paz, C. (1999) Cerebellar morphological alterations in rats induced by prenatal ozone exposure. Neurosci. Lett. 276: 37-40.

Rose, R. C.; Richer, S. P.; Bode, A. M. (1998) Ocular oxidants and antioxidant protection. Proc. Soc. Exp. Biol. Med. 217: 397-407.

Savov, J. D.; Whitehead, G. S.; Wang, J.; Liao, G.; Usuka, J.; Peltz, G.; Foster, W. M.; Schwartz, D. A. (2004) Ozone-induced acute pulmonary injury in inbred mouse strains. Am. J. Respir. Cell Mol. Biol. 31: 69-77.

Schelegle, E. S.; Alfaro, M. F.; Putney, L.; Stovall, M.; Tyler, N.; Hyde, D. M. (2001) Effect of C-fiber-mediated, ozone-induced rapid shallow breathing on airway epithelial injury in rats. J. Appl. Physiol. 91: 1611-1618.

Schelegle, E. S.; Miller, L. A.; Gershwin, L. J.; Fanucchi, M. V.; Van Winkle, L. S.; Gerriets, J. E.; Walby, W. F.; Mitchell, V.; Tarkington, B. K.; Wong, V. J.; Baker, G. L.; Pantle, L. M.; Joad, J. P.; Pinkerton, K. E.; Wu, R.; Evans, M. J.; Hyde, D. M.; Plopper, C. G. (2003a) Repeated episodes of ozone inhalation amplifies the effects of allergen sensitization and inhalation on airway immune and structural development in Rhesus monkeys. Toxicol. Appl. Pharmacol. 191: 74-85.

Schelegle, E. S.; Walby, W. F.; Alfaro, M. F.; Wong, V. J.; Putney, L.; Stovall, M. Y.; Sterner-Kock, A.; Hyde, D. M.; Plopper, C. G. (2003b) Repeated episodes of ozone inhalation attenuates airway injury/repair and release of substance P, but not adaptation. Toxicol. Appl. Pharmacol. 186: 127-142.

Schlesinger, R. B. (1995) Interaction of gaseous and particulate pollutants in the respiratory tract: mechanisms and modulators. Toxicology 105: 315-325.

Schlesinger, R. B.; Cohen, M. D.; Gordon, T.; Nadziejko, C.; Zelikoff, J. T.; Sisco, M.; Regal, J. F.; Menache, M. G. (2002a) Ozone differentially modulates airway responsiveness in atopic versus nonatopic guinea pigs. Inhalation Toxicol. 14: 431-457.

Schlesinger, R. B.; Cohen, M.; Gordon, T.; Nadziejko, C.; Zelikoff, J. T.; Sisco, M.; Regal, J. F.; Menache, M. G. (2002b) Ozone-induced modulation of airway hyperresponsiveness in guinea pigs. Boston, MA: Health Effects Institute; research report no. 109.

Schwartz, D. A. (2002) TLR4 and LPS hyporesponsiveness in humans. Int. J. Hyg. Environ. Health 205: 221-227.

Segura, P.; Montaño, L. M.; Bazán-Perkins, B.; Gustin, P.; Vargas, M. H. (1997) Ozone at high-pollution urban levels causes airway hyperresponsiveness to substance P but not to other agonists. Environ. Toxicol. Pharmacol. 3: 91-95.

Sen, S.; Dulchavsky, S. A.; Dutta, S. (1993) Effects of triiodothyronine (T3) supplementation upon ozone-induced lung injury. Free Radic. Res. Commun. 18: 299-308.

Shore, S. A.; Abraham, J. H.; Schwartzman, I. N.; Murthy, G. G.; Laporte, J. D. (2000) Ventilatory responses to ozone are reduced in immature rats. J. Appl. Physiol. 88: 2023-2030.

Shore, S. A.; Johnston, R. A.; Schwartzman, I. N.; Chism, D.; Krishna Murthy, G. G. (2002) Ozone-induced airway hyperresponsiveness is reduced in immature mice. J. Appl. Physiol. 92: 1019-1028.

Shore, S. A.; Rivera-Sanchez, Y. M.; Schwartzman, I. N.; Johnston, R. A. (2003) Responses to ozone are increased in obese mice. J. Appl. Physiol. 95: 938-945.

Sindhu, R. K.; Mautz, W. J.; Kikkawa, Y. (1998) Chronic exposure to ozone and nitric acid vapor results in increased levels of rat pulmonary putrescine. Arch. Toxicol. 72: 445-449.

Slade, R.; Watkinson, W. P.; Hatch, G. E. (1997) Mouse strain differences in ozone dosimetry and body temperature changes. Am. J. Physiol. 272: L73-L77.

Sorace, A.; De Acetis, L.; Alleva, E.; Santucci, D. (2001) Prolonged exposure to low doses of ozone: short- and long-term changes to behavioral performance in mice. Environ. Res. 85: 122-134.

Spannhake, E. W. (1996) Down-regulation of canine airway mast cell function following exposure to ozone in vivo. Exp. Lung Res. 22: 163-178.

Sterner-Kock, A.; Kock, M.; Braun, R.; Hyde, D. M. (2000) Ozone-induced epithelial injury in the ferret is similar to nonhuman primates. Am. J. Respir. Crit. Care Med. 162: 1152-1156.

Sun, J.; Chung, K. F. (1997) Interaction of ozone exposure with airway hyperresponsiveness and inflammation induced by trimellitic anhydride in sensitized guinea pigs. J. Toxicol. Environ. Health 51: 77-87.

Sun, J.; Koto, H.; Chung, K. F. (1997) Interaction of ozone and allergen challenges on bronchial responsiveness and inflammation in sensitised guinea pigs. Int. Arch. Allergy Immunol. 112: 191-195.

Szarek, J. L.; Stewart, N. L.; Zhang, J. Z.; Webb, J. A.; Valentovic, M. A.; Catalano, P. (1995) Contractile responses and structure of small bronchi isolated from rats after 20 months' exposure to ozone. Fundam. Appl. Toxicol. 28: 199-208.

Tager, I. B. (1999) Air pollution and lung function growth. Is it ozone? [editorial]. Am. J. Respir. Crit. Care Med. 160: 387-389.

Takahashi, T.; Miura, M.; Katsumata, U.; Ichinose, M.; Kimura, K.; Inoue, H.; Takishima, T.; Shirato, K. (1993) Involvement of superoxide in ozone-induced airway hyperresponsiveness in anesthetized cats. Am. Rev. Respir. Dis. 148: 103-106.

Takahashi, N.; Yu, X.-Y.; Schofield, B. H.; Kleeberger, S. R.; Scott, A. L.; Hasegawa, S.; Spannhake, E. W. (1995a) Expression of ICAM-1 in airway epithelium after acute ozone exposure in the mouse. J. Appl. Physiol. 79: 1753-1761.

Takahashi, M.; Kleeberger, S. R.; Croxton, T. L. (1995b) Genetic control of susceptibility to ozone-induced changes in mouse tracheal electrophysiology. Am. J. Physiol. 269: L6-L10.

Takebayashi, T.; Abraham, J.; Murthy, G. G. K.; Lilly, C.; Rodger, I.; Shore, S. A. (1998) Role of tachykinins in airway responses to ozone in rats. J. Appl. Physiol. 85: 442-450.

Tankersley, C. G.; Kleeberger, S. R. (1994) Ozone-induced inflammation and altered ventilation in genetically susceptible mice: a comparison of acute and subacute exposures. Toxicol Lett. 72: 279-289.

Tankersley, C. G.; Fitzgerald, R. S.; Mitzner, W. A.; Kleeberger, S. R. (1993) Hypercapnic ventilatory responses in mice differentially susceptible to acute ozone exposure. J. Appl. Physiol. 75: 2613-2619.

Tesfaigzi, J.; Hotchkiss, J. A.; Harkema, J. R. (1998) Expression of the Bcl-2 protein in nasal epithelia of F344/N rats during mucous cell metaplasia and remodeling. Am. J. Respir. Cell Mol. Biol. 18: 794-799.

Thiele, J. J. (2001) Oxidative targets in the stratum corneum. A new basis for antioxidative strategies. Skin Pharmacol. Appl. Skin Physiol. 14(suppl. 1): 87-91.

Thiele, J. J.; Traber, M. G.; Packer, L. (1998) Depletion of human stratum corneum vitamin E: an early and sensitive in vivo marker of UV induced photo-oxidation. J. Invest. Dermatol. 110: 756-761.

Thiele, J. J.; Hsieh, S. N.; Briviba, K.; Sies, H. (1999) Protein oxidation in human stratum corneum: susceptibility of keratins to oxidation *in vitro* and presence of a keratin oxidation gradient *in vivo*. J. Invest. Dermatol. 113: 335-339.

Tsai, J.-J.; Lin, Y.-C.; Kwan, Z.-H.; Kao, H.-L. (1998) Effects of ozone on ovalbumin sensitization in guinea pigs. J. Microbiol. Immunol. Infect. 31: 225-232.

U.S. Environmental Protection Agency. (1993) Air quality criteria for oxides of nitrogen. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report nos. EPA/600/8-91/049aF-cF. 3v. Available from: NTIS, Springfield, VA; PB95-124533, PB95-124525, and PB95-124517.

U.S. Environmental Protection Agency. (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available online at: www.epa.gov/ncea/ozone.htm.

Uhlson, C.; Harrison, K.; Allen, C. B.; Ahmad, S.; White, C. W.; Murphy, R. C. (2002) Oxidized phospholipids derived from ozone-treated lung surfactant extract reduce macrophage and epithelial cell viability. Chem. Res. Toxicol. 15: 896-906.

Ulrich, M. M. W.; Alink, G. M.; Kumarathasan, P.; Vincent, R.; Boere, A. J.; Cassee, F. R. (2002) Health effects and time course of particulate matter on the cardiopulmonary system in rats with lung inflammation. J. Toxicol. Environ. Health Part A 65: 1571-1595.

Valacchi, G.; Weber, S. U.; Luu, C.; Cross, C. E.; Packer, L. (2000) Ozone potentiates vitamin E depletion by ultraviolet radiation in the murine stratum corneum. FEBS Lett. 466: 165-168.

Valacchi, G.; Pagnin, E.; Okamoto, T.; Corbacho, A. M.; Olano, E.; Davis, P. A.; Van der Vliet, A.; Packer, L.; Cross, C. E. (2003) Induction of stress proteins and MMP-9 by 0.8 ppm of ozone in murine skin. Biochem. Biophys. Res. Commun. 305: 741-746.

Valverde, M.; del Carmen Lopez, M.; Lopez, I.; Sanchez, I.; Fortoul, T. I.; Ostrosky-Wegman, P.; Rojas, E. (1997) DNA damage in leukocytes and buccal and nasal epithelial cells of individuals exposed to air pollution in Mexico City. Environ. Mol. Mutagen. 30: 147-152.

Van Bree, L.; Dormans, J. A. M. A.; Boere, A. J. F.; Rombout, P. J. A. (2001) Time study on development and repair of lung injury following ozone exposure in rats. Inhalation Toxicol. 13: 703-717.

Van Bree, L.; Dormans, J. A. M. A.; Koren, H. S.; Devlin, R. B.; Rombout, P. J. A. (2002) Attenuation and recovery of pulmonary injury in rats following short-term, repeated daily exposure to ozone. Inhalation Toxicol. 14: 883-900.

Van Hoof, I. H. J. M.; Van Bree, L.; Bast, A. (1996) Changes in receptor function by oxidative stress in guinea pig tracheal smooth muscle. Cent. Eur. J. Public Health 4(suppl.): 3-5.

Van Hoof, H. J. M.; Van Acker, F. A. A.; Voss, H.-P.; Van Bree, L.; Bast, A. (1997a) Acute exposure to ozone does not influence neuroreceptor density and sensitivity in guinea pig lung. Toxicol. Lett. 90: 53-60.

Van Hoof, H. J. M.; Voss, H.-P.; Kramer, K.; Boere, A. J. F.; Dormans, J. A. M. A.; Van Bree, L.; Bast, A. (1997b) Changes in neuroreceptor function of tracheal smooth muscle following acute ozone exposure of guinea pigs. Toxicology 120: 159-169.

Vanda, B.; de Buen, N.; Jasso, R.; Valero, G.; Vargas, M. H.; Olmos, R.; Arreola, J. L.; Santillán, P.; Alonso, P. (1998) Inflammatory cells and ferruginous bodies in bronchoalveolar lavage in urban dogs. Acta Cytol. 42: 939-944.

Vargas, M. H.; Romero, L.; Sommer, B.; Zamudio, P.; Gustin, P.; Montaño, L. M. (1998) Chronic exposure to ozone causes tolerance to airway hyperresponsiveness in guiea pigs: lack of SOD role. J. Appl. Physiol. 84: 1749-1755.

Vesely, D. L.; Giordano, A. T.; Raska-Emery, P.; Montgomery, M. R. (1994a) Ozone increases amino- and carboxy-terminal atrial natriuretic factor prohormone peptides in lung, heart, and circulation. J. Biochem. Toxicol. 9: 107-112.

Vesely, D. L.; Giordano, A. T.; Raska-Emery, P.; Montgomery, M. R. (1994b) Increase in atrial natriuretic factor in the lungs, heart, and circulatory system owing to ozone. Chest 105: 1551-1554.

Vesely, D. L.; Giordano, A. T.; Raska-Emery, P.; Montgomery, M. R. (1994c) Ozone increases atrial natriuretic peptides in heart, lung and circulation of aged vs. adult animals. Gerontology (Basel) 40: 227-236.

Vesely, K. R.; Schelegle, E. S.; Stovall, M. Y.; Harkema, J. R.; Green, J. F.; Hyde, D. M. (1999a) Breathing pattern response and epithelial labeling in ozone-induced airway injury in neutrophil-depleted rats. Am. J. Respir. Cell Mol. Biol. 20: 699-709.

Vesely, K. R.; Hyde, D. M.; Stovall, M. Y.; Harkema, J. R.; Green, J. F.; Schelegle, E. S. (1999b) Capsaicin-sensitive C-fiber-mediated protective responses in ozone inhalation in rats. J. Appl. Physiol. 86: 951-962.

Vincent, R.; Janzen, E. G.; Chen, G.; Kumarathasan, P.; Haire, D. L.; Guénette, J.; Chen, J. Z.; Bray, T. M. (1996) Spin trapping study in the lungs and liver of F344 rats after exposure to ozone. Free Radical Res. 25: 475-488.

Vincent, R.; Bjarnason, S. G.; Adamson, I. Y. R.; Hedgecock, C.; Kumarathasan, P.; Guénette, J.; Potvin, M.; Goegan, P.; Bouthillier, L. (1997) Acute pulmonary toxicity of urban particulate matter and ozone. Am. J. Pathol. 151: 1563-1570.

Vyskocil, A.; Viau, C.; Lamy, S. (1998) Peroxyacetyl nitrate: review of toxicity. Hum. Exp. Toxicol. 17: 212-220.

Wagner, J. G.; Hotchkiss, J. A.; Harkema, J. R. (2001a) Effects of ozone and endotoxin coexposure on rat airway epithelium: potentiation of toxicant-induced alterations. Environ. Health Perspect. 109(suppl. 4): 591-598.

Wagner, J. G.; Van Dyken, S. J.; Hotchkiss, J. A.; Harkema, J. R. (2001b) Endotoxin enhancement of ozone-induced mucous cell metaplasia is neutrophil-dependent in rat nasal epithelium. Toxicol. Sci. 60: 338-347.

Wagner, J. G.; Hotchkiss, J. A.; Harkema, J. R. (2002) Enhancement of nasal inflammatory and epithelial responses after ozone and allergen coexposure in brown Norway rats. Toxicol. Sci. 67: 284-294.

Wagner, J. G.; Van Dyken, S. J.; Wierenga, J. R.; Hotchkiss, J. A.; Harkema, J. R. (2003) Ozone exposure enhances endotoxin-induced mucous cell metaplasia in rat pulmonary airways. Toxicol. Sci. 74: 437-446.

Wang, G.; Umstead, T. M.; Phelps, D. S.; Al-Mondhiry, H.; Floros, J. (2002) The effect of ozone exposure on the ability of human surfactant protein A variants to stimulate cytokine production. Environ. Health Perspect. 110: 79-84.

Watkinson, W. P.; Wiester, M. J.; Highfill, J. W. (1995) Ozone toxicity in the rat. I. Effect of changes in ambient temperature on extrapulmonary physiological parameters. J. Appl. Physiol. 78: 1108-1120.

Watkinson, W. P.; Campen, M. J.; Nolan, J. P.; Costa, D. L. (2001) Cardiovascular and systemic responses to inhaled pollutants in rodents: effects of ozone and particulate matter. Environ. Health Perspect. 109(suppl. 4): 539-546.

Watkinson, W. P.; Campen, M. J.; Wichers, L. B.; Nolan, J. P.; Costa, D. L. (2003) Cardiac and thermoregulatory responses to inhaled pollutants in healthy and compromised rodents: modulation via interaction with environmental factors. Environ. Res. 92: 35-47.

Watt, K. C.; Plopper, C. G.; Weir, A. J.; Tarkington, B.; Buckpitt, A. R. (1998) Cytochrome P450 2E1 in rat tracheobronchial airways: response to ozone exposure. Toxicol. Appl. Pharmacol. 149: 195-202.

Wattiez, R.; Noël-Georis, I.; Cruyt, C.; Broeckaert, F.; Bernard, A.; Falmagne, P. (2003) Susceptibility to oxidative stress: proteomic analysis of bronchoalveolar lavage from ozone-sensitive and ozone-resistant strains of mice. Proteomics 3: 658-665.

Weber, S. U.; Thiele, J. J.; Cross, C. E.; Packer, L. (1999) Vitamin C, uric acid, and glutathione gradients in murine stratum corneum and their susceptibility to ozone exposure. J. Invest. Dermatol. 113: 1128-1132.

Weber, S. U.; Jothi, S.; Thiele, J. J. (2000) High-pressure liquid chromatography analysis of ozone-induced depletion of hydrophilic and lipophilic antioxidants in murine skin. Methods Enzymol. 319: 536-546.

Weber, S. U.; Han, N.; Packer, L. (2001) Ozone: an emerging oxidative stressor to skin. Curr. Probl. in Dermatol. 29: 52-61.

Weller, B. L.; Crapo, J. D.; Slot, J.; Posthuma, G.; Plopper, C. G.; Pinkerton, K. E. (1997) Site- and cell-specific alteration of lung copper/zinc and manganese superoxide dismutases by chronic ozone exposure. Am. J. Respir. Cell Mol. Biol. 17: 552-560.

Weller, B. L.; Witschi, H.; Pinkerton, K. E. (2000) Quantitation and localization of pulmonary manganese superoxide dismutase and tumor necrosis factor α following exposure to ozone and nitrogen dioxide. Toxicol. Sci. 54: 452-461.

Wells, C. A.; Ravasi, T.; Faulkner, G. J.; Carninci, P.; Okazaki, Y.; Hayashizaki, Y.; Sweet, M.; Wainwright, B. J.; Hume, D. A. (2003) Genetic control of the innate immune response. BMC Immunol. 4: 5. Available: http://www.biomedcentral.com/1471-2172/4/5 [18 February, 2003]

Wiester, M. J.; Watkinson, W. P.; Costa, D. L.; Crissman, K. M.; Richards, J. H.; Winsett, D. W.; Highfill, J. W. (1996) Ozone toxicity in the rat. III. Effect of changes in ambient temperature on pulmonary parameters. J. Appl. Physiol. 81: 1691-1700.

Wilkins, C. K.; Clausen, P. A.; Wolkoff, P.; Larsen, S. T.; Hammer, M.; Larsen, K.; Hansen, V.; Nielsen, G. D. (2001) Formation of strong irritants in mixtures of isoprene/ozone and isoprene/ozone/nitrogen dioxide. Environ. Health Perspect. 109: 937-941.

Witschi, H.; Espiritu, I.; Pinkerton, K. E.; Murphy, K.; Maronpot, R. R. (1999) Ozone carcinogenesis revisited. Toxicol. Sci. 52: 162-167.

Wu, Z.-X.; Morton, R. F.; Lee, L.-Y. (1997) Role of tachykinins in ozone-induced airway hyperresponsiveness to cigarette smoke in guinea pigs. J. Appl. Physiol. 83: 958-965.

Yamauchi, T.; Shima, M.; Kuwaki, T.; Ando, M.; Ohmichi, M.; Fukuda, Y.; Adachi, M. (2002) Acute effects of ozone exposure on lung function in mice sensitized to ovalbumin. Toxicology (Ireland) 172: 69-78.

Yu, M.; Pinkerton, K. E.; Witschi, H. (2002) Short-term exposure to aged and diluted sidestream cigarette smoke enhances ozone-induced lung injury in B6C3F1 mice. Toxicol. Sci. 65: 99-106.

Zhang, L.-Y.; Levitt, R. C.; Kleeberger, S. R. (1995) Differential susceptibility to ozone-induced airways hyperreactivity in inbred strains of mice. Exp. Lung Res. 21: 503-518.

Zhao, Q.; Simpson, L. G.; Driscoll, K. E.; Leikauf, G. D. (1998) Chemokine regulation of ozone-induced neutrophil and monocyte inflammation. Am. J. Physiol. 274: L39-L46.

6. CONTROLLED HUMAN EXPOSURE STUDIES OF OZONE AND RELATED PHOTOCHEMICAL OXIDANTS

6.1 INTRODUCTION

In the previous chapter, results of ozone (O_3) studies in laboratory animals and in vitro test systems were presented. The extrapolation of results from animal studies is one mechanism by which information on potential adverse human health effects from exposure to O_3 is obtained. More direct evidence of human health effects due to O_3 exposure can be obtained through controlled human exposure studies of volunteers or through field and epidemiologic studies of populations exposed to ambient O_3 (*see Chapter 7*). Controlled human exposure studies typically use fixed concentrations of O_3 under carefully regulated environmental conditions and subject activity levels. This chapter discusses studies in which volunteers were exposed for up to 8 h to between 0.08 to 0.75 ppm O_3 while at rest or during varying intensities of exercise.

The majority of controlled human studies have investigated the effects of exposure to O_3 in young nonsmoking healthy adults (18 to 35 years of age) performing continuous exercise (CE) or intermittent exercise (IE). Varied combinations of O_3 concentration, exercise routine, and exposure duration have been used in these studies. The responses to ambient O_3 concentrations include decreased inspiratory capacity; mild bronchoconstriction; rapid, shallow breathing patterns during exercise; and symptoms of cough and pain on deep inspiration. Reflex inhibition of inspiration results in a decrease in forced vital capacity (FVC) and total lung capacity (TLC) and, in combination with mild bronchoconstriction, contributes to a decrease in the forced expiratory volume in 1 s (FEV_1). In addition to physiological pulmonary responses and respiratory symptoms, O_3 has been shown to result in airway hyperresponsiveness, epithelial permeability, and inflammation.

The most salient observations from studies reviewed in the 1996 EPA Ozone Air Quality Criteria Document or O_3 AQCD (U.S. Environmental Protection Agency, 1996) were that: (1) young healthy adults exposed to O_3 concentrations $\geq$0.08 ppm develop significant reversible, transient decrements in pulmonary function if minute ventilation ($\dot{V}_E$) or duration of exposure is

increased sufficiently, (2) children experience similar spirometric responses but lesser symptoms from O_3 exposure relative to young adults, (3) O_3-induced spirometric responses are decreased in the elderly relative to young adults, (4) there is a large degree of intersubject variability in physiologic and symptomatic responses to O_3 but responses tend to be reproducible within a given individual over a period of several months, and (5) subjects exposed repeatedly to O_3 for several days develop a tolerance to successive exposures, as demonstrated by an attenuation of responses, which is lost after about a week without exposure.

There are several important limitations associated with these clinical studies: (1) the ability to study only short-term, acute effects; (2) difficulties in trying to link short-term effects with long-term consequences; (3) the use of a small number of volunteers that may not be representative of the general population; and (4) the statistical limitations associated with the small sample size. Sample size affects the power of a study, and having a small number of samples causes a risk of Type II error, i.e., the incorrect conclusion that no difference exists between treatments or groups when comparisons are not significantly different. This affects the confidence in estimates of a minimum O_3 concentration at which some degree of pulmonary impairment will occur in both the general population and susceptible subpopulations. As a result, the conclusions drawn from many of the studies cited in this chapter may underestimate the presence of responses at low O_3 concentrations and low activity levels.

Most of the scientific information summarized in this chapter comes from the literature published since the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). In addition to further study of physiological pulmonary responses and symptoms of breathing discomfort, much of this literature has focused on mechanisms of inflammation and cellular responses to injury induced by O_3 inhalation. A more thorough discussion and review of this literature appears in Annex AX6 of this document. In summarizing the literature, effects are described if they are statistically significant at a probability (p-value) of less than 0.05; otherwise, trends are noted as such.

As spirometry typically *improves* in healthy young adults with exercise exposures to filtered air (FA), the term "O_3-induced" is used herein and in the annex to designate effects that have been corrected for responses during FA exposures. For healthy adults, an O_3-induced change in lung function is the difference between the *decrement* experienced with O_3 exposure and the *improvement* observed with FA exposure. However, the distinction between an O_3-

induced change and a post- versus preexposure change is particularly important in individuals with respiratory disease who may experience exercise-induced *decrements* in pulmonary function during both FA and O_3 exposures. Hence, in subjects with respiratory disease, exercise-induced responses could be mistaken for O_3-induced responses in the absence of a correction for FA responses.

6.2 PULMONARY FUNCTION EFFECTS OF OZONE EXPOSURE IN HEALTHY SUBJECTS

6.2.1 Introduction

As reviewed in the 1986 and 1996 O_3 AQCD's (U.S. Environmental Protection Agency, 1986, 1996), 0.5 ppm is the lowest O_3 concentration at which statistically significant reductions in FVC and FEV_1 have been reported in sedentary subjects. On average, young adults (n = 23; mean age, 22 yrs) exposed at rest for 2 h to 0.5 ppm O_3 had O_3-induced decrements of ~4% in FVC and ~7% in FEV_1 (Folinsbee et al., 1978; Horvath et al., 1979). During exercise, spirometric and symptoms responses are observed at lower O_3 concentrations. For acute exposures of 2 h or less to ≥0.12 ppm O_3 , if $\dot{V}_E$ is sufficiently increased by exercise, healthy human subjects generally experience decreases in TLC, inspiratory capacity (IC), FVC, FEV_1, mean forced expiratory flow from 25% to 75% of FVC (FEF_{25-75}), and tidal volume (V_T) and increases in specific airways resistance (sRaw), breathing frequency (f_B), and airway responsiveness. These exposures also cause symptoms of cough, pain on deep inspiration, shortness of breath, throat irritation, and wheezing. With exposures of 4- to 8-h in duration, statistically significant pulmonary function and symptoms responses are observed at lower O_3 concentrations and lower $\dot{V}_E$ than in shorter duration studies.

6.2.2 Acute Exposure for Up to 2 h

With heavy CE ($\dot{V}_E$ = 89 L/min), an O_3-induced decrement of 9.7% in FEV_1 has been reported for healthy young adults (n = 17; age, 24 ± 3 yrs) exposed for only 1 h to 0.12 ppm O_3 (Gong et al., 1986). With moderate-to-heavy IE (15 min intervals of rest and exercise [$\dot{V}_E$ = 68 L/min]), McDonnell et al. (1983) reported a physiologically small, but

significant, O_3-induced decrement of 3.4% in FEV_1 for young healthy adults (n = 22, age, 22 ± 3 yrs) exposed for 2 h to 0.12 ppm O_3. Using the same 2 h IE exposure protocol, Linn et al. (1986) found no statistically significant spirometic responses at O_3 concentrations of 0.16 ppm and lower. However, the subjects in the Linn et al. (1986) study were potentially exposed concurrently in Los Angeles to ambient O_3 levels of between 0.12 and 0.16 ppm and were on average 3 yrs older than the subjects in the McDonnell et al. (1983) study. (*The attenuating effects of increasing age and repeated O_3 exposures are discussed in Sections 6.5.1 and 6.6, respectively.*) The disparities between the Linn et al. (1986) and McDonnell et al. (1983) studies demonstrate the difficulty in determining a no-effect-level for O_3 based on relatively small study populations.

Studies analyzing large data sets (≥300 subjects) provide better predictive ability of acute changes in FEV_1 at low levels of O_3 and $\dot{V}_E$ than possible via comparisons between smaller studies. Such an analysis was performed by McDonnell et al. (1997), who examined FEV_1 responses in 485 healthy white males (18 to 36 years of age; subjects recruited from the area around Chapel Hill, NC) exposed once for 2 h to O_3 concentrations of up to 0.40 ppm at rest or with IE. Decrements in FEV_1 were modeled by sigmoid-shaped curve as a function of subject age, O_3 concentration, $\dot{V}_E$, and duration of exposure. Regarding applicability to the general population, the McDonnell et al. (1997) model has an apparent limitation of considering only data for white males. However, two other large studies (n = 372; 18 to 35 yrs of age; subjects recruited from the area around Chapel Hill, NC) found no significant gender or race effects on spirometric responses to O_3 exposure (Seal et al., 1993, 1996).

Ultman et al. (2004) recently reported pulmonary responses in 60 young heathy nonsmoking adults (32 M, 28 F) exposed to 0.25 ppm O_3 for 1 h with CE at a target $\dot{V}_E$ of 30 L/min. Consistent with findings reported in the 1996 O_3 criteria document, considerable intersubject variability in FEV_1 decrements was reported by Ultman et al. (2004) with responses ranging from a 4% improvement to a 56% decrement. One-third of the subjects had FEV_1 decrements of >15% and 7% of the subjects had decrements of >40%. It should be pointed out that the McDonnell et al. (1997) model predicts only *average* responses. In a more recent study, McDonnell et al. (1999) also reported a model predicting average symptom responses from O_3 exposure. Unfortunately, neither of these papers (McDonnell et al., 1997, 1999) provide

predictions of intersubject variability in response. (*Section 6.4 of this Chapter discusses intersubject variability in response to O_3 exposure*).

In addition to the overt effects of O_3 exposure on the large airways as indicated by spirometric responses, O_3 exposure also affects the function of the small airways and parenchymal lung. Foster et al. (1993, 1997) examined the effect of O_3 on ventilation distribution in healthy adult males. In healthy nonsmoking males (26.7 ± 7 years old) exposed to FA or 0.33 ppm O_3 for 2 h with IE, there was a significant reduction in the ventilation to the lower-lung (31% of lung volume) and significant increases in ventilation to the upper- and middle-lung regions relative to the FA values in 7 of the 9 subjects (Foster et al., 1993). In another study, 15 healthy nonsmoking males (25.4 ± 2 years old) were exposed to FA or 0.35 ppm O_3 for 2.2 h with IE (Foster et al., 1997). Following O_3 exposure, an inert gas washout was delayed and resembled a two-compartment washout, whereas pre-O_3 exposure a log-linear gas clearance as a function of expired volume resembled a single-compartment washout. The pronounced slow phase of gas washout occurring post-O_3, represented a 24% decrease in the washout rate relative to pre-O_3. At 24-h post-O_3, 6 of the 12 subjects still had [or developed] a delayed washout relative to the pre-O_3 maneuver. This suggests a prolonged O_3 effect on the small airways and ventilation distribution in some individuals.

6.2.3 Prolonged Ozone Exposures

In the exposure range of 0.08 to 0.16 ppm O_3, a number of studies using moderate quasi continuous exercise (QCE; 50 min exercise and 10 min rest per h) for 4 to 8 h have shown significant responses under the following conditions: 0.16 ppm for 4 h with QCE at $\dot{V}_E \approx 40$ L/min (Folinsbee et al., 1994), 0.08 to 0.12 ppm for 6.6 h with QCE at $\dot{V}_E \approx 35$ to 40 L/min (Adams, 2002; Adams, 2003a; Folinsbee et al., 1988; Horstman et al., 1990), and 0.12 ppm for 8 h of IE (30 min per h) at $\dot{V}_E \approx 40$ L/min (Hazucha et al., 1992). Symptoms and spirometric responses increased with duration of exposure, O_3 concentration, and total $\dot{V}_E$. Airway resistance is only modestly affected with moderate or even heavy exercise combined with O_3 exposure (Folinsbee et al., 1978; McDonnell et al., 1983; Seal et al., 1993).

6.2.3.1 Effect of Exercise Ventilation Rate on FEV_1 Response to 6.6 h Ozone Exposure

It is well established that response to O_3 exposure is a function of $\dot{V}_E$ in studies of 2 h or less in duration (*See Section AX6.2.2*). It is reasonable to expect that response to a prolonged 6.6-h O_3 exposure is also a function of $\dot{V}_E$, although quantitative analyses are lacking. Data from five similar prolonged exposure studies are available for evaluation of FEV_1 responses as a function of exercise $\dot{V}_E$ (Adams, 2000; Adams and Ollison, 1997; Folinsbee et al., 1988, 1994; Horstman et al., 1990). Each of these studies exposed similarly aged subjects (mean ages 22 to 25 yrs) to 0.12 ppm O_3 for 6.6 h. In total, ten sets of mean FEV_1 decrements were available for exercise $\dot{V}_E$ ranging from 20 to 43 L/min, although no data were available for $\dot{V}_E$ between 20 and 30 L/min (*data illustrated in Figure AX6-2*). As in 2 h exposure studies, FEV_1 decrements are a function of $\dot{V}_E$ in prolonged 6.6-h exposure studies as demonstrated by a significant correlation between these variables (Pearson, $r = 0.95$, $p < 0.001$; Spearman, $r = 0.84$, $p < 0.01$).

6.2.3.2 Exercise Ventilation Rate as a Function of Body/Lung Size on FEV_1 Response to 6.6 h Ozone Exposure

Based on the assumption that the total inhaled O_3 dose (product of O_3 concentration, exposure duration, and $\dot{V}_E$) is proportional to the lung size, exercise $\dot{V}_E$ is typically selected to be a multiple of body surface area (BSA) or FVC. Data from several recent studies do not support the contention that $\dot{V}_E$ should be normalized. In an analysis of data from 485 young adults, McDonnell et al. (1997) found that any effect of BSA, height, or baseline FVC on percent decrement in FEV_1 was small to nonexistent. This is consistent with Messineo and Adams (1990), who compared pulmonary function responses in young adult women having small ($n = 14$) or large ($n = 14$) lung sizes (mean FVC of 3.74 and 5.11 L, respectively) and found no significant group difference in FEV_1 decrements. For 30 subjects (15M, 15F) exposed to 0.12 ppm O_3 for 6.6 h, Adams (2000) also reported that FEV_1 responses were more closely related to $\dot{V}_E$ than to $\dot{V}_E$ normalized to BSA. The O_3 dosimetry study of Bush et al. (1996) suggested that normalization of the O_3 dose might more appropriately be a function of anatomic dead space. Ozone penetrates deeper into the lungs of individuals with larger conducting airway

volumes, however, FEV_1 responses in subjects exposed for 2 h to 0.25 ppm O_3 did not appear to be associated with O_3 uptake (Ultman et al., 2004).

6.2.3.3 Comparison of 2 h IE to 6.6 h O_3 Exposure Effects on Pulmonary Function

Adams (2003b) examined whether prolonged 6.6-h QCE exposure to a relatively low O_3 concentration (0.08 ppm) and the 2-h IE exposure at a relatively high O_3 concentration (0.30 ppm) elicited consistent individual subject FEV_1 responses. Individual subject O_3 exposure reproducibility was first examined via a regression plot of the postexposure FEV_1 response to the 6.6-h chamber exposure as a function of postexposure FEV_1 response to the 2-h IE chamber exposure. The R^2 of 0.40, although statistically significant, was substantially less than that observed in a comparison of individual FEV_1 response to the two 2-h IE exposures by chamber and face mask, respectively ($R^2 = 0.83$). The Spearman rank order correlation for the chamber 6.6-h and chamber 2-h exposure comparison was also substantially less (0.49) than that obtained for the two 2-h IE exposures (0.85). The primary reason for the greater variability in the chamber 6.6-h exposure FEV_1 response as a function of that observed for the two 2-h IE exposures is very likely related to the increased variability in response upon repeated exposure to O_3 concentrations lower than 0.18 ppm ($R = 0.57$, compared to a mean R of 0.82 at higher concentrations) reported by McDonnell et al. (1985a). This rationale is supported by the lower r^2 (0.40) observed by Adams (2003b) for the FEV_1 responses found in 6.6 h chamber and face mask exposures to 0.08 ppm O_3, compared to an r^2 of 0.83 observed for responses found at 0.30 ppm O_3.

6.2.4 Triangular Ozone Exposures

To further explore the factors that determine responsiveness to O_3, Hazucha et al. (1992) designed a protocol to examine the effect of varying, rather than constant, O_3 concentrations. Subjects were exposed to an O_3 level that increased linearly from 0 to 0.24 ppm for the first 4 h and then decreased linearly from 0.24 to 0 ppm over the second 4 h of the 8 h exposure (triangular concentration profile) and to a constant level exposure of 0.12 ppm O_3 for 8 h. While total inhaled O_3 doses for the constant and the triangular concentration profile were almost identical, the FEV_1 response was dissimilar. For the constant 0.12 ppm O_3 exposure, FEV_1 declined ~5% by the fifth hour and then remained at that level. With the triangular O_3

concentration profile, there was minimal FEV_1 response over the first 3 h followed by a rapid decrease in FEV_1 (−10.3%) over the next 3 h. During the seventh and eighth hours, mean FEV_1 decrements improved to −6.3% as the O_3 concentration decreased from 0.12 to 0.00 ppm (mean = 0.06 ppm).

More recently, Adams (2003a) used a less abrupt triangular O_3 exposure profile at concentrations assumed to be typical of outdoor ambient conditions (beginning at 0.03 ppm, increasing steadily to 0.15 ppm in the fourth hour and decreasing steadily to 0.05 ppm at 6.6 h (mean = 0.08 ppm). Postexposure values for FEV_1 and symptoms were not significantly different between the 6.6 h triangular and a square-wave 0.08 ppm O_3 exposure. During the triangular exposure, however, FEV_1 responses became statistically significant after 4.6 h, whereas, they were not significant until 6.6 h during the square wave exposure (Adams, 2003a). Perhaps due to the lower O_3 concentrations, evidence of FEV_1 response recovery with the triangular exposure was less pronounced than as observed by Hazucha et al. (1992). Figure 6-1 illustrates the average O_3-induced FEV_1 responses and the O_3 exposure schemes for the Adams (2003a) and Hazucha et al. (1992) studies. For completeness, other studies have also used a triangular exposure profile but for a shorter duration of only 130 minutes (Foster and Stetkiewicz, 1996; Foster et al., 1996).

With square-wave O_3 exposures between 0.08 to 0.12 ppm, FEV_1 decrements may increase with time of exposure (and O_3 dose) or reach a plateau (Horstman et al., 1990; McDonnell et al., 1991). For the triangular exposures used by Hazucha et al. (1992) and Adams (2003a), maximal FEV_1 responses occurred 1 h to 2 h after peak O_3 concentration and 1 h to 2 h before the maximal O_3 dose occurred (at the end of the O_3 exposure). These two studies suggest that depending upon the profile of the exposure, the triangular exposure can potentially lead to higher FEV_1 responses than square wave exposures at overall equivalent ozone doses.

6.2.5 Mechanisms of Pulmonary Function Responses

Inhalation of O_3 for several hours while physically active elicits both subjective respiratory tract symptoms and acute pathophysiologic changes. The typical symptomatic response consistently reported in studies is that of tracheobronchial airway irritation. Depending on the individual's responsiveness to O_3, this is accompanied by several pathophysiologic changes such as decrements in lung capacities and volumes, bronchoconstriction, airway hyperresponsiveness,

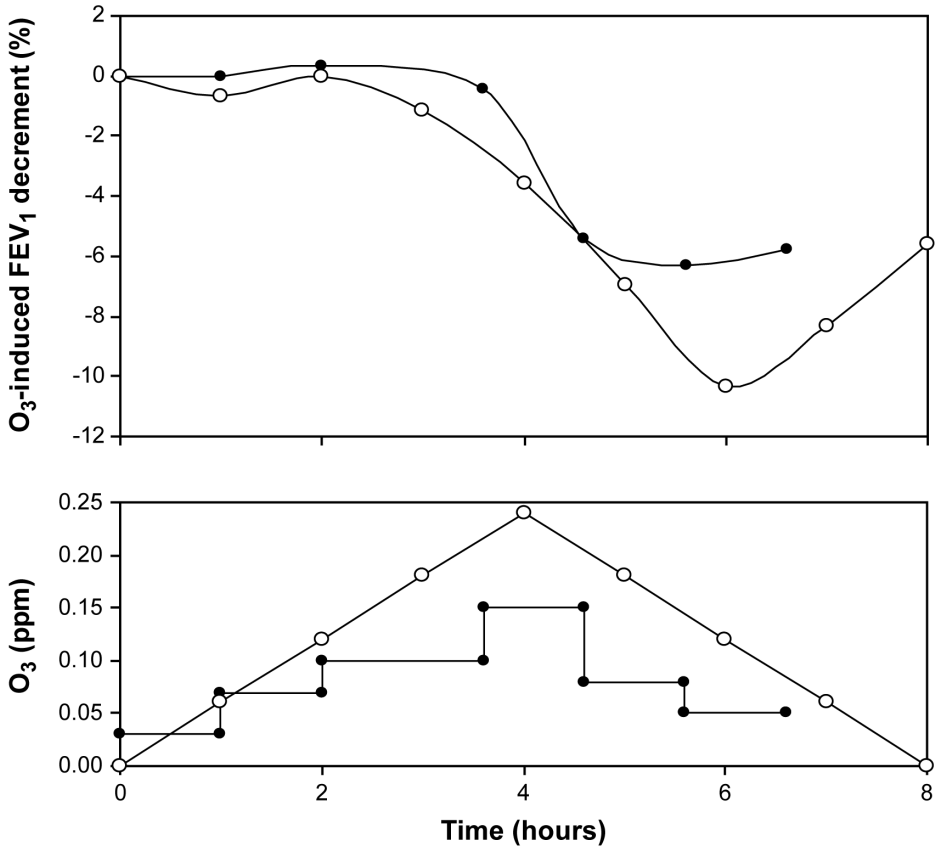


Figure 6-1. Triangular exposure profile – O_3-induced FEV_1 decrements (top panel) and O_3 concentrations (bottom panel) as a function of exposure duration. Open (○) and closed (●) circles illustrate average data from Hazucha et al. (1992) and Adams (2003a), respectively. For clarification, the "O_3-induced FEV_1 decrement" is the FEV_1 response following O_3 exposure minus the FEV_1 response following FA.

airway inflammation, immune system activation, and epithelial injury. The severity of symptoms and the magnitude of response depend on inhaled dose, O_3 sensitivity of an individual, and the extent of tolerance resulting from the individual's previous exposures. The development of effects is time-dependent during both exposure and recovery periods with considerable overlap of evolving and receding effects. The time sequence, magnitude and the type of responses of this complex series of events, both in terms of development and recovery, indicate that several mechanisms, activated at different times of exposure, must contribute to the overall lung function response (U.S. Environmental Protection Agency, 1996).

Available information on recovery from O_3 exposure indicates that an initial phase of recovery proceeds relatively rapidly, and some 40 to 65% of the acute spirometric and symptom response appears to occur within about 2 h (Folinsbee and Hazucha, 1989). Following a 2 h exposure to 0.4 ppm O_3 with IE, Nightingale et al. (2000) observed a 13.5% decrement in FEV_1. By 3 h postexposure, however, only a 2.7% FEV_1 decrement persisted as illustrated in Figure 6-2. A similar postexposure recovery in FVC was also observed. Gerrity et al. (1993) suggested that for healthy young adults transient increases in mucus clearance (mediated by cholinergic receptors) due to O_3 exposure may be coincident to pulmonary function responses, i.e., the transient increases in clearance and decrements in lung function return to baseline values within 2 to 3 h postexposure. However, there is some indication that the spirometric responses, especially at higher O_3 concentrations, are not fully recovered within 24 h (Folinsbee and Horvath, 1986; Folinsbee et al., 1998). In hyperresponsive individuals, the recovery takes longer, as much as 48 hours, to return to baseline values. Collectively, these observations suggest that there is a rapid recovery of O_3-induced spirometric responses and symptoms, which may occur during resting exposure to O_3 (Folinsbee et al., 1977) or as O_3 concentration is reduced during exposure (Hazucha et al., 1992), and a slower phase, which may take at least 24 h to complete (Folinsbee and Hazucha, 2000). Repeated exposure studies at higher concentrations typically show that FEV_1 response to O_3 is enhanced on the second of several days of exposure (Table AX6-8). This enhanced response suggests a residual effect of the previous exposure, about 22 h earlier, even though the preexposure spirometry may be the same as on the previous day. The absence of the enhanced response with repeated exposure at lower O_3 concentrations may be the result of a more complete recovery or less damage to pulmonary tissues (Folinsbee et al., 1994).

6.2.5.1 Pathophysiologic Mechanisms

Breathing pattern changes

Human studies consistently report that inhalation of O_3 alters the breathing pattern without significantly affecting minute ventilation. A progressive decrease in tidal volume and a "compensatory" increase in frequency of breathing to maintain steady minute ventilation during exposure suggests a direct modulation of ventilatory control. These changes parallel a response of many animal species exposed to O_3 and other lower airway irritants (Tepper et al., 1990).

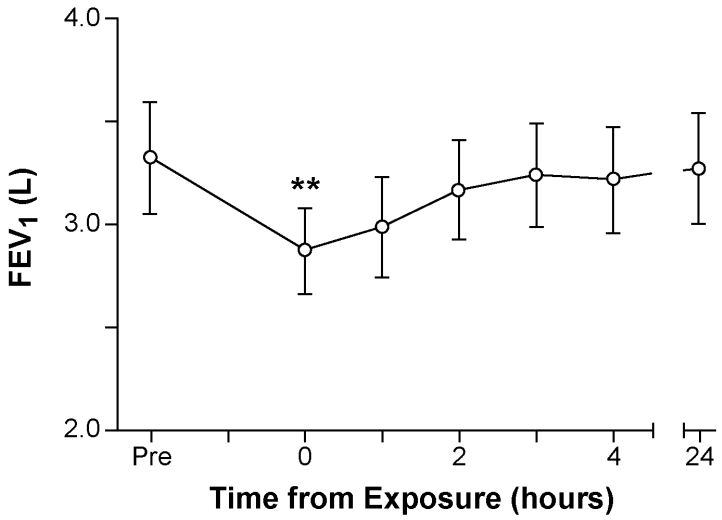


Figure 6-2. Recovery of FEV_1 responses following a 2 h exposure to 0.4 ppm O_3 with IE. Immediately postexposure, FEV_1 was significantly (p < 0.001) decreased. At 3 h postexposure, FEV_1 was at 97% of the preexposure value.**

Adapted from Nightingale et al. (2000).

Bronchial C-fibers and rapidly adapting receptors appear to be the primary vagal afferents responsible for O_3-induced changes in ventilatory rate and depth in both humans (Folinsbee and Hazucha, 2000) and animals (Coleridge et al., 1993; Hazucha and Sant'Ambrogio, 1993; Schelegle et al., 1993).

The potential modulation of breathing pattern by activation of sensory afferents located in extrathoracic airways by O_3 has not yet been studied in humans. Nasal only O_3 exposure of rats produces changes in breathing pattern that are similar to changes observed in humans (Kleinman et al., 1999).

Symptoms and lung function changes

As discussed, in addition to changes in ventilatory control, O_3 inhalation by humans will also induce a variety of symptoms, reduce vital capacity (VC) and related functional measures, and increase airway resistance.

Schelegle et al. (2001) demonstrated that the reduction in VC due to O_3 exposure is a reflex action and not a voluntary termination of inspiration as result of discomfort. They reported

that O_3-induced symptom responses (mediated in part by bronchial C-fibers) are substantially reduced by inhaled topical anesthetic. However, the anesthetic had a minor and irregular effect on pulmonary function decrements and tachypnea. Since respiratory symptom responses were largely abolished, these findings support reflex inhibition of VC due to stimulation of both bronchial and pulmonary C-fibers.

The involvement of nociceptive bronchial C-fibers modulated by opioid receptors in limiting maximal inspiration and eliciting subjective symptoms in humans was studied by Passannante et al. (1998). Sufentanil (an opioid agonist and analgesic) rapidly reversed O_3-induced symptom responses and reduced spirometric decrements in "strong" responders. The incomplete recovery in FEV_1 following sufentanil administration, however, suggests involvement of non-opioid receptor modulated mechanisms as well. Interestingly, naloxone (opioid receptor antagonist) had no significant effect on FEV_1 decrements in "weak" responders. Plasma levels of β-endorphin (a potent pain suppressor) were not related with O_3 responses.

Airway hyperreactivity

In addition to limitation of maximal inspiration and its effects on other spirometric endpoints, activation of airway sensory afferents also plays a role in receptor-mediated bronchoconstriction and an increase in airway resistance. Despite this common mechanism, post-O_3 pulmonary function changes and either early or late bronchial hyperresponsiveness (BHR) to inhaled aerosolized methacholine or histamine are poorly correlated either in time or magnitude. Fentanyl and indomethacin, the drugs that have been shown to attenuate O_3-induced lung function decrements in humans, did not prevent induction of BHR when administered to guinea pigs prior to O_3 exposure (Yeadon et al., 1992). Neither does post-O_3 BHR seem to be related to airway baseline reactivity. These findings imply that the mechanisms are either not related or are activated independently in time. Animal studies (with limited support from human studies) have suggested that an early post-O_3 BHR is, at least in part, vagally mediated (Freed, 1996) and that stimulation of C-fibers can lead to increased responsiveness of bronchial smooth muscle independently of systemic and inflammatory changes which may be even absent (Joad et al., 1996). In vitro study of isolated human bronchi have reported that O_3-induced airway sensitization involves changes in smooth muscle excitation-contraction coupling (Marthan,

1996). Characteristic O_3-induced inflammatory airway neutrophilia which at one time was considered a leading BHR mechanism, has been found in a murine model, to be only coincidentally associated with BHR, i.e., there was no cause and effect relationship (Zhang et al., 1995). However, this observation does not rule out involvement of other cells such as eosinophils or T-helper cells in BHR modulation. There is some evidence that release of inflammatory mediators by these cells can sustain BHR and bronchoconstriction. In vitro and animal studies have also suggested that airway neutral endopeptidase activity can be a strong modulator of BHR (Marthan et al., 1996; Yeadon et al., 1992). Late BHR observed in some studies is plausibly due to a sustained damage of the airway epithelium and continual release of inflammatory mediators (Foster et al., 2000). Thus, O_3-induced BHR appears to be a product of many mechanisms acting at different time periods and levels of the bronchial smooth muscle signaling pathways (*The effects of O_3 on BHR are described in Section 6.8*).

6.2.5.2 Mechanisms at a Cellular and Molecular Level

Stimulation of vagal afferents by O_3 and reactive products, the primary mechanism of lung function impairment, is enhanced and sustained by what can be considered in this context to be secondary mechanisms activated at a cellular and molecular level. The complexity of these mechanisms is beyond the scope of this section and the reader is directed to Section 6.9 of this chapter for greater detail. A comprehensive review by Mudway and Kelly (2000) discusses the cellular and molecular mechanisms of O_3-induced pulmonary response in great detail.

Stimulation of bronchial C-fibers by O_3 not only inhibits maximal inspiration but, through local axon reflexes, induces neurogenic inflammation. This pathophysiologic process is characterized by release of tachykinins and other proinflammatory neuropeptides. Ozone exposure has been shown to elevate the C-fiber-associated tachykinin, substance P, in human bronchial lavage fluid (Hazbun et al. 1993) and to deplete neuropeptides synthesized and released from C-fibers in human airway epithelium rich in substance P-immunoreactive axons. Substance P and other transmitters are known to induce granulocyte adhesion and subsequent transposition into the airways, increase vascular permeability and plasma protein extravasation, cause bronchoconstriction, and promote mucus secretion (Solway and Leff, 1991). Although the initial pathways of neurogenic, antigen-induced, and innate immune-mediated inflammation are not the same, they eventually converge leading to further amplification of airway inflammatory

processes by subsequent release of cytokines, eicosanoids, and other mediators. Significantly negative correlations between O_3-induced leukotriene ($LTC_4/D_4/E_4$) production and spirometric decrements (Hazucha et al., 1996), and an increased level of postexposure PGE_2, a mediator known to stimulate bronchial C-fibers, show that these mediators play an important role in attenuation of lung function due to O_3 exposure (Mohammed et al., 1993; Hazucha et al., 1996). Moreover, because the density of bronchial C-fibers is much lower in the small than large airways, the reported post O_3 dysfunction of small airways assessed by decrement in FEF_{25-75} (Weinman et al., 1995; Frank et al., 2001) may be due in part to inflammation. Also, because of the relative slowness of inflammatory responses as compared to reflex effects, O_3-triggered inflammatory mechanisms are unlikely to initially contribute to progressive lung function reduction. It is plausible, however, that when fully activated, they sustain and possibly further aggravate already impaired lung function. Indeed, a prolonged recovery of residual spirometric decrements following the initial rapid improvement after exposure termination could be due to slowly resolving airway inflammation. Bronchial biopsies performed 6 h postexposure have shown that O_3 caused a significant decrease in immunoreactivity to substance P in the submucosa (Krishna et al., 1997). A strong negative correlation with FEV_1 also suggests that the release of substance P may be a contributing mechanism to persistent post-O_3 bronchoconstriction (Krishna et al., 1997). Persistent spirometry changes observed for up to 48 h postexposure could plausibly be sustained by the inflammatory mediators, many of which have bronchoconstrictive properties (Blomberg et al., 1999).

6.3 SUBJECTS WITH PREEXISTING DISEASE

Individuals with respiratory disease are of primary concern in evaluating the health effects of O_3 because even a small change in function is likely to have more impact on a person with reduced reserve, i.e., O_3-induced effects are superimposed on preexisting pulmonary impairment.

6.3.1 Subjects with Chronic Obstructive Pulmonary Disease

For patients with COPD performing light to moderate IE, no decrements in pulmonary function were observed after 1- and 2-h exposures to $\leq$0.30 ppm O_3 (Kehrl et al., 1985; Linn et al., 1982a, 1983a; Solic et al., 1982), and only small decreases in forced expiratory volume

were observed for 3-h exposures of chronic bronchitics to 0.41 ppm O_3 (Kulle et al., 1984). More recently, Gong et al. (1997a) found no significant difference in response between age-matched controls and COPD patients to a 4 h exposure to 0.24 ppm O_3 with IE. Although the clinical significance is uncertain, small transient decreases in arterial blood oxygen saturation have also been observed in some of these studies.

6.3.2 Subjects with Asthma

Based on studies reviewed in the 1996 criteria document (U.S. Environmental Protection Agency, 1996), asthmatic subjects appear to be at least as sensitive to acute effects of O_3 as healthy nonasthmatic subjects.

Several recent studies support a tendency for slightly increased spirometric responses in mild asthmatic versus healthy subjects. Alexis et al. (2000) reported reductions in FVC (12%, 10%) and FEV_1 (13%, 11%) for 13 mild asthmatic and 9 healthy subjects, respectively, exposed to 0.4 ppm O_3 for 2 h with IE ($\dot{V}_E$ = 30 L/min). The FVC and FEV_1 responses were attenuated by indomethacin in the healthy subjects but not the asthmatics. As assessed by the magnitude of reductions in mid-flows (viz. FEF_{25}, FEF_{50}, FEF_{60p}, FEF_{75}) following O_3 exposure, the small airways tended to be more affected in asthmatics than healthy subjects. In a larger study, Jörres et al. (1996) exposed 24 asthmatics, 12 allergic rhinitics, and 10 healthy subjects to 0.25 ppm O_3 for 3 h with IE. The O_3-induced FEV_1 decrements tended to be greater in the diseased populations (allergic rhinitis, 14.1%; asthmatic, 12.5%; healthy controls, 10.2%). Scannell et al. (1996) exposed 18 asthmatics to 0.2 ppm O_3 for 4 h with IE ($\dot{V}_E \approx 25$ L/min/m^2 BSA). An O_3-induced increase in sRaw tended to be greater in the asthmatics compared to 81 healthy subjects who underwent similar experimental protocols (Aris et al., 1995; Balmes et al., 1996).

Increased sensitivity of asthmatics to O_3 was also demonstrated in the epidemiological study by Höppe et al. (2003). Relevant pulmonary function responses (>10% drop in FEV_1, FVC, or PEF, and/or >20% increase in sRaw) subsequent to O_3 exposure were experienced by 22 of 43 young asthmatics (mean age, 15 yrs) versus only 6 of 43 young athletes (mean age, 18 yrs). Participants were asked to engage in their normal activities for 2 h in the afternoon (61-62 ppb O_3, on average) prior to pulmonary function testing. The estimated activity level during O_3 exposures was lower in the asthmatics ($\dot{V}_E \approx 25$ L/min) than the athletes ($\dot{V}_E \approx 80$ L/min). As discussed in Sections 6.2.2 and 6.2.3.1, responses to O_3 increase

with $\dot{V}_E$. Hence, in the absence of some underlying susceptibility to adverse O_3 effects, the asthmatics would actually be expected to respond far less than the athletes who had a 3.2-fold greater $\dot{V}_E$.

Similar O_3-induced spirometric responses are suggested by some studies. The Scannell et al. (1996) study of 18 asthmatics reported FEV_1 and FVC decrements that were similar to 81 healthy subjects (Aris et al., 1995; Balmes et al., 1996). Similar group decrements in FEV_1 and FVC were reported by Hiltermann et al. (1995), who exposed 6 asthmatics and 6 healthy subjects to 0.4 ppm O_3 for 2 h with light IE. Basha et al. (1994) also reported similar spirometric responses between 5 asthmatic and 5 healthy subjects exposed to 0.2 ppm O_3 for 6 h with IE. The lack of significant differences in the Hiltermann et al. (1995) and Basha et al. (1994) studies is not compelling given the extremely small sample sizes and corresponding lack of statistical power. The Basha et al. (1994) study was also confounded by the asthmatics having an average preexposure FEV_1 that was about 430 mL lower (a 12% difference) on the O_3-day relative to the air-day. Hence, only the Scannell et al. (1996) study supports similar O_3-induced spirometric responses in asthmatics versus healthy subjects.

One study has reported that asthmatics tend to have smaller O_3-induced FEV_1 decrements relative healthy subjects (3% versus 8%, respectively) when exposed to 0.2 ppm O_3 for 2 h with IE (Mudway et al., 2001). However, the asthmatics in the Mudway et al. (2001) study also tended to be older than the healthy subjects, which could partially explain their lesser response.

In a longer exposure duration (7.6 h) study, Horstman et al. (1995) exposed 17 mild-to-moderate asthmatics and 13 healthy controls to 0.16 ppm O_3 or FA with quasi continuous exercise ($\dot{V}_E \approx 30$ L/min). The FEV_1 decrement observed in the asthmatics was significantly greater than in the healthy subjects (19% versus 10%, respectively). There was also tendency for a greater O_3-induced decrease in FEF_{25-75} in asthmatics relative to the healthy subjects (24% versus 15%, respectively). A significant positive correlation in asthmatics was also reported between O_3-induced spirometric responses and baseline lung function, i.e., responses increased with severity of disease.

With repeated O_3 exposures asthmatics, like healthy subjects (*see Section 6.6*), develop tolerance. Gong et al. (1997b) exposed 10 asthmatics to 0.4 ppm O_3, 3 h per day with IE ($\dot{V}_E \approx 32$ L/min), for 5 consecutive days. Symptom and spirometric responses were greatest on the first (-35 % FEV_1) and second (-34 % FEV_1) exposure days, and progressively

diminished toward baseline levels (−6 % FEV_1) by the fifth exposure day. Similar to healthy subjects, asthmatics lost their tolerance 4 and 7 days later.

Some, but not all, studies have reported that asthmatics have a somewhat exaggerated inflammatory response to acute O_3 exposure relative to healthy controls (e.g., McBride et al., 1994; Basha et al., 1994; Peden et al., 1995, 1997; Peden, 2001a; Scannell et al., 1996; Hiltermann et al., 1997, 1999; Michelson et al., 1999; Vagaggini et al., 1999; Newson et al., 2000; Holz et al., 2002) also (*see Section 6.9 and Tables AX6-3 and -12*). For example, at 18-h post-O_3 exposure (0.2 ppm, 4-h with IE) and corrected for FA responses, Scannell et al. (1996) found significantly increased neutrophils in 18 asthmatics (12%) compared to 20 healthy subjects (4.5%). This inflammatory response difference was observed despite no group differences in spirometric responses to O_3. Inflammatory responses do not appear to be correlated with lung function responses in either asthmatic or healthy subjects (Balmes et al., 1996, 1997; Holz et al., 1999). This lack of correlations between inflammatory and spirometric responses may be due to differences in the time kinetics of these responses (Stenfors et al., 2002). In addition, airway responsiveness to inhaled allergens is increased by O_3 exposure in subjects with allergic asthma for up to 24 h (*see Section 6.8*).

6.3.3 Subjects with Allergic Rhinitis

Allergic rhinitis is a condition defined by inflammation of the nasal membranes. Nayak (2003) recently reviewed the commonalities between asthma and allergic rhinitis. Clinically, greater than 60% of asthmatics have allergic rhinitis and slightly less than 40% of allergic rhinitics have asthma. Leukotrienes and histamine are well-recognized mediators of responses (viz., inflammation, hyperresponsiveness, and bronchoconstriction) in both asthma and allergic rhinitis. Although, rhinitis and asthma are distinguished as affecting the upper and lower airways, respectively, it has been suggested that these diseases are manifestations of the same disease entity.

Given the prevalence of concomitant asthma and rhinitis and their common response mediators, it should be expected that allergic rhinitics might respond more similarly to asthmatics than healthy individuals. Regarding spirometric responses, Jörres et al. (1996) provide the only data demonstrating a trend in support of this supposition.

Studies demonstrating the interaction between air pollutants and allergic processes in the human nasal airways and rhinoconjunctival tissue have been reviewed by Peden (2001b) and Riediker et al. (2001), respectively. Ozone exposure of subjects with allergic rhinitis has been shown to induce nasal inflammation and increase airway responsiveness to nonspecific bronchoconstrictors.

Peden et al. (1995), who studied allergic asthmatics exposed to O_3, found that O_3 causes an increased response to nasal allergen challenge in addition to nasal inflammatory responses. Their data suggested that allergic subjects have an increased immediate response to allergen after O_3 exposure. In a follow-up study, Michelson et al. (1999) reported that 0.4 ppm O_3 did not promote early-phase-response mediator release or enhance the response to allergen challenge in the nasal airways of mild, asymptomatic dust mite-sensitive asthmatic subjects. Ozone did, however, promote an inflammatory cell influx, which helps induce a more significant late-phase response in this population.

Jörres et al. (1996) found that O_3 causes an increased response to bronchial allergen challenge in subjects with allergic rhinitis. This study also measured responses in healthy subjects and mildly allergic asthmatics (*see Sections AX6.3.2 and AX6.8*). All subjects were exposed to 0.25 ppm O_3 for 3 h with IE. Statistically significant O_3-induced decrements in FEV_1 occurred in rhinitics (14.1%), asthmatics (12.5%), and the healthy controls (10.2%), but these responses did not differ statistically between groups. Methacholine responsiveness was significantly increased in asthmatics, but not in subjects with allergic rhinitis. Airway responsiveness to an individual's historical allergen (either grass and birch pollen, house dust mite, or animal dander) was significantly increased after O_3 exposure when compared to FA exposure. The authors concluded that subjects with allergic rhinitis, but without asthma, could be at risk if a high O_3 exposure is followed by a high dose of allergen.

Holz et al. (2002) extended the results of Jörres et al. (1996) by demonstrating that repeated daily exposure to lower concentrations of O_3 (0.125 ppm for 4 days) causes an increased response to bronchial allergen challenge in subjects with preexisting allergic airway disease, with or without asthma. These investigators observed no major difference in the pattern of bronchial allergen response between asthmatics or rhinitics, except for a 10-fold increase in the dose of allergen required to elicit a similar response ($\geq$20% decrease in FEV_1) in the asthmatic subjects. Early phase responses were more consistent in subjects with rhinitis and

late-phase responses were more pronounced in subjects with asthma. There also was a tendency towards a greater effect of O_3 in subjects with greater baseline response to specific allergens (chosen on the basis of skin prick test and history, viz., grass, rye, birch, or alder pollen, house dust mite, or animal dander). These data suggest that the presence of allergic bronchial sensitization, but not a history of asthma, may be a key determinant of increased airway allergen responsiveness following exposure to O_3 (*for a more complete discussion of airway responsiveness*) see Section AX6.8.

6.3.4 Subjects with Cardiovascular Disease

Possibly due to the age of subjects studied, O_3 exposure does not appear to result in significant pulmonary function impairment or evidence of cardiovascular strain in patients with cardiovascular disease relative to healthy controls. Gong et al. (1998) exposed 10 hypertensive and 6 healthy adult males, 41 to 78 years of age, to 0.3 ppm O_3 for 3 h with IE at 30 L/min. For all subjects combined (no significant group differences), there was an O_3-induced decrement of 7% in FEV_1 and an 70% increase in the alveolar-arterial oxygen tension gradient. The overall results did not indicate any major acute cardiovascular effects of O_3 in either the hypertensive or normal subjects. Gong et al. (1998) suggested that by impairing alveolar-arterial oxygen transfer, the O_3 exposure could potentially lead to adverse cardiac events by decreasing oxygen supply to the myocardium. However, the subjects in their study had sufficient functional reserve so as to not experience significant ECG changes or myocardial ischemia and/or injury (*see Section 6.10 for additional discussion*).

6.4 INTERSUBJECT VARIABILITY AND REPRODUCIBILITY OF RESPONSE

Analysis of factors that contribute to intersubject variability is important for the understanding of individual responses, mechanisms of response, and health risks associated with acute O_3 exposures. A large intersubject variability in response to O_3 has been reported by numerous investigators (Adams et al., 1981; Aris et al., 1995; Folinsbee et al., 1978; Kulle et al., 1985; McDonnell et al., 1983). The magnitude of individual variability in FEV_1 response in 2 h IE exposures increases at higher O_3 concentrations (Kulle et al., 1985; McDonnell et al., 1983).

McDonnell (1996) examined the FEV_1 response data from three 6.6-h exposure studies conducted at the EPA Health Effects Research Laboratory and showed that the FEV_1 responses in FA were small with most tightly grouped around zero. With increasing O_3 concentrations between 0.08 and 0.12 ppm, the mean response became asymmetrical with a few individuals experiencing quite large decrements in FEV_1 (*Intersubject variability observed in O3 dosimetry studies is discussed in Chapter 4.2*).

As an example of the variation in spirometric responses to O_3 exposure, Hazucha et al. (2003) analyzed the distribution of O_3 responsiveness in 240 subjects (18 to 60 years of age) exposed to 0.42 ppm O_3 (on 3 occasions) for 1.5 h with IE at $\dot{V}_E$ = 20 L/min/m^2 BSA. Across all ages, 18% of subjects were weak responders (≤5% FEV_1 decrement), 39% were moderate responders, and 43% were strong responders (≥15% FEV_1 decrement). Younger subjects (≤35 years of age) were predominately strong responders, whereas, older subjects (>35 years of age) were mainly weak responders. The influence of age on intersubject variability was also noted by Passannante et al. (1998) who found that subjects under 35 years of age were more like to be strong responders than older individuals. In contrast to these clinical studies, Höppe et al. (2003) observed relevant pulmonary function responses (>10% drop in FEV_1, FVC, or PEF, and/or >20% increase in sRaw) subsequent to O_3 exposure in 27% of elderly adults (n = 41; mean age, 81 yrs; $\dot{V}_E \approx$ 10 L/min) versus only 14% of young athletes (n = 43; mean age, 18 yrs; $\dot{V}_E \approx$ 80 L/min). In the absence of some underlying susceptibility to adverse O_3 effects, the elderly adults would be expected to respond far less than the athletes who had an estimated 8-fold greater $\dot{V}_E$.

For repeated exposures, Hazucha et al. (2003) reported that the reproducibility of FEV_1 responses was related to the length of time between exposures. The Spearman correlation coefficient of 0.54 was found between responses for exposures separated by 105 days (median), whereas, a correlation coefficient of 0.85 was found between responses for exposures separated by only 7 days (median). The more reproducible the subject's response, the more precisely it indicates his/her intrinsic responsiveness. In 2 h IE O_3 exposures, McDonnell et al. (1985b) found a relatively poor FEV_1 reproducibility (R = 0.58) at the lowest concentration, 0.12 ppm, due, in part, to a lack of specific O_3 response or a uniformly small response in the majority of subjects. It was concluded that for 2 h IE O_3 exposures equal to or greater than 0.18 ppm, the intersubject differences in magnitude of change in FVC and FEV_1 are quite reproducible over

time (21 to 385 days; mean = 33 days) and are due primarily to differences in intrinsic responsiveness of individual subjects to O_3 exposure.

Intersubject variability, mechanisms of response, and health risks associated with acute O_3 exposures are complicated by a poor association between various O_3-induced responses. In a retrospective study of 485 male subjects (ages 18 to 36 yrs) exposed to one of six O_3 concentrations at one of three activity levels for 2 h, McDonnell et al. (1999) observed significant, but low, Spearman rank order correlations between FEV_1 response and symptoms of cough (R = 0.39), shortness of breath (R = 0.41), and pain on deep inspiration (R = 0.30). These authors concluded that these responses are related mechanistically to some degree, but indicated that there does not appear to be a single factor which is responsible for the observed individual differences in O_3 responsiveness across the spectrum of symptom and lung function responses.

The effect of large intersubject variability on the ability to predict individual responsiveness to O_3 was demonstrated by McDonnell et al. (1993). These investigators analyzed the data of 290 male subjects (18 to 32 years of age) who underwent repeat 2 h IE exposures to one or more O_3 concentrations ranging from 0.12 to 0.40 ppm. They attempted to identify personal characteristics (i.e., age, height, baseline pulmonary function, presence of allergies, and past smoking history) that might predict individual differences in FEV_1 response. Only age contributed significantly to intersubject responsiveness (younger subjects were more responsive), accounting for just 4% of the observed variance. Interestingly, O_3 concentration accounted for only 31% of the variance, strongly suggesting the importance of as yet undefined individual characteristics that determine FEV_1 responsiveness to O_3. The authors concluded that much individual variability in FEV_1 response to O_3 remains unexplained.

6.5 FACTORS MODIFYING RESPONSIVENESS TO OZONE

6.5.1 Influence of Age

Children, adolescents, and young adults (<18 yrs of age) appear, on average, to have nearly equivalent spirometric responses to O_3, but have greater responses than middle-aged and older adults when exposed to a comparable O_3 doses (U.S. Environmental Protection Agency, 1996). Symptomatic responses to O_3 exposure, however, appear to increase with age until early adulthood and then gradually decrease with increasing age (U.S. Environmental Protection

Agency, 1996). In contrast to young adults, the diminished symptomatic responses in children and the elderly may put them at an increased risk for continued exposure. Although no new laboratory studies investigating O_3 responses in children have been published since the last O_3 AQCD, the epidemiological studies published during the last decade *(see section 7.2.3.1 for details*) are generally in agreement with the earlier laboratory studies.

The ensuing discussion in this section will provide information on average FEV_1 responses to O_3 exposure as a function of age in healthy adults ranging from 18 to 50 years of age. As was specifically addressed in Section 6.4, however, there is considerable intersubject variability in responses and this between subject variability increases with increasing O_3 dose (*see Figure AX6-6*). Epidemiological studies also report such intersubject variability. For instant, Höppe et al. (2003) observed relevant pulmonary function responses (>10% drop in FEV_1, FVC, or PEF, and/or >20% increase in sRaw) subsequent to O_3 exposure in 50% of healthy children (n = 44; mean age, 7 yrs; $\dot{V}_E \approx 30$ L/min), 14% of young athletes (n = 43; mean age, 18 yrs; $\dot{V}_E \approx 80$ L/min), and 27% of elderly adults (n = 41; mean age, 81 yrs; $\dot{V}_E \approx 10$ L/min).

Beyond approximately 18 years of age, spirometric and symptom responses to O_3 exposure begin to decline with increasing age. In healthy individuals, the rate of decline in O_3 responsiveness appears to be greater in younger (18 to 35 yrs) versus middle aged (35 to 55 yrs) individuals (Passannante et al., 1998; Hazucha et al., 2003). Beyond this age (>55 yrs), acute O_3 exposure elicits minimal spirometric changes. An average FEV_1 decrement of ~3% has been reported by Gong et al. (1997a) for this older population under a "worst case" exposure scenario (0.24 ppm O_3 with 4 h IE). Although Gong et al. (1997a) and others have examined responses to O_3 exposure in subjects of various ages, the exposure conditions differ between most studies so that age effects remain uncertain.

Three recent studies, which analyzed large data sets ($\geq$240 subjects) of similarly exposed subjects, show clearly discernable changes in FEV_1 responses to O_3 as a function of age. Seal et al. (1996) analyzed O_3-induced spirometric responses in 371 young nonsmokers (18 to 35 years of age) exposed for 2.3 h during IE at a $\dot{V}_E$ of 25 L/min/m^2 BSA. On average, for the same O_3 concentration (C), the response of 25, 30, and 35 year old individuals are predicted to be 83, 65, and 48%, respectively, of the response in a 20 year old. For example, a 5.4% decrement in FEV_1 is predicted for 20 year old exposed to 0.12 ppm O_3 for 2.3 h

IE ($\dot{V}_E$ = 25 L/min/m^2 BSA), whereas, a similarly exposed 35 yr old is predicted to have only a 2.6% decrement.

McDonnell et al. (1997) examined FEV_1 responses in 485 healthy white males (18 to 36 years of age) exposed once for 2 h to an O_3 concentration of 0.0, 0.12, 0.18, 0.24, 0.30, or 0.40 ppm at rest or one of two levels of IE ($\dot{V}_E$ of 25 and 35 L/min/m^2 BSA). For the same exposure conditions (C, $\dot{V}_E$, and duration), the average responses of 25, 30, and 35 year old individuals are predicted to be 69, 48, and 33%, respectively, of the response in 20 year olds. Hazucha et al. (2003) analyzed the distribution of O_3 responsiveness in 240 subjects (18 to 60 years of age) exposed to 0.42 ppm O_3 for 1.5 h with IE at $\dot{V}_E$ = 20 L/min/m^2 BSA. In males, the FEV_1 responses of 25, 35, and 50 year olds are predicted to be 94, 83, and 50% , respectively, of the average response in 20 year old males. In females, the FEV_1 responses of 25, 35, and 50 year olds are predicted to be 82, 46, and 18%, respectively, of the average response in 20 year old females.

For subjects aged 18 to 36 yrs, McDonnell et al. (1999) recently reported that symptom responses from O_3 exposure also decrease with increasing age. Whether the same age-dependent pattern of O_3 sensitivity decline also holds for airway reactivity or inflammatory endpoints has not been determined.

6.5.2 Gender and Hormonal Influences

Several studies have suggested that physiological differences between the genders may predispose females to a greater susceptibility to O_3. Lower plasma and nasal lavage fluid levels of uric acid (the most prevalent antioxidant) in females relative to males may be a contributing factor (Housley et al., 1996). Consequently, reduced absorption of O_3 in the upper airways may promote its deeper penetration. Dosimetric measurements have shown that the absorption distribution of O_3 is independent of gender when absorption is normalized to anatomical dead space (Bush et al., 1996). More recently, Ultman et al. (2004) reported that the whole lung uptake fraction of O_3 was significantly greater in males (91.4%) than females (87.1%). But, this increase in O_3 uptake in the males was consistent with their larger tidal volume and slower breathing frequency relative to the females. Furthermore, O_3 uptake was not correlated with spirometric responses. Thus, a differential removal of O_3 by uric acid seems to have minimal

effect. In general, the spirometric responses of young healthy females to O_3 exposure appears comparable to the responses of young males (Hazucha et al., 2003). Although, during the follicular phase of the menstrual cycle, lung function response to O_3 is enhanced (Fox et al., 1993).

6.5.3 Racial, Ethnic, and Socioeconomic Status Factors

A few epidemiologic studies have implied that minorities are more responsive to O_3 than caucasians. However, this may be more of a consequence of the overall quality of health care and socioeconomic status (SES) than an innate sensitivity to oxidants (Gwynn and Thurston, 2001; Seal et al, 1996). The paucity of data has prevented making any definitive conclusions on the influence of race, ethnic or other related factors on the responsiveness to O_3.

6.5.4 Influence of Physical Activity

Any physical activity will increase minute ventilation and therefore the dose of inhaled O_3. Consequently, the intensity of physiological response following an acute exposure will be strongly associated with minute ventilation (*see Figures 6-3 and AX6-2*).

6.5.5 Environmental Factors

Since the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996) few human laboratory studies have examined the potential influence of environmental factors such as rural versus urban environment, passive cigarette smoke exposure, and bioactive agents such as endotoxin on healthy individual's pulmonary function changes due to O_3.

New controlled human exposure studies have confirmed that smokers are less responsive to O_3 than nonsmokers. Spirometric and plethysmographic pulmonary function decline, nonspecific airway hyperreactivity, and inflammatory response of smokers to O_3 were all weaker than those reported for nonsmokers. Although all these responses are intrinsically related, the functional association between them, as in nonsmokers, has been weak. Similarly, the time course of development and recovery of these effects, as well their reproducibility, was not different from nonsmokers. Chronic airway inflammation with desensitization of bronchial nerve endings and an increased production of mucus may plausibly explain the pseudo-protective effect of smoking (Frampton et al., 1997; Torres et al., 1997).

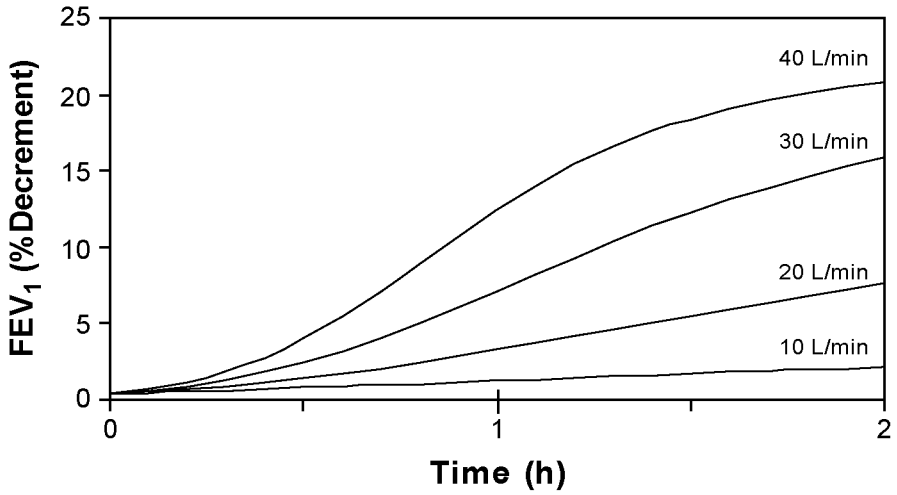


Figure 6-3. Predicted O_3-induced decrements in FEV_1 as a function of exposure duration and level of IE (line labels are $\dot{V}_E$ levels) in young healthy adults (20 yrs of age) exposed to 0.3 ppm O_3. The illustrated activity levels range from rest ($\dot{V}_E$ = 10 L/min) to moderate exercise ($\dot{V}_E$ = 40 L/min). Predictions are for Model 1 coefficients in Table 3 of McDonnell et al. (1997).

Source: Based on McDonnell et al. (1997).

The effect of environmental tobacco smoke (ETS) on O_3 responses has received very little attention. In one study, preexposure of mice to sidestream cigarette smoke (ETS surrogate) elicited no immediate effects, but potentiated subsequent O_3-induced inflammatory responses (Yu et al., 2002) (*See Chapter 5.4.2 for additional ETS details*). Endotoxin is a biologically active component of both mainstream and sidestream tobacco smoke (Hasday et al., 1999) which might contribute to the potentiation of O_3 effects.

The influence of ambient temperature on pulmonary effects induced by O_3 exposure in humans has been studied infrequently under controlled laboratory conditions. Several experimental human studies have reported additive effects of heat and O_3 exposure (see U.S. Environmental Protection Agency, 1986, 1996). Foster et al. (2000) exposed 9 young healthy subjects for 130 min (IE 10 min at 36 to 39 l/min) to filtered air and to ramp profile O_3 at 22 °C and 30 °C, 45-55% RH. The O_3 exposure started at 0.12 ppm, reached the peak of 0.24 ppm midway through and subsequently declined to 0.12 ppm at the end of exposure. At the end of exposure FEV_1 decreased significantly ($p < 0.5$) by ~8% at 22 °C and ~6.5% at 30 °C. One day

(19 h) later, the decline of 2.3% from baseline was still significant ($p < 0.05$) at both temperatures. FVC decrements were smaller and significant only for the 22 °C condition immediately postexposure. There was a decline in specific airway conductance (sGaw; $p < 0.05$) at 30 °C but not at 22 °C. The nonspecific bronchial responsiveness to methacloline assessed as PC_{50} sGaw was significantly ($p < 0.05$) higher one day following O_3 exposure at both temperatures but more so at 30 °C. Thus, these findings suggest that elevated temperature may partially attenuate spirometric responses but enhance airway reactivity.

6.5.6 Oxidant-Antioxidant Balance

The first line of defense against oxidative stress is antioxidant present in epithelial lining fluid (ELF) which scavenge free radicals and limit lipid peroxidation. Exposure to O_3 depletes the antioxidant level in nasal ELF probably due to scrubbing of O_3 (Mudway et al., 1999), however, the concentration and the activity of antioxidant enzymes either in ELF or plasma do not appear to be related to O_3 responsiveness (Avissar et al., 2000; Blomberg et al., 1999; Samet et al., 2001). Carefully controlled studies of dietary antioxidant supplementation have demonstrated some protective effects of α-tocopherol and ascorbate on spirometric lung function from O_3 but not on the intensity of subjective symptoms and inflammatory response including cell recruitment, activation and release of mediators (Samet et al., 2001; Trenga et al., 2001). Dietary antioxidants have also afforded partial protection to asthmatics by attenuating post-exposure bronchial hyperresponsiveness (Trenga et al., 2001). The field studies performed in Mexico City (*described in Section 7.2.3.1*) and animal studies (*described in Section 5.2.1.3*) have also demonstrated the protective effects of ELF antioxidants during O_3 exposures.

6.5.7 Genetic Factors

Several recent studies (Yang et al., 2005; David et al., 2003; Romieu et al., 2004) have reported that genetic polymorphism of antioxidant enzymes and inflammatory genes may modulate pulmonary function and inflammatory response to O_3 challenge. It appears that healthy carriers of NAD(P)H:quinone oxidoreductase wild type (NQO1wt) in combination with glutathione S-transferase μ-1 (GSTM1null) genotype are more responsive to O_3. The authors have implied that the interindividual variability in O_3 responsiveness (FEV_1 changes) is related

to the polymorphism of these enzymes. Adults with GSTM1null only genotype did not show O_3 hyperresponsiveness (Bergamaschi et al., 2001). A subsequent study from the same laboratory reported a positive association between O_3 responsiveness, as characterized by the level of oxidative stress and inflammatory mediators (8-isoprostane, LTB_4 and TBARS) in EBC fluid, and the antioxidant enzyme polymorphism. However, none of the spirometric lung function endpoints were affected by ozone exposure (Corradi et al., 2002). It is of interest to note, that human nasal mucosa biopsies of GSTM1 deficient subjects showed higher antioxidant enzymes activity than biopsies of GSTM1 positive individuals when exposed to ozone (Otto-Knapp et al., 2003).

Asthmatic children with a genetic deficiency of GSTM1 were reported to be more responsive to ambient O_3 exposure, as assessed by decrements in FEF_{25-75}, in this field study. Antioxidant supplementation (vit. C and E) attenuated post-ozone lung function response in these children (Romieu et al., 2004). More specific genotyping has shown that ozone responsiveness of asthmatic children may be related to the presence of variant Ser allele for NQO1. The presence of at least one NQO1 Ser allele in combination with GSTM1 null genotype lowered the risk of asthma in ozone exposed asthmatic children relative to Pro/Pro genotype (David et al., 2003).

The influence of functional polymorphism in TNF-α, lymphotoxin- α (LTA), TLR4, SOD2 and GPX1 genes on ozone-induced lung function changes in healthy individuals, mild asthmatics and subjects with rhinitis was varied. Of the inflammatory genes studied only TNF-α has appeared to show some promise as one of the genetic factors of susceptibility. However, as the authors stated "the functional significance of individual TNF-α polymorphisms remains controversial" (Yang et al., 2005).

These recent studies have shown that individual's innate susceptibility to ozone may be linked to genetic background of an individual. Although a number of potential ozone susceptibility genes have been identified, additional better designed and controlled studies are needed to ascertain the link between susceptibility and polymorphism.

6.6 REPEATED O_3 EXPOSURE EFFECTS

Based on studies reviewed here and in the previous O_3 criteria documents (U.S. Environmental Protection Agency, 1986, 1996), several conclusions can be drawn about repeated 1 to 2-h O_3 exposures. Repeated exposures to O_3 can cause an enhanced (i.e., greater) pulmonary function response on the second day of exposure (*see Tables AX6-8 and AX6-9 for added detail*). This enhancement appears to be dependent on the interval between the exposures (24 h is associated with the greatest increase) and is absent with intervals >3 days (Bedi et al., 1985; Folinsbee and Horvath, 1986; Schonfeld et al., 1989). An enhanced response also appears to depend to some extent on the magnitude of the initial response (Horvath et al., 1981). Small responses to the first O_3 exposure are less likely to result in an enhanced response on the second day of O_3 exposure (Folinsbee et al., 1994). With continued daily exposures (i.e., beyond the second day) there is an attenuation of pulmonary function responses, typically after 3 to 5 days of repeated exposure. This attenuated response persists for less than 1 week (Kulle et al., 1982; Linn et al., 1982b) or as long as 2 weeks (Horvath et al., 1981). In temporal conjunction with pulmonary function changes, symptoms induced by O_3, such as cough, pain on deep inspiration, and chest discomfort, are increased on the second exposure day and attenuated with repeated exposure thereafter (Folinsbee et al., 1980, 1998; Foxcroft and Adams, 1986; Linn et al., 1982b). O_3-induced changes in airway responsiveness persist longer and attenuate more slowly than pulmonary function and symptoms responses (Dimeo et al., 1981; Kulle et al., 1982), although this has been studied only on a limited basis (Folinsbee et al., 1994). In longer-duration (4 h to 6.6 h), lower-concentration studies that do not cause an enhanced second-day response, the attenuation of response to O_3 appears to proceed more rapidly (Folinsbee et al., 1994) [*Effects of repeated exposures on inflammatory responses are discussed in Section 6.9.4*).

6.7 EFFECTS ON EXERCISE PERFORMANCE

The effects of acute O_3 inhalation on endurance exercise performance have been examined in numerous controlled laboratory studies. These studies were discussed in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996) and can be divided into two categories: (1) those that examined the effects of acute O_3 inhalation on maximal oxygen uptake ($\dot{V}O_{2max}$) and

(2) those that examined the effects of acute O_3 inhalation on the ability to complete strenuous continuous exercise protocols of up to 1 h in duration.

In brief, endurance exercise performance and $\dot{V}O_{2max}$ may be limited by acute exposure to O_3 (Adams and Schelegle, 1983; Schelegle and Adams, 1986; Gong et al., 1986; Foxcroft and Adams, 1986; Folinsbee et al., 1977; Linder et al., 1988). Gong et al. (1986) and Schelegle and Adams (1986) found that significant reductions in maximal endurance exercise performance may occur in well-conditioned athletes while they perform CE ($\dot{V}_E$ > 80 L/min) for 1 h at O_3 concentrations ≥0.18 ppm. Reports from studies of exposure to O_3 during high-intensity exercise indicate that breathing discomfort associated with maximal ventilation may be an important factor in limiting exercise performance in some, but not all, subjects.

6.8 EFFECTS ON AIRWAY RESPONSIVENESS

Airway or bronchial hyperresponsiveness (BHR) refers to a condition in which the propensity for the airways to bronchoconstrict due to a variety of stimuli becomes augmented. Airway responsiveness is typically quantified by measuring the decrement in pulmonary function (i.e., spirometry or plethysmography) following the inhalation of small amounts of an aerosolized bronchoconstrictor agent (specific [antigen, allergen] or nonspecific [methacholine, histamine]) or a measured stimulus (e.g., exercise, cold air).

Ozone exposure causes an increase in nonspecific airway responsiveness as indicated by a reduction in the concentration of methacholine or histamine required to produce a given reduction in FEV_1 or increase in SRaw. Increased airway responsiveness is an important consequence of exposure to O_3 because its presence means that the airways are predisposed to narrowing on inhalation of a variety of stimuli (e.g., specific allergens, SO_2, cold air).

Ozone exposure of asthmatic subjects, who characteristically have increased airway responsiveness at baseline, can cause further increases in responsiveness (Kreit et al., 1989). Similar relative changes in airway responsiveness are seen in asthmatics exposed to O_3 despite their markedly different baseline airway responsiveness. Several studies (Jörres et al., 1996; Kehrl et al., 1999; Molfino et al., 1991) have been published suggesting an increase in specific (i.e., allergen-induced) airway reactivity. An important aspect of increased airway

responsiveness after O_3 exposure is that this represents a plausible link between ambient O_3 exposure and increased hospital admissions for asthma.

Changes in airway responsiveness after O_3 exposure appear to be resolved more slowly than changes in FEV_1 or respiratory symptoms (Folinsbee and Hazucha, 2000). Furthermore, in studies of repeated exposure to O_3, changes in airway responsiveness tend to be somewhat less susceptible to attenuation with consecutive exposures than changes in FEV_1 (Dimeo et al., 1981; Folinsbee et al., 1994; Gong et al., 1997b; Kulle et al., 1982). Increases in airway responsiveness do not appear to be strongly associated with decrements in lung function or increases in symptoms.

The mechanism of O_3-induced increases in airway responsiveness is only partially understood, but it appears to be associated with a number of cellular and biochemical changes in airway tissue. Although inflammation could play a role in the increase in airway responsiveness, cyclooxygenase inhibitors have not been effective at blocking the O_3-induced influx of PMNs into bronchoalveolar lavage (BAL) fluid (Hazucha et al., 1996; Ying et al., 1990). Therefore, O_3-induced airway responsiveness may not be due to the presence of PMNs in the airway or to the release of arachidonic acid metabolites. Rather, it seems likely that the mechanism for this response is multifactorial, possibly involving the presence of cytokines, prostanoids, or neuropeptides; activation of macrophages, eosinophils, or mast cells; and epithelial damage that increases direct access of mediators to the smooth muscle or receptors in the airways that are responsible for reflex bronchoconstriction.

6.9 EFFECTS ON INFLAMMATION AND HOST DEFENSE

6.9.1 Introduction

Short-term exposure of humans to O_3 can cause acute inflammation and long-term exposure of laboratory animals results in a chronic inflammatory state (*see Chapter 5*). The relationship between repetitive bouts of acute inflammation in humans caused by O_3 and the development of chronic respiratory disease is unknown.

The presence of neutrophils (PMNs) in the lung has long been accepted as a hallmark of inflammation and is an important indicator that O_3 causes inflammation in the lungs. It is apparent, however, that inflammation within airway tissues may persist beyond the point that

inflammatory cells are found in BAL fluid (BALF). Soluble mediators of inflammation such as the cytokines (IL-6, IL-8) and arachidonic acid metabolites (e.g., PGE_2, $PGF_{2\alpha}$, thromboxane, and leukotrienes [LTs] such as LTB_4) have been measured in the BAL fluid of humans exposed to O_3. In addition to their role in inflammation, many of these compounds have bronchoconstrictive properties and may be involved in increased airway responsiveness following O_3 exposure.

Some recent evidence suggests that changes in small airways function may provide a sensitive indicator of O_3 exposure and effect, despite the fact that inherent variability in their measurement by standard spirometric approaches make their assessment difficult (Frank et al., 2001). Observations of increased functional responsiveness of these areas relative to the more central airways, and of persistent effects following repeated exposure, may indicate that further investigation of inflammatory processes in these regions is warranted.

6.9.2 Inflammatory Responses in the Upper Respiratory Tract

The nasal passages constitute the primary portal for inspired air at rest and, therefore, the first region of the respiratory tract to come in contact with airborne pollutants. Nikasinovic et al. (2003) recently reviewed the literature of laboratory-based nasal inflammatory studies published since 1985. Nasal lavage (NL) has provided a useful tool for assessing O_3-induced inflammation in the nasopharynx. Increased levels of PMNs in the NL fluid of humans exposed to 0.5 ppm O_3 at rest for 4 h has been reported (Graham et al., 1988; Bascom et al., 1990).

Graham and Koren (1990) compared inflammatory mediators present in both the NL and BAL fluids of humans exposed to 0.4 ppm O_3 for 2 h. Similar increases in PMN were observed in NL and BAL, suggesting a qualitative correlation between inflammatory changes in the lower airways (BAL) and the upper respiratory tract (NL). Torres et al. (1997) compared NL and BAL in smokers and nonsmokers exposed to 0.22 ppm O_3 for 4 h. In contrast to Graham and Koren (1990), they did not find a relationship between numbers or percentages of PMNs in the nose and the lung, perhaps in part due to the variability observed in their NL recoveries. Albumin, a marker of epithelial cell permeability, was increased 18 h later, but not immediately after exposure, as seen by Bascom et al. (1990).

McBride et al. (1994) reported that asthmatic subjects were more sensitive than nonasthmatics to upper airway inflammation at an O_3 concentration (0.24 ppm for 1.5 h with

light IE) that did not affect pulmonary function. In the asthmatics, there was a significant increase in the number of PMNs in NL fluid both immediately and 24 h after exposure. Peden et al. (1995) also found that exposure to 0.4 ppm O_3 had a direct nasal inflammatory effect and a priming effect on response to nasal allergen challenge. A subsequent study in dust mite-sensitive asthmatic subjects indicated that O_3 at this concentration enhanced eosinophil influx in response to allergen but did not promote early mediator release or enhance the nasal response to allergen (Michelson et al., 1999). Similar to observations made in the lower airways, the presence of O_3 molecular "targets" in nasal lining fluid is likely to provide some level of local protection against exposure. In a study of healthy subjects exposed to 0.2 ppm O_3 for 2 h, Mudway and colleagues (1999) observed a significant depletion of uric acid in NL fluid at 1.5 h following exposure.

6.9.3 Inflammatory Response in the Lower Respiratory Tract

As reviewed in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996), acute exposure to O_3 results in an inflammatory reaction, increased epithelial cell permeability, and may stimulate fibrogenic processes. Inflammatory markers are observed in BALF of healthy subjects by 1 h post-O_3 exposure and may persist for at least 18 to 24 h. Not all inflammatory markers, however, follow the same time course. Studies published since the 1996 O_3 AQCD support these earlier findings.

Inflammatory effects have been assessed *in vivo* by lavage (proximal airway and bronchoalveolar), bronchial biopsy, and more recently, induced sputum. *In vitro* studies of human alveolar macrophages (AM) and airway epithelial cells exposed to O_3 suggest that most mediators found in the BALF of O_3-exposed humans are produced by epithelial cells (U.S. Environmental Protection Agency, 1996). Recent evidence suggests that the release of mediators from AMs may be modulated by the products of O_3-induced oxidation of airway lining fluid components, such as human surfactant protein A (Wang et al., 2002).

Spirometric responses to O_3 are independent from inflammatory responses and markers of epithelial injury (Balmes et al., 1996; Blomberg et al., 1999; Hazucha et al., 1996; Torres et al., 1997). Significant inflammatory responses to O_3 exposures that did not elicit significant spirometric responses have been reported (Holz et al., 2005; McBride et al., 1994). A meta-

analysis of 21 studies (Mudway and Kelly, 2004), showed that PMN influx in health subjects is associated with total O_3 dose (product of O_3 concentration, exposure duration, and $\dot{V}_E$).

The time course of the inflammatory response to O_3 in humans has not been fully characterized. From a review of the literature by Mudway and Kelly (2000), Figure 6-4 illustrates a plausible time course of acute O_3 responses. As the figure shows, different markers have peak responses at different times. Studies in which lavages were performed 1 h after O_3 exposure (1 h at 0.4 ppm or 4 h at 0.2 ppm) have demonstrated that the inflammatory responses are quickly initiated (Devlin et al., 1996; Schelegle et al., 1991; Torres et al., 1997). Inflammatory mediators and cytokines such as IL-8, IL-6, and PGE_2 are greater at 1 h than at 18 h post-O_3 exposure (Devlin et al., 1996; Torres et al., 1997). However, IL-8 still remains elevated at 18 h post-O_3 (4 h at 0.2 ppm O_3 versus FA) in healthy subjects and more so in asthmatics (Balmes et al., 1996; Scannell et al., 1996). Schelegle et al. (1991) found increased PMNs in the "proximal airway" lavage at 1, 6, and 24 h after O_3 exposure (4 h at 0.2 ppm O_3), with a peak response at 6 h. Although, at 18 to 24 h after O_3 exposure, PMNs remain elevated relative to 1 h postexposure (Schelegle et al., 1991; Torres et al., 1997). In addition to the influx of PMNs and (in allergic asthmatics) eosinophils, lymphocyte numbers in BALF are elevated significantly at 6 h following exposure (2 h at 0.2 ppm O_3) of healthy subjects (Blomberg et al., 1997). Flow cytometry also indicated the increased presence of CD3+, CD4+ and CD8+ T cell subsets. This same laboratory later demonstrated that within 1.5 h following exposure of healthy subjects to the same O_3 regimen, expression of human leukocyte antigen (HLA)-DR on lavaged macrophages underwent a significant, 2.5-fold increase (Blomberg et al., 1999).

The inflammatory responses to O_3 exposure have also been studied in asthmatic subjects (Basha et al., 1994; Scannell et al., 1996; Peden et al., 1997). In these studies, asthmatics showed significantly more neutrophils in the BALF (18 h post-exposure) than similarly exposed healthy individuals. In one of these studies (Peden et al., 1997), which included only allergic asthmatics who tested positive for Dematophagoides farinae antigen, there was an eosinophilic inflammation (2-fold increase), as well as neutrophilic inflammation (3-fold increase). In a study of subjects with intermittent asthma exposed to 0.4 ppm O_3 for 2 h, increases in eosinophil cationic protein, neutrophil elastase and IL-8 were found to be significantly increased 16 h post-exposure and comparable in induced sputum and BALF (Hiltermann et al, 1999). Scannell et al. (1996) also reported that IL-8 tends to be higher in the BALF of asthmatics compared to

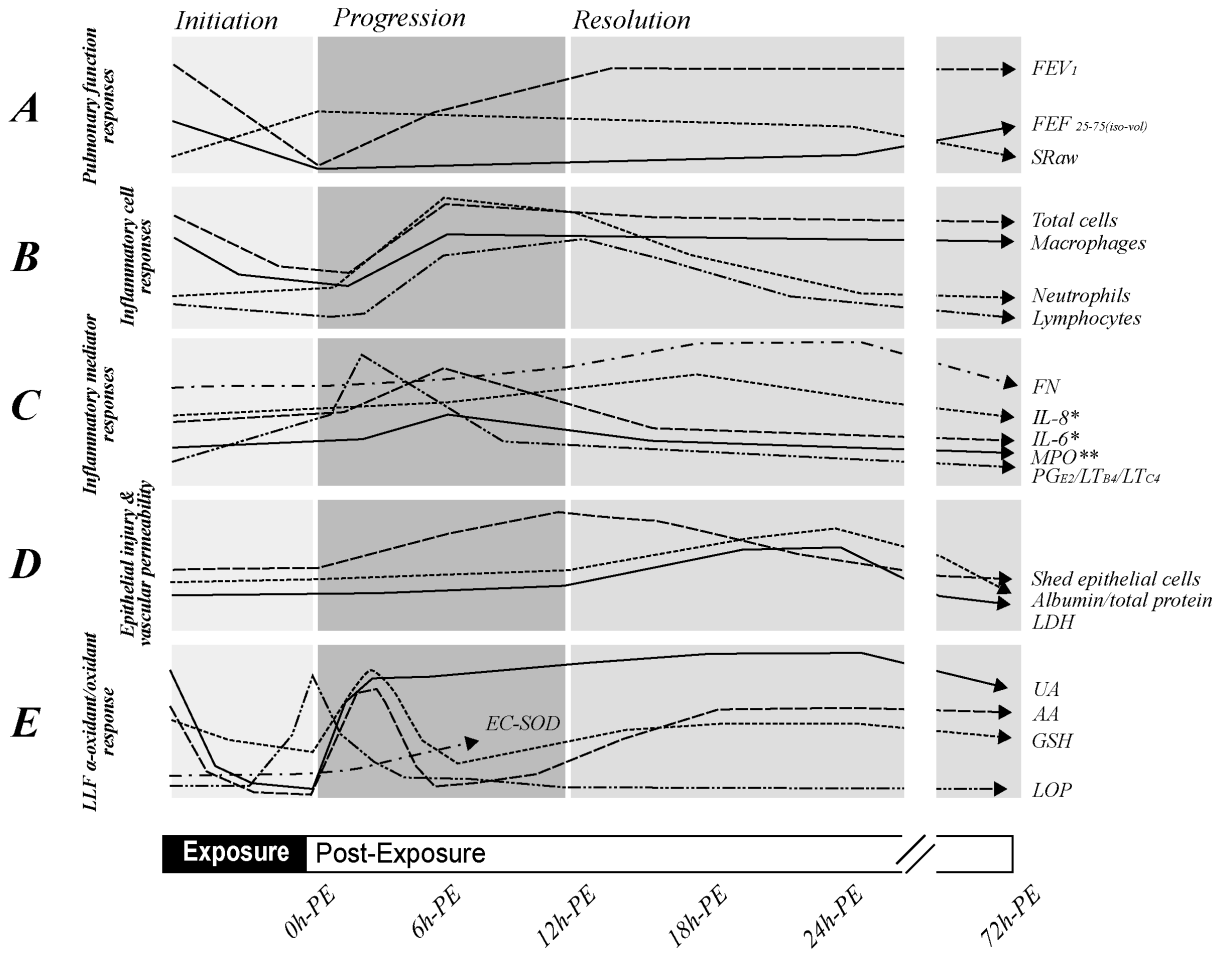


Figure 6-4. Time course of acute responses seen in humans exposed to O_3. Responses are divided into three phases: initiation, during O_3 exposure; progression, where responses develop post-O_3 exposure; and resolution, during which responses return baseline levels. *The IL-8 response is shown with a progressive increase peaking at 18 h postexposure (18h-PE). The IL-8 literature is inconclusive and neutrophils have been demonstrated in the absence of an IL-8 increase. **Few studies have measured MPO, however, a trend for an increase at 6h-PE has been reported.

Abbreviations: LOP (lipid ozonation products), GSH (reduced glutathione), AA (ascorbic acid), UA (uric acid), LDH (lactate dehydrogenase), PG_{E2} (prostaglandin E2), LT_{B4} (leukotriene B4), LT_{C4} (leukotriene C4), MPO (myeloperoxidase).

Source: Reprinted from Mudway and Kelly (2000) with permission from Elsevier.

nonasthmatics following O_3 exposure, suggesting a possible mediator for the significantly increased neutrophilic inflammation in those subjects. Bosson et al. (2003) found significantly greater the epithelial expression of IL-5, IL-8, granulocyte-macrophage colony-stimulating factor (GM-CSF) and epithelial cell-derived neutrophil-activating peptide 78 (ENA-78) in asthmatics compared to healthy subjects following exposure to 0.2 ppm O_3 for 2 h. Stenfors and colleagues (2002) were unable to detect a difference in the increased neutrophil numbers between 15 mild asthmatic and 15 healthy subjects by bronchial wash at the 6 h post-exposure time point. However, the asthmatics were on average 5 years older than the healthy subjects in this study and it is not yet known how age affects inflammatory responses. It is also possible that the time course of neutrophil influx differs between healthy and asthmatic individuals.

Vagaggini et al. (2002) investigated the effect of prior allergen challenge on responses in mild asthmatics exposed for 2 h to 0.27 ppm O_3 or filtered air. At 6 h post-exposure, eosinophil numbers in induced sputum were found to be significantly greater after O_3 than after air. Studies such as these suggest that the time course of eosinophil and neutrophil influx following O_3 exposure can occur to levels detectable within the airway lumen by as early as 6 h. They also suggest that the previous or concurrent activation of proinflammatory pathways within the airway epithelium may enhance the inflammatory effects of O_3. For example, in an *in vitro* study of epithelial cells from the upper and lower respiratory tract, cytokine production induced by rhinovirus infection was enhanced synergistically by concurrent exposure to O_3 at 0.2 ppm for 3 h (Spannhake et al, 2002).

Although the release of mediators has been demonstrated to occur at exposure concentrations and times that are minimally cytotoxic to airway cells, potentially detrimental latent effects have been demonstrated in the absence of cytotoxicity. These include the generation of DNA single strand breaks (Kozumbo et al., 1996), the loss of cellular replicative activity (Gabrielson et al., 1994) in bronchial epithelial cells exposed *in vitro*, and the formation of protein and DNA adducts. A highly toxic aldehyde formed during O_3-induced lipid peroxidation is 4-hydroxynonenal (HNE). Healthy human subjects exposed to 0.4 ppm O_3 for 1 h underwent BAL 6 h later. Analysis of lavaged AMs by Western Blot indicated increased levels of a 32-kDa HNE-protein adduct, as well as 72-kDa heat shock protein and ferritin in O_3-versus air-exposed subjects (Hamilton et al., 1998). In a recent study of healthy subjects exposed to 0.1 ppm O_3 for 2 h (Corradi et al., 2002), formation of 8-hydroxy-2'-deoxyguanosine

(8-OHdG), a biomarker of reactive oxidant species (ROS)-DNA interaction, was measured in peripheral blood lymphocytes. At 18 h post exposure, 8-OHdG was significantly increased in cells compared to pre-exposure levels, presumably linked to concurrent increases in chemical markers of ROS. Of interest, the increase in 8-OHdG was only significant in a subgroup of subjects with the wild genotype for NQ01 and the null genotype for GSTM1, suggesting that polymorphisms in redox enzymes may confer "susceptibility' to O_3 in some individuals.

The generation of ROS following exposure to O_3 has been shown to be associated with a wide range of responses. In a recent study, ROS production by alveolar macrophages lavaged from subjects exposed to 0.22 ppm for 4 h was assessed by flow cytometry (Voter et al., 2001). Levels were found to be significantly elevated 18 h post exposure and associated with several markers of increased permeability. An *in vitro* study of human tracheal epithelial cells exposed to O_3 indicated that generation of ROS resulted in decrease in synthesis of the bronchodilatory prostaglandin, PGE_2, as a result of inactivation of prostaglandin endoperoxide G/H synthase 2 (Alpert et al., 1997).

6.9.4 Adaptation of Inflammatory Responses

Physiologic and symptomatic responses in humans following repeated exposure to O_3 were discussed in Section 6.6. Inflammatory responses upon repeated O_3 exposures are discussed in this section. Animal studies suggest that while inflammation may be diminished with repeated exposure, underlying damage to lung epithelial cells continues (Tepper et al., 1989). Markers from BALF following both 2-h (Devlin et al., 1997) and 4-h (Christian et al., 1998; Jörres et al., 2000) repeated O_3 exposures (up to 5 days) indicate that there is ongoing cellular damage irrespective of the attenuation of some cellular inflammatory responses of the airways, pulmonary function, and symptom responses.

Devlin et al. (1997) examined the inflammatory responces of humans repeatedly exposed to 0.4 ppm O_3 for 5 consecutive days. Several indicators of inflammation (e.g., PMN influx, IL-6, PGE_2, BAL protein, fibronectin) were attenuated after 5 days of exposure (i.e., values were not different from FA). Several markers (LDH, IL-8, total protein, epithelial cells) did not show attenuation, indicating that tissue damage probably continues to occur during repeated exposure. The recovery of the inflammatory response occurred for some markers after 10 days, but some responses were not normalized even after 20 days. The continued presence of cellular injury

markers indicates a persistent effect that may not necessarily be recognized due to the attenuation of spirometric and symptom responses.

Christian et al. (1998) randomly subjected heathy subjects to a single exposure and to 4 consecutive days of exposure to 0.2 ppm O_3 for 4 h. Both "bronchial" and "alveolar" fractions of the BAL showed decreased numbers of PMNs and fibronectin concentration at day 4 versus the single exposure, and a decrease in IL-6 levels in the alveolar fraction. Following a similar study design and exposure parameters, Jörres et al. (2000) found both functional and BAL cellular responses to O_3 were abolished at 24 h postexposure following the fourth exposure day. However, levels of total protein, IL-6, IL-8, reduced glutathione and ortho-tyrosine were still increased significantly. In addition, visual scores for bronchitis, erythema and the numbers of neutrophils in the mucosal biopsies were increased. Their results indicate that, despite reduction of some markers of inflammation in BAL and measures of large airway function, inflammation within the airways persists following repeated exposure to O_3.

Holz et al. (2002) made a comparison of early and late responses to allergen challenge following O_3 in subjects with allergic rhinitis or allergic asthma. With some variation, both early and late FEV_1 and cellular responses in the two subject groups were significantly enhanced by 4 consecutive days of exposure to 0.125 ppm O_3 for 3 h.

In another study, Frank and colleagues (2001) exposed healthy subjects to FA and to O_3 (0.25 ppm, 2 h) on 4 consecutive days each, with pulmonary function measurements being made prior to and following each exposure. BAL was performed on day 5, 24 h following the last exposure. On day 5, PMN numbers remained significantly higher following O_3 compared to FA. Of particular note in this study was the observation that small airway function, assessed by grouping values for isovolumetric FEF_{25-75}, Vmax50 and Vmax75 into a single value, showed persistent reduction from day 2 through day 5. These data suggest that techniques monitoring the function in the small peripheral airway regions, the primary sites of O_3 uptake in the lung, may provide important information regarding both acute and cumulative effects of O_3 exposure.

6.9.5 Effect of Anti-Inflammatory and Other Mitigating Agents

Pretreatment of healthy subjects with non-steroidal anti-inflammatory drugs (ibuprofen, etc.) has been found to partially suppress development of airway inflammation and pulmonary function changes (U.S. Environmental Protection Agency, 1996). Although atropine blocked the

increase in Raw in response to O_3 exposure, it did not alter the spirometric or symptom responses (Beckett et al., 1985). Similarly, albuterol and salbutamol, which had no effect on O_3-induced changes in spirometry, also had no effect of symptom responses (McKenzie et al., 1987; Gong et al., 1988). The anti-inflammatory medications indomethacin and ibuprofen, which partially inhibit the spirometric responses to O_3 exposure, also cause a reduction in respiratory symptoms (Schelegle et al., 1987; Hazucha et al., 1994). Indomethacin attenuates decrements in FEV_1 and FVC in healthy subjects, but not asthmatics (Alexis et al., 2000). In contrast, inhalation of the corticosteroid budesonide does not prevent or even attenuate O_3-induced responses in healthy subjects as assessed by measurements of lung function, bronchial reactivity and airway inflammation (Nightingale et al., 2000). In asthmatic subjects, budesonide decreases airway neutrophil influx following O_3 exposure (Vagaggini et al., 2001). This suggests that corticosteroids may be effective only when the inflammation is already present, such as in asthmatics.

Holz et al. (2005) studied inflammatory responses in healthy ozone-responders (>10% increase in sputum neutrophils from O_3) pretreated with single doses (the highest shown to be safe and well tolerated) of inhaled fluticasone and oral prednisolone. The O_3 exposure caused small changes in FEV_1 ($-3.6\% \pm 6.8\%$) that were not significantly different from baseline or between treatment groups (i.e., prescreening, placebo, fluticasone, and prednisolone). Relative to placebo, the inhaled or oral corticosteroids significantly reduced O_3-induced neutrophil levels. These authors note that their study design was intended to test the anti-inflammatory effects of the steroids and that such high-dose regimens should not be considered for potential long-term patient treatment.

6.9.6 Changes in Host Defense Capability Following Ozone Exposures

A number of studies clearly show that a single acute exposure (1 to 4 h) of humans to moderate concentrations of O_3 (0.2 to 0.6 ppm) while exercising at moderate to heavy levels results in a number of cellular and biochemical changes in the lung including an inflammatory response characterized by increased numbers of PMNs, increased permeability of the epithelial cells lining the respiratory tract, cell damage, and production of proinflammatory cytokines and prostaglandins. This response can be detected as early as 1 h after exposure (Koren et al., 1991; Schelegle et al., 1991) and persists for at least 18 h (Aris et al., 1993; Koren et al., 1989). The

response profile of these mediators is not defined adequately, although it is clear that the time course of response varies for different mediators and cells (Devlin et al., 1997; Schelegle et al., 1991). These changes also occur in humans exposed to 0.08 and 0.10 ppm O_3 for 6 to 8 h (Devlin et al., 1991; Peden et al., 1997). Ozone also causes inflammatory changes in the nose, as indicated by increased levels of PMNs and albumin, a marker for increased epithelial cell permeability. Nasal lavage analyses, however, are not necessarily parallel to BAL analyses.

There appears to be no strong correlation between any of the measured cellular and biochemical changes and changes in lung function measurements, suggesting that different mechanisms may be responsible for these processes (Balmes et al., 1996; Devlin et al., 1991). The idea of different mechanisms is supported by a study in which ibuprofen, a cyclooxygenase inhibitor, blunted the O_3-induced decrements in lung function without altering the O_3-induced increase in PMNs or epithelial cell permeability (Hazucha et al., 1996). In vitro studies suggest that epithelial cells are the primary target of O_3 in the lung and that O_3 induces them to produce many of the mediators found in the BAL fluid of humans exposed to O_3. Although O_3 does not induce AMs to produce these compounds in large quantities, it does directly impair the ability of AMs to phagocytize and kill microorganisms.

A number of studies have found that O_3 exposures increases epithelial cell permeability through direct (technetium-99m labeled diethylene triamine pentaacetic acid, ^{99m}Tc-DTPA, clearance) and indirect (e.g, increased BAL albumin, protein) techniques. Kehrl et al. (1987) showed increased ^{99m}Tc-DTPA clearance in healthy young adults at 75 minutes postexposure to 0.4 ppm O_3 for 2 h. More recently, Foster and Stetkiewicz (1996) have shown that increased ^{99m}Tc-DTPA clearance persists for at least 18-20 h post-O_3 exposure (130 min to average O_3 concentration of 0.24 ppm) and the effect is greater at the lung apices than at the base. Increased BAL protein, suggesting O_3-induced changes in epithelial permeability, have also been reported at 1 h and 18 h postexposure (Balmes et al., 1996; Devlin et al., 1997). A recent meta-analysis of results from 21 publications (Mudway and Kelly, 2004), showed that increased BAL protein is associated with total ozone dose (product of O_3 concentration, exposure duration, and $\dot{V}_E$). Changes in permeability associated with acute inflammation may provide increased access of inhaled antigens, particles, and other substances to the smooth muscle, interstitial cells, and the blood.

In addition to affecting epithelial permeability and AM-mediated clearance in the respiratory region of the lung, mucociliary clearance of the tracheobronchial airways is also affected by O_3 exposure. Only two studies (Foster et al., 1987; Gerrity et al., 1993) have investigated the effect of O_3 exposure on mucociliary particle clearance in humans. Foster et al. (1987) measured clearance during and after a 2 h exposure to 0.4 ppm O_3. Gerrity et al. (1993) measured clearance at 2 h postexposure (0.4 ppm O_3), by which time, sRaw had returned to baseline and FVC was within 5% of baseline (versus an 11% decrement immediately postexposure). Foster et al. (1987) found a stimulatory effect of acute O_3 exposure on mucociliary clearance. Gerrity et al. (1993), who observed no effect on clearance, suggested that transient clearance increases are coincident to pulmonary function responses. Investigators in both studies suggested that O_3-induced increases in mucociliary clearance could be mediated by cholinergic receptors.

6.10 EXTRAPULMONARY EFFECTS OF OZONE

Ozone reacts rapidly on contact with respiratory system tissue and is not absorbed or transported to extrapulmonary sites to any significant degree as such. Human exposure studies discussed in the previous criteria documents (U.S. Environmental Protection Agency, 1986, 1996) failed to demonstrate any consistent extrapulmonary effects. More recently, some human exposure studies have attempted to identify specific markers of exposure to O_3 in blood. Foster et al. (1996) found a reduction in the serum levels of the free radical scavenger α-tocopherol after O_3 exposure. Liu et al. (1997, 1999) used a salicylate metabolite, 2,3, dehydroxybenzoic acid (DHBA), to indicate increased levels of hydroxyl radical which hydroxylates salicylate to DHBA. Increased DHBA levels after exposure to 0.12 and 0.40 ppm suggest that O_3 increases production of hydroxyl radical. The levels of DHBA were correlated with changes in spirometry.

Gong et al. (1998) observed a statistically significant O_3-induced increase the alveolar-to-arterial PO_2 gradient in both healthy (n = 6) and hypertensive (n = 10) adult males (41-78 years old) exposed for 3 h with IE ($\dot{V}_E \approx 30$ L/min) to 0.3 ppm O_3. The mechanism for the decrease in arterial oxygen tension in the Gong et al. (1998) study could be due to an O_3-induced ventilation-perfusion mismatch. Foster et al. (1993) has demonstrated that even in relatively young healthy

adults (26.7 ± 7 years old), O_3 exposure can cause ventilation to shift away from the well perfused basal lung. This effect of O_3 on ventilation distribution [and by association the small airways] may persist beyond 24-h post-exposure (Foster et al., 1997). Gong et al. (1998) suggested that by impairing alveolar-arterial oxygen transfer, the O_3 exposure could potentially lead to adverse cardiac events by decreasing oxygen supply to the myocardium. The subjects in the Gong et al. (1998) study had sufficient functional reserve so as to not experience significant ECG changes or myocardial ischemia and/or injury.

Effects of O_3 exposure on alveolar-arterial oxygen gradients may be more pronounced in patients with preexisting obstructive lung diseases. Relative to healthy elderly subjects, COPD patients have reduced gas exchange and low SaO_2. Any inflammatory or edematous responses due to O_3 delivered to the well ventilated regions of the COPD lung could further inhibit gas exchange and reduce oxygen saturation. In addition, O_3-induced vasoconstriction could also acutely induce pulmonary hypertension. Inducing pulmonary vasoconstriction and hypertension in these patients would perhaps worsen their condition, especially if their right ventricular function was already compromised.

6.11 EFFECTS OF OZONE MIXED WITH OTHER POLLUTANTS

Over the past 10 years only a handful of human controlled studies have examined the effects of pollutant mixtures containing O_3. The studies summarized in this section complement the studies reviewed in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). (*The complexities of O_3 and co-pollutant exposures in animal studies are discussed in Section 5.4.4*).

The results of a controlled study on children (Linn et al., 1997), designed to approximate exposure conditions of an epidemiologic study (Neas et al., 1995) by matching the population and exposure atmosphere (0.1 ppm O_3, 0.1 ppm SO_2 and 101 $\mu g/m^2$ H_2SO_4), did not support the findings of this epidemiologic study. The study points out the difficulties in attempting to link the outcomes of epidemiologic and controlled studies. Another vulnerable population, asthmatics, demonstrated enhanced airway reactivity to house dust mite following exposures to O_3, NO_2, and the combination of the two gases. Spirometric response, however, was impaired only by O_3, and O_3 + NO_2 at higher concentrations (Jenkins et al., 1999). Continuous exposure

to SO_2 and NO_2 increases inhaled bolus O_3 absorption, while continuous exposure to O_3 decreases O_3 bolus absorption (Rigas et al., 1997). Inhalation of a mixture of $PM_{2.5}$ and O_3 by healthy subjects increased brachial artery tone and reactivity (Brook et al., 2002). Since no other cardiovascular endpoints were affected by the exposure, the pathophysiological importance of this observation remains uncertain. However, acute pulmonary hypertension due to O_3-induced vasoconstriction could pose a risk to individuals with cardiovascular disease (*see Section 6-10*).

All in all, the contention that air pollutant mixtures elicit stronger pathophysiologic effects than individual pollutants of the mix is only weakly supported by human studies of either healthy or at-risk population.

6.12 CONTROLLED STUDIES OF AMBIENT AIR EXPOSURES

A large amount of informative O_3 exposure-effects data has been obtained in controlled laboratory exposure studies under a variety of different experimental conditions. However, laboratory simulation of the variable pollutant mixtures present in ambient air is not practical. Thus, the exposure effects of one or several artificially generated pollutants (i.e., a simple mixture) on pulmonary function and symptoms may not explain responses to ambient air where complex pollutant mixtures exist.

6.12.1 Mobile Laboratory Studies

Quantitatively useful information on the effects of acute exposure to photochemical oxidants on pulmonary function and symptoms responses from field studies using a mobile laboratory were presented in prior criteria documents (U.S. Environmental Protection Agency, 1986, 1996). Relative to controlled exposure studies, mobile laboratory ambient air studies suffer the additional limation of a dependence on ambient outdoor conditions. Consistent with controlled exposure studies, mobile studies in California demonstrated that pulmonary effects from exposure to ambient air in Los Angeles are related to O_3 concentration and level of exercise. Healthy subjects with a history of allergy also appeared to be more responsive to O_3 than "nonallergic" subjects (Linn et al., 1980, 1983b), although a standardized evaluation of atopic status was not performed.

6.12.2 Aircraft Cabin Studies

Respiratory symptoms and pulmonary function effects resulting from exposure to O_3 in commercial aircraft flying at high altitudes, and in altitude-simulation studies, have been assessed previously (U.S. Environmental Protection Agency, 1986, 1996). Commercial aircraft cabin O_3 levels were reported to be very low (average concentration 0.01 to 0.02 ppm) during 92 randomly selected smoking and nonsmoking flights in 1989 (Nagda et al., 1989). None of these flights recorded O_3 concentrations exceeding the 3-h time-weighted average (TWA) standard of 0.10 ppm promulgated by the U.S. Federal Aviation Administration (FAA, 1980), probably due to the use of O_3-scrubbing catalytic filters (Melton, 1990).

Ozone contamination aboard high-altitude aircraft also has been an interest to the U.S. Air Force because of complaints by crew members of frequent symptoms of dryness and irritation of the eyes, nose, and throat and an occasional cough (Hetrick et al., 2000). Despite the lack of ventilation system modifications as used in commercial aircraft, the O_3 concentrations never exceeded the FAA ceiling limit of 0.25 ppm and exceeded the 3-h TWA of 0.10 ppm only 7% of the total monitored flight time (43 h). The authors concluded that extremely low average relative humidity (12%) during flight operations was most likely responsible for the reported symptoms.

6.13 SUMMARY

Responses in humans exposed to ambient O_3 concentrations include decreased inspiratory capacity; mild bronchoconstriction; rapid, shallow breathing pattern during exercise; and symptoms of cough and pain on deep inspiration. Reflex inhibition of inspiration results in a decrease in forced vital capacity (FVC) and, in combination with mild bronchoconstriction, contributes to a decrease in the forced expiratory volume in 1 s (FEV_1). In addition to physiological pulmonary responses and symptoms of breathing discomfort, O_3 exposure also results in airway hyperresponsiveness, inflammation, immune system activation, and epithelial injury. With repeated O_3 exposures over several days, spirometric and symptom responses become attenuated, but this tolerance is lost after about a week without exposure. Airway responsiveness also appears to be attenuated with repeated O_3 exposures, but less than FEV_1.

Unlike spirometric and symptom responses, airway inflammation and small airways dysfunction may not become attenuated by repeated O_3 exposures.

Young healthy adults exposed to O_3 concentrations $\geq$0.08 ppm develop significant reversible, transient decrements in pulmonary function if minute ventilation ($\dot{V}_E$) or duration of exposure are increased sufficiently. The pattern of FEV_1 response appears to depend on the O_3 exposure profile. Triangular exposure profiles can potentially lead to greater FEV_1 responses than square wave exposures at equivalent average O_3 doses. O_3-induced decrements in FEV_1 do not appear to depend on gender, race, body surface area, height, lung size, or baseline FVC in young healthy adults. Healthy children experience similar spirometric responses but lesser symptoms from O_3 exposure relative to young adults. On average, spirometric and symptom responses to O_3 exposure appear to decline with increasing age beyond approximately 18 years of age. There is a large degree of intersubject variability in physiologic and symptomatic responses of heathy adults exposed to O_3. However, responses tend to be reproducible within a given individual over a period of several months. With increasing O_3 concentration, the distribution of FEV_1 decrements becomes asymmetrical with a few individuals experiencing large decrements.

There is a tendency for slightly increased spirometric responses in mild asthmatics and allergic rhinitics relative to healthy young adults. Spirometric responses in asthmatics appear to be affected by baseline lung function, i.e., responses increase with disease severity. With repeated daily O_3 exposures, spirometric responses of asthmatics become attenuated; however, airway responsiveness becomes increased in subjects with preexisting allergic airway disease (with or without asthma). Possibly due to patient age, O_3 exposure does not appear to cause significant pulmonary function impairment or evidence of cardiovascular strain in patients with cardiovascular disease or chronic obstructive pulmonary disease relative to healthy subjects.

Available information on recovery from O_3 exposure indicates that an initial phase of recovery in healthy individuals proceeds relatively rapidly, with acute spirometric and symptom responses resolving within about 2 to 4 h. Small residual lung function effects are almost completely resolved within 24 hours. Effects of O_3 on the small airways, assessed by persistent decrement in FEF_{25-75} and altered ventilation distribution, may be due in part to inflammation. Indeed, a prolonged recovery of residual spirometric decrements following the initial rapid recovery could be due to slowly resolving airway inflammation. In hyperresponsive individuals,

this recovery takes longer, as much as 48 hours, to return to baseline values. Persistent spirometry changes observed for up to 48 h postexposure could plausibly be sustained by the inflammatory mediators. Cellular responses (e.g., release of immunomodulatory cytokines) appear to still be active as late as 20 h postexposure. More slowly developing inflammatory and cellular changes may persist for up to 48 h, but the time course for these parameters in humans has not been explored fully.

Soluble mediators of inflammation such as the cytokines (IL-6, IL-8) and arachidonic acid metabolites (e.g., PGE_2, $PGF_{2\alpha}$, thromboxane, and leukotrienes [LTs] such as LTB_4) have been measured in the BAL fluid of humans exposed to O_3. Many of these compounds have bronchoconstrictive properties and may be involved in increased airway responsiveness following O_3 exposure. Some indicators of inflammation (e.g., PMN influx, IL-6, PGE_2, fibronectin) are attenuated with repeated O_3 exposures. However, indicating that tissue damage probably continues to occur during repeated O_3 exposure, other markers (LDH, IL-8, total protein, epithelial cells) do not show attenuation. There appears to be no strong correlation between any of the measured cellular and biochemical changes and changes in lung function measurements. A limited number of studies suggest that inflammatory responses may be detected following O_3 exposures that are insufficient to cause decrements in pulmonary function. Whether airway reactivity or inflammatory responses to O_3 are dependent on the age of the exposed individual, such as spirometric responses, has not been determined.

Dietary antioxidant supplementation attenuates O_3-induced spirometric responses but not the intensity of subjective symptoms nor inflammatory responses. Dietary antioxidants also afforded partial protection to asthmatics by attenuating postexposure bronchial hyperresponsiveness.

REFERENCES

Adams, W. C. (2000) Ozone dose-response effects of varied equivalent minute ventilation rates. J. Exposure Anal. Environ. Epidemiol. 10: 217-226.

Adams, W. C. (2002) Comparison of chamber and face-mask 6.6-hour exposures to ozone on pulmonary function and symptoms responses. Inhalation Toxicol. 14: 745-764.

Adams, W. C. (2003a) Comparison of chamber and face mask 6.6-hour exposure to 0.08 ppm ozone via square-wave and triangular profiles on pulmonary responses. Inhalation Toxicol. 15: 265-281.

Adams, W. C. (2003b) Relation of pulmonary responses induced by 6.6-h exposures to 0.08 ppm ozone and 2-h exposures to 0.30 ppm ozone via chamber and face-mask inhalation. Inhalation Toxicol. 15: 745-759.

Adams, W. C.; Ollison, W. M. (1997) Effects of prolonged simulated ambient ozone dosing patterns on human pulmonary function and symptomatology. Presented at: 90th annual meeting of the Air & Waste Management Association; June; Toronto, Ontario, Canada. Pittsburgh, PA: Air & Waste Management Association; paper no. 97-MP9.02.

Adams, W. C.; Schelegle, E. S. (1983) Ozone and high ventilation effects on pulmonary function and endurance performance. J. Appl. Physiol.: Respir. Environ. Exercise Physiol. 55: 805-812.

Adams, W. C.; Savin, W. M.; Christo, A. E. (1981) Detection of ozone toxicity during continuous exercise via the effective dose concept. J. Appl. Physiol.: Respir. Environ. Exercise Physiol. 51: 415-422.

Alexis, N.; Urch, B.; Tarlo, S.; Corey, P.; Pengelly, D.; O'Byrne, P.; Silverman, F. (2000) Cyclooxygenase metabolites play a different role in ozone-induced pulmonary function decline in asthmatics compared to normals. Inhalation Toxicol. 12: 1205-1224.

Alpert, S. E.; Walenga, R. W.; Jaspers, I.; Qu, Q.; Chen, L. C. (1997) Ozone inactivates cyclooxygenase in human tracheal epithelial cells without altering PGHS-2 mRNA or protein. Am. J. Physiol. 272: L879-L887.

Aris, R. M.; Christian, D.; Hearne, P. Q.; Kerr, K.; Finkbeiner, W. E.; Balmes, J. R. (1993) Ozone-induced airway inflammation in human subjects as determined by airway lavage and biopsy. Am. Rev. Respir. Dis. 148: 1363-1372.

Aris, R. M.; Tager, I.; Christian, D.; Kelly, T.; Balmes, J. R. (1995) Methacholine responsiveness is not associated with O_3-induced decreases in FEV_1. Chest 107: 621-628.

Avissar, N. E.; Reed, C. K.; Cox, C.; Frampton, M. W.; Finkelstein, J. N. (2000) Ozone, but not nitrogen dioxide, exposure decreases glutathione peroxidases in epithelial lining fluid of human lung. Am. J. Respir. Crit. Care Med. 162: 1342-1347.

Balmes, J. R.; Chen, L. L.; Scannell, C.; Tager, I.; Christian, D.; Hearne, P. Q.; Kelly, T.; Aris, R. M. (1996) Ozone-induced decrements in FEV_1 and FVC do not correlate with measures of inflammation. Am. J. Respir. Crit. Care Med. 153: 904-909.

Balmes, J. R.; Aris, R. M.; Chen, L. L.; Scannell, C.; Tager, I. B.; Finkbeiner, W.; Christian, D.; Kelly, T.; Hearne, P. Q.; Ferrando, R.; Welch, B. (1997) Effects of ozone on normal and potentially sensitive human subjects. part I: airway inflammation and responsiveness to ozone in normal and asthmatic subjects. Cambridge, MA: Health Effects Institute. Research report no. 78; pp 1-37, 81-99.

Bascom, R.; Naclerio, R. M.; Fitzgerald, T. K.; Kagey-Sobotka, A.; Proud, D. (1990) Effect of ozone inhalation on the response to nasal challenge with antigen of allergic subjects. Am. Rev. Respir. Dis. 142: 594-601.

Basha, M. A.; Gross, K. B.; Gwizdala, C. J.; Haidar, A. H.; Popovich, J., Jr. (1994) Bronchoalveolar lavage neutrophilia in asthmatic and healthy volunteers after controlled exposure to ozone and filtered purified air. Chest 106: 1757-1765.

Beckett, W. S.; McDonnell, W. F.; Horstman, D. H.; House, D. E. (1985) Role of the parasympathetic nervous system in acute lung response to ozone. J. Appl. Physiol. 59: 1879-1885.

Bedi, J. F.; Drechsler-Parks, D. M.; Horvath, S. M. (1985) Duration of increased pulmonary function sensitivity to an initial ozone exposure. Am. Ind. Hyg. Assoc. J. 46: 731-734.

Bergamaschi, E.; De Palma, G.; Mozzoni, P.; Vanni, S.; Vettori, M. V.; Broeckaert, F.; Bernard, A.; Mutti, A. (2001) Polymorphism of quinone-metabolizing enzymes and susceptibility to ozone-induced acute effects. Am. J. Respir. Crit. Care Med. 163: 1426-1431.

Blomberg, A.; Helleday, R.; Pourazar, J.; Stenfors, N.; Kelly, F. J.; Frew, A. J.; Holgate, S. T.; Sandström, T. (1997) Early airway and peripheral blood cell responses to 0.20 ppm ozone in healthy human subjects. Eur. Respir. J. 10(suppl 25): 274S.

Blomberg, A.; Mudway, I. S.; Nordenhäll, C.; Hedenström, H.; Kelly, F. J.; Frew, A. J.; Holgate, S. T.; Sandström, T. (1999) Ozone-induced lung function decrements do not correlate with early airway inflammatory or antioxidant responses. Eur. Respir. J. 13: 1418-1428.

Bosson, J.; Stenfors, N.; Bucht, A.; Helleday, R.; Pourazar, J.; Holgate, S. T.; Kelly, F. J.; Sandström, T.; Wilson, S.; Frew, A. J.; Blomberg, A. (2003) Ozone-induced bronchial epithelial cytokine expression differs between healthy and asthmatic subjects. Clin. Exp. Allergy 33: 777-782.

Brook, R. D.; Brook, J. R.; Urch, B.; Vincent, R.; Rajagopalan, S.; Silverman, F. (2002) Inhalation of fine particulate air pollution and ozone causes acute arterial vasoconstriction in healthy adults. Circulation 105: 1534-1536.

Bush, M. L.; Asplund, P. T.; Miles, K. A.; Ben-Jebria, A.; Ultman, J. S. (1996) Longitudinal distribution of O_3 absorption in the lung: gender differences and intersubject variability. J. Appl. Physiol. 81: 1651-1657.

Christian, D. L.; Chen, L. L.; Scannell, C. H.; Ferrando, R. E.; Welch, B. S.; Balmes, J. R. (1998) Ozone-induced inflammation is attenuated with multiday exposure. Am. J. Respir. Crit. Care Med. 158: 532-537.

Coleridge, J. C. G.; Coleridge, H. M.; Schelegle, E. S.; Green, J. F. (1993) Acute inhalation of ozone stimulates bronchial C-fibers and rapidly adapting receptors in dogs. J. Appl. Physiol. 74: 2345-2352.

Corradi, M.; Alinovi, R.; Goldoni, M.; Vettori, M.; Folesani, G.; Mozzoni, P.; Cavazzini, S.; Bergamaschi, E.; Rossi, L.; Mutti, A. (2002) Biomarkers of oxidative stress after controlled human exposure to ozone. Toxicol. Lett. 134: 219-225.

David, G. L.; Romieu, I.; Sienra-Monge, J. J.; Collins, W. J.; Ramirez-Aguilar, M.; Del Rio-Navarro, B. E.; Reyes-Ruiz, N. I.; Morris, R. W.; Marzec, J. M.; London, S. J. (2003) Nicotinamide adenine dinucleotide (Phosphate) reduced:quinone oxidoreductase and glutathione s-transferase m1 polymorphism and childhood asthma. Am. J. Respir. Crit. Care Med. 168: 1199-1204.

Devlin, R. B.; McDonnell, W. F.; Mann, R.; Becker, S.; House, D. E.; Schreinemachers, D.; Koren, H. S. (1991) Exposure of humans to ambient levels of ozone for 6.6 hours causes cellular and biochemical changes in the lung. Am. J. Respir. Cell Mol. Biol. 4: 72-81.

Devlin, R. B.; McDonnell, W. F.; Becker, S.; Madden, M. C.; McGee, M. P.; Perez, R.; Hatch, G.; House, D. E.; Koren, H. S. (1996) Time-dependent changes of inflammatory mediators in the lungs of humans exposed to 0.4 ppm ozone for 2 hr: a comparison of mediators found in bronchoalveolar lavage fluid 1 and 18 hr after exposure. Toxicol. Appl. Pharmacol. 138: 176-185.

Devlin, R. B.; Folinsbee, L. J.; Biscardi, F.; Hatch, G.; Becker, S.; Madden, M. C.; Robbins, M.; Koren, H. S. (1997) Inflammation and cell damage induced by repeated exposure of humans to ozone. Inhalation Toxicol. 9: 211-235.

Dimeo, M. J.; Glenn, M. G.; Holtzman, M. J.; Sheller, J. R.; Nadel, J. A.; Boushey, H. A. (1981) Threshold concentration of ozone causing an increase in bronchial reactivity in humans and adaptation with repeated exposures. Am. Rev. Respir. Dis. 124: 245-248.

Federal Aviation Administration. (1980) Airplane cabin ozone contamination. F. R. (January 21) 45: 3880-3883.

Folinsbee, L. J.; Hazucha, M. J. (1989) Persistence of ozone-induced changes in lung function and airway responsiveness. In: Schneider, T.; Lee, S. D.; Wolters, G. J. R.; Grant, L. D., eds. Atmospheric ozone research and its policy implications: proceedings of the 3rd US-Dutch international symposium; May 1988; Nijmegen, The Netherlands. Amsterdam, The Netherlands: Elsevier Science Publishers; pp. 483-492. (Studies in environmental science 35).

Folinsbee, L. J.; Hazucha, M. J. (2000) Time course of response to ozone exposure in healthy adult females. Inhalation Toxicol. 12: 151-167.

Folinsbee, L. J.; Horvath, S. M. (1986) Persistence of the acute effects of ozone exposure. Aviat. Space Environ. Med. 57: 1136-1143.

Folinsbee, L. J.; Silverman, F.; Shephard, R. J. (1977) Decrease of maximum work performance following ozone exposure. J. Appl. Physiol.: Respir. Environ. Exercise Physiol. 42: 531-536.

Folinsbee, L. J.; Drinkwater, B. L.; Bedi, J. F.; Horvath, S. M. (1978) The influence of exercise on the pulmonary function changes due to exposure to low concentrations of ozone. In: Folinsbee, L. J.; Wagner, J. A.; Borgia, J. F.; Drinkwater, B. L.; Gliner, J. A.; Bedi, J. F., eds. Environmental stress: individual human adaptations. New York, NY: Academic Press; pp. 125-145.

Folinsbee, L. J.; Bedi, J. F.; Horvath, S. M. (1980) Respiratory responses in humans repeatedly exposed to low concentrations of ozone. Am. Rev. Respir. Dis. 121: 431-439.

Folinsbee, L. J.; McDonnell, W. F.; Horstman, D. H. (1988) Pulmonary function and symptom responses after 6.6-hour exposure to 0.12 ppm ozone with moderate exercise. JAPCA 38: 28-35.

Folinsbee, L. J.; Horstman, D. H.; Kehrl, H. R.; Harder, S.; Abdul-Salaam, S.; Ives, P. J. (1994) Respiratory responses to repeated prolonged exposure to 0.12 ppm ozone. Am. J. Respir. Crit. Care Med. 149: 98-105.

Folinsbee, L. J.; Devlin, R. B.; Robbins, M. K.; Biscardi, F. H.; Abdul-Salaam, S.; Koren, H. S. (1998) Repeated exposure of humans to ozone: pulmonary function and symptom responses. Research Triangle Park, NC: U.S. Environmental Protection Agency; National Center for Environmental Assessment; unpublished data.

Foster, W. M.; Stetkiewicz, P. T. (1996) Regional clearance of solute from the respiratory epithelia: 18-20 h postexposure to ozone. J. Appl. Physiol. 81: 1143-1149.

Foster, W. M.; Costa, D. L.; Langenback, E. G. (1987) Ozone exposure alters tracheobronchial mucociliary function in humans. J. Appl. Physiol. 63: 996-1002.

Foster, W. M.; Silver, J. A.; Groth, M. L. (1993) Exposure to ozone alters regional function and particle dosimetry in the human lung. J. Appl. Physiol. 75: 1938-1945.

Foster, W. M.; Wills-Karp, M.; Tankersley, C. G.; Chen, X.; Paquette, N. C. (1996) Bloodborne markers in humans during multiday exposure to ozone. J. Appl. Physiol. 81: 794-800.

Foster, W. M.; Weinmann, G. G.; Menkes, E.; Macri, K. (1997) Acute exposure of humans to ozone impairs small airway function. Ann. Occup. Hyg. 41(suppl. 1): 659-666.

Foster, W. M.; Brown, R. H.; Macri, K.; Mitchell, C. S. (2000) Bronchial reactivity of healthy subjects: 18-20 h postexposure to ozone. J. Appl. Physiol. 89: 1804-1810.

Fox, S. D.; Adams, W. C.; Brookes, K. A.; Lasley, B. L. (1993) Enhanced response to ozone exposure during the follicular phase of the menstrual cycle. Environ. Health Perspect. 101: 242-244.

Foxcroft, W. J.; Adams, W. C. (1986) Effects of ozone exposure on four consecutive days on work performance and $\dot{V}O_{2max}$. J. Appl. Physiol. 61: 960-966.

Frampton, M. W.; Morrow, P. E.; Torres, A.; Cox, C.; Voter, K. Z.; Utell, M. J.; Gibb, F. R.; Speers, D. M. (1997) Ozone responsiveness in smokers and nonsmokers. Am. J. Respir. Crit. Care Med. 155: 116-121.

Frank, R.; Liu, M. C.; Spannhake, E. W.; Mlynarek, S.; Macri, K.; Weinmann, G. G. (2001) Repetitive ozone exposure of young adults: evidence of persistent small airway dysfunction. Am. J. Respir. Crit. Care Med. 164: 1253-1260.

Freed, A. N.; Chou, C. L.; Fuller, S. D.; Croxton, T. L. (1996) Ozone-induced vagal reflex modulates airways reactivity in rabbits. Respir. Physiol. 105: 95-102.

Gabrielson, E. W.; Yu, X.-Y.; Spannhake, E. W. (1994) Comparison of the toxic effects of hydrogen peroxide and ozone on cultured human bronchial epithelial cells. Environ. Health Perspect. 102: 972-974.

Gerrity, T. R.; Bennett, W. D.; Kehrl, H.; DeWitt, P. J. (1993) Mucociliary clearance of inhaled particles measured at 2 h after ozone exposure in humans. J. Appl. Physiol. 74: 2984-2989.

Gong, H., Jr.; Bradley, P. W.; Simmons, M. S.; Tashkin, D. P. (1986) Impaired exercise performance and pulmonary function in elite cyclists during low-level ozone exposure in a hot environment. Am. Rev. Respir. Dis. 134: 726-733.

Gong, H., Jr.; Bedi, J. F.; Horvath, S. M. (1988) Inhaled albuterol does not protect against ozone toxicity in nonasthmatic athletes. Arch. Environ. Health 43: 46-53.

Gong, H., Jr.; Shamoo, D. A.; Anderson, K. R.; Linn, W. S. (1997a) Responses of older men with and without chronic obstructive pulmonary disease to prolonged ozone exposure. Arch. Environ. Health 52: 18-25.

Gong, H., Jr.; McManus, M. S.; Linn, W. S. (1997b) Attenuated response to repeated daily ozone exposures in asthmatic subjects. Arch. Environ. Health 52: 34-41.

Gong, H., Jr.; Wong, R.; Sarma, R. J.; Linn, W. S.; Sullivan, E. D.; Shamoo, D. A.; Anderson, K. R.; Prasad, S. B. (1998) Cardiovascular effects of ozone exposure in human volunteers. Am. J. Respir. Crit. Care Med. 158: 538-546.

Graham, D. E.; Koren, H. S. (1990) Biomarkers of inflammation in ozone-exposed humans: comparison of the nasal and bronchoalveolar lavage. Am. Rev. Respir. Dis. 142: 152-156.

Graham, D.; Henderson, F.; House, D. (1988) Neutrophil influx measured in nasal lavages of humans exposed to ozone. Arch. Environ. Health 43: 228-233.

Gwynn, R. C.; Thurston, G. D. (2001) The burden of air pollution: impacts among racial minorities. Environ. Health Perspect. Suppl. 109(4): 501-506.

Hamilton, R. F.; Li, L.; Eschenbacher, W. L.; Szweda, L.; Holian, A. (1998) Potential involvement of 4-hydroxynonenal in the response of human lung cells to ozone. Am. J. Physiol. 274: L8-L16.

Hasday, J. D.; Bascom, R.; Costa, J. J.; Fitzgerald, T.; Dubin, W. (1999) Bacterial endotoxin is an active component of cigarette smoke. Chest 115: 829-835.

Hazbun, M. E.; Hamilton, R.; Holian, A.; Eschenbacher, W. L. (1993) Ozone-induced increases in substance P and 8-epi-prostaglandin $F_{2\alpha}$ in the airways of human subjects. Am. J. Respir. Cell Mol. Biol. 9: 568-572.

Hazucha, M. J.; Sant'Ambrogio, G. (1993) Effects of ozone on the activity of slowly (SAR) and rapidly adapting (RAR) receptors in cats. FASEB J. 7: 407A.

Hazucha, M. J.; Folinsbee, L. J.; Seal, E., Jr. (1992) Effects of steady-state and variable ozone concentration profiles on pulmonary function. Am. Rev. Respir. Dis. 146: 1487-1493.

Hazucha, M. J.; Folinsbee, L. J.; Seal, E.; Bromberg, P. A. (1994) Lung function response of healthy women after sequential exposures to NO_2 and O_3. Am. J. Respir. Crit. Care Med. 150: 642-647.

Hazucha, M. J.; Madden, M.; Pape, G.; Becker, S.; Devlin, R.; Koren, H. S.; Kehrl, H.; Bromberg, P. A. (1996) Effects of cyclo-oxygenase inhibition on ozone-induced respiratory inflammation and lung function changes. Eur. J. Appl. Physiol. Occup. Med. 73: 17-27.

Hazucha, M. J.; Folinsbee, L. J.; Bromberg, P. A. (2003) Distribution and reproducibility of spirometric response to ozone by gender and age. J. Appl. Physiol. 95: 1917-1925.

Hetrick, S. M.; Gould, W. D.; Christensen, D. E. (2000) Inflight cabin ozone aboard long duration C-5 airlift missions: a historical issue revisited. Aviat. Space Environ. Med. 71: 408-414.

Hiltermann, T. J. N.; Stolk, J.; Hiemstra, P. S.; Fokkens, P. H. B.; Rombout, P. J. A.; Sont, J. K.; Sterk, P. J.; Dijkman, J. H. (1995) Effect of ozone exposure on maximal airway narrowing in non-asthmatic and asthmatic subjects. Clin. Sci. 89: 619-624.

Hiltermann, T. J. N.; de Bruijne, C. R.; Stolk, J.; Zwinderman, A. H.; Spieksma, F. Th. M.; Roemer, W.; Steerenberg, P. A.; Fischer, P. H.; van Bree, L.; Hiemstra, P. S. (1997) Effects of photochemical air pollution and allergen exposure on upper respiratory tract inflammation in asthmatics. Am. J. Respir. Crit. Care Med. 156: 1765-1772.

Hiltermann, J. T. N.; Lapperre, T. S.; Van Bree, L.; Steerenberg, P. A.; Brahim, J. J.; Sont, J. K.; Sterk, P. J.; Hiemstra, P. S.; Stolk, J. (1999) Ozone-induced inflammation assessed in sputum and bronchial lavage fluid from asthmatics: a new noninvasive tool in epidemiologic studies on air pollution and asthma. Free Radical Biol. Med. 27: 1448-1454.

Holz, O.; Jörres, R. A.; Timm, P.; Mücke, M.; Richter, K.; Koschyk, S.; Magnussen, H. (1999) Ozone-induced airway inflammatory changes differ between individuals and are reproducible. Am. J. Respir. Crit. Care Med. 159: 776-784.

Holz, O.; Mücke, M.; Paasch, K.; Böhme, S.; Timm, P.; Richter, K.; Magnussen, H.; Jörres, R. A. (2002) Repeated ozone exposures enhance bronchial allergen responses in subjects with rhinitis or asthma. Clin. Exp. Allergy. 32: 681-689.

Holz, O.; Tal-Singer, R.; Kanniess, F.; Simpson, K. J.; Gibson, A.; Vessey, R. S. J.; Janicki, S.; Magnussen, H.; Jörres, R. A.; Richter, K. (2005) Validation of the human ozone challenge model as a tool for assessing anti-inflammatory drugs in early development. J. Clin. Pharmacol. 45: 498-503.

Höppe, P.; Peters, A.; Rabe, G.; Praml, G.; Lindner, J.; Jakobi, G.; Fruhmann, G.; Nowak, D. (2003) Environmental ozone effects in different population subgroups. Int. J. Hyg. Environ. Health 206: 505-516.

Horstman, D. H.; Folinsbee, L. J.; Ives, P. J.; Abdul-Salaam, S.; McDonnell, W. F. (1990) Ozone concentration and pulmonary response relationships for 6.6-hour exposures with five hours of moderate exercise to 0.08, 0.10, and 0.12 ppm. Am. Rev. Respir. Dis. 142: 1158-1163.

Horstman, D. H.; Ball, B. A.; Brown, J.; Gerrity, T.; Folinsbee, L. J. (1995) Comparison of pulmonary responses of asthmatic and nonasthmatic subjects performing light exercise while exposed to a low level of ozone. Toxicol. Ind. Health 11: 369-385.

Horvath, S. M.; Gliner, J. A.; Matsen-Twisdale, J. A. (1979) Pulmonary function and maximum exercise responses following acute ozone exposure. Aviat. Space Environ. Med. 50: 901-905.

Horvath, S. M.; Gliner, J. A.; Folinsbee, L. J. (1981) Adaptation to ozone: duration of effect. Am. Rev. Respir. Dis. 123: 496-499.

Housley, D. G.; Eccles, R.; Richards, R. J. (1996) Gender difference in the concentration of the antioxidant uric acid in human nasal lavage. Acta Oto-Laryngol. 116: 751-754.

Jenkins, H. S.; Devalia, J. L.; Mister, R. L.; Bevan, A. M.; Rusznak, C.; Davies, R. J. (1999) The effect of exposure to ozone and nitrogen dioxide on the airway response of atopic asthmatics to inhaled allergen: dose- and time-dependent effects. Am. J. Respir. Crit. Care Med. 160: 33-39.

Joad, J. P.; Kott, K. S.; Bric, J. M. (1996) The local C-fiber contribution to ozone-induced effects on the isolated guinea pig lung. Toxicol. Appl. Pharmacol. 141: 561-567.

Jörres, R.; Nowak, D.; Magnussen, H.; Speckin, P.; Koschyk, S. (1996) The effect of ozone exposure on allergen responsiveness in subjects with asthma or rhinitis. Am. J. Respir. Crit. Care Med. 153: 56-64.

Jörres, R. A.; Holz, O.; Zachgo, W.; Timm, P.; Koschyk, S.; Müller, B.; Grimminger, F.; Seeger, W.; Kelly, F. J.; Dunster, C.; Frischer, T.; Lubec, G.; Waschewski, M.; Niendorf, A.; Magnussen, H. (2000) The effect of repeated ozone exposures on inflammatory markers in bronchoalveolar lavage fluid and mucosal biopsies. Am. J. Respir. Crit. Care Med. 161: 1855-1861.

Kehrl, H. R.; Hazucha, M. J.; Solic, J. J.; Bromberg, P. A. (1985) Responses of subjects with chronic obstructive pulmonary disease after exposures to 0.3 ppm ozone. Am. Rev. Respir. Dis. 131: 719-724.

Kehrl, H. R.; Vincent, L. M.; Kowalsky, R. J.; Horstman, D. H.; O'Neil, J. J.; McCartney, W. H.; Bromberg, P. A. (1987) Ozone exposure increases respiratory epithelial permeability in humans. Am. Rev. Respir. Dis. 135: 1124-1128.

Kehrl, H. R.; Peden, D. B.; Ball, B. A.; Folinsbee, L. J.; Horstman, D. H. (1999) Increased specific airway reactivity of persons with mild allergic asthma after 7.6 hours of exposure to 0.16 ppm ozone. J. Allergy. Clin. Immunol. 104: 1198-1204.

Kleinman, M. T.; Mautz, W. J.; Bjarnason, S. (1999) Adaptive and non-adaptive responses in rats exposed to ozone, alone and in mixtures, with acidic aerosols. Inhalation Toxicol. 11: 249-264.

Koren, H. S.; Devlin, R. B.; Graham, D. E.; Mann, R.; McGee, M. P.; Horstman, D. H.; Kozumbo, W. J.; Becker, S.; House, D. E.; McDonnell, W. F.; Bromberg, P. A. (1989) Ozone-induced inflammation in the lower airways of human subjects. Am. Rev. Respir. Dis. 139: 407-415.

Koren, H. S.; Devlin, R. B.; Becker, S.; Perez, R.; McDonnell, W. F. (1991) Time-dependent changes of markers associated with inflammation in the lungs of humans exposed to ambient levels of ozone. Toxicol. Pathol. 19: 406-411.

Kozumbo, W. J.; Hanley, N. M.; Agarwal, S.; Thomas, M. J.; Madden, M. C. (1996) Products of ozonized arachidonic acid potentiate the formation of DNA single strand breaks in cultured human lung cells. Environ. Mol. Mutagen. 27: 185-195.

Kreit, J. W.; Gross, K. B.; Moore, T. B.; Lorenzen, T. J.; D'Arcy, J.; Eschenbacher, W. L. (1989) Ozone-induced changes in pulmonary function and bronchial responsiveness in asthmatics. J. Appl. Physiol. 66: 217-222.

Krishna, M. T.; Springall, D.; Meng, Q.-H.; Withers, N.; Macleod, D.; Biscione, G.; Frew, A.; Polak, J.; Holgate, S. (1997) Effects of ozone on epithelium and sensory nerves in the bronchial mucosa of healthy humans. Am. J. Respir. Crit. Care Med. 156: 943-950.

Kulle, T. J.; Sauder, L. R.; Kerr, H. D.; Farrell, B. P.; Bermel, M. S.; Smith, D. M. (1982) Duration of pulmonary function adaptation to ozone in humans. Am. Ind. Hyg. Assoc. J. 43: 832-837.

Kulle, T. J.; Milman, J. H.; Sauder, L. R.; Kerr, H. D.; Farrell, B. P.; Miller, W. R. (1984) Pulmonary function adaptation to ozone in subjects with chronic bronchitis. Environ. Res. 34: 55-63.

Kulle, T. J.; Sauder, L. R.; Hebel, J. R.; Chatham, M. D. (1985) Ozone response relationships in healthy nonsmokers. Am. Rev. Respir. Dis. 132: 36-41.

Linder, J.; Herren, D.; Monn, C.; Wanner, H.-U. (1988) Die Wirkung von Ozon auf die körperliche Leistungsfähigkeit [The effect of ozone on physical activity]. Schweiz Z. Sportmed. 36: 5-10.

Linn, W. S.; Jones, M. P.; Bachmayer, E. A.; Spier, C. E.; Mazur, S. F.; Avol, E. L.; Hackney, J. D. (1980) Short-term respiratory effects of polluted ambient air: a laboratory study of volunteers in a high-oxidant community. Am. Rev. Respir. Dis. 121: 243-252.

Linn, W. S.; Fischer, D. A.; Medway, D. A.; Anzar, U. T.; Spier, C. E.; Valencia, L. M.; Venet, T. G.; Hackney, J. D. (1982a) Short-term respiratory effects of 0.12 ppm ozone exposure in volunteers with chronic obstructive pulmonary disease. Am. Rev. Respir. Dis. 125: 658-663.

Linn, W. S.; Medway, D. A.; Anzar, U. T.; Valencia, L. M.; Spier, C. E.; Tsao, F. S.-D.; Fischer, D. A.; Hackney, J. D. (1982b) Persistence of adaptation to ozone in volunteers exposed repeatedly for six weeks. Am. Rev. Respir. Dis. 125: 491-495.

Linn, W. S.; Shamoo, D. A.; Venet, T. G.; Spier, C. E.; Valencia, L. M.; Anzar, U. T.; Hackney, J. D. (1983a) Response to ozone in volunteers with chronic obstructive pulmonary disease. Arch. Environ. Health 38: 278-283.

Linn, W. S.; Avol, E. L.; Hackney, J. D. (1983b) Effects of ambient oxidant pollutants on humans: a movable environmental chamber study. In: Lee, S. D.; Mustafa, M. G.; Mehlman, M. A., eds. International symposium on the biomedical effects of ozone and related photochemical oxidants; March 1982; Pinehurst, NC. Princeton, NJ: Princeton Scientific Publishers, Inc.; pp. 125-137. (Advances in modern environmental toxicology: v. 5).

Linn, W. S.; Avol, E. L.; Shamoo, D. A.; Spier, C. E.; Valencia, L. M.; Venet, T. G.; Fischer, D. A.; Hackney, J. D. (1986) A dose-response study of healthy, heavily exercising men exposed to ozone at concentrations near the ambient air quality standard. Toxicol. Ind. Health 2: 99-112.

Linn, W. S.; Gong, H., Jr.; Shamoo, D. A.; Anderson, K. R.; Avol, E. L. (1997) Chamber exposures of children to mixed ozone, sulfur dioxide, and sulfuric acid. Arch. Environ. Health 52: 179-187.

Liu, L.; Leech, J. A.; Urch, R. B.; Silverman, F. S. (1997) *In vivo* salicylate hyroxylation: a potential biomarker for assessing acute ozone exposure and effects in humans. Am. J. Respir. Crit. Care Med. 156: 1405-1412.

Liu, L.; Leech, J. A.; Urch, R. B.; Poon, R.; Zimmerman, B.; Kubay, J. M.; Silverman, F. S. (1999) A comparison of biomarkers of ozone exposure in human plasma, nasal lavage, and sputum. Inhalation Toxicol. 11: 657-674.

Marthan, R.; Roux, E.; Savineau, J.-P. (1996) Human bronchial smooth muscle responsiveness after *in vitro* exposure to oxidizing pollutants. Cell Biol. Toxicol. 12: 245-249.

McBride, D. E.; Koenig, J. Q.; Luchtel, D. L.; Williams, P. V.; Henderson, W. R., Jr. (1994) Inflammatory effects of ozone in the upper airways of subjects with asthma. Am. J. Respir. Crit. Care Med. 149: 1192-1197.

McDonnell, W. F. (1996) Individual variability in human lung function responses to ozone exposure. Environ. Toxicol. Pharmacol. 2: 171-175.

McDonnell, W. F.; Horstman, D. H.; Hazucha, M. J.; Seal, E., Jr.; Haak, E. D.; Salaam, S. A.; House, D. E. (1983) Pulmonary effects of ozone exposure during exercise: dose-response characteristics. J. Appl. Physiol.: Respir. Environ. Exercise Physiol. 54: 1345-1352.

McDonnell, W. F., III; Chapman, R. S.; Leigh, M. W.; Strope, G. L.; Collier, A. M. (1985a) Respiratory responses of vigorously exercising children to 0.12 ppm ozone exposure. Am. Rev. Respir. Dis. 132: 875-879.

McDonnell, W. F., III; Horstman, D. H.; Abdul-Salaam, S.; House, D. E. (1985b) Reproducibility of individual responses to ozone exposure. Am. Rev. Respir. Dis. 131: 36-40.

McDonnell, W. F.; Kehrl, H. R.; Abdul-Salaam, S.; Ives, P. J.; Folinsbee, L. J.; Devlin, R. B.; O'Neil, J. J.; Horstman, D. H. (1991) Respiratory response of humans exposed to low levels of ozone for 6.6 hours. Arch. Environ. Health 46: 145-150.

McDonnell, W. F.; Muller, K. E.; Bromberg, P. A.; Shy, C. M. (1993) Predictors of individual differences in acute response to ozone exposure. Am. Rev. Respir. Dis. 147: 818-825.

McDonnell, W. F.; Stewart, P. W.; Andreoni, S.; Seal, E., Jr.; Kehrl, H. R.; Horstman, D. H.; Folinsbee, L. J.; Smith, M. V. (1997) Prediction of ozone-induced FEV_1 changes: effects of concentration, duration, and ventilation. Am. J. Respir. Crit. Care Med. 156: 715-722.

McDonnell, W. F.; Stewart, P. W.; Smith, M. V.; Pan, W. K.; Pan, J. (1999) Ozone-induced respiratory symptoms: exposure-response models and association with lung function. Eur. Respir. J. 14: 845-853.

McKenzie, D. C.; Stirling, D. R.; Fadl, S.; Allen, M. (1987) The effects of salbutamol on pulmonary function in cyclists exposed to ozone: a pilot study. Can. J. Sport Sci. 12: 46-48.

Melton, C. E. (1990) Airliner cabin ozone: an updated review. Washington, DC: Federal Aviation Administration, Office of Aviation Medicine; report no. DOT/FAA/AM-89/13. Available from: NTIS, Springfield, VA; AD-A219 264.

Messineo, T. D.; Adams, W. C. (1990) Ozone inhalation effects in females varying widely in lung size: comparison with males. J. Appl. Physiol. 69: 96-103.

Michelson, P. H.; Dailey, L.; Devlin, R. B.; Peden, D. B. (1999) Ozone effects on the immediate-phase response to allergen in the nasal airways of allergic asthmatic subjects. Otolaryngol. Head Neck Surg. 120: 225-232.

Mohammed, S. P.; Higenbottam, T. W.; Adcock, J. J. (1993) Effects of aerosol-applied capsaicin, histamine and prostaglandin-E2 on airway sensory receptors of anaesthetized cats. J. Physiol. Lond. 469: 51-66.

Molfino, N. A.; Wright, S. C.; Katz, I.; Tarlo, S.; Silverman, F.; McClean, P. A.; Szalai, J. P.; Raizenne, M.; Slutsky, A. S.; Zamel, N. (1991) Effect of low concentrations of ozone on inhaled allergen responses in asthmatic subjects. Lancet 338(8761): 199-203.

Mudway, I. S.; Kelly, F. J. (2000) Ozone and the lung: a sensitive issue. Mol. Aspects. Med. 21: 1-48.

Mudway, I. S.; Kelly, F. J. (2004) An investigation of inhaled ozone dose and the magnitude of airway inflammation in healthy adults. Am. J. Respir. Crit. Care Med. 169: 1089-1095.

Mudway, I. S.; Blomberg, A.; Frew, A. J.; Holgate, S. T.; Sandström, T.; Kelly, F. J. (1999) Antioxidant consumption and repletion kinetics in nasal lavage fluid following exposure of healthy human volunteers to ozone. Eur. Respir. J. 13: 1429-1438.

Mudway, I. S.; Stenfors, N.; Blomberg, A.; Helleday, R.; Dunster, C.; Marklund, S. L.; Frew, A. J.; Sandström, T.; Kelly, F. J. (2001) Differences in basal airway antioxidant concentrations are not predictive of individual responsiveness to ozone: a comparison of healthy and mild asthmatic subjects. Free Radical Biol. Med. 31: 962-974.

Nagda, N. L.; Fortmann, R. C.; Koontz, M. D.; Baker, S. R.; Ginevan, M. E. (1989) Airliner cabin environment: contaminant measurements, health risks, and mitigation options. Washington, DC: U.S. Department of Transportation, Office of the Secretary. Available from: NTIS, Springfield, VA; PB91-159384.

Nayak, A. S. (2003) The asthma and allergic rhinitis link. Allergy Asthma Proc. 24: 395-402.

Neas, L. M.; Dockery, D. W.; Koutrakis, P.; Tollerud, D. J.; Speizer, F. E. (1995) The association of ambient air pollution with twice daily peak expiratory flow rate measurements in children. Am. J. Epidemiol. 141: 111-122.

Newson, E. J.; Krishna, M. T.; Lau, L. C. K.; Howarth, P. H.; Holgate, S. T.; Frew, A. J. (2000) Effects of short-term exposure to 0.2 ppm ozone on biomarkers of inflammation in sputum, exhaled nitric oxide, and lung function in subjects with mild atopic asthma. J. Occup. Environ. Med. 42: 270-277.

Nightingale, J. A.; Rogers, D. F.; Chung, K. F.; Barnes, P. J. (2000) No effect of inhaled budesonide on the response to inhaled ozone in normal subjects. Am. J. Respir. Crit. Care Med. 161: 479-486.

Nikasinovic, L.; Momas, I.; Seta, N. (2003) Nasal epithelial and inflammatory response to ozone exposure: a review of laboratory-based studies published since 1985. J. Toxicol. Environ. Health B 6: 521-568.

Otto-Knapp, R.; Jurgovsky, K.; Schierhorn, K.; Kunkel, G. (2003) Antioxidative enzymes in human nasal mucosa after exposure to ozone. Possible role of GSTM1 deficiency. Inflamm. Res. 52: 51-55.

Passannante, A. N.; Hazucha, M. J.; Bromberg, P. A.; Seal, E.; Folinsbee, L.; Koch, G. (1998) Nociceptive mechanisms modulate ozone-induced human lung function decrements. J. Appl. Physiol. 85: 1863-1870.

Peden, D. B. (2001a) Air pollution in asthma: effect of pollutants on airway inflammation. Ann. Allergy Asthma Immunol. 87(suppl. 3): 12-17.

Peden, D. B. (2001b) Effect of pollutants in rhinitis. Curr. Allergy Asthma Rep. 1: 242-246.

Peden, D. B.; Setzer, R. W., Jr.; Devlin, R. B. (1995) Ozone exposure has both a priming effect on allergen-induced responses and an intrinsic inflammatory action in the nasal airways of perennially allergic asthmatics. Am. J. Respir. Crit. Care Med. 151: 1336-1345.

Peden, D. B.; Boehlecke, B.; Horstman, D.; Devlin, R. (1997) Prolonged acute exposure to 0.16 ppm ozone induces eosinophilic airway inflammation in asthmatic subjects with allergies. J. Allergy Clin. Immunol. 100: 802-808.

Riediker, M.; Monn, C.; Koller, T.; Stahel, W. A.; Wüthrich, B. (2001) Air pollutants enhance rhinoconjunctivitis symptoms in pollen-allergic individuals. Ann. Allergy Asthma Immunol. 87: 311-318.

Rigas, M. L.; Ben-Jebria, A.; Ultman, J. S. (1997) Longitudinal distribution of ozone absorption in the lung: effects of nitrogen dioxide, sulfur dioxide, and ozone exposures. Arch. Environ. Health 52: 173-178.

Romieu, I.; Sienra-Monge, J. J.; Ramírez-Aguilar, M.; Moreno-Macias, H.; Reyes-Ruiz, N. I.; Estela del Rio-Navarro, B.; Hernández-Avila, M.; London, S. J. (2004) Genetic polymorphism of *GSTM1* and antioxidant supplementation influence lung function in relation to ozone exposure in asthmatic children in Mexico City. Thorax 59: 8-10.

Samet, J. M.; Hatch, G. E.; Horstman, D.; Steck-Scott, S.; Arab, L.; Bromberg, P. A.; Levine, M.; McDonnell, W. F.; Devlin, R. B. (2001) Effect of antioxidant supplementation on ozone-induced lung injury in human subjects. Am. J. Respir. Crit. Care Med. 164: 819-825.

Scannell, C.; Chen, L.; Aris, R. M.; Tager, I.; Christian, D.; Ferrando, R.; Welch, B.; Kelly, T.; Balmes, J. R. (1996) Greater ozone-induced inflammatory responses in subjects with asthma. Am. J. Respir. Crit. Care Med. 154: 24-29.

Schelegle, E. S.; Adams, W. C. (1986) Reduced exercise time in competitive simulations consequent to low level ozone exposure. Med. Sci. Sports Exercise 18: 408-414.

Schelegle, E. S.; Adams, W. C.; Siefkin, A. D. (1987) Indomethacin pretreatment reduces ozone-induced pulmonary function decrements in human subjects. Am. Rev. Respir. Dis. 136: 1350-1354.

Schelegle, E. S.; Siefkin, A. D.; McDonald, R. J. (1991) Time course of ozone-induced neutrophilia in normal humans. Am. Rev. Respir. Dis. 143: 1353-1358.

Schelegle, E. S.; Carl, M. L.; Coleridge, H. M.; Coleridge, J. C. G.; Green, J. F. (1993) Contribution of vagal afferents to respiratory reflexes evoked by acute inhalation of ozone in dogs. J. Appl. Physiol. 74: 2338-2344.

Schelegle, E. S.; Eldridge, M. W.; Cross, C. E.; Walby, W. F.; Adams, W. C. (2001) Differential effects of airway anesthesia on ozone-induced pulmonary responses in human subjects. Am. J. Respir. Crit. Care Med. 163: 1121-1127.

Schonfeld, B. R.; Adams, W. C.; Schelegle, E. S. (1989) Duration of enhanced responsiveness upon re-exposure to ozone. Arch. Environ. Health 44: 229-236.

Schwartz, L. W.; Dungworth, D. L.; Mustafa, M. G.; Tarkington, B. K.; Tyler, W. S. (1976) Pulmonary responses of rats to ambient levels of ozone: effects of 7-day intermittent or continuous exposure. Lab. Invest.

Seal, E., Jr.; McDonnell, W. F.; House, D. E.; Salaam, S. A.; Dewitt, P. J.; Butler, S. O.; Green, J.; Raggio, L. (1993) The pulmonary response of white and black adults to six concentrations of ozone. Am. Rev. Respir. Dis. 147: 804-810.

Seal, E., Jr.; McDonnell, W. F.; House, D. E. (1996) Effects of age, socioeconomic status, and menstrual cycle on pulmonary response to ozone. Arch. Environ. Health 51: 132-137.

Solic, J. J.; Hazucha, M. J.; Bromberg, P. A. (1982) The acute effects of 0.2 ppm ozone in patients with chronic obstructive pulmonary disease. Am. Rev. Respir. Dis. 125: 664-669.

Solway, J.; Leff, A. R. (1991) Sensory neuropeptides and airway function. J. Appl. Physiol. 71: 2077-2087.
Spannhake, E. W.; Reddy, S. P. M.; Jacoby, D. B.; Yu, X.-Y.; Saatian, B.; Tian, J. (2002) Synergism between rhinovirus infection and oxidant pollutant exposure enhances airway epithelial cell cytokine production. Environ. Health Perspect. 110: 665-670.
Stenfors, N.; Pourazar, J.; Blomberg, A.; Krishna, M. T.; Mudway, I.; Helleday, R.; Kelly, F. J.; Frew, A. J.; Sandström, T. (2002) Effect of ozone on bronchial mucosal inflammation in asthmatic and healthy subjects. Respir. Med. 96: 352-358.
Tepper, J. S.; Costa, D. L.; Lehmann, J. R.; Weber, M. F.; Hatch, G. E. (1989) Unattenuated structural and biochemical alterations in the rat lung during functional adaptation to ozone. Am. Rev. Respir. Dis. 140: 493-501.
Tepper, J. S.; Wiester, M. J.; Weber, M. F.; Ménache, M. G. (1990) Measurements of cardiopulmonary response in awake rats during acute exposure to near-ambient concentrations of ozone. J. Appl. Toxicol. 10: 7-15.
Torres, A.; Utell, M. J.; Morow, P. E.; Voter, K. Z.; Whitin, J. C.; Cox, C.; Looney, R. J.; Speers, D. M.; Tsai, Y.; Frampton, M. W. (1997) Airway inflammation in smokers and nonsmokers with varying responsiveness to ozone. Am. J. Respir. Crit. Care Med. 156: 728-736.
Trenga, C. A.; Koenig, J. Q.; Williams, P. V. (2001) Dietary antioxidants and ozone-induced bronchial hyperresponsiveness in adults with asthma. Arch. Environ. Health 56: 242-249.
U.S. Environmental Protection Agency. (1986) Air quality criteria for ozone and other photochemical oxidants. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report nos. EPA-600/8-84-020aF-eF. 5v. Available from: NTIS, Springfield, VA; PB87-142949.
U.S. Environmental Protection Agency. (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: http://cfpub2.epa.gov/ncea/.
Ultman, J. S.; Ben-Jebria, A.; Arnold, S. F. (2004) Uptake distribution of ozone in human lungs: intersubject variability in physiologic response. Boston, MA: Health Effects Institute; research report no. 125. Available: http://www.healtheffects.org/Pubs/Ultman.pdf [29 July, 2005].
Vagaggini, B.; Carnevali, S.; Macchioni, P.; Taccola, M.; Fornai, E.; Bacci, E.; Bartoli, M. L.; Cianchetti, S.; Dente, F. L.; Di Franco, A.; Giannini, D.; Paggiaro. P. L. (1999) Airway inflammatory response to ozone in subjects with different asthma severity. Eur. Respir. J. 13: 274-280.
Vagaggini, B.; Taccola, M.; Conti, I.; Carnevali, S.; Cianchetti, S.; Bartoli, M. L.; Bacci, E.; Dente, F. L.; Di Franco, A.; Giannini, D.; Paggiaro, P. L. (2001) Budesonide reduces neutrophilic but not functional airway response to ozone in mild asthmatics. Am. J. Respir. Crit. Care Med. 164: 2172-2176.
Vagaggini, B.; Taccola, M.; Clanchetti, S.; Carnevali, S.; Bartoli, M. L.; Bacci, E.; Dente, F. L.; Di Franco, A.; Giannini, D.; Paggiaro, P. L. (2002) Ozone exposure increases eosinophilic airway response induced by previous allergen challenge. Am. J. Respir. Crit. Care Med. 166: 1073-1077.
Voter, K. Z.; Whitin, J. C.; Torres, A.; Morrow, P. E.; Cox, C.; Tsai, Y.; Utell, M. J.; Frampton, M. W. (2001) Ozone exposure and the production of reactive oxygen species by bronchoalveolar cells in humans. Inhalation Toxicol. 13: 465-483.
Wang, G.; Umstead, T. M.; Phelps, D. S.; Al-Mondhiry, H.; Floros, J. (2002) The effect of ozone exposure on the ability of human surfactant protein A variants to stimulate cytokine production. Environ. Health Perspect. 110: 79-84.
Weinmann, G. G.; Weidenbach-Gerbase, M.; Foster, W. M.; Zacur, H.; Frank, R. (1995) Evidence for ozone-induced small-airway dysfunction: lack of menstrual-cycle and gender effects. Am. J. Respir. Crit. Care Med. 152: 988-996.
Yang, I. A.; Holz, O.; Jörres, R. A.; Magnussen, H.; Barton, S. J.; Rodríguez, S.; Cakebread, J. A.; Holloway, J. W.; Holgate, S. T. (2005) Association of tumor necrosis factor-α polymorphisms and ozone-induced change in lung function. Am. J. Respir. Crit. Care Med. 171: 171-176.
Yeadon, M.; Wilkinson, D.; Darley-Usmar, V.; O'Leary, V. J.; Payne, A. N. (1992) Mechanisms contributing to ozone-induced bronchial hyperreactivity in guinea-pigs. Pulm. Pharmacol. 5: 39-50.
Ying, R. L.; Gross, K. B.; Terzo, T. S.; Eschenbacher, W. L. (1990) Indomethacin does not inhibit the ozone-induced increase in bronchial responsiveness in human subjects. Am. Rev. Respir. Dis. 142: 817-821.
Yu, M.; Pinkerton, K. E.; Witschi, H. (2002) Short-term exposure to aged and diluted sidestream cigarette smoke enhances ozone-induced lung injury in B6C3F1 mice. Toxicol. Sci. 65: 99-106.
Zhang, L.-Y.; Levitt, R. C.; Kleeberger, S. R. (1995) Differential susceptibility to ozone-induced airways hyperreactivity in inbred strains of mice. Exp. Lung Res. 21: 503-518.

7. EPIDEMIOLOGIC STUDIES OF HUMAN HEALTH EFFECTS ASSOCIATED WITH AMBIENT OZONE EXPOSURE

7.1 INTRODUCTION

This chapter evaluates current epidemiologic literature on health and physiological effects of ambient O_3 exposure. Epidemiologic studies linking community ambient O_3 concentrations to health effects were reported in the 1996 Ozone Air Quality Criteria Document (O_3 AQCD; U.S. Environmental Protection Agency, 1996a). Many of those studies reported that pulmonary function decrements, respiratory symptoms, and hospital and emergency department admissions in human populations were associated with ambient levels of O_3. Numerous more recent epidemiologic studies discussed in this chapter evaluate the relationship of ambient O_3 to morbidity and mortality, and thereby provide an expanded basis for assessment of health effects associated with exposures to O_3 at concentrations currently encountered in the United States.

As discussed elsewhere in this document (Chapters 5 and 6), a substantial amount of experimental evidence links O_3 exposure unequivocally with respiratory effects in laboratory animals and humans. These include structural changes in the bronchiolar-alveolar transition (centriacinar) region of the lung, biochemical evidence of acute cellular/tissue injury, inflammation, increased frequency and severity of experimental bacterial infection, and temporary reductions in mechanical lung function. These effects have been observed with exposure to O_3 at ambient or near-ambient concentrations. Thus, many of the reported epidemiologic associations of ambient O_3 with respiratory health effects have considerable biological credibility. Accordingly, the new epidemiologic studies of ambient O_3 assessed here are best considered in combination with information from the other chapters on ambient O_3 concentration and exposure (Chapter 3), and toxicological effects of O_3 in animals and humans (Chapters 5 and 6, respectively). The epidemiologic studies constitute important information on associations between health effects and exposures of human populations to "real-world" O_3 and also help to identify susceptible subgroups and associated risk factors. A wide variety of oxidants in both the gaseous and particulate phases have not been examined in relation to health

outcomes in the literature. Therefore, discussion in this chapter is limited to studies of human health effects associated with ambient O_3 exposure.

7.1.1 Approach to Identifying Ozone Epidemiologic Studies

Numerous O_3 epidemiologic papers have been published since completion of the 1996 O_3 AQCD. The U.S. Environmental Protection Agency (NCEA-RTP) has implemented a systematic approach to identify relevant epidemiologic studies for consideration in this chapter. In general, an ongoing search has been employed in conjunction with other strategies to identify O_3 epidemiologic literature pertinent to developing criteria for the O_3 National Ambient Air Quality Standards (NAAQS). A publication base was established using Medline, Pascal, BIOSIS, NTIS, and Embase, and a set of search terms proven by prior use to identify pertinent literature. The search strategy was reexamined and modified to enhance identification of published papers. PubMed was added to the search regime.

While the above search regime provided good coverage of the relevant literature, additional approaches augmented the traditional search methods. First, a Federal Register Notice was issued requesting information and published papers from the public at large. Next, non-EPA chapter authors, expert in this field, identified literature on their own. NCEA-RTP staff also identified publications as an element of their assessment and interpretation of the literature. Finally, additional potentially relevant publications were included following external review as a result of comments from both the public and CASAC. The combination of these approaches is believed to produce a comprehensive collection of studies appropriate for review and assessment here. The principal objective criteria used for selecting literature for the present assessment is to include all identified studies that evaluated the relationship between measured ambient O_3 levels and a human health outcome. New studies accepted for publication through December 2004, as identified using the approaches above, have been included in this AQCD and additional efforts have been made to assess more recent studies.

7.1.2 Approach to Assessing Epidemiologic Evidence

Definitions of the various types of epidemiologic studies assessed have been provided in an earlier PM AQCD (U.S. Environmental Protection Agency, 1996b). Briefly, epidemiologic studies are generally divided into two groups, *morbidity* studies and *mortality* studies. *Morbidity*

studies evaluate O_3 effects on a wide range of health endpoints, including the following: changes in pulmonary function, respiratory symptoms, self-medication in asthmatics, and airway inflammation; changes in cardiovascular physiology/functions; and cardiopulmonary emergency department visits and hospital admissions. *Mortality* studies investigate O_3 effects on total (nonaccidental) mortality and cause-specific mortality, providing evidence related to a clearly adverse endpoint. The epidemiologic strategies most commonly used in O_3 health studies are prospective cohort studies, ecologic studies, time-series semi-ecologic studies, and case-crossover studies. All of these are observational studies rather than experimental studies.

The approach to assessing epidemiologic evidence has been stated most recently in the 2004 PM AQCD (U.S. Environmental Protection Agency, 2004) and is summarized here. The critical assessment of epidemiologic evidence presented in this chapter is conceptually based upon consideration of salient aspects of the evidence of associations so as to reach fundamental judgments as to the likely causal significance of the observed associations (see Hill, 1965). The general evaluation of the strength of the epidemiologic evidence reflects consideration not only of the magnitude and precision of reported O_3 effect estimates and their statistical significance, but also of the robustness of the effects associations. Statistical significance corresponds to the allowable rate of error (Type I error) in the decision problem constructed from assuming that a simple null hypothesis of no association is true. It is a conditional probability; for statistical significance, typically there is a less than 0.05 chance of rejecting the null hypothesis given that it is true. Robustness of the associations is defined as stability in the effect estimates after considering a number of factors, including alternative models and model specifications, potential confounding by copollutants, as well as issues related to the consequences of measurement error.

Consideration of the consistency of the effects associations, as discussed in the following sections, involves looking across the results of multiple- and single-city studies conducted by different investigators in different places and times. Relevant factors are known to exhibit much variation across studies, including, for example, the presence and levels of copollutants, the relationships between central measures of O_3 and exposure-related factors, relevant demographic factors related to sensitive subpopulations, and climatic and meteorological conditions. Thus, in this case, consideration of consistency and the related heterogeneity of effects are appropriately understood as an evaluation of the similarity or general concordance of results, rather than an expectation of finding quantitative results within a very narrow range.

Looking beyond the epidemiologic evidence, evaluation of the biological plausibility of the O_3-health effects associations observed in epidemiologic studies reflects consideration of both exposure-related factors and dosimetric/toxicologic evidence relevant to identification of potential biological mechanisms. Similarly, coherence of health effects associations reported in the epidemiologic literature reflects consideration of information pertaining to the nature of the various respiratory- and cardiac-related mortality and morbidity effects and biological markers evaluated in toxicologic and human clinical studies. These broader aspects of the assessment are only touched upon in this chapter but are more fully integrated in the discussion presented in Chapter 8.

In assessing the relative scientific quality of epidemiologic studies reviewed here and to assist in interpreting their findings, the following considerations were taken into account:

(1) To what extent are the aerometric data/exposure metrics used of adequate quality and sufficiently representative to serve as credible exposure indicators, well-reflecting geographic or temporal differences in study population pollutant exposures in the range(s) of pollutant concentrations evaluated?

(2) Were the study populations well defined and adequately selected so as to allow for meaningful comparisons between study groups or meaningful temporal analyses of health effects results?

(3) Were the health endpoint measurements meaningful and reliable, including clear definition of diagnostic criteria utilized and consistency in obtaining dependent variable measurements?

(4) Were the statistical analyses used appropriate, and properly performed and interpreted?

(5) Were likely important covariates (e.g., potential confounders or effect modifiers) adequately controlled for or taken into account in the study design and statistical analyses?

(6) Were the reported findings internally consistent, biologically plausible, and coherent in terms of consistency with other known facts?

These guidelines provide benchmarks for judging the relative quality of various studies and in assessing the body of epidemiologic evidence. Detailed critical analysis of all epidemiologic studies on O_3 health effects, especially in relation to all of the above questions, is beyond the scope of this document. Of most importance for present purposes are those studies which

provide useful qualitative or quantitative information on concentration-response relationships for health effects associated with ambient air levels of O_3 likely to be encountered in the U.S. among healthy and susceptible populations.

7.1.3 Considerations in the Interpretation of Epidemiologic Studies of Ozone Health Effects

Prior to discussing results from recent O_3 epidemiologic studies, issues and questions arising from the study designs and analysis methods used in the assessment of O_3 effect estimates will be briefly presented. Study design can restrict the health effect parameters that can be estimated. Separate considerations need to be made for acute versus chronic effect studies as well as individual versus aggregate-level analyses. Time-series studies and panel studies are most frequently conducted in air pollution epidemiologic research. Aggregate-level exposure and/or outcome data are often used in these types of studies. Analyses using administrative health outcome data (e.g., numbers of deaths and emergency hospital admissions) have inherent limitations as well as strengths (Virnig and McBean, 2001). The impact of study design or the loss of information due to aggregation largely depends upon exposure variation (Sheppard et al., 2005).

This section mainly focuses on the topics of exposure assessment and model specification in air pollution epidemiologic studies. Potential biases that may result from O_3 exposure measurement error, and choice of exposure index and lag period are first presented. A discussion of model specification issues and potential confounding by temporal factors, meteorological effects, seasonal trends, and copollutants follow.

7.1.3.1 Exposure Assessment and Measurement Error in Epidemiologic Studies

In many air pollution epidemiologic studies, especially time-series studies with administrative data on mortality and hospitalization outcomes, data from central ambient monitoring sites generally are used as the estimate of exposure. Personal exposures of individual study participants generally are not directly observed in epidemiologic studies. The use of O_3 concentrations from ambient monitors as surrogate measures for personal O_3 exposures was discussed previously in Section 3.9. Routinely collected ambient monitor data, though readily

available and convenient, may not represent true personal exposure, which includes both ambient and non-ambient source exposures.

In several studies focused on evaluating exposure to O_3, measurements were made in a variety of indoor environments, including homes (Lee et al., 2004), schools (Linn et al., 1996), and the workplace (Liu et al., 1995). Indoor O_3 concentrations were, in general, approximately one-tenth of the outdoor concentrations in these studies. Few indoor sources of O_3 exist, possible sources being office equipment (e.g., photocopiers, laser printers) and air cleaners. As described in Section 3.8 of this document, O_3 in the indoor environment is largely dependent on the outdoor ambient O_3 concentration. Other factors that influence the O_3 concentration indoors include the air exchange rate, outdoor infiltration, indoor circulation rate, and O_3 removal process.

Sheppard (2005) states that non-ambient exposures typically vary across individuals but are not likely to have strong temporal correlations. In contrast, ambient concentrations for individuals should be highly correlated as they vary over time similarly for everyone because of changes in source generation, weather, and season. The independence of ambient and non-ambient exposure sources has important implications for selection of study designs that are most effective for estimating health effects (Sheppard, 2005). In an ideal situation, studies of air pollution health effects would be conducted at the individual level, with information on personal exposure to the various pollutants. However, determining accurate personal exposure information is difficult and generally impractical. A simulation study by Sheppard et al. (2005) examining non-reactive pollutants observed that there was no noticeable difference between effect estimates using either total personal exposure or ambient concentration data when non-ambient source exposures were independent of ambient source exposures in time-series studies. Sheppard (2005) concludes that for estimating acute effects, ambient concentration measurements are adequate in time-series studies. In the case of O_3, there are limited non-ambient sources; thus, ambient concentrations of O_3 are also likely to be adequate in the analysis of O_3 health effects in time-series studies. Even with the lower exposure variation when using only ambient concentration data, the large sample sizes and longer study duration make time-series studies quite powerful.

As discussed thoroughly in the 2004 PM AQCD (Section 8.4.5), the resulting exposure measurement error and its effect on the estimates of relative risk must be considered. In theory,

there are three components to exposure measurement error in time-series studies as described by Zeger et al. (2000): (1) the use of average population rather than individual exposure data; (2) the difference between average personal ambient exposure and ambient concentrations at central monitoring sites; and (3) the difference between true and measured ambient concentrations. Zeger et al. indicated that the first and third error components were largely Berksonian errors; although they would increase the standard errors, they would not bias the risk estimate. However, the second error component resulting from the difference between average personal ambient exposure and outdoor ambient concentration levels might attenuate the risk estimate.

The impact of exposure measurement error on O_3 effect estimates was demonstrated in a study by Navidi et al. (1999). In this study, a simulation was conducted using data from the University of Southern California Children's Health Study of the long-term effects of air pollutants on children. The effect estimate from computed "true" O_3 exposure was compared to effect estimates from exposure determined using several methods: (1) ambient stationary monitors; (2) the microenvironmental approach (multiply concentrations in various microenvironments by time present in each microenvironment); and (3) personal sampling. Effect estimates based on all three exposure measures were biased towards the null. The bias that results when using the microenvironmental and personal sampling approach is due to nondifferential measurement error. Use of ambient monitors to determine exposure will generally overestimate true personal O_3 exposure (assumes that subjects are outdoors 100% of their time and not in close proximity to sources that reduce O_3 levels such as NO emissions from mobile sources), thus generally their use will result in effect estimates that are biased towards the null.

Zidek (1997) notes that a statistical analysis must balance bias and imprecision (error variance). Ignoring measurement error in air pollution epidemiologic studies often will result in underestimated risk estimates. In a reanalysis of the study by Burnett et al. (1994) on the acute respiratory effects of ambient air pollution, Zidek et al. (1998) observed that accounting for measurement error, as well as a few additional changes to the analysis, resulted in qualitatively similar conclusions. However, while the original analysis by Burnett et al. found that 5% of daily respiratory admissions in the summer months was attributable to O_3, Zidek et al. calculated that O_3 was associated with a 14% increase in respiratory admissions. Available data and

analysis limit our ability to weigh the importance of uncertainty due to measurement error relative to other sources in the studies reviewed.

As discussed in Section 3.9, there is suggestive evidence that ambient O_3 concentrations from central monitors may serve as valid surrogate measures for aggregate personal O_3 exposures in time-series studies. However, using ambient concentrations to determine exposure generally overestimates true personal O_3 exposures, resulting in biased descriptions of underlying concentration-response relationships. These effect estimates, though conservative from a testing perspective, must be evaluated and used with caution as they may lead to an underestimation of the overall impact of air pollution on health effects. A better understanding of the relationship between ambient concentrations and personal exposures, and the factors that affect the relationship will improve the interpretation of ambient concentration-population health response associations observed in epidemiologic studies.

7.1.3.2 Ozone Exposure Indices Used

The O_3-related effect estimates for mortality and morbidity health outcomes are usually presented in this document as a relative risk, or risk rate relative to a baseline mortality or morbidity rate. These relative risks are based on an incremental change in exposure. To enhance comparability between studies, presenting these relative risks by a uniform exposure increment is needed. However, determining a standard increment is complicated by the use of different O_3 exposure indices in the existing health studies. The three daily O_3 exposure indices that most often appear in the literature are 1-h average maximum (1-h max), 8-h average maximum (8-h max), and 24-h average (24-h avg) concentrations. As levels are lower and less variable for the longer averaging times, relative risks of adverse health outcomes for a specific numeric concentration range are not directly comparable across metrics. Using the nationwide distributional data for O_3 monitors in U.S. Metropolitan Statistical Areas, increments representative of a low-to-high change in O_3 concentrations were approximated based on annual mean to 95th percentile differences (Langstaff, 2003), as follows:

Daily Exposure Index	Exposure Increment (ppb)
1-h max O_3	40
8-h max O_3	30
24-h avg O_3	20

In the following discussion sections, efforts were made to standardize the O_3 excess risks using these increments, except as noted, so that risk estimates could be compared across studies. Note that in the Annex Tables, effect estimates are not standardized; results are presented in the tables as they are reported in the papers.

7.1.3.3 Lag Time: Period between Ozone Exposure and Observed Health Effect

Lags of exposure may reflect the distribution of effects across time in a population and the potential mechanisms of effects. The choice of lag days for the relationship between exposure and health effects depends on the hypothesis being tested and the mechanism involved in the expression of the outcome. Effects can occur acutely with exposure on the same or previous day, cumulatively over several days, or after a delayed period of a few days. With knowledge of the mechanism of effect, the choice of lag days can be determined prior to analysis. For example, one can expect cough to occur acutely after exposure with a lag of 0 or 1 day, as O_3 can act as a short-term irritant. However, an O_3-related inflammatory response may not lead to asthma exacerbation until several days later. An asthmatic may be impacted by O_3 on the first day of exposure, have effects triggered further on the second day, then report to the emergency room for an asthmatic attack three days after exposure. Further, within a population of asthmatics, exacerbation of asthma symptoms may be observed over a period of several days, since each asthmatic has individual aspects of the disease and may be affected by the exposure differently depending on his/her sensitivity and disease severity. The results from controlled human studies may be useful in assessing the adequacy of lags for some respiratory health outcomes.

Some studies attempted to examine the overall impact of O_3 through distributed lag models. Schildcrout and Heagerty (2005) compared regression analyses using single-day versus distributed lag models. The single-day lag model calculates a risk estimate that assumes dependence only on exposure from the specified day. In contrast, the distributed lag model provides an estimate that is a summary measure of the cumulative distributed lag effect from all included lag days. The standard error of the cumulative sum of the individual distributed lag coefficients takes into consideration the variance-covariance of the multiple lags, and is therefore larger than the standard error of the single-day lag coefficient. Thus, if the underlying O_3-health outcome relationship was a single-day effect, then modeling the relationship with a distributed

lag model would make the estimate less significant. On the other hand, if the effect of O_3 on health outcomes persisted over several days, then applying a single-day lag model would result in an underestimation of the multiday effects. The correct choice requires balancing variance and bias.

As the parameters estimated from single-day lag versus multiday lag models are not the same, interpretation and comparison of these results may be difficult. When comparing the impacts of these different models, the nuance of increments used in calculating the estimates is different depending on the model. For example, an excess percent mortality risk "per 20 ppb increase in 24-h avg O_3" in a distributed lag model including lag 0- through 6-days tacitly means a 20 ppb increase in each of the seven days. The difference in the exposure scenarios in the single-day versus multiday lag model (i.e., 20 ppb increase in one day versus several consecutive days) complicates a simple comparison of risk estimates from two different models using "the same increment."

Only a limited number of studies have hypothesized a priori the lag structure to be examined. Most of the O_3 time-series studies examined relatively small numbers of single-day lag models, typically lags of 0 through 3 days. Sheppard et al. (1999) notes that when considering single-day lag estimates it is important to consider the effect estimate in the context of the pattern of adjacent lags as these estimates contain information from the adjacent days owing to serial correlation of the pollutant series. In many cases, a pattern of positive associations across several lag days were reported. For the respiratory and cardiovascular outcomes investigated, the "most significant" lags were generally 0- or 1-day lags, suggesting that the majority of the single-day associations are immediate, not a random pattern in which associations can be observed on any of the lags examined with equal probabilities. For example, two recent meta-analyses of O_3-mortality effects observed that the combined estimate from 0-day lag models was larger than the estimate from longer lag days (Bell et al., 2005; Levy et al., 2005).

Bias resulting from the selection of lags has not been examined specifically for O_3 effects. However, the issue of lags has been investigated for PM and the results of this analysis are most likely of relevance for O_3. Lumley and Sheppard (2000) performed a simulation study to examine model selection bias in air pollution epidemiology using $PM_{2.5}$ as an example. Sheppard et al. (1999; reanalysis Sheppard, 2003) had investigated the association between

asthma hospital admissions and ambient $PM_{2.5}$ concentrations over an eight-year period in Seattle, WA. Note that the results from Lumley and Sheppard (2000) and Sheppard et al. (1999) were based on GAM using default convergence criteria (see Section 7.1.3.7). A negative control analysis, using simulated data with no association between PM exposure and the health outcome, and a positive control analysis, in which a specified non-zero excess risk is added to the simulation, were performed for comparison. The bias from selection of best of seven lags (0 to 6 days) and residual seasonal confounding in the negative control analysis (median log relative risk of 0.0013) was approximately half the log relative risk estimated from the observed data (0.0027), after adjusting for season and temperature. In the positive control model (true log relative risk of 0.0083), the bias was small (median log relative risk of 0.0080). Results from these simulations indicate that bias from selection of lags may be small, but of the same magnitude as the estimated health impacts.

Selection of lag periods should depend on the hypothesis of the study and the potential mechanism of the effect. When the mechanism of the health effect is unknown, investigating the association between outcome and exposure using cumulative distributed lag models may be informative. Analyzing a large number of lags and simply choosing the largest and most significant results may bias the air pollution risk estimates away from the null. Most studies have shown that O_3 has a fairly consistent, immediate effect on health outcomes, including respiratory hospitalizations and mortality. Several studies also observed significant O_3 effects over longer cumulative lag periods, suggesting that in addition to single-day lags, multiday lags should be investigated to fully capture a delayed O_3 effect on health outcomes. In this document, discussion largely focuses on effect estimates from 0- and 1-day lags, with some consideration of cumulative, multiday lag effects. It is not straightforward to compare and contrast results from single-day versus multiday lag models because the parameters estimated from these models are not the same. These complications need to be taken into consideration when interpreting results from various lag models.

7.1.3.4 Model Specification to Adjust for Temporal Trends and Meteorologic Effects

Several challenges present themselves with respect to designing and interpreting time-series studies. The principal challenge facing the analyst in the daily time-series context is avoiding bias due to confounding by short-term temporal factors operating over time scales from

days to seasons. In the current regression models used to estimate short-term effects of air pollution, two major potential confounders need to be considered: (1) seasonal trend and other "long-wave" temporal trends; and (2) weather effects. Both of these variables tend to predict a significant fraction of fluctuations in time-series. Unfortunately, as O_3 has strong seasonal cycles and is formed more at higher temperatures, both terms are also highly correlated with O_3. The correlation of O_3 with these confounding terms tends to be higher than that for PM or other gaseous pollutants. In the U.S., the mass concentration of $PM_{2.5}$ generally does not have strong seasonal cycles like O_3 because $PM_{2.5}$ tends to reflect both primary emissions (throughout the year, but often higher in winter in most U.S. cities) and secondary aerosols (higher in summer). Therefore, $PM_{2.5}$ and O_3 effect estimates from studies primarily designed to examine $PM_{2.5}$ health effects may not be comparable as model specifications that may be appropriate for $PM_{2.5}$ may not necessarily be adequate for O_3.

An examination of recent time-series studies indicates that several types of fitting approaches have been used to adjust for temporal trends and weather effects. The use of parametric and nonparametric smoothers with varying degrees of freedom per year has emerged as the prevailing approach. The use of larger degrees of freedom to adjust for potential confounding by time-varying factors may inadvertently result in ascribing more effects to these unmeasured potential confounders and mask the air pollution effect. Often smaller pollution effect estimates are observed when more degrees of freedom are used. Currently, the degrees of freedom used to adjust for temporal trends in time-series studies generally range from 4 to 12 degrees of freedom per year using either nonparametric or parametric smoothers. Statistical diagnostics such as Akaike's Information Criteria, residual autocorrelation, or dispersion of the regression model often are used to choose or evaluate the adequacy of the degrees of freedom for temporal trend. However, these diagnostics do not guarantee "adequate" control for temporal confounding, as choosing the appropriate extent of smoothing requires prior knowledge of the nature of the confounding (e.g., shape and duration of influenza epidemics).

The issue of model specifications to adjust for temporal trends and weather variables in time-series studies was a consideration of several researchers that conducted sensitivity analyses of PM data (Health Effects Institute, 2003). The sensitivity of O_3 coefficients to model specifications for temporal trend adjustment has not been as well-studied. Recent multicity studies examined the sensitivity of O_3 coefficients to the extent of smoothing for adjustment of

temporal trends and meteorologic factors (Bell et al., 2004; Huang et al., 2005; Ito et al., 2005). Most, if not all, O_3 studies used the same model specifications to estimate the excess risks for PM and other gaseous pollutants. The model specification designed to control confounding by meteorological and temporal factors for PM may not be necessarily adequate for O_3. As noted above, O_3 is expected to have the strongest correlation with both temporal (seasonal) trend and weather effects. The strong annual cycle in O_3 concentrations presents a unique problem in time-series analyses where time trends are fitted simultaneously with pollution and other model terms (i.e., co-adjustment). In this setting, the annual O_3 cycle itself may compete with the smooth function of time to explain some of the annual, cyclic behavior in the health outcome, which can result in biased effect estimates for O_3 when data for all seasons are analyzed together.

Current weather models used in time-series analyses can be classified into: (1) quantile (e.g., quartile, quintile) indicators; (2) parametric functional forms such as V- or U-shape functions; and (3) parametric (e.g., natural splines) or nonparametric (e.g., locally estimated smoothing splines [LOESS]) smoothing functions. More recent studies tend to use smoothing functions. While these methods provide flexible ways to fit health outcomes as a function of temperature and other weather variables, there are two major issues that need further examination to enable more meaningful interpretation of O_3 morbidity and mortality effects.

The first issue is the interpretation of weather or temperature effects. Most researchers agree about the morbidity and mortality effects of extreme temperatures (i.e., heat waves or cold spells). However, as extreme hot or cold temperatures, by definition, happen rarely, much of the health effects occur in the mild or moderate temperature range. Given the significant correlation between O_3 and temperature, ascribing the association between temperature and health outcomes solely to temperature effects may underestimate the effect of O_3. The second issue is that weather model specifications are fitted for year-round data in most studies. Such models will ignore the correlation structure that can change across seasons, resulting in inefficiency and model mis-specification. This is particularly important for O_3, which appears to change its relationship with temperature as well as with other pollutants across seasons.

This changing relationship between O_3 and temperature, as well as O_3 and other pollutants across seasons, and its potential implications to health effects modeling have not been examined thoroughly in the time-series literature. Even the flexible smoother-based adjustments for

seasonal and other time-varying variables cannot fully take into account these complex relationships. One obvious way to alleviate or avoid this complication is to analyze data by season. While this practice reduces sample size, its extent would not be as serious as PM (which is collected only every sixth day in most locations) because O_3 is collected daily, though only in warm seasons in some states. An alternative approach is to include separate O_3 concentration variables for each season (by multiplying O_3 concentrations by a season indicator variable).

In locations where seasonal variability may be a factor, O_3 effect estimates calculated using year-round data can be misleading, as the changing relationship between O_3, temperature, and other pollutants across seasons may have a significant influence on the estimates. Analyses have indicated that confounding from seasonal variability may be controlled effectively by stratifying the data by season.

7.1.3.5 Confounding Effects of Copollutants

Extensive discussions on the issues related to confounding effects among air pollutants in time-series studies are provided in Section 8.4.3 of the 2004 PM AQCD. Since the general issues discussed in that document are applicable to all pollutants, such discussions are not repeated here. What was not discussed in the 2004 PM AQCD was the issue of changing relationships among air pollutants across seasons. Compared to other pollutants, O_3 has strong seasonal cycles. Ambient O_3 levels are typically higher in the summer or warm season, often referred to as the O_3 season. In the winter or colder months, O_3 levels tend to be much lower compared to the summer months. During the winter in some urban locations, O_3 mainly comes from the free troposphere and can be considered a tracer for relatively clean air (i.e., cold, clear air coming down from the upper atmosphere), as discussed in Chapter 3 of this AQCD. The clean air is associated with the passage of cold fronts and the onset of high-pressure conditions, which occur with colder temperatures. Thus, sunny clear winter days following a high-pressure system are the days when air pollution levels from primary emissions (e.g., NO_2, SO_2, and PM from local sources) tend to be lower and O_3 is relatively higher. This can lead to negative correlations between O_3 and the primary pollutants in the winter. As shown in Figure 3-24 in the Chapter 3 Annex, the relationship between O_3 and $PM_{2.5}$ was U-shaped for the year-round data in Fort Meade, MD. The negative $PM_{2.5}/O_3$ slope was in the range of O_3 concentrations less than 30 ppb, providing supporting evidence of the aforementioned winter phenomenon. Thus, the

correlation between O_3 and PM for year-round data may be misleading. The high reactivity of O_3 with certain copollutants further complicates the analysis. For example, the reaction between NO (emitted from motor vehicles) and O_3 results in reduced O_3 levels but increased NO_2 levels during high traffic periods.

Multipollutant regression models often are used to assess potential confounding by copollutants; however, there are limitations to these models. Zidek et al. (1996) examined, through simulation, the joint effects of multicollinearity and measurement error in a Poisson regression model. The results illustrated the transfer of effects from the "causal" variable to the confounder. However, in order for the confounder to have a larger effect size than the true predictor, the correlation between the two covariates had to be very high ($r \geq 0.9$), with moderate error ($\alpha > 0.5$) for the true predictor and no error for the confounder in their scenarios. The transfer-of-causality effect was lessened when the confounder also became subject to error. Another interesting finding was the behavior of the standard errors of the coefficients. When the correlation between the covariates was high ($r = 0.9$) and both covariates had no error, the standard errors for both coefficients were inflated by a factor of two; however, this phenomenon disappeared when the confounder had error. The effect of multicollinearity is generally even more complex when analyzing real data. For further discussion, see the 2004 PM AQCD (Sections 8.4.3 and 8.4.5).

Uncertainty remains as to the use of multipollutant regression models to assess the independent health effects of pollutants that are correlated. Particularly in the case of O_3, concern remains as to whether multipollutant regression models for year-round data can adjust for potential confounding adequately due to the changing relationship between O_3 and other pollutants. Despite these limitations, multipollutant models are still the prevailing approach in most, if not all, studies of O_3 health effects and serve as an important tool in addressing the issue of confounding by copollutants, especially in season-stratified analyses.

7.1.3.6 Model Uncertainty from Multiple Hypothesis Testing

Epidemiologic studies that investigated the association between various measures of O_3 (multiple lags, different metrics, etc.) and various health outcomes often found significant effects. A major question is: Are these significant associations an artifact of model selection due to multiple testing and does this lead to overestimation of the effect estimates?

Multiple testing occurs when multiple health outcomes are examined, several lags are tested, different metrics of O_3 exposure are used, and many sub-populations are tested. Statistically testing a null hypothesis (i.e., there is no effect of O_3) requires one to calculate the value of a test statistic (i.e., t-value). If the observed test statistic exceeds a critical value (oftentimes the 95th percentile) or is outside a range of values, the null hypothesis is rejected. However, when multiple testing is done using a critical value determined for a single test, the chance that at least one of the hypotheses is significant may be greater than the expected 5% error rate. This uncertainty clouds the interpretation and weakens the evidence against any of the null hypotheses. Still, multiple hypotheses testing may be of great value. For example, developing a few hypotheses a priori allows researchers to explore more throughly potential associations for an O_3-related health effect. Sensitivity analyses, which are critical for model validation, also involve multiple testing. There are two types of sensitivity testing. One tests for the consistency of the effect when different adjustments are made for seasonal effects, or other covariates. Another tests for sensitive subpopulations and other specific conditions. In the former case, one should guard against a multiple testing error by restricting the inferences to consistency of the effect and not treat the hypotheses generated for sensitivity analyses as being confirmatory.

Recent attention has focused on Bayesian model averaging as a method to address model uncertainty from multiple hypothesis testing. In Bayesian model averaging, predictions and inferences are based on a set of models rather than a single model, and each model contributes proportionally to the support it receives from the observed data (Clyde, 1999). In addition to the uncertainty of effect estimation, Bayesian model averaging can incorporate uncertainty regarding the choice of confounding variables, pollutants, and lags. Koop and Tole (2004) used Bayesian model averaging to analyze the effect of various air pollutants, including O_3, SO_2, CO, NO, NO_2, $PM_{10-2.5}$, and $PM_{2.5}$, on mortality in Toronto, Canada. The 50+ explanatory variables required the fitting of an enormous number of potential models. Clyde et al. (2000) and Clyde (2000) also used Bayesian model averaging to analyze the relationship between PM and mortality. Clyde (2000) noted that Bayesian model averaging did not take into consideration factors that might bias the estimated effect toward the null. For example, measurement error in the exposure variables was not considered. In addition, the Poisson model (similar to many other regression models) assumed that all individuals in a population had equal risks, including potentially

susceptible populations such as those with respiratory illnesses and outdoor workers. While Bayesian model averaging can theoretically be used to take into account uncertainty, claims of causality based on observational studies may be highly sensitive to the choice of prior distributions and class of models under consideration (Clyde et al., 2000). Another limitation of Bayesian model averaging is that the estimated posterior effects may be diluted (i.e., result in smaller coefficients) when variables are highly correlated, as may be the case for air pollution studies (George, 1999 in comments to Hoeting et al., 1999).

Additional methods to control for model uncertainty resulting from multiple hypothesis testing are by a priori deciding hypotheses that are confirmatory and exploratory, and limiting the number of confirmatory tests. For example, Dominici et al. (2003) used a minimum number of tests in the U.S. 90 cities study, which reduced the uncertainty associated with multiple testing. In addition, they performed sensitivity analyses to examine the consistency and robustness of the effects. Another approach is to partition the data into two sets, one for model identification and a second for model confirmation.

The summary of health effects in this chapter is vulnerable to the errors of publication bias and multiple testing. Recent studies (Bell et al., 2005; Ito et al., 2005; Lumley and Sheppard, 2000) have found indications of what the magnitudes of these errors might be in some instances. Some researchers have used methods to protect their estimates against these errors. Efforts have been made to reduce the impact of multiple testing errors on the conclusions in this document. To address multiple hypothesis testing in this chapter, emphasis will be on a priori hypotheses. As identifying a priori hypotheses is difficult in the majority of the studies, the most common hypotheses will be considered. For example, although many studies examined multiple single-day lag models, priority would be given to the effects observed at 0- or 1-day lags rather than at longer lags. Both single- and multiple-pollutant models that include O_3 will be considered and examined for robustness of results. Analyses of multiple model specifications for adjustment of temporal or meteorological trends will be considered sensitivity analyses. Sensitivity analyses shall not be granted the same inferential weight as the original hypothesis-driven analysis; however, these analyses will be discussed in this chapter as appropriate given their valuable insights that may lead scientific knowledge in new directions.

7.1.3.7 Impact of GAM Convergence Issue on Ozone Risk Estimates

Generalized Additive Models (GAM) have been widely utilized for epidemiologic analysis of the health effects attributable to air pollution. The impact of the GAM convergence issue was thoroughly discussed in Section 8.4.2 of the 2004 PM AQCD. Reports have indicated that using the default convergence criteria in the Splus software package for the GAM function can lead to biased regression estimates for PM and an underestimation of the standard error of the effect estimate (Dominici et al., 2002; Ramsay et al., 2003). GAM default convergence criteria has a convergence precision of 10^{-3} and a maximum number of 10 iterations. The more stringent convergence criteria refers to increased stringency of both the convergence precision and number of iterations. The default convergence criteria was found to be a problem when the estimated relative risks were small and two or more nonparametric smoothing curves were included in the GAM (Dominici et al., 2002). The magnitude and direction of the bias depend in part on the concurvity of the independent variables in the GAM and the magnitude of the risk estimate. Recent focus has been on the influence of the GAM function on effect estimates for PM. However, because O_3 covaries more strongly with both weather and time factors than does PM, the issue of GAM convergence criteria for O_3 also needs to be considered.

A meta-analysis by Stieb et al. (2003) found some difference in O_3-mortality risk estimates between the GAM studies and non-GAM studies. GAM studies were defined as studies that analyzed effect estimates using nonparametric smoothing functions of time or weather. Non-GAM studies were all other studies, including those using Generalized Linear Models (GLM) and Generalized Estimating Equations (GEE) in their analysis. In the single-pollutant models, the O_3-mortality risk estimates for the non-GAM studies (10 estimates) and GAM studies (15 estimates) were 1.8% (95% CI: 0.5, 3.1) and 2.2% (95% CI: 1.4, 2.8), respectively, per 40 ppb daily 1-h max O_3. In the multipollutant models, the pooled risk estimate was 1.0% (95% CI: −0.5, 2.6) for non-GAM studies (7 estimates) and 0.5% (95% CI: −1.0, 1.9) for GAM studies (4 estimates).

Results from recent meta-analyses of O_3-mortality effects suggest that there are no substantial differences between GAM-affected estimates and non-GAM-affected estimates (Bell et al., 2005; Ito et al., 2005; Levy et al., 2005). GAM-affected studies included those that used default convergence criteria. Non-GAM-affected studies included GAM studies that used stringent convergence criteria or those that used other modeling techniques. Ito et al. (2005)

found that the single-pollutant combined estimate for the GAM-affected studies (15 estimates) and non-GAM-affected studies (28 estimates) were 1.92% (95% CI: 1.02, 2.81) and 1.40% (95% CI: 0.78, 2.02), respectively, per 20 ppb increase in 24-h avg O_3. In the analysis by Levy et al. (2005), the single-pollutant combined estimate for the GAM-affected studies (29 estimates) and non-GAM-affected studies (17 estimates) were 1.56% (95% CI: 1.01, 2.11) and 1.80% (95% CI: 1.17, 2.43), respectively, per 40 ppb increase in 1-h max O_3. Bell et al. (2005) also reported that the pooled estimate was larger for the studies that were not GAM-affected.

A few GAM studies reanalyzed O_3 risk estimates using more stringent convergence criteria or GLM. Reanalysis of an asthma hospital admissions study in Seattle, WA (Sheppard et al., 1999; reanalysis Sheppard, 2003) indicated that there were only slight changes in the risk estimates when using more stringent convergence precision (10^{-8}) in GAM. The original GAM analysis indicated an excess risk of 9% (95% CI: 3, 17) whereas the stringent GAM analysis found an excess risk of 11% (95% CI: 3, 19) per 30 ppb increase in 8-h max O_3 at a 2-day lag. Similar results were found using GLM with natural splines, 11% (95% CI: 2, 20). In the reanalysis of Santa Clara County, CA data, Fairley (1999; reanalysis Fairley, 2003) used the same methods as the original analysis except the convergence precision (ϵ) was increased from 10^{-4} to 10^{-12} and the maximum number of iterations (M) were increased from 10 to 10^7. The O_3-mortality risk estimate slightly increased from 2.8% (95% CI not provided) using default GAM parameters to 2.9% (95% CI: −0.3, 6.0) using stringent GAM parameters per 30 ppb increase in 8-h max O_3 at a 0-day lag. The O_3-mortality risk estimates further increased to 3.0% (95% CI: −0.3, 6.3) using GLM with natural cubic splines. In the reanalysis of the Netherlands data by Hoek et al. (2000; reanalysis Hoek, 2003), the O_3 nonaccidental mortality risk estimates increased from 1.3% (95% CI: 0.8, 1.9) using default GAM to 1.5% (95% CI: 1.0, 2.1) using stringent GAM ($\epsilon = 10^{-8}$, $M = 10^3$) and 1.6% (95% CI: 0.9, 2.4) using GLM with natural splines per 30 ppb increase in 8-h avg O_3 (12 p.m. - 8 p.m.) at a 1-day lag.

In the limited number of studies that have reanalyzed O_3 risk estimates, there is little evidence that default GAM analyses resulted in positively biased estimates as observed for PM. Generally it appears that the use of default convergence criteria in GAM tends to bias risk estimates towards the null, in addition to underestimating the standard errors. However, one study by Cifuentes et al. (2000) in Santiago, Chile observed a large difference in the O_3-mortality excess risks calculated using default GAM (0.9% [95%CI: 0.2, 1.6] per 40 ppb

increase in 1-h max O_3) and GLM (0.1% [95% CI: −0.6, 0.8]). The GAM convergence problem appears to vary depending on data sets, and likely depends upon the intercorrelation among covariates and the magnitude of the risk estimate; thus, its impact on the results of individual studies cannot be known without a reanalysis. In uniformity with the approach used in the 2004 PM AQCD, the results from studies that analyzed data using GAM with default convergence criteria and at least two nonparametric smoothing terms are generally not considered in this chapter, with some exceptions as noted.

7.1.4 Approach to Presenting Ozone Epidemiologic Evidence

To produce a thorough appraisal of the evidence, key information (including study design, analysis, mean O_3 concentrations, and health outcome results) from important new studies is presented in summary tables in Chapter 7 of the Annex. Each section of the chapter starts by concisely highlighting important points derived from the 1996 O_3 AQCD assessment. In the main body of the chapter, particular emphasis is focused on studies and analyses that provide pertinent information for the critical assessment of health risks from O_3 exposure. Not all studies should be accorded equal weight in the overall interpretive assessment of evidence regarding O_3-associated health effects. Among well-conducted studies with adequate control for confounding, increasing scientific weight should be accorded in proportion to the precision of their effect estimates. Small-scale studies without a wide range of exposures generally produce less precise estimates compared to larger studies with a broad exposure gradient. The size of the study, as indicated by the length of the study period and total number of events, and the variability of O_3 exposures are important components of the precision of the health effect estimates. More weight should be accorded to estimates from studies with narrow confidence bands.

Emphasis is placed on text discussion of (1) new multicity studies that employ standardized methodological analyses for evaluating O_3 effects across several or numerous cities and often provide overall effect estimates based on combined analyses of information pooled across multiple cities; (2) studies that consider O_3 as a component of a complex mixture of air pollutants including PM and other gaseous criteria pollutants (CO, NO_2, SO_2); and (3) North American studies conducted in the U.S. or Canada. Multicity studies are of particular interest and value due to their evaluation of a wider range of O_3 exposures and large numbers of

observations. They generally provide more precise effect estimates than most smaller scale studies of single cities. Compared to meta-analyses of multiple "independent" studies, a potential advantage of multicity studies is consistency in data handling and model specifications which eliminates variation due to analysis approach. Also, unlike meta-analyses, they do not suffer from potential omission of nonsignificant results due to "publication bias." Furthermore, geographic patterns of air pollution effects have the potential to provide especially valuable evidence regarding relative homogeneity and/or heterogeneity of O_3 health effects relationships across geographic locations. Due to the potential for confounding by copollutants, preference is given to studies with effect estimates from multipollutant models, i.e., models with both O_3 and PM rather than O_3-only models. The potential impacts of different health care systems and the underlying health status of populations also need to be accounted for in the assessment (Hubbell et al., 2005; Levy et al., 2001); thus, U.S. studies are emphasized over non-U.S. studies. In accordance to the emphasis placed on the O_3 epidemiologic studies in this chapter, the tables in the Chapter 7 Annex were organized by region with multicity studies in each region presented first.

In the coming sections, field/panel studies and studies of emergency department visits and hospital admissions, which contributed to the establishment of the revised 1997 NAAQS for O_3, are presented first. This is followed by a discussion of O_3-related mortality and effects of chronic exposures to O_3. The chapter ends with an integrative discussion providing a summary and conclusions.

7.2 FIELD STUDIES ADDRESSING ACUTE EFFECTS OF OZONE

7.2.1 Summary of Key Findings on Field Studies of Acute Ozone Effects From the 1996 O_3 AQCD

In the 1996 O_3 AQCD, individual-level camp and exercise studies provided useful quantitative information on the concentration-response relationships linking human lung function declines with ambient O_3 concentrations. The available body of evidence supported a dominant role of O_3 in the observed lung function decrements. Extensive epidemiologic evidence of pulmonary function responses to ambient O_3 came from camp studies. Six studies from three separate research groups provided a combined database on individual

concentration-response relationships for 616 children (mostly healthy, nonasthmatic) ranging in age from 7 to 17 years, each with at least six sequential measurements of FEV_1 (forced expiratory volume in 1 second) while attending summer camps (Avol et al., 1990; Higgins et al., 1990; Raizenne et al., 1987, 1989; Spektor et al., 1988a, 1991). In the combined reanalysis by Kinney et al. (1996a) using consistent analytical methods, these data yielded an average relationship between afternoon FEV_1 and concurrent-hour O_3 concentration of −0.50 mL/ppb (95% CI: −0.63, −0.36), with study-specific slopes ranging from −1.29 to −0.19 mL/ppb. Exposure in camp studies usually extended for multiple hours. Although the regression results noted above were based on one-hour O_3 levels, single- and multiple-hour averages were observed to be highly correlated; thus, these results might represent, to some extent, the influence of multihour exposures. In addition to the camp study results, two studies involving lung function measurements before and after well-defined exercise events in adults yielded concentration-response slopes of −0.4 mL/ppb (95% CI: −0.7, −0.1) (Selwyn et al., 1985) and −1.35 mL/ppb (95% CI: −2.04, −0.66) (Spektor et al., 1988b). Ozone concentrations during exercise events of approximately ½-hour duration ranged from 4 to 135 ppb in these studies.

Results from other field panel studies also supported a consistent relationship between ambient O_3 exposure and acute respiratory morbidity in the population. Respiratory symptoms (or exacerbation of asthma) and decrements in peak expiratory flow (PEF) occurred with increased ambient O_3 concentrations, especially in asthmatic children (Lebowitz et al., 1991; Krzyzanowski et al., 1992). The results showed greater responses in asthmatic individuals than in nonasthmatics (Lebowitz et al., 1991; Krzyzanowski et al., 1992), indicating that asthmatics might constitute a sensitive group in epidemiologic studies of oxidant air pollution. Since the 1996 O_3 AQCD, new research has examined a broad scope of field studies which are presented next.

7.2.2 Introduction to Recent Field Studies of Acute Ozone Effects

Numerous field studies carried out over the past decade have tested for and, in many cases, observed acute associations between measures of respiratory ill-health and O_3 concentrations in groups of subjects (Table AX7-1 in Chapter 7 Annex). Acute field studies are distinguished from time-series study designs in that they recruit and collect data from individual human subjects instead of utilizing administrative data on aggregate health outcomes such as daily

mortality, hospital admissions, or emergency department visits. Although individual-level health outcome data are collected in field studies, ambient O_3 concentrations from centrally located monitoring stations are generally used to assess exposure. Because of the logistical burden associated with direct data collection from individual subjects, field/panel studies tend to be small in both numbers of subjects and in duration of follow-up. While this may limit the statistical power of field studies as compared with time-series studies, the ability to determine individual-level information on health outcomes and potentially confounding factors adds scientific value.

The most common outcomes measured in acute field studies on the effects of air pollution exposure are lung function and various respiratory symptoms. Other respiratory outcomes examined on a limited basis include inflammation and generation of hydroxyl radicals in the upper airways, and school absences. Several studies examined cardiovascular outcomes including heart rate variability (HRV) and risk of myocardial infarctions (MI). The first group of studies provided varying degrees of evidence supporting the conclusion that elevated O_3 levels could have negative impacts on lung function and symptoms, confirming and adding to the body of knowledge that was presented in the 1996 O_3 AQCD. Some emphasis has been placed in examining the independent role of O_3 in the presence of PM and other pollutants. The other new studies contribute information on cardiopulmonary outcomes which have not been as well documented previously.

7.2.3 Acute Ozone Exposure and Lung Function

As discussed in the 1996 O_3 AQCD and in the earlier chapter of this document on controlled human exposure studies (Chapter 6), a large body of literature from clinical and field studies has clearly and consistently demonstrated reversible decrements in pulmonary function following acute O_3 exposure. Significant O_3-induced spirometric and symptom responses have been observed in clinical studies of exercising healthy young adults (see Section 6.2) and in some potentially susceptible subpopulations, namely asthmatics and children (see Sections 6.3.2 and 6.5.1). Field studies of acute O_3 exposure that examine pulmonary function fall into two distinct groupings, those that conduct spirometry (measuring FEV_1, FVC [forced vital capacity], and other spirometric indices) and those that measure PEF using peak flow meters. Results from the previous O_3 AQCD and Chapter 6 of this document support the conclusion that the

spirometric parameter FEV_1 is a strong and consistent measure of lung function and may be used in the assessment of asthma (Fuhlbrigge et al., 2001). PEF is a closely related but different metric of lung function. PEF measurements have been shown to be more variable than FEV_1 in some studies (Vaughan et al., 1989; Cross and Nelson, 1991), and can have an element of uncertain reliability when self-administered by study subjects. However, Lippmann and Spektor (1998) state that PEF measurements from small, inexpensive flow meters, which are more feasible to use in field studies, have been shown to produce similar results to PEF measured spirometrically.

Studies of FEV_1 will be presented first, followed by a discussion of PEF studies. Other dividing aspects within these two major types of lung function studies include health status of subjects (e.g., healthy, mildly asthmatic, severely asthmatic), age group, time spent outdoors, and exertion levels. Several studies brought these factors together to produce informative data. Some FEV_1 studies involved both increased outdoor O_3 exposure and higher exertion levels. The results from this group of subjects may be comparable to those from exercising subjects in the clinical studies discussed in Chapter 6.

7.2.3.1 Acute Ozone Studies with Spirometry (FEV_1)

Studies published over the past decade have provided some new insights on the acute effects of O_3 on FEV_1. The results of all studies that investigated quantitative O_3-related effects on FEV_1 are summarized in the following tables. Tables 7-1a,b,c present changes in FEV_1 associated with O_3 exposure in adults while Tables 7-2a,b,c present effects in children. Tables 7-1b and 7-2b present the effect of O_3 on FEV_1 measured either in the morning or afternoon; Tables 7-1c and 7-2c present O_3 effects on changes in FEV_1 across the day (afternoon FEV_1 – morning FEV_1). Studies that did not provide quantitative O_3 data were not included in the tables (Cuijpers et al., 1994; Delfino et al., 2004; Frischer et al., 1997). The data presented in Höppe et al. (1995a) were further analyzed in a subsequent paper (Höppe et al., 2003); results from the latter paper are included in the tables. In general, the O_3 effect estimates showed decrements for FEV_1 across studies, especially in children. The studies presented in the tables are discussed in further detail, starting with the O_3 effect on individuals with elevated exertion levels and increased exposure due to time spent outdoors, followed by its effect on other potential risk groups.

Table 7-1a. Field Studies that Investigated the Association Between Acute Ambient O_3 Exposure and Changes in FEV_1 in Adults

Reference	Study Location	Study Period	Mean O_3 (SD) Level, ppb	O_3 Index
Korrick et al. (1998)	Mount Washington, NH	Summers 1991, 1992	40 (12)	8-h avg
Brauer et al. (1996)	Fraser Valley, British Columbia, Canada	Jun-Aug 1993	40.3 (15.2)	1-h max
Schindler et al. (2001)	Eight communities in Switzerland	May-Sep 1991	46.6 (1.5-127.6) [a]	8-h avg
Höppe et al. (2003)	Munich, Germany	Apr-Sep 1992-1995	65.9 - 70.4 [b]	½-h max
Romieu et al. (1998)	Mexico City	Mar-May 1996; Jun-Aug 1996	123 (40)	1-h max

[a] Range of 8-h avg concentrations is presented by Schindler et al. (2001).
[b] Range of mean ½-h max O_3 concentrations on high O_3 days is presented for Höppe et al. (2003).

Exercise and outdoor worker panels

The current 8-hour NAAQS for O_3 was originally based on results from controlled human exposure studies, as discussed in Chapter 6. These field studies with subjects at elevated exertion levels are of particular interest due to their similarities to the human chamber studies. The majority of human chamber studies have examined the effects of O_3 exposure in subjects performing continuous or intermittent exercise for variable periods of time (see Chapter 6 of this O_3 AQCD).

A study by Brauer and colleagues (1996) reported unusually large O_3 effects on lung function among outdoor workers. This study presented O_3 effects during an extended outdoor exposure period combined with elevated levels of exertion. The investigators repeatedly measured spirometric lung function before and after outdoor summer work shifts over 59 days on a group of 58 berry pickers in Fraser Valley, British Columbia, Canada. The subjects, both male and female native Punjabi-speakers, ranged in age from 10 to 69 years old, with a mean age of 44 years. Outdoor work shifts averaged 11 hours in duration. The mean 1-h max O_3 concentration was 40.3 ppb (SD 15.2). Exertion levels were estimated using portable heart rate monitors carried over a period of four or more hours by a representative subset of subjects during 16 work shifts. Heart rates over the work shift averaged 36% higher than resting levels.

Table 7-1b. Percent Changes in FEV_1 (95% CI) Associated with Acute Ambient O_3 Exposures in Adults, Ordered by Size of the Estimate [a]

	Reference	Study Population/Analysis	N	% Change in FEV_1
1	Brauer et al. (1996) [b]	Berry pickers, next morning	58	−6.36 (−8.02, −4.70)
2	Brauer et al. (1996) [b]	Berry pickers, afternoon	58	−5.40 (−6.51, −4.28)
3	Romieu et al. (1998) [c]	Street workers on placebo, 1st phase (lag 0-1)	19	−3.55 (−6.28, −0.82)
4	Schindler et al. (2001)	Adults who never smoked (lag 0)	3912	−2.96 (−5.11, −0.76)
5	Romieu et al. (1998) [c]	Street workers on placebo, 1st phase (lag 0)	19	−2.17 (−3.45, −0.89)
6	Höppe et al. (2003) [b]	Athletes, afternoon (lag 0)	43	−1.26 (−2.63, 0.10)
7	Romieu et al. (1998) [c]	Street workers on supplement, 1st phase (lag 0-1)	22	−1.25 (−4.36, 1.86)
8	Romieu et al. (1998) [c]	Street workers on supplement, 1st phase (lag 0)	22	−0.53 (−2.08, 1.01)
9	Romieu et al. (1998) [c]	Street workers on placebo, 2nd phase (lag 0)	23	−0.40 (−1.94, 1.14)
10	Romieu et al. (1998) [c]	Street workers on placebo, 2nd phase (lag 0-1)	23	−0.36 (−2.93, 2.20)
11	Höppe et al. (2003)	Elderly, morning (lag 2)	41	−0.22 (−3.86, 3.42)
12	Romieu et al. (1998) [c]	Street workers on supplement, 2nd phase (lag 0)	19	0.18 (−0.72, 1.08)
13	Höppe et al. (2003) [b]	Athletes, afternoon (lag 2)	43	0.24 (−0.64, 1.12)
14	Höppe et al. (2003) [b]	Athletes, afternoon (lag 1)	43	0.48 (−0.97, 1.94)
15	Höppe et al. (2003) [b]	Athletes, morning (lag 2)	43	0.62 (−0.45, 1.68)
16	Höppe et al. (2003) [b]	Athletes, morning (lag 1)	43	0.71 (−0.65, 2.07)
17	Höppe et al. (2003)	Elderly, afternoon (lag 0)	41	0.75 (−2.08, 3.58)
18	Romieu et al. (1998) [c]	Street workers on supplement, 2nd phase (lag 0-1)	19	0.82 (−0.77, 2.42)
19	Höppe et al. (2003)	Elderly, afternoon (lag 1)	41	1.16 (−1.26, 3.58)
20	Höppe et al. (2003)	Elderly, morning (lag 1)	41	1.82 (−2.19, 5.84)
21	Höppe et al. (2003)	Elderly, afternoon (lag 2)	41	2.88 (−0.24, 6.00)

[a]Change in FEV_1 is per standard unit ppb O_3 (40 ppb for ½-h max O_3 and 1-h max O_3, 30 ppb for 8-h max O_3, and 20 ppb for 24-hr avg O_3).
[b]Brauer et al. (1996) and Höppe et al. (2003) studies also included children. The study population for Brauer et-al. ranged in age from 10 to 69 years (mean age 44 years). For Höppe et al. (2003), the athletes ranged in age from 13 to 38 years (mean age 18 years).
[c]Romieu et al. (1998) present change in FEV_1 (mL). The data from Romieu et al. (1998) were transformed to percent change by dividing the estimates by 3,300 mL (average FEV_1 for 40 year old Mexican-American males by Hankinson et al., 1999).

Table 7-1c. Cross-day Percent Changes in FEV_1 (95% CI) Associated with Acute Ambient O_3 Exposures in Adults, Ordered by Size of the Estimate [a]

	Reference	Study Population/Analysis	N	Cross-day % Change in FEV_1
1	Korrick et al. (1998)	Hikers with wheeze or asthma (post–pre-hike)	40	−4.47 (−7.65, −1.29)
2	Korrick et al. (1998)	Hikers who hiked 8-12 hours (post–pre-hike)	265	−2.07 (−3.78, −0.36)
3	Korrick et al. (1998)	Hikers age 28-37 years (post–pre-hike)	185	−2.01 (−3.42, −0.60)
4	Korrick et al. (1998)	Hikers who never smoked (post–pre-hike)	405	−1.77 (−3.24, −0.30)
5	Korrick et al. (1998)	Hikers male (post–pre-hike)	375	−1.65 (−3.12, −0.18)
6	Korrick et al. (1998)	Hikers age 38-47 years (post–pre-hike)	142	−1.59 (−3.12, −0.06)
7	Korrick et al. (1998)	All hikers (post–pre-hike)	530	−1.53 (−2.82, −0.24)
8	Korrick et al. (1998)	All hikers, with $PM_{2.5}$ and acidity in model (post–pre-hike)	530	−1.44 (−3.32, 0.44)
9	Korrick et al. (1998)	Hikers age 18-27 years (post–pre-hike)	135	−1.29 (−2.88, 0.30)
10	Korrick et al. (1998)	Hikers female (post–pre-hike)	155	−1.17 (−3.46, 1.12)
11	Korrick et al. (1998)	Hikers age 48-64 years (post–pre-hike)	68	−1.14 (−3.08, 0.80)
12	Korrick et al. (1998)	Hikers without wheeze or asthma (post–pre-hike)	490	−1.08 (−2.49, 0.33)
13	Korrick et al. (1998)	Hikers who hiked 2-8 hours (post–pre-hike)	265	−0.99 (−2.70, 0.72)
14	Korrick et al. (1998)	Hikers who formerly smoked (post–pre-hike)	125	−0.72 (−3.07, 1.63)
15	Brauer et al. (1996) [b]	Berry pickers (post–pre-work shift)	58	0.00 (−1.66, 1.66)

[a]Change in FEV_1 is per standard unit ppb O_3 (40 ppb for ½-h max O_3 and 1-h max O_3, 30 ppb for 8-h max O_3, and 20 ppb for 24-h avg O_3).
[b]Brauer et al. (1996) study also included children. The study population ranged in age from 10 to 69 years (mean age 44 years).

Post-shift FEV_1 and FVC decreased as a function of O_3 concentration and the effects of O_3 remained significant after adjusting for $PM_{2.5}$ in the analysis. Declines in lung function also were observed on the morning following high O_3 exposure. The effects seen in this study are larger than have been reported previously in studies with briefer exposure durations. For example, afternoon FEV_1 was 3.8 mL (95% CI: −4.6, −3.0) lower per 1 ppb increase in O_3 concentrations, compared to the decline of 0.4 mL/ppb and 1.35 mL/ppb observed in the earlier adult exercise studies (Spektor et al., 1988b; Selwyn et al., 1985). These results are consistent

Table 7-2a. Field Studies that Investigated the Association Between Acute Ambient O_3 Exposure and Changes in FEV_1 in Children

Reference	Study Location	Study Period	Mean O_3 (SD) Level, ppb	O_3 Index
Linn et al. (1996)	Rubidoux, Upland, and Torrance, CA	Fall-spring 1992-1993, 1993-1994	23 (12)	24-h avg
Scarlett et al. (1996)	Surrey, England	Jun-Jul 1994	50.7 (24.48)	8-h max
Höppe et al. (2003)	Munich, Germany	Apr-Sep 1992-1995	65.9 - 70.4 [a]	½-h max
Ulmer et al. (1997)	Freudenstadt and Villingen, Germany	Mar-Oct 1994	Freudenstadt: 50.6 (22.5-89.7) [b] Villingen: 32.1 (0.5-70.1) [b]	½-h max ½-h max
Castillejos et al. (1995)	SW Mexico City	Aug 1990-Oct 1991	112.3 (0-365) [c]	1-h max
Romieu et al. (2002)	Mexico City	Oct 1998-Apr 2000	102 (47)	1-h max
Chen et al. (1999)	Sanchun, Taihsi, and Linyuan, Taiwan	May 1995-Jan 1996	19.7 - 110.3 [c]	1-h max

[a] Range of mean ½-h max O_3 concentrations on high O_3 days is presented for Höppe et al. (2003).
[b] Median and 90th percentile interval are presented for Ulmer et al. (1997).
[c] Range of 1-h max O_3 concentrations are presented by Castillejos et al. (1995) and Chen et al. (1999).

with the interpretation that extended exposures to O_3 produce more marked effects on lung function. Further, when data were restricted to days with 1-h max O_3 concentrations under 40 ppb, the O_3 effects on afternoon FEV_1 did not change in magnitude and remained significant. However, a possible role of copollutants cannot be completely excluded.

In a Mexico City study of 47 outdoor street workers (Romieu et al., 1998), spirometry was performed repeatedly at the end of the work shift over a two-month period. Subjects were exposed to outdoor ambient O_3 levels for a mean of 7.4 hours during the workday. Among those who had never taken an antioxidant supplement (subjects who received a placebo during the first phase of the study), same day O_3 concentrations were associated with decreases in FEV_1. A mean change of −71.6 mL (95% CI: −113.9, −29.3) (approximately a 4% decline) was observed per 40 ppb increase in 1-h max O_3. The results from this study, in addition to those from the Canadian study of berry pickers (Brauer et al., 1996), indicate that outdoor workers are a potentially vulnerable population that may need protection from O_3 exposures.

Table 7-2b. Percent Changes in FEV_1 (95% CI) Associated with Acute Ambient O_3 Exposures in Children, Ordered by Size of the Estimate [a]

	Reference	Study Population/Analysis	N	% Change in FEV_1
1	Ulmer et al. (1997) [b]	School children in Freudenstadt (lag 1)	57	−4.60 (−7.54, −1.67)
2	Ulmer et al. (1997) [b]	School boys in Freudenstadt and Villingen (lag 1)	67	−3.23 (−6.47, 0.00)
3	Ulmer et al. (1997) [b]	School children in Freudenstadt and Villingen (lag 1)	135	−2.98 (−5.33, −0.63)
4	Ulmer et al. (1997) [b]	School girls in Freudenstadt and Villingen (lag 1)	68	−2.32 (−5.53, 0.88)
5	Höppe et al. (2003) [c]	Asthmatics, afternoon (lag 2)	43	−2.08 (−6.24, 2.08)
6	Chen et al. (1999)	Children, with NO_2 in model (lag 1)	895	−1.97 (−3.51, −0.43)
7	Chen et al. (1999)	Children (lag 1)	895	−1.48 (−2.84, −0.12)
8	Romieu et al. (2002) [b]	Moderate to severe asthmatic children on placebo (lag 1)	35	−0.99 (−1.80, −0.18)
9	Romieu et al. (2002) [b]	Moderate to severe asthmatic children on placebo, with NO_2 and PM_{10} in model (lag 1)	35	−0.97 (−1.87, −0.07)
10	Chen et al. (1999)	Children (lag 2)	895	−0.93 (−2.56, 0.71)
11	Ulmer et al. (1997) [b]	School children in Villingen (lag 1)	78	−0.79 (−3.93, 2.34)
12	Chen et al. (1999)	Children (lag 7)	895	−0.72 (−1.81, 0.37)
13	Höppe et al. (2003) [c]	Asthmatics, afternoon (lag 1)	43	−0.56 (−4.61, 3.50)
14	Linn et al. (1996) [b]	School children, next morning	269	−0.27 (−0.79, 0.24)
15	Linn et al. (1996) [b]	School children, afternoon	269	−0.19 (−0.73, 0.35)
16	Romieu et al. (2002) [b]	All asthmatic children on placebo (lag 1)	78	−0.19 (−0.71, 0.33)
17	Höppe et al. (2003)	Children, afternoon (lag 0)	44	−0.14 (−2.71, 2.42)
18	Höppe et al. (2003) [c]	Asthmatics, afternoon (lag 0)	43	−0.10 (−6.59, 6.39)
19	Romieu et al. (2002) [b]	Moderate to severe asthmatic on supplement (lag 1)	47	−0.04 (−0.80, 0.72)
20	Romieu et al. (2002) [b]	Moderate to severe asthmatic on supplement, with NO_2 and PM_{10} in model (lag 1)	47	−0.01 (−0.82, 0.80)
21	Scarlett et al. (1996) [d]	School children (lag 1)	154	0.01 (−0.20, 0.22)
22	Romieu et al. (2002) [b]	All asthmatic children on supplement (lag 1)	80	0.04 (−0.52, 0.60)
23	Höppe et al. (2003) [c]	Asthmatics, morning (lag 1)	43	0.30 (−3.93, 4.53)
24	Höppe et al. (2003)	Children, morning (lag 1)	44	0.83 (−0.53, 2.20)

Table 7-2b (cont'd). Percent Changes in FEV_1 (95% CI) Associated with Acute Ambient O_3 Exposures in Children, Ordered by Size of the Estimate [a]

	Reference	Study Population/Analysis	N	% Change in FEV_1
25	Höppe et al. (2003)	Children, afternoon (lag 1)	44	0.93 (−0.80, 2.66)
26	Höppe et al. (2003)	Children, morning (lag 2)	44	1.17 (−0.36, 2.70)
27	Höppe et al. (2003)	Children, afternoon (lag 2)	44	1.20 (−0.12, 2.52)
28	Höppe et al. (2003) [c]	Asthmatics, morning (lag 2)	43	1.40 (−3.69, 6.49)

[a]Change in FEV_1 is per standard unit ppb O_3 (40 ppb for ½-h max O_3 and 1-h max O_3, 30 ppb for 8-h max O_3, and 20 ppb for 24-h avg O_3).
[b]Linn et al. (1996), Romieu et al. (2002), and Ulmer et al. (1997) present change in FEV_1 (mL). The data were transformed to percent change by dividing the estimates by 1,900 mL (average FEV_1 among 8 to 10 year olds by Hankinson et al., 1999).
[c]Höppe et al. (2003) study also included young adults. The study population age for the asthmatics ranged from 12 to 23 years (mean age 15 years).
[d]$FEV_{0.75}$ results are presented in Scarlett et al. (1996).

Table 7-2c. Cross-day Percent Changes in FEV_1 (95% CI) Associated with Acute Ambient O_3 Exposures in Children, Ordered by Size of the Estimate [a]

	Reference	Study Population/Analysis	N	Cross-day % Change in FEV_1
1	Linn et al. (1996) [b]	School children (p.m.−a.m.)	269	−0.61 (−1.09, −0.14)
2	Castillejos et al. (1995)	Private primary school (post−pre-exercise)	40	−0.48 (−0.72, −0.24)

[a]Change in FEV_1 is per standard unit ppb O_3 (40 ppb for ½-h max O_3 and 1-h max O_3, 30 ppb for 8-h max O_3, and 20 ppb for 24-h avg O_3).
[b]Linn et al. (1996) present change in FEV_1 (mL). The data were transformed to percent change by dividing the estimates by 1,900 mL (average FEV_1 among 8 to 10 year olds by Hankinson et al., 1999).

Höppe et al. (1995a) examined forestry workers (n = 41) for changes in pulmonary function attributable to O_3 exposure in Munich, Germany. In addition, athletes (n = 43) were monitored in the afternoon following a two-hour outdoor training period. Pulmonary function tests were conducted on days of both "high" (mean ½-h max O_3 of 64 to 74 ppb) and "low" (mean ½-h max O_3 of 32 to 34 ppb) ambient O_3 concentrations. From the average activity levels, ventilation rates were estimated. Athletes, who had a fairly high ventilation rate of 80 L/min, experienced a significant decrease of 60.8 mL (95% CI: 6.4, 115.2) in FEV_1 per 40 ppb increase

in ½-h max O_3. Among the forestry workers, a similar O_3-related decline in FEV_1 also was observed (−56.0 mL [95% CI: −118.4, 6.4]). In a subsequent study, Höppe et al. (2003) reanalyzed the results of the athletes after stratifying the spirometric data by time of day (morning versus afternoon) and at different lag periods (lags of 0 to 2 days). The reanalysis indicated that O_3-related decrements were observed only with the afternoon FEV_1 at a 0-day lag, −1.26% (95% CI: −2.63, 0.10) change in FEV_1 per 35 ppb increase in 3-h avg O_3.

One FEV_1 study clearly demonstrated small but measurable effects of multihour O_3 exposures on adults exercising outdoors. In Korrick et al. (1998), adult hikers (n = 530) of Mount Washington, NH performed spirometry before and after hiking for a mean of 8 hours (range 2–12). The mean hourly O_3 concentration ranged from 21 to 74 ppb. After the hike, all subjects combined experienced a small mean decline of 1.5% (95% CI: 0.2, 2.8) in FEV_1 and 1.3% (95% CI: 0.5, 2.1) in FVC per 30 ppb increase in the mean of the hourly O_3 concentration during the hike. In addition, Korrick et al. (1998) compared hikers who hiked 8 to 12 hours to those who hiked 2 to 8 hours. Among those who hiked longer, the percent change in FEV_1 was more than twofold greater per ppb exposure compared to those who hiked only for 2 to 8 hours. Each hour hiked, which may reflect dose, was associated with a decline of 0.3% ($p = 0.05$) in FEV_1, after adjusting for O_3.

In a Mexico City study, the O_3 effect attributable to exercise was determined using a group of school children (n = 40) chronically exposed to moderate to high levels of O_3 (Castillejos et al., 1995). Children were tested up to 8 times between August 1990 and October 1991. Spirometry was performed by the children before and after a one-hour intermittent exercise session outdoors. Outdoor O_3 levels ranged up to 365 ppb, with a mean of 112.3 ppb. Linear trend analyses indicated a relationship between quintiles of O_3 and percent change in lung function. However, stratified analyses indicated that significant changes were observed only with higher quintiles of O_3 exposure (72-125 ppb and 183-365 ppb). Therefore, children exercising at higher O_3 levels experienced declines in pulmonary function despite the repeated daily exposure to moderate and high levels of O_3 in Mexico City.

Collectively, the above studies confirm and extend clinical observations that prolonged exposure periods, combined with elevated levels of exertion or exercise, may magnify the effect of O_3 on lung function. The most representative data come from the Korrick et al. (1998) hiker study. This U.S. study provided outcome measures stratified by several factors (e.g., gender,

age, smoking status, presence of asthma) within a population capable of more than normal exertion.

Panel studies of children, elderly, and asthmatics

Höppe et al. (1995a,b) examined several potentially susceptible populations for changes in pulmonary function attributable to O_3 exposure in Munich, Germany. The forestry workers and athletes were discussed in the previous section. Senior citizens (n = 41) and juvenile asthmatics (n = 43) were also monitored on "low" O_3 and "high" O_3 days. Subjects were requested to stay outdoors for at least 2 hours just before the afternoon pulmonary function test. Clerks (n = 40) were considered the nonrisk control group. Although clerks spent the majority of their time indoors, their outdoor exposures on high O_3 days were similar to that of the four other risk groups. The results showed no significant O_3 effects on the senior citizens. Clinical studies also have consistently shown that seniors are less responsive to O_3 (Bedi et al., 1989; Drechsler-Parks, 1995). Asthmatics and clerks experienced slight reductions in FEV_1 on high O_3 days. Among all risk groups, juvenile asthmatics experienced the largest O_3-related decline in FEV_1, −84.0 mL (95% CI: −196.4, 28.4) per 40 ppb increase in ½-h max O_3. To further examine their hypotheses on characteristics of O_3 risk groups, Höppe et al. (2003) conducted a different analysis on a more expanded data base than utilized in the earlier study. Children were examined as an additional risk group. Höppe et al. (2003) presented both group mean values and analyses on an individual basis. For the group mean values, consistent O_3 effects were not detectable. On an individual basis, a potential pattern of O_3 sensitivity was observed (see Table AX7-1 in the Annex for details). About 20% of the children and asthmatics were regarded as O_3 responders (i.e., individuals with greater than 10% change in FEV_1) compared to only 5% of the elderly and athletes. These results indicated that while the majority of the population did not react to O_3 exposure, a small group of susceptible individuals experienced health effects from O_3. The sample size limits quantitative extrapolation to larger populations, but may allow cautious first estimates.

Several other panel studies performed spirometry in children, another potentially susceptible group (Avol et al., 1998; Chen et al., 1999; Cuijpers et al., 1994; Frischer et al., 1997; Linn et al., 1996; Romieu et al., 2002; Scarlett et al., 1996; Ulmer et al., 1997). All studies, with the exception of Avol et al. (1998) and Scarlett et al. (1996), observed a

decrease in FEV_1 associated with O_3 exposure. One large study measured spirometric lung function in 895 school children in three towns in Taiwan (Chen et al., 1999). Lung function was measured only once for each subject. The authors reported significant associations between diminished FEV_1 and FVC with a 1-day lag of O_3 concentrations. Effect sizes were typical of those observed in past studies, i.e., 0.5 to 1.0 mL decline in FEV_1 per ppb increase in O_3 concentration. Ozone was the only air pollutant associated with changes in lung function in multipollutant models including SO_2, CO, PM_{10}, and NO_2.

Linn et al. (1996) repeatedly measured spirometric lung function among 269 school children in three southern California communities (Rubidoux, Upland, and Torrance). Lung function was measured over five consecutive days, once in each of three seasons over two school years. Between-week variability was controlled in the analysis by seasonal terms in the model. Statistical power was limited by the narrow range of exposures that were experienced within each week. In addition, the study was restricted to the school year, eliminating most of the "high" O_3 season from consideration. During the study period, 24-h avg O_3 levels at the central monitoring site ranged up to 53 ppb (mean 23 ppb) while personal measurements ranged up to 16 ppb (mean 5 ppb). A mean change of −11.6 mL (95% CI: −20.6, −2.6) (approximately a 1% decline) in FEV_1 was observed from morning to afternoon per 20 ppb increase in 24-h avg O_3. Other associations (involving individual morning or afternoon FVC and FEV_1 measurements) went in the plausible direction but the O_3 effect estimates were considerably smaller.

Ulmer et al. (1997) examined 135 children aged 8 to 11 years in two towns in Germany from March to October 1994 for O_3 effects on pulmonary function at four time periods. The cross-sectional results at each of the four time points showed limited FVC and no FEV_1 associations. However, the longitudinal analysis, which combined data from all four periods yielded a mean change of −87.5 mL (95% CI: −143.2, −31.7) (approximately a 5% decline) in FEV_1 per 40 ppb increase in ½-h max O_3 for the town with the higher O_3 levels (median ½-h max of 50.6 ppb versus 32.1 ppb). In the cross-sectional analysis, only between-person variability was analyzed. The longitudinal analysis, in which the subjects provided multiple days of measurements, provided information on both between- and within-subject responses.

There are a limited number of new epidemiologic studies examining the effects of O_3 on FEV_1; however, results from these studies indicate that acute exposure to O_3 is associated with declines in FEV_1 in children. These results further support the negative effects of O_3 on lung

function observed in the meta-analysis on children attending summer camp (Kinney et al., 1996a) and in the clinical literature.

7.2.3.2 Acute Ozone Studies of PEF

Many studies of the acute effect of O_3 on PEF examined self-administered PEF levels daily, both in the morning and afternoon. PEF follows a circadian rhythm with the highest values found during the late afternoon and lowest values during the night and early morning. Due to the diurnal variation in PEF, most studies analyzed their data after stratifying by time of day. The peak flow studies examined both asthmatic panels and healthy individuals. The asthma panels are discussed first.

Asthma panels

The effects of acute O_3 exposure on PEF in asthmatics were examined in several panel studies. Figures 7-1 and 7-2 present percent changes in morning and evening PEF outcomes from seven panel studies of children, mostly asthmatic, ranging in age from 5 to 13 years. The effect estimates from all single-day and multiday lag models are presented. Only single-city results with analyses stratified by morning and afternoon are included in the figure. Studies that examined cross-day changes and daily variability in PEF (e.g., Just et al., 2002; Thurston et al., 1997) are not included in the figure since such outcomes are not directly comparable. Collectively, nearly all of the studies indicated decrements of peak flow but most of the individual estimates were not statistically significant. The results from the individual studies are further discussed below.

In Mexico City, two studies of asthmatic school children were carried out simultaneously in the northern (Romieu et al., 1996) and southwestern sections of the city (Romieu et al., 1997). In the northern study, 71 mildly asthmatic school children aged 5 to 13 years old, were followed over time for daily morning (before breakfast) and afternoon (bedtime) PEF. In single-pollutant models, O_3 concentrations at 0-, 1-, and 2-day lags were associated with diminished morning and afternoon PEF, but only the 0-day lag morning effect was significant. The O_3 effect became nonsignificant when $PM_{2.5}$ was added to the model. In the southwestern study, 65 mildly asthmatic children aged 5 to 13 years old were followed during the summer and winter for daily morning and afternoon PEF. Ozone concentrations at a 0- and 1-day lag were associated with

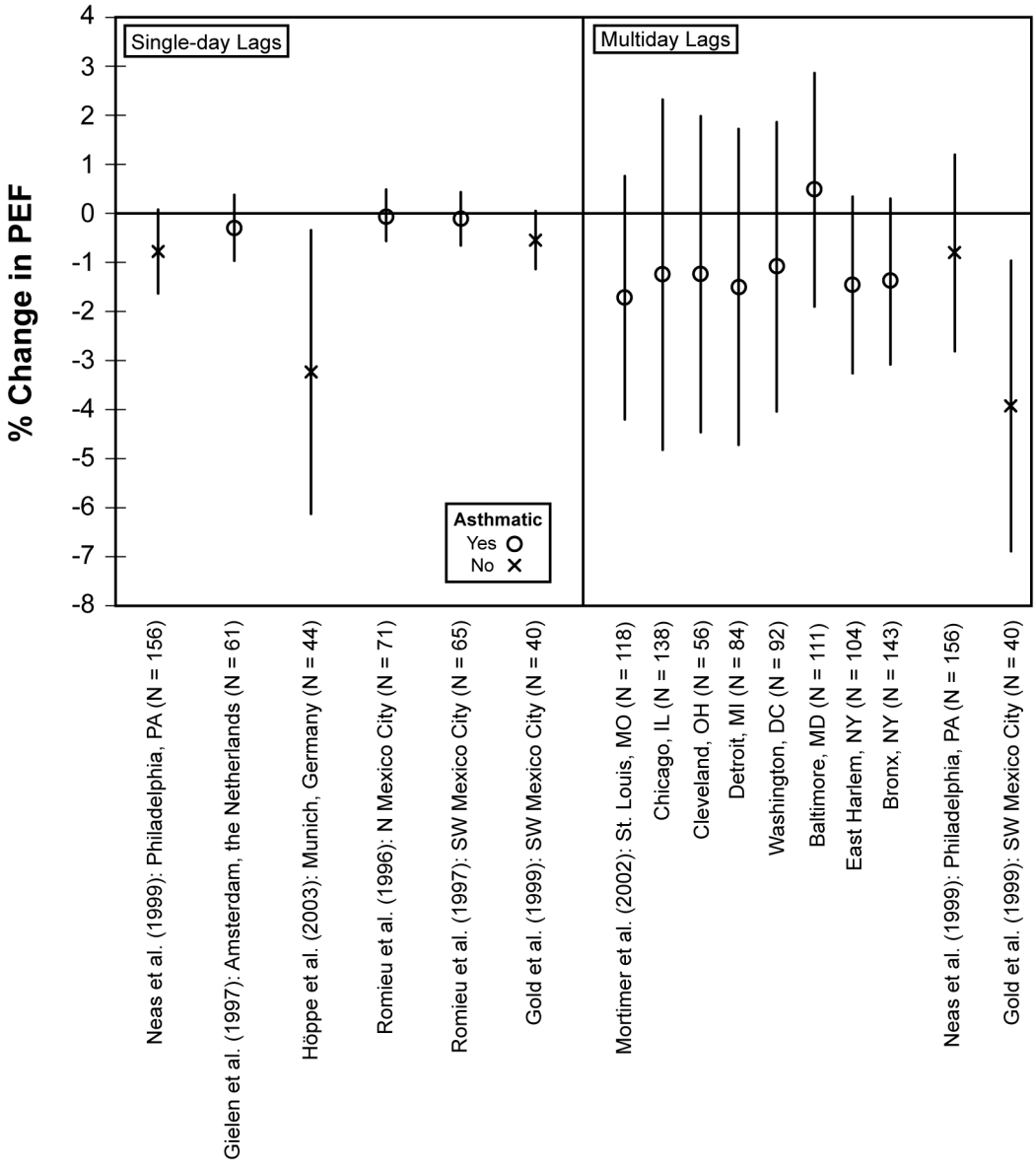


Figure 7-1. Percent change (95% CI) in <u>morning PEF</u> in children per standardized increment (see Section 7.1.3.2). For single-day lag models, previous day O_3 effects are shown. For multiday lag models, the cumulative effects of a 1- to 5-day lag are shown for Mortimer et al. (2002) and Neas et al. (1999), and the effect of a 1- to 10-day lag is shown for Gold et al. (1999).

afternoon PEF, with larger effects at a 1-day lag. Associations involving O_3 were stronger than those involving PM_{10}. Several additional studies, both in the U.S. and in other countries, reported significant associations between O_3 exposure and decrements in PEF among asthmatics

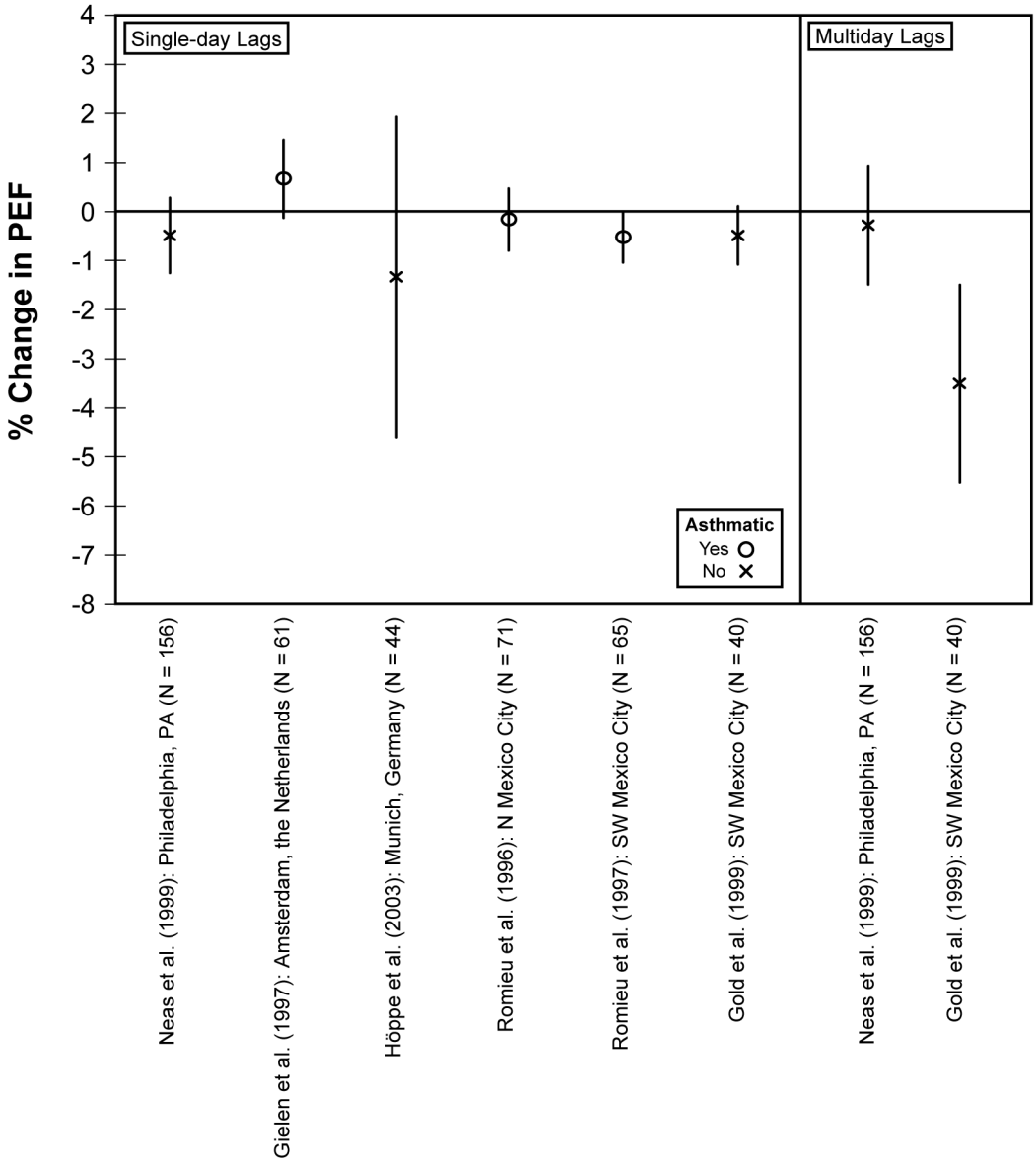


Figure 7-2. Percent change (95% CI) in <u>afternoon PEF</u> in children per standardized increment (see Section 7.1.3.2). For single-day lag models, current day O_3 effects are shown. For multiday lag models, the cumulative effect of a 1- to 5-day lag is shown for Neas et al. (1999) and a 1- to 9-day lag is shown for Gold et al. (1999).

(Gielen et al., 1997; Jalaludin et al., 2000; Just et al., 2002; Ross et al., 2002; Thurston et al., 1997).

Other epidemiologic studies did not find a significant O_3 effect on the lung function of asthmatics. Delfino et al. (1997a) examined morning and evening PEF among 22 asthmatics ranging in age from 9 to 46 years, living in Alpine, CA. Daily ambient 12-h avg O_3 (8 a.m.-8 p.m.) concentrations ranged from 34 to 103 ppb, with a mean value of 64 ppb. Unique to this study, personal O_3 exposures were measured using 12-h passive O_3 samplers that were worn by the subjects. The personal 12-h avg O_3 (8 a.m.-8 p.m.) concentrations, which had a mean value of 18 ppb, were much lower than the fixed-site ambient levels. Quantitative O_3 results were not reported but researchers stated that no O_3 effects were observed on morning and evening PEF. In Hiltermann et al. (1998), 60 nonsmoking adults aged 18 to 55 years in Bilthoven, the Netherlands, were followed between July and October 1995 with morning and afternoon PEF measurements. Although negative associations were observed between O_3 and cross-day changes in PEF, the results were not significant.

Mortimer et al. (2002) examined 846 asthmatic children from the National Cooperative Inner-City Asthma Study (NCICAS) for O_3-related changes in PEF. Children from eight urban areas in the U.S. (St. Louis, MO; Chicago, IL; Detroit, MI; Cleveland, OH; Washington, DC; Baltimore, MD; East Harlem, NY; and Bronx NY) were monitored from June through August 1993. This study provides representative data for the U.S. as children from multiple cities throughout the East and Midwest were examined. Asthmatic children from urban areas are an important subgroup of potentially at-risk populations. Study children either had physician-diagnosed asthma and symptoms in the past 12 months or respiratory symptoms consistent with asthma that lasted more than 6 weeks during the previous year.

Mortimer et al. (2002) examined O_3-related changes in PEF for single-day lags from 1 to 6 days and a multiday lag period of 5 days. Of all the pollutants examined, including O_3, PM_{10}, NO_2, and SO_2, none were associated with evening PEF. Only O_3 was found to be associated with morning PEF. The effect estimates of the association between O_3 and morning PEF for the single-day and multiday lags are depicted as error density curves in Figure 7-3 (for description of error density curves, see Annex Section AX7-2). Small morning effects were observed at 1- and 2-day lags. The effect of O_3 on morning outcomes increased over several days. The strongest association between O_3 and PEF was found with a multiday lag period (cumulative lag of 1 to 5 days). Unrestricted lag models suggested that the O_3 exposure from 3 to 5 days prior had a greater impact on morning % PEF than more immediate exposures. Mortimer et al. discussed

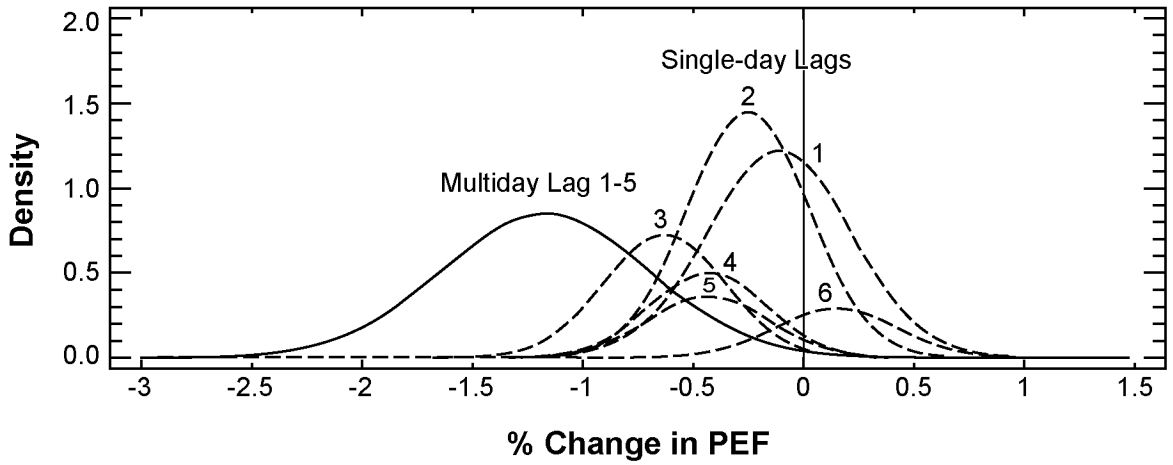


Figure 7-3. Comparison of single-day lags (1-, 2-, 3-, 4-, 5-, and 6-day) to a cumulative multiday lag (1- to 5-day) for percent changes in PEF per 30 ppb increase in 8-h avg O_3 in urban children.

Source: Derived from Mortimer et al. (2002).

biological mechanisms for delayed effects on pulmonary function, which included increased bronchial reactivity secondary to airway inflammation associated with irritant exposure. Animal toxicology and human chamber studies (see Chapters 5 and 6) provide further evidence that exposure to O_3 may augment cellular infiltration and cellular activation, enhance release of cytotoxic inflammatory mediators, and alter membrane permeability.

Figure 7-4 illustrates the probability density curves of the results from the city-stratified analysis and that from the pooled analysis of all eight cities. The error density curve for the all-cities analysis is a graphical presentation of the all-cities regression analysis presented by Mortimer et al. (2002), a change in morning PEF of −1.18% (95% CI: −2.10, −0.26) per 30 ppb increase in 8-h avg O_3 (10 a.m.-6 p.m.) with a cumulative lag of 1 to 5 days. The summary density curve for the city-stratified analysis was calculated by summing together eight normal distribution functions, one for each of the study cities, then taking the derivative of the summed function (see Annex Section AX7-2 for further explanation of summary density curves). The area under the density curve and to the left of a value on the x-axis is an estimate of the probability that the effect estimate will be less than or equal to that value. For example, the area under the density curve to the left of 0% change in PEF is 99% in the all-cities analysis. A wider

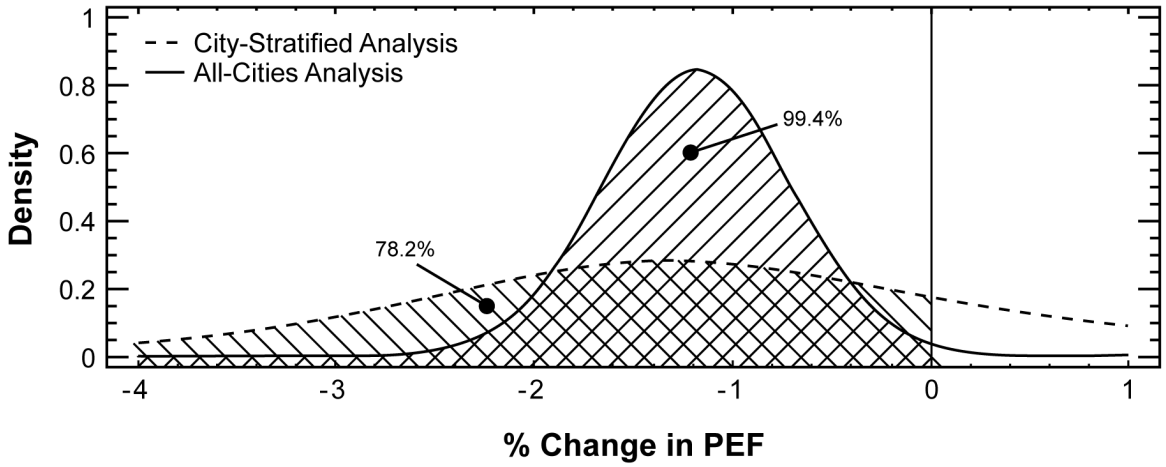


Figure 7-4. Density curves of the percent change in PEF per 30 ppb increase in 8-h avg O_3 with a cumulative lag of 1 to 5 days for the individual eight NCICAS cities and the pooled average of all cities. Note that 99% and 78% of the areas under the curves are less than zero for the pooled cities analysis and individual cities analysis, respectively.

Source: Derived from Mortimer et al. (2002).

distribution was observed in the city-stratified analysis, with only 78% of the area less than zero. The all-cities analysis likely had a smaller standard error compared to the city-specific analysis as it was based upon more subjects and considered differences between cities to vary about the same mean effect. The regression analysis by Mortimer et al. (2002) suggested a lack of heterogeneity by city, as indicated by the nonsignificant interaction term between O_3 effect and city. As shown in Figure 7-4, the summary density curve of the city-stratified analysis has a peak at about the same value as the curve of the all-cities analysis, suggesting a common O_3 effect for all eight cities and small variation among them. The unimodal shape of the density curve of the city-stratified analysis also indicates the absence of outlying cities.

Mortimer et al. (2002) further noted that small declines in morning PEF may be of uncertain clinical significance, thus they calculated the incidence of $\geq$10% declines in PEF. A 5 to 15% change in FEV_1 has been expressed as having clinical importance to asthma morbidity (American Thoracic Society, 1991; Lebowitz et al., 1987; Lippmann, 1988). Although greater variability is expected in PEF measurements, a $\geq$10% change in PEF also may have clinical significance. In Mortimer et al. (2002), O_3 was associated with an increased

incidence of ≥10% declines in morning PEF (odds ratio of 1.30 [95% CI: 1.04, 1.61] per 30 ppb increase in 8-h avg O_3 for a 5-day cumulative lag). This finding suggests that exposure to O_3 might be related to clinically important changes in PEF in asthmatic children. This study also observed that excluding days when 8-h avg O_3 levels were greater than 80 ppb provided effect estimates which were similar to those when all days were included in the analysis, indicating that the negative effect of O_3 on morning PEF persisted at levels below 80 ppb. There is some concern, however, regarding the lack of an association between O_3 and afternoon PEF.

Results from the multicities study by Mortimer et al. (2002), as well as those from several regional studies provide evidence of a significant relationship between O_3 concentrations and PEF among asthmatics. Collectively, these studies indicate that O_3 may be associated with declines in lung function in this potentially susceptible population.

Panels of healthy subjects

The effect of O_3 on PEF in healthy subjects also was investigated in several studies. A study of 162 children (9 years of age) in England examined the relationship between O_3 and PEF in the winter and summer seasons (Ward et al., 2002). The O_3 effect estimates were generally positive in the winter and negative in the summer. Single-day lags of 0- to 3-days were examined; however, the strongest association was found with a multiday lag period. During the summer, a decline of 11.10 L/min (95% CI: 0.18, 21.98) was observed in morning PEF per 20 ppb increase in 24-h avg O_3 with a 7-day cumulative lag. Smaller O_3 effects were observed on afternoon PEF.

During the summer of 1990, Neas et al. (1995) examined 83 children in Uniontown, PA and reported twice daily PEF measurements. Researchers found that evening PEF was associated with O_3 levels weighted by hours spent outdoors. Using a similar repeated measures design, Neas et al. (1999) saw evidence for effects due to ambient O_3 exposure among 156 children attending two summer day camps in the Philadelphia, PA area. Associations were found between afternoon PEF (recorded before leaving camp) and same-day O_3 concentrations, and between morning PEF (recorded upon arrival at camp) and previous-day O_3 concentrations. However, as in the case of Ward et al. (2002), the relationship between PEF and O_3 was significant only when a multiday lag period was considered. Naeher et al. (1999), in a sample of

473 nonsmoking women (age 19 to 43 years) living in Vinton, VA, also showed the strongest association between O_3 and evening PEF with a 5-day cumulative lag exposure.

Another study in southwestern Mexico City analyzed morning and afternoon PEF data collected from 40 school children aged 8 to 11 years (Gold et al., 1999). Subjects provided measurements upon arriving and before departing from school each day. A negative effect of O_3 on PEF was observed, −1.60 mL/s (95% CI: −3.56, 0.36) and −1.80 mL/s (95% CI: −3.76, 0.16) per 20 ppb increase in 24-h avg O_3 on the same day afternoon and next day morning PEF, respectively. A greater effect was observed for PEF regressed on O_3 concentrations with a cumulative 10-day lag period (−3.50 mL/s [95% CI: −5.52, −1.49] on same day afternoon). These results suggest a longer, cumulative effect of O_3 on PEF. Alternatively, the associations observed at the 10-day lag period may reflect confounding by other time-varying factors or be a chance finding from an exploratory analysis.

In a recent study of 43 mail carriers in Taichung City, Taiwan, PEF was monitored twice daily during a six-week period (Chan and Wu, 2005). The mean 8-h avg O_3 (9 a.m.-5 p.m.) concentration during their work shift was 35.6 ppb (SD 12.1). Associations were observed between evening PEF and O_3 concentrations at lags of 0, 1 and 2 days. The greatest effect was observed at a lag of 1 day, a 2.07% decline in PEF per 30 ppb increase in 8-h avg O_3 (quantitative results for 95% CI not provided). Similar O_3 effects on morning PEF were observed. The effect of O_3 on PEF was robust to adjustment for copollutants; no association with PEF was observed for PM_{10} and NO_2 in multipollutant models.

7.2.4 Respiratory Symptoms

Studies published over the past decade represent an improved new body of data on the symptom effects of O_3. Respiratory symptoms in acute air pollution field studies are usually measured using questionnaire forms or "daily diaries" that are filled out by study subjects, usually without the direct supervision of research staff. Questions address the daily experience of coughing, wheezing, shortness of breath (or difficulty breathing), production of phlegm, and others. While convenient and potentially useful in identifying acute episodes of morbidity, measurements of daily symptoms are prone to a variety of errors. These include misunderstanding of the meaning of symptoms, variability in individual interpretation of symptoms, inability to remember symptoms if not recorded soon after their occurrence, reporting

bias if days of high air pollution levels are identifiable by subjects, and the possibility of falsified data. In spite of these potential problems, the ease of data collection has made daily symptom assessment a common feature of field studies. Many of the studies reviewed above for lung function results also included measurements of daily symptoms. Pearce et al. (1998) reports that one advantage in the case of asthma panels is that the population is usually already familiar with symptom terms such as wheezing and cough. Delfino et al. (1998a) further states that the use of repeated daily symptom diaries has additional advantages of reducing recall bias given the proximity of events and allowing health effects to be modeled with each subject serving as their own control over time. Also, study design can blind the participants from the air pollution aspect of the study. Careful efforts by study staff can help ensure that the symptom diaries will provide information that is less affected by the potential problems noted.

Similar to studies of lung function, respiratory symptom studies can be divided into two groups, asthma panels or healthy subjects. Asthma panel studies are presented first.

Asthma panels

Most studies examining respiratory symptoms related to O_3 exposure focused on asthmatic children. Among the health outcomes, of particular interest were those associated with asthma, including cough, wheeze, shortness of breath, and increased medication use. Figures 7-5 and 7-6 present the odds ratios for O_3-related cough and medication use among asthmatic children from six studies (Gielen et al., 1997; Jalaludin et al., 2004; Just et al., 2002; Ostro et al., 2001; Romieu et al., 1996, 1997). Only single city/region studies that present odds ratios are included in the figure for consistency. Studies that present change in severity of symptoms, another informative health outcome, are not included in the figure since this symptom outcome differs from indicating simple presence of symptoms. The study by Gent et al. (2003) also is not included in this figure as odds ratios for cough and mediation use were analyzed for quintiles of O_3 concentrations using the lowest quintile as the reference. These studies are discussed separately.

The various effect estimates for the association between O_3 concentrations and cough are depicted in Figure 7-5. Despite the variability in the individual effect estimates, there is some consistency in the O_3 effects. In general, the majority of the odds ratios appear to be greater than one among the single-day lag models, suggesting an association between acute exposure to O_3 and increased cough among asthmatic children. Figure 7-6 presents the odds ratios for

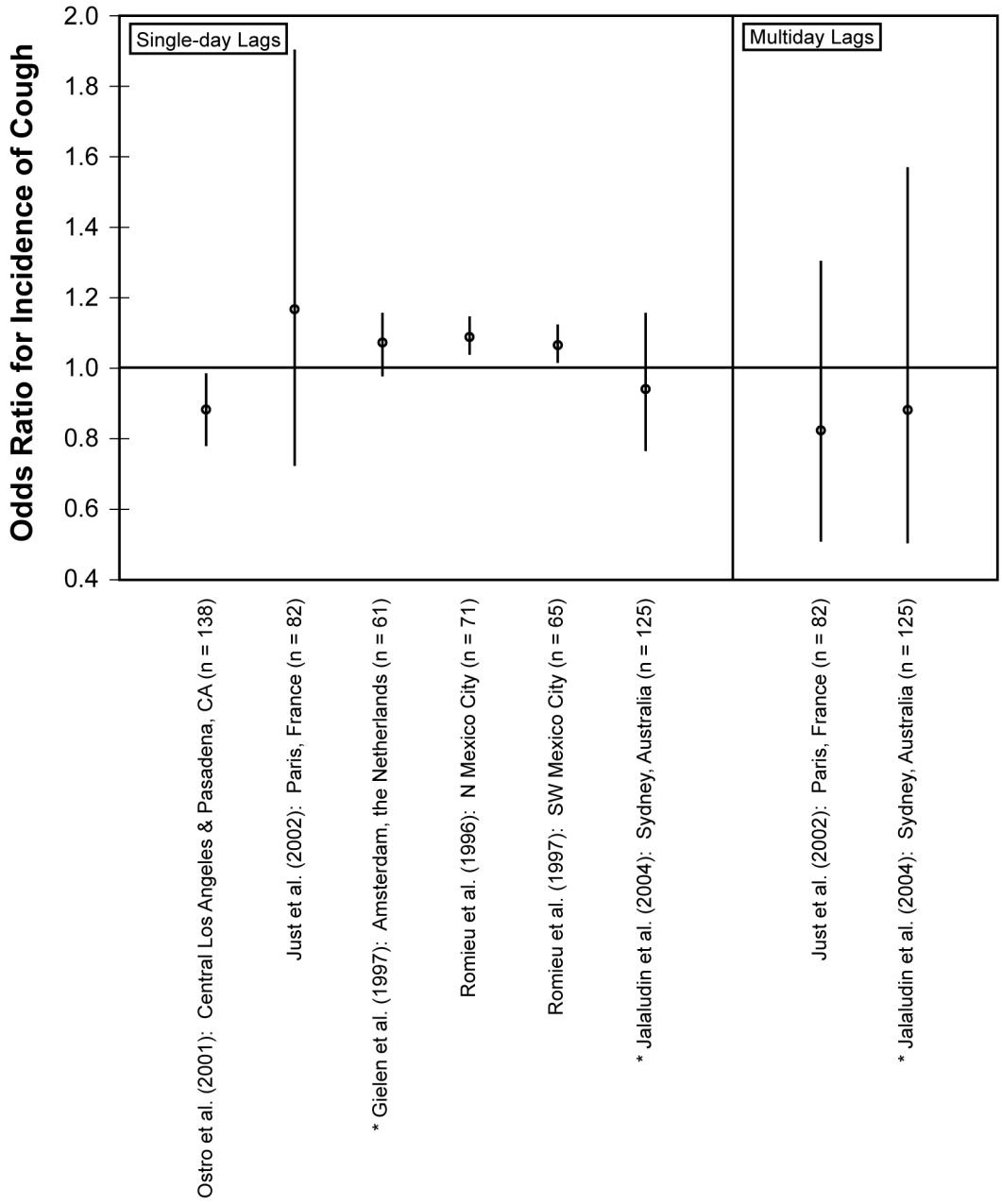


Figure 7-5. Odds ratios for the incidence of cough among asthmatic children per standardized increment (see Section 7.1.3.2). For single-day lag models, current day O_3 effects are shown with the exception of Ostro et al. (2001) which only presented results from a 3-day lag. For multiday lag models, the cumulative effects of a 0- to 4-day lag are shown. *Note that Gielen et al. (1997) and Jalaludin et al. (2004) presented results for prevalence of cough.

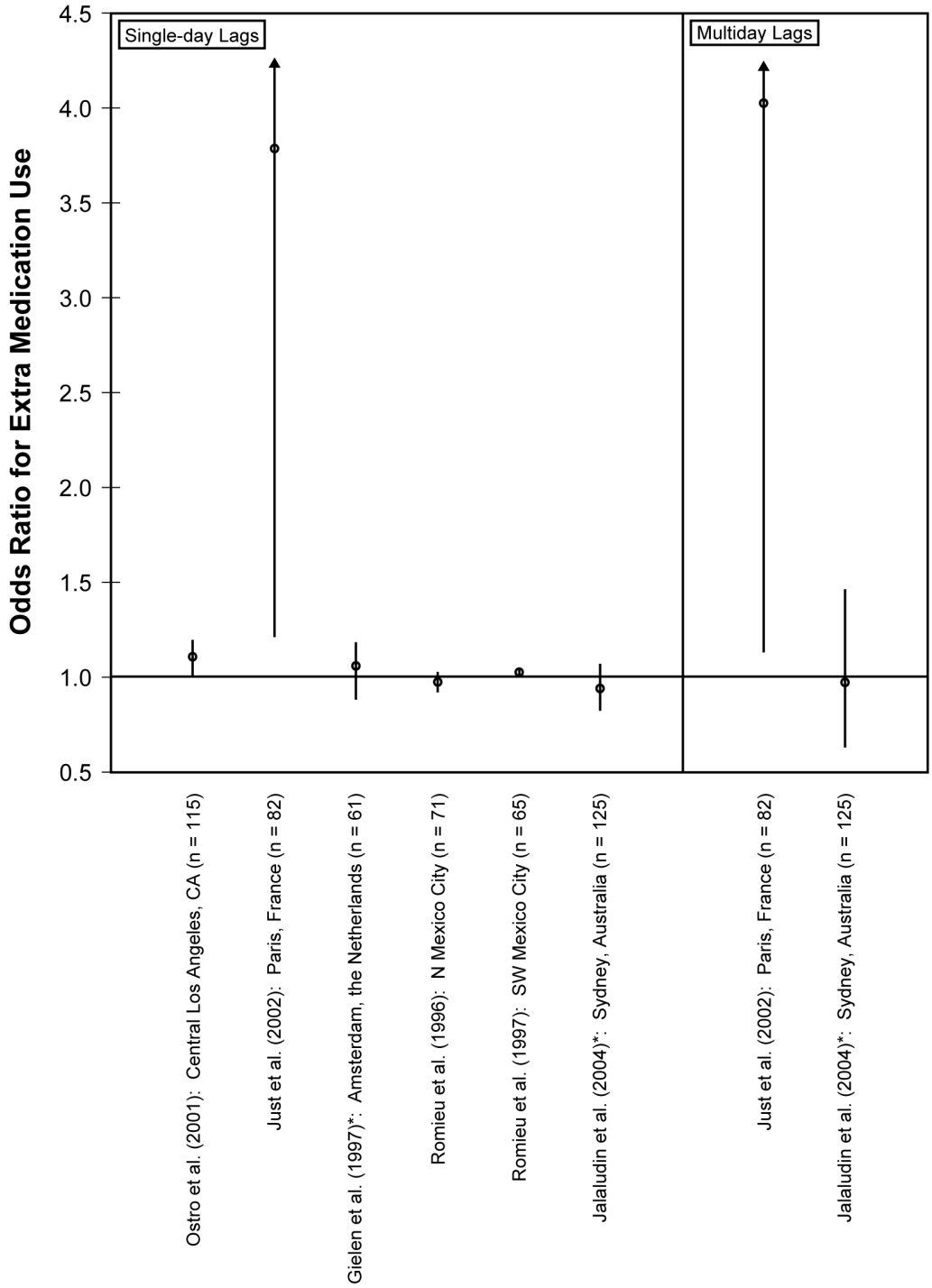


Figure 7-6. Odds ratios for extra medication use among asthmatic children per standardized increment (see Section 7.1.3.2). For single-day lag models, current day O_3 effects are shown. For multiday lag models, the cumulative effects of a 0- to 4-day lag are shown.

O_3-associated bronchodilator use. The results from medication use are less consistent than those from cough; one study by Just et al. (2002) observed strong positive associations, but had wide confidence intervals.

Among the studies reporting results for daily symptoms and asthma medication use, several observed associations with O_3 concentrations that appeared fairly robust (Delfino et al., 2003; Desqueyroux et al., 2002a,b; Gent et al., 2003; Hilterman et al., 1998; Just et al., 2002; Mortimer et al., 2000, 2002; Newhouse et al., 2004; Romieu et al., 1996, 1997; Ross et al., 2002; Thurston et al., 1997). Mortimer et al. (2002) reported morning symptoms in 846 asthmatic children from eight urban areas of the U.S. to be most strongly associated with a cumulative 4-day lag of O_3 concentrations in the NCICAS. The NCICAS used standard protocols which included instructing caretakers of the subjects to record symptoms in the daily diary by observing or asking the child (Mitchell et al., 1997). Symptoms reported included cough, chest tightness, and wheeze. In the analysis pooling data from all eight cities, the odds ratio for the incidence of symptoms was 1.35 (95% CI: 1.04, 1.69) per 30 ppb increase in 8-h avg O_3 (10 a.m.-6 p.m.). Excluding days when 8-h avg O_3 was greater than 80 ppb, the odds ratio was 1.37 (95% CI: 1.02, 1.82) for incidence of morning symptoms. Figure 7-7 presents the probability density curves of the odds ratios for the incidence of symptoms from the city-stratified analysis and that from the all-cities analysis. This figure confirms the regression results that there is a significant increase in odds for incidence of symptoms, as the area under the density curve with an odds ratio greater than one is 99%. Mortimer et al. (2002) did not observe significant interactions among the eight cities, indicating that there was no heterogeneity among the city-specific estimates. The unimodal distribution of the city-stratified summary density curve also suggests a lack of significant heterogeneity in O_3 effects among the cities. It should be noted that other pollutants, including PM_{10} (monitored in 3 cities), NO_2 (in 7 cities), and SO_2 (in all 8 cities), also were associated with increased incidence of morning symptoms. In multipollutant models, the O_3 effect was shown to be slightly diminished.

Another one of the larger studies was that of Gent and colleagues (2003), where 271 asthmatic children under age 12 and living in southern New England were followed over 6 months (April through September) for daily symptoms. The data were analyzed for two separate groups of subjects, 130 who used maintenance asthma medications during the follow-up period and 141 who did not. The need for regular medication was considered to be a proxy for

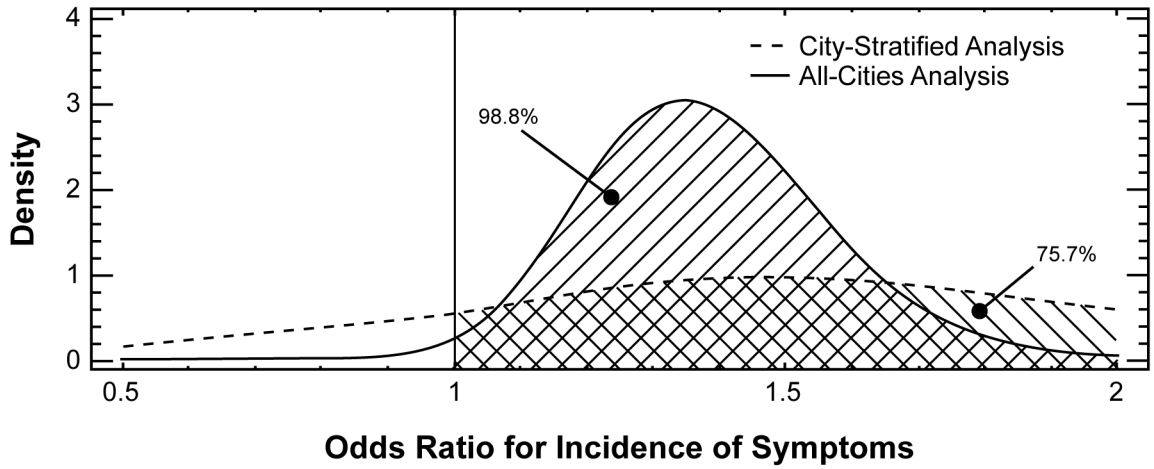


Figure 7-7. Density curves of the odds ratios for the incidence of symptoms per 30 ppb increase in 8-h avg O_3 with a cumulative lag of 1 to 4 days for the individual eight cities and the pooled average of all cities. Note that 99% and 76% of the areas under the curves are greater than one for the pooled cities and individual cities analyses, respectively.

Source: Derived from Mortimer et al. (2002).

more severe asthma. Not taking any medication on a regular basis and not needing to use a bronchodilator would suggest the presence of very mild asthma. Effects of 1-day lag O_3 were observed on a variety of respiratory symptoms only in the medication user group. Both daily 1-h max and 8-h max O_3 concentrations were similarly related to symptoms such as chest tightness and shortness of breath. Effects of O_3, but not $PM_{2.5}$, remained significant and even increased in magnitude in two-pollutant models. Some of the associations were noted at 1-h max O_3 levels below 60 ppb. In contrast, no effects were observed among asthmatics not using maintenance medication. In terms of person-days of follow-up, this is one of the larger studies currently available that address symptom outcomes in relation to O_3, and provides supportive evidence for effects of O_3 independent of $PM_{2.5}$. Study limitations include limited control for meteorological factors and the post-hoc nature of the population stratification by medication use.

Some international studies have reported significant symptoms associations with O_3. The incidence of asthma attacks was associated with O_3 concentrations in a group of 60 severe asthmatics (mean age 55 years) followed over a 13-month period in Paris (Desqueyroux et al.,

2002a). In a similar study, Desqueyroux et al. (2002b) observed O_3-associated exacerbation of symptoms in 39 adult patients (mean age 67 years) with chronic obstructive pulmonary disease (COPD). Interestingly, in contrast to the controlled human studies (see Section 6.3.1, Subjects with COPD), the O_3 effect appeared larger among subjects who smoked and those with more severe COPD. However, the low O_3 concentrations experienced during this study (summer mean 8-h max O_3 of approximately 21 ppb [SD 9]) raise plausibility questions. In a study of 60 nonsmoking asthmatic adults (aged 18 to 55 years) in Bilthoven, the Netherlands, Hilterman and colleagues (1998) reported associations between O_3 and daily symptoms of shortness of breath and pain upon deep inspiration. The O_3 associations were stronger than those of PM_{10}, NO_2, SO_2, and black smoke (BS). No differences in response were evident between subgroups of subjects defined on the basis of steroid use or airway hyperresponsiveness. Daily use of bronchodilators or steroid inhalers was not found to be associated with O_3 in this study.

Other studies showed only limited or a lack of evidence for symptom increases associated with O_3 exposure (Avol et al., 1998; Chen et al., 1998; Delfino et al., 1996, 1997a, 1998a; Gielen et al., 1997; Jalaludin et al., 2004; Ostro et al., 2001; Taggart et al., 1996). Avol et al. (1998) studied symptoms in asthmatic, wheezy, and healthy children aged 10 to 12 years in southern California. Some symptom associations were noted but they were inconsistent. For example, children with wheeze were at increased risk of difficulty breathing and wheezing at low O_3 concentrations, but not at higher O_3 concentrations. Authors noted that O_3 concentrations were relatively low and that children studied did not spend substantial time outdoors engaged in physical activities. Ostro et al. (2001) reported no associations between daily symptoms and ambient O_3 concentrations in a cohort of 138 African-American children with asthma followed over 3 months (August to October) in Central Los Angeles and Pasadena, CA. However, the use of extra asthma medication was associated with 1-h max O_3 concentrations at a 1-day lag. Delfino and colleagues (1996) followed 12 asthmatic teens living in San Diego, CA for respiratory symptoms over a two-month period and saw no relationship with central site ambient O_3. Personal O_3 exposures measured with passive diffusion monitors were associated with the composite symptom score and β_2-agonist inhaler use, but the relationship with symptom score disappeared when weekday/weekend differences were controlled in the statistical analysis. Study power was likely compromised by the small sample size. This observation of stronger associations with O_3 levels from personal monitors implies that gains in power may be achieved

if exposure misclassification is reduced through the use of personal exposure measurements rather than central site ambient O_3 concentrations. A similar study of 22 asthmatics in Alpine, CA observed no effects of O_3 on symptoms when personal O_3 exposure was used as the exposure metric (Delfino et al., 1997a). However, a later study in the same location involving 24 subjects (Delfino et al., 1998a) did find an association between respiratory symptoms and ambient O_3 exposure, with stronger O_3 effects experienced by asthmatics not on anti-inflammatory medication. In this study, a binary symptom score was used, whereas the earlier study used a linear symptom score of 0 through 6.

In conclusion, the various studies seem to indicate that O_3 concentrations are associated with respiratory symptoms and increased medication use in asthmatics. The multicities study by Mortimer et al. (2002) provides an asthmatic population most representative of the U.S., and several single-city studies also add to the knowledge base. However, there are a number of well-conducted, albeit relatively smaller studies that have not found these effects.

Panels of healthy subjects

Fewer studies examined the effect of O_3 on respiratory symptoms in healthy individuals. Neas et al. (1995) reported that in school children, evening cough was associated with O_3 levels weighted by hours spent outdoors. The study by Linn and colleagues (1996) of 269 school children in southern California reported no associations between respiratory symptoms and O_3, but subjects were exposed to fairly low O_3 concentrations as determined using personal monitors. Gold et al. (1999) examined symptoms in 40 healthy children in southwest Mexico City. Pollutant exposures were associated with increased production of phlegm in the morning, although the effects of the air pollutants ($PM_{2.5}$, PM_{10}, and O_3) could not be separated in multipollutant models. Hoek and Brunekreef (1995) did not find a consistent association between ambient O_3 levels and the prevalence or incidence of respiratory symptoms in children living in two rural towns in the Netherlands. Collectively, these studies indicate that there is no consistent evidence of an association between O_3 and respiratory symptoms among healthy children.

7.2.5 Acute Airway Inflammation

Acute airway inflammation has been shown to occur among adults exposed to 80 ppb O_3 over 6.6 hours with exercise in controlled chamber studies (Devlin et al., 1991). Kopp and colleagues (1999) attempted to document inflammation of the upper airways in response to summer season O_3 exposures by following a group of 170 school children in two towns in the German Black Forest from March to October of 1994. To assess inflammation, the investigators collected nasal lavage samples at 11 time points spanning the follow-up period. The nasal lavage samples were analyzed for markers of inflammation, including eosinophil cationic protein, albumin, and leukocyte counts. Subjects who were sensitized to inhaled allergens were excluded. When analyzed across the entire follow-up period, no association was detected between upper airway inflammation and O_3 concentrations. More detailed analysis showed that the first significant O_3 episode of the summer was followed by a rise in eosinophil cationic protein levels, however, subsequent and even higher O_3 episodes had no effect. These findings suggest an adaptive response of inflammation in the nasal airways that is consistent with controlled human studies (see Section 6.9, Effects of Inflammation and Host Defense).

Frischer and colleagues (1993) collected nasal lavage samples from 44 school children in Umkirch, Germany the morning after "low" O_3 days (<140 $\mu g/m^3$ or approximately 72 ppb) and "high" O_3 days (>180 $\mu g/m^3$ or approximately 93 ppb) to measure levels of biochemical markers of inflammation. The researchers found that higher O_3 levels were associated with increased polymorphonuclear leukocyte counts in all children, and increases in myeloperoxydases and eosinophilic cation proteins among children without symptoms of rhinitis (n = 30). These results indicated that O_3 was associated with inflammation in the upper airways. Frischer et al. (1997) further investigated whether hydroxyl radical attacks played a role in mediating the O_3-associated inflammatory response of the airways. O*rtho*- and *para*-tyrosine levels were measured in the nasal lavage samples and the *ortho*/*para* radical ratio was used to determine the generation of hydroxyl radicals. Significant increases in the *ortho*/*para* ratio were observed on days following high ambient O_3 levels. However, the *ortho*/*para* ratio was not related to polymorphonuclear leukocyte counts, suggesting that there was no detectable relationship between hydroxyl radical attacks and the inflammatory response seen in these children. Similar to the study by Kopp et al. (1999), the *ortho*/*para* ratio decreased at the end of the summer although O_3 concentrations were still high, providing additional evidence for a possible adaptive

response. These findings, however, do not preclude the possibility that other unmeasured effects, including cell damage or lower airway responses, may have occurred with ongoing summer season exposures. In fact, a study of joggers repeatedly exposed to O_3 while exercising over the summer in New York City suggested that cell damage may occur in the absence of ongoing inflammation (Kinney et al., 1996b).

In two Mexico City studies by Romieu et al. (1998, 2002), the effect of antioxidant supplements on the association between O_3 and lung function in outdoor workers and asthmatic children was investigated. Romieu and colleagues (1998) observed significant inverse associations between O_3 and lung function parameters, including FVC, FEV_1, and FEF_{25-75} (forced expiratory flow at 25 to 75% of FVC), among outdoor workers who were on the placebo, but not among those taking the antioxidant supplement during the first phase of testing. Likewise, O_3 concentrations were associated with declines in lung function among children with moderate-to-severe asthma who were on the placebo, but no associations were found among those who were taking the vitamin C and E supplement (Romieu et al., 2002). These results indicate that supplementation with antioxidants may modulate the impact of O_3 exposure on the small airways of two potentially at-risk populations, outdoor workers and children with moderate-to-severe asthma. In a further analysis, genetic factors were found to contribute to the variability between individuals in the effects of O_3 on lung function (Romieu et al., 2004). Individuals with polymorphism of the glutathione S-transferase gene (GSTM1 null genotype) lack glutathione transferase enzyme activity, which plays an important role in protecting cells against oxidative damage. Results from this analysis indicate that asthmatic children with GSTM1 null genotype were found to be more susceptible to the impact of O_3 exposure on small airways function. Romieu et al. (2004) noted that supplementation with the antioxidant vitamins C and E above the minimum daily requirement might compensate for the genetic susceptibility.

7.2.6 Acute Ozone Exposure and School Absences

The association between school absenteeism and ambient air pollution was assessed in a few studies (Chen et al., 2000; Gilliland et al., 2001; Park et al., 2002). In the study by Chen and colleagues (2000), daily school absenteeism was examined in 27,793 students (kindergarten to sixth grade) from 57 elementary school students in Washoe County, NV over a two-year period. One major limitation of this study was that the percent of total daily absences was the outcome

of interest, not illness-related absences, as reasons for absences were not noted in all schools. In models adjusting for PM_{10} and CO, ambient O_3 levels were associated with school absenteeism. With a distributed lag of 1 to 14 days, O_3 concentrations were associated with a 10.41% (95% CI: 2.73, 18.09) excess rate of school absences per 40 ppb increase in 1-h max O_3. PM_{10} and CO concentrations also were associated with school absenteeism, however, the effect estimate for PM_{10} was negative. The inverse relationship between O_3 and PM_{10} may have partially attributed to the negative association observed between PM_{10} and school absenteeism.

Ozone-related school absences also were examined in a study of 1,933 fourth grade students from 12 southern California communities participating in the Children's Health Study (Gilliland et al., 2001). Due to its comprehensive characterization of health outcomes, this study is valuable in assessing the effect of O_3 on illness-related school absenteeism in children. The study spanned a period, January through June 1996, that captured a wide range of exposures while staying mostly below the highest levels observed in the summer season. All school absences that occurred during this period were followed up with phone calls to determine whether they were illness-related. For illness-related absences, further questions assessed whether the illness was respiratory or gastrointestinal, with respiratory symptoms including runny nose/sneeze, sore throat, cough, earache, wheezing, or asthma attacks. Multiple pollutants were measured at a central site in each of the 12 communities. The statistical analysis controlled for temporal cycles, day of week, and temperature, and expressed exposure as a distributed lag out to 30 days. Associations were found between the 30-day distributed lag of 8-h avg O_3 (10 a.m.-6 p.m.) and all absence categories. Larger O_3 effects were seen for respiratory causes (147% [95% CI: 6, 478] increase in absences per 30 ppb increase in 8-h avg O_3) than for nonrespiratory causes (61% [95% CI: 9, 138] increase). Among the respiratory absences, larger effects were seen for lower respiratory diseases than for upper respiratory diseases. Multipollutant analyses were not performed; however, in single-pollutant models neither PM_{10} or NO_2 were associated with any respiratory or nonrespiratory illness-related absences. Some concern exists regarding the possibility of residual seasonal confounding given the six-month time span of the monitoring period and the long lag periods of exposure, which are likely to capture seasonally changing factors such as pollen episodes. Further, the biological relevance of O_3 concentrations lagged 30 days present an interpretive challenge.

Park et al. (2002) examined the association between air pollution and school absenteeism in 1,264 students, first to sixth grade, attending school in Seoul, Korea. The study period extended from March 1996 to December 1999, with 8-h avg O_3 concentrations ranging from 3.13 ppb to 69.15 ppb (mean 22.86 ppb). Note that analysis was performed using Poisson GAM with default convergence criteria. Same day O_3 concentrations were positively associated with illness-related absences, but inversely associated with non-illness-related absences. PM_{10} concentrations also were positively associated with illness-related absences. In two-pollutant models containing O_3 and PM_{10}, both estimates were robust, with a slightly greater effect seen for O_3.

Results from Chen et al. (2000), Gilliland et al. (2001), and Park et al. (2002) suggest that ambient O_3 concentrations, on the same day as well as accumulated over two to four weeks, may be associated with school absenteeism, particularly illness-related absences. Further replication is needed before firm conclusions can be reached regarding the effect of O_3 on school absences.

7.2.7 Cardiovascular Endpoints

Several air pollution studies have examined various cardiovascular endpoints (Table AX7-2 in Chapter 7 Annex). The earlier studies focused on PM effects. For a more thorough discussion of these PM studies and their health endpoints, refer to the 2004 PM AQCD (Section 8.3.1). More recently studies have examined associations of O_3 and other gaseous pollutants with various measures of heart beat rhythms in panels of elderly subjects, as discussed below. Other studies examined the increased risk of MI related to air pollutants exposures.

7.2.7.1 Cardiac Autonomic Control

Alterations in heart rate and/or rhythm are thought to reflect pathophysiologic changes that may represent possible mechanisms by which ambient air pollutants such as O_3 may exert acute effects on human health. Decreased HRV has been identified as a predictor of increased cardiovascular morbidity and mortality. Brook et al. (2004) state that HRV, resting heart rate, and blood pressure are modulated by a balance between the two determinants of autonomic tone (the sympathetic and parasympathetic nervous systems). They note that decreased HRV predicts an increased risk of cardiovascular morbidity and mortality in the elderly and those with significant heart disease, which is generally determined by analyses of time (e.g., standard

deviation of normal R-R intervals) and frequency domains (e.g., low frequency/high frequency ratio by power spectral analysis, reflecting autonomic balance) measured during 24 hours of electrocardiography. Decreased parasympathetic input to the heart may provide an important mechanistic link between air pollution and cardiovascular mortality by promoting fatal tachyarrhythmias.

The potentially adverse effects of air pollutants on cardiac autonomic control were examined in a large population-based study, among the first in this field. Liao et al. (2004) investigated short-term associations between ambient pollutants and cardiac autonomic control from the fourth cohort examination (1996-1998) of the population-based Atherosclerosis Risk in Communities Study (ARIC). PM_{10}, O_3, and other gaseous air pollutants were examined in this study. PM_{10} (24-h avg) and O_3 exposures (8-h avg, 10 a.m.-6 p.m.) one day prior to the randomly allocated examination date were used. They calculated 5-minute HRV indices between 8:30 a.m. and 12:30 p.m., and used logarithmically-transformed data on high-frequency (0.15 to 0.40 Hz) and low-frequency (0.04 to 0.15 Hz) power, standard deviation of normal R-R intervals, and mean heart rate. The effective sample sizes for O_3 and PM_{10} were 5,431 and 4,899, respectively, from three U.S. study centers in North Carolina, Minnesota, and Mississippi. PM_{10} concentrations measured one day prior to the HRV measurements were inversely associated with both frequency- and time-domain HRV indices. Ambient O_3 concentrations were inversely associated with high-frequency power among whites. Consistently more pronounced associations were suggested between PM_{10} and HRV among persons with a history of hypertension. Liao et al. note that these findings may represent potentially important arrhythmogenic mechanisms of ambient air pollution. The acute adverse effect of air pollution on cardiac autonomic control hypothesizes that increased air pollution levels may stimulate the autonomic nervous system and lead to an imbalance of cardiac autonomic control characterized by sympathetic activation unopposed by parasympathetic control. Such an imbalance of cardiac autonomic control may predispose susceptible people to greater risk of life-threatening arrhythmias and acute cardiac events. The findings from Liao et al. were cross-sectionally derived from a population-based sample and reflect the short-term effects of air pollution on HRV. When the regression coefficients for each individual pollutant model were compared, the effects for PM_{10} were considerably larger than the effects for gaseous pollutants such as O_3. Because of the population-based sample, this study does have better generalizability than other

smaller panel studies. The findings are suggestive of short-term effects of air pollutants, including O_3, on HRV at the population level.

Another population-based study of air pollutants and HRV was conducted in Boston on 497 men from the VA Normative Aging Study (NAS) (Park et al., 2005). Ozone showed several associations with HRV outcomes. Stronger associations were reported with $PM_{2.5}$. In two-pollutant models, the magnitude of the percent changes for both $PM_{2.5}$ and O_3 diminished slightly. In analyses by ischemic heart disease, hypertension, and diabetes status, stronger associations of HRV with O_3 and $PM_{2.5}$ were observed for individuals with ischemic heart disease and hypertension. These results are consistent with a Mexico City study (n = 34) by Holguín et al. (2003) which reported an HRV effect for O_3 in subjects with hypertension. The association of O_3 exposure with reduced low-frequency power in the full cohort seemed to be driven by subjects not taking calcium-channel blockers (Park et al., 2005). This suggests that this drug is blocking effects of O_3 on the sympathetic pathway. This study cohort consists of all males and almost all whites. This population-based study suggests that short-term exposures to O_3 are predictors of alteration in cardiac autonomic function as measured by HRV among older male adults.

Two related studies in Boston, MA, examined the association between air pollution and the incidence of ventricular arrhythmias (Dockery et al., 2005; Rich et al., 2005). A total of 203 patients with implanted cardioverter defibrillators who lived within 25 miles of the ambient monitoring site at the Harvard School of Public Health were monitored. They had a total of 635 person-years of follow-up or an average of 3.1 years per subject. In the analysis by Dockery et al. (2005), positive associations were observed between ventricular arrhythmias within three days of a prior event and a two-day mean of several air pollutants, including $PM_{2.5}$, black carbon, NO_2, CO, and SO_2. No associations were observed with O_3. There was, however, a suggestion of increasing risk with increasing quintiles of O_3 ($p < 0.05$). The analysis by Rich et al. (2005) observed stronger O_3 effects on ventricular arrhythmias using a case-crossover study design. Case periods were defined by the time each arrhythmic event began; for each case, three to four control periods were selected by matching on weekday and hour of the day within the same calender month. For a 20 ppb increase in 24-hour moving average O_3, a 27% (95% CI: 0, 60) increased risk of ventricular arrhythmias was estimated. Significant effects also were found for $PM_{2.5}$, NO_2, and SO_2. In two-pollutant models, the O_3 effect was found to be generally

robust. Stratified analysis by the presence of a recent ventricular arrhythmia within the previous three days indicated that O_3 was associated with increased risk among subjects without a recent event (37% [95% CI: 6, 79]), but not among those with recent events (5% [95% CI: −27, 49]). Rich et al. explained that the use of the case-crossover study design and conditional analysis might have contributed to the stronger associations observed in their study compared to Dockery et al. In addition, the use of a 24-hour moving average instead of a calender-day air pollution concentration might have reduced exposure misclassification, resulting in larger effect estimates.

Other studies do not provide evidence for an O_3 effect on HRV and cardiac arrhythmias (Peters et al., 2000a; Rich et al., 2004; Vedal et al., 2004). These studies, however, may have had limited power to examine subtle effects. Gold et al. (2000; reanalysis Gold et al., 2003) reported results that suggest that O_3 exposure may decrease vagal tone, leading to reduced HRV. Schwartz et al. (2005) reported a weak association of O_3 with the root mean squared differences between adjacent R-R intervals in a study of 28 elderly subjects and noted that lack of personal exposure measurements may render such studies less able to assess autonomic functions. This study reported the strongest effects for black carbon.

7.2.7.2 Acute Myocardial Infarction

The effect of O_3 on the incidence of MI was examined in a limited number of studies. Acute MI was studied in relation to air pollution in Toulouse, France based on the existence of an acute MI registry (Monitoring Trend and Determinants in Cardiovascular Disease [MONICA]) and an air quality network covering the same population (Ruidavets et al., 2005). After adjustment for temperature, relative humidity, and influenza epidemics, the relative risk of acute MI occurrence was 1.76 (95% CI: 1.12, 2.45) for current day O_3 concentrations. The increased risk of MI was more evident in the oldest group, 55 to 64 years of age. Further, the oldest subjects without a personal history of ischemic heart disease were more susceptible to an acute event when O_3 levels increased. No PM data was reported in this study.

In a case-crossover study (n = 772) in Boston, MA, Peters et al. (2001) reported an odds ratio of 1.27 (95% CI: 0.87, 1.88) per 40 ppb increase in 2-h avg O_3 (1 hour before onset of event). Stronger effects on the incidence of MI were observed for $PM_{2.5}$ and PM_{10}.

7.2.7.3 Cardiovascular Endpoints in Human Clinical Studies

In a controlled human exposure study discussed in Chapter 6, Sections 6.3.4 and 6.10, Gong et al. (1998a) studied 10 nonmedicated hypertensive and 6 healthy male adults exposed to 0.3 ppm O_3 with intermittent exercise in relation to various cardiovascular effects. The overall results did not indicate acute cardiovascular effects of O_3 in either the hypertensive or control subjects. The authors observed an increase in rate-pressure product and heart rate, a decrement for FEV_1, and a >10 mm Hg increase in the alveolar/arterial pressure difference for O_2 following O_3 exposure. These findings suggest that O_3 can exert cardiovascular effects indirectly by impairing alveolar-arterial O_2 transfer and potentially reducing O_2 supply to the myocardium. Ozone exposure may increase myocardial work and impair pulmonary gas exchange to a degree that may be clinically important in persons with significant pre-existing cardiovascular impairment.

7.2.7.4 Summary of Field Studies with Cardiovascular Outcomes

A limited epidemiologic database examining cardiovascular outcomes in relation to O_3 exposures is available. Among these studies, three were population-based and involved cohorts such as the ARIC (Liao et al., 2004), MONICA (Ruidavets et al., 2005), and NAS (Park et al., 2005). Such studies may offer more informative results based on their large subject-pool and design. Results from these three studies were suggestive of an association between O_3 exposure and the cardiovascular endpoints studied. As in the case of respiratory disease outcomes, Brook et al. (2004) state that the increase in relative risk for cardiovascular disease due to air pollution is small compared with the impact of the established cardiovascular risk factors. However, because of the enormous number of people affected, even conservative risk estimates can translate into a substantial increase in mortality due to cardiovascular disease within the population. The impact of air pollution on cardiovascular disease therefore may represent a serious public health problem.

7.2.8 Summary of Field Studies Assessing Acute Ozone Effects

- Results from recent field/panel studies support the evidence from clinical studies that acute O_3 exposure is associated with a significant effect on lung function, as indicated by decrements in FEV_1, FVC, and PEF. The declines in lung function were noted particularly in children and asthmatics.

- Limited evidence suggests that more time spent outdoors, higher levels of exertion, and the related increase in O_3 exposure may potentiate the risk of respiratory effects. In addition to children and asthmatics, adults who work or exercise outdoors may be particularly vulnerable to O_3-associated health effects.

- Many new studies have examined the association between O_3 concentrations and a wide variety of respiratory symptoms (e.g., cough, wheeze, production of phlegm, and shortness of breath). Collectively, the results suggest that acute exposure to O_3 is associated with increased respiratory symptoms and increased as-needed medication use in asthmatic children.

- Additional panel studies investigated the effect of O_3 on other health outcomes, including school absences, and markers of inflammation and oxidative damage. Ozone exposure was associated with increases in respiratory-related school absences, as well as increased inflammation and generation of hydroxyl radicals in the upper airways. Use of antioxidant supplements was found to diminish the O_3 effect on lung function.

- Some field studies have examined the association between O_3 and cardiac physiologic outcomes. The current evidence is rather limited but supportive of a potential effect on HRV, ventricular arrhythmias, and the incidence of MI. Additional studies need to be performed before any conclusions can be made regarding an O_3 effect on cardiovascular outcomes.

7.3 ACUTE EFFECTS OF OZONE ON DAILY EMERGENCY DEPARTMENT VISITS AND HOSPITAL ADMISSIONS

7.3.1 Summary of Key Findings on Studies of Emergency Department Visits and Hospital Admissions from the 1996 O_3 AQCD

In the 1996 O_3 AQCD, aggregate population time-series studies of O_3-related health effects provided relevant evidence of acute responses, even below a 1-h max O_3 of 0.12 ppm. Emergency room visits and hospital admissions were examined as possible outcomes following exposure to O_3. In the case of emergency room visits, the evidence was limited (Bates et al., 1990; Cody et al., 1992; Weisel et al., 1995; White et al., 1994), but results generally indicated an O_3 effect on morbidity. The strongest and most consistent evidence of O_3 effects, at levels both above and below 1-h max O_3 levels of 0.12 ppm, was provided by the multiple studies that had been conducted on summertime daily hospital admissions for respiratory causes in various locales in eastern North America (Bates and Sizto, 1983, 1987, 1989; Burnett et al., 1994;

Lipfert and Hammerstrom, 1992; Thurston et al., 1992, 1994). These studies consistently demonstrated that O_3 air pollution was associated with increased hospital admissions, accounting for roughly one to three excess respiratory hospital admissions per million persons with each 100 ppb increase in 1-h max O_3. This association had been shown to remain even after statistically controlling for the possible confounding effects of temperature and copollutants (e.g., H^+, SO_4^{-2}, PM_{10}), as well as when considering only days with 1-h max O_3 concentrations below 0.12 ppm. Overall, the aggregate population time-series studies considered in the 1996 O_3 AQCD provided strong evidence that ambient exposures to O_3 can cause significant exacerbations of preexisting respiratory disease in the general public.

7.3.2 Review of Recent Studies of Emergency Department Visits for Respiratory Diseases

Emergency department visits represent an important acute outcome that may be affected by O_3 exposures. Morbidities that result in emergency department visits are closely related to, but are generally less severe than, those that result in unscheduled hospital admissions. In many cases, acute health problems are successfully treated in the emergency department; a subset of more severe cases that present initially to the emergency department may require admission to the hospital.

Several studies have been published in the past decade examining the temporal associations between O_3 exposures and emergency department visits for respiratory diseases (Table AX7-3 in Chapter 7 Annex). Total respiratory causes for emergency room visits may include asthma, pneumonia, bronchitis, emphysema, upper and lower respiratory infections such as influenza, and a few other minor categories. Asthma visits typically dominate the daily incidence counts. Chronic bronchitis and emphysema often are combined to define COPD, which is a prominent diagnosis among older adults with lung disease. Figure 7-8 presents percent changes in emergency department visits for asthma from single-pollutant models, with results expressed in standardized increments. The lags presented in the figure vary depending on reported results. Most studies reported effect estimates from a short lag period (0 to 2 days). Results from Weisel et al. (2002) are not included as comparable risks estimates for O_3 are not presented. Among the U.S. studies, there was one multicity study which examined three cities in Ohio (Jaffe et al., 2003). Several presented Atlanta, GA data. In general, O_3

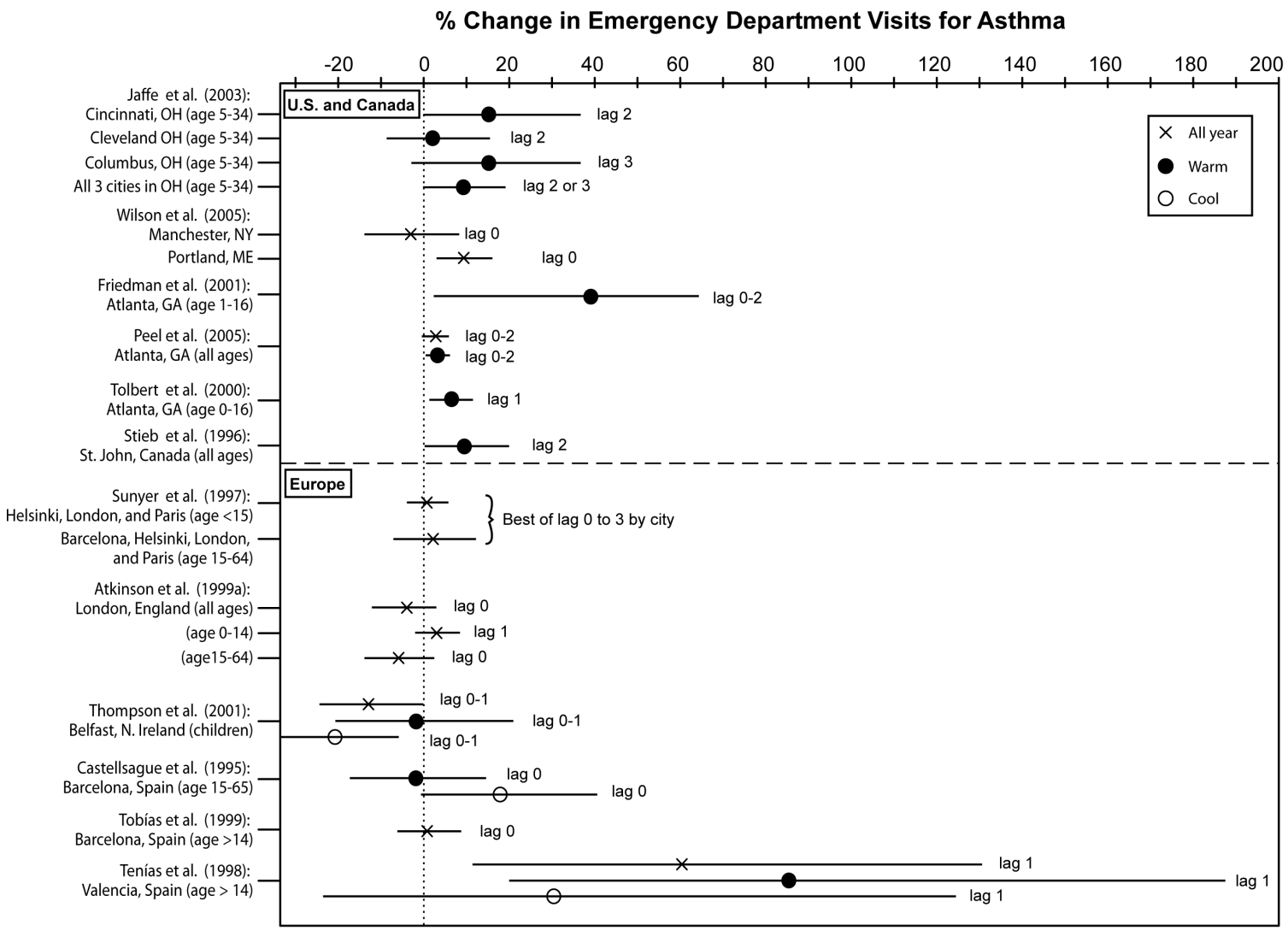


Figure 7-8. Ozone-associated percent change (95% CI) in emergency department visits for asthma per standardized increment (see Section 7.1.3.2).

effect estimates from warm season only analyses tended to be positive and larger compared to results from cool season or all year analyses.

Among studies with adequate controls for seasonal patterns, many reported at least one positive association with O_3. These studies examined emergency department visits for total respiratory complaints (Delfino et al., 1997b, 1998b; Herñandez-Gardûno et al., 1997; Ilabaca et al., 1999; Jones et al., 1995; Lin et al., 1999), asthma (Friedman et al., 2001; Jaffe et al., 2003; Stieb et al., 1996; Tenías et al., 1998; Tobías et al., 1999; Tolbert et al., 2000; Weisel et al., 2002), and COPD (Tenías et al., 2002).

One recent study examined emergency department visits for total and cause-specific respiratory diseases in Atlanta, GA over an 8-year period (Peel et al., 2005). A distributed lag of 0 to 2 days was specified a priori. Ozone concentrations were associated with emergency department visits for total respiratory diseases and upper respiratory infections in all ages. A marginally significant association was observed with asthma visits (2.6% [95% CI: −0.5, 5.9] excess risk per 30 ppb increase in 8-h max O_3), which became stronger when analysis was restricted to the warm months (3.1% [95% CI: 0.2, 6.2] excess risk). In multipollutant models adjusting for PM_{10}, NO_2 and CO, O_3 was the only pollutant that remained significantly associated with upper respiratory infections. Another large asthma emergency department study was carried out during the months of May through September from 1984 to 1992 in St. John, New Brunswick, Canada (Stieb et al., 1996). Effects were examined separately among children aged less than 15 years and in persons aged 15 years and older. A significant effect of O_3 on emergency department visits was reported among persons 15 years and older. There was suggestion of a threshold somewhere in the range below a 1-h max O_3 of 75 ppb. A study in Valencia, Spain from 1994 to 1995 observed that emergency room visits for asthma among persons over 14 years old were robustly associated with relatively low O_3 levels (median 1-h max O_3 of 62.8 $\mu g/m^3$ or approximately 32.4 ppb) (Tenías et al., 1998). The excess risk of asthma emergency room visits was larger in the warm season (May to October), 85% (95% CI: 20, 188) excess risk per 40 ppb increase in 1-h max O_3, compared to the cool season (November-April), 31% (95% CI: −24, 125) excess risk (Tenías et al., 1998).

Among the studies that observed a positive association between O_3 and emergency department visits for respiratory outcomes, O_3 effects were found to be robust to adjustment for PM_{10}, NO_2, SO_2, and BS (Lin et al., 1999; Peel et al., 2005; Tenías et al., 1998). One study by

Tolbert and colleagues (2000) observed that the significant univariate effects of both O_3 and PM_{10} on pediatric asthma emergency department visits in Atlanta, GA became nonsignificant in two-pollutant regressions, reflecting the high correlation between the two pollutants (r = 0.75).

For several other studies with total respiratory and asthma outcomes, inconsistencies confound an interpretation of likely causal effects. For example, in a Montreal, Canada study, O_3 effects on total respiratory emergency department visits were seen in a short data series from the summer of 1993 but not in a similar data series from the summer of 1992 (Delfino et al., 1997b). The significant 1993 results were seen only for persons older than 64 years. A very similar analysis of two additional summers (1989 and 1990) revealed an O_3 association only for 1989 and again only in persons over 64 years old (Delfino et al., 1998b). An analysis of data on respiratory emergency department visits from June to August of 1990 in Baton Rouge, LA reported O_3 effects in adults, but not in children or among the elderly (Jones et al., 1995).

Tobías and colleagues (1999) showed that regression results for asthma emergency department visits could be quite sensitive to methods used to control for asthma epidemics. Ozone was associated with the outcome variable in only one of eight models tested. An Atlanta, GA study by Zhu et al. (2003) examined asthma emergency department visits in children during three summers using Bayesian hierarchical modeling to address model variability. Data were analyzed at the zip code level to account for spatially misaligned longitudinal data. Results indicated a positive, but nonsignificant relationship between O_3 and emergency room visits for asthma.

Other studies also reported no association between O_3 and emergency department visits for respiratory causes (Atkinson et al., 1999a; Castellsague et al., 1995; Chew et al., 1999; Hwang and Chan, 2002; Sunyer et al., 1997). Using Bayesian hierarchical modeling, Hwang and Chan (2002) examined the effect of air pollutants on daily clinic visits for lower respiratory illnesses across 50 cities in Taiwan. All pollutants except O_3 were associated with daily clinic visits. In a pooled analysis of emergency admissions for asthma in four European cities as part of the Air Pollution on Health: European Approach (APHEA) study, there was no overall effect of O_3 observed (Sunyer et al., 1997). Atkinson et al. (1999a) in London, England also did not find an association between O_3 and emergency department visits at a mean 8-h max O_3 concentration of 17.5 ppb. One study by Thompson and colleagues (2001) in Belfast, Northern Ireland observed a decreased risk of childhood asthma admissions (−21% [95% CI: −33, −6] per 20 ppb increase

in 24-h avg O_3) in the cold season (November-April). After adjusting for benzene levels, O_3 was no longer associated with asthma emergency department visits. The inverse relationship of O_3 with benzene concentrations ($r = -0.65$), and perhaps with other pollutants, might have produced the apparent protective effect of O_3. No significant O_3 effect was found in the warm season (May-October). The O_3 levels were low in both seasons, with a mean 24-h avg O_3 concentration of 18.7 ppb in the warm season and 17.1 ppb in the cold season. A study by Hajat et al. (1999, 2002) of physician consultations for asthma, lower respiratory diseases, and upper respiratory diseases in London reported negative associations with O_3, which was suggestive of residual confounding by copollutants or weather factors (note that data were analyzed using Poisson GAM with default convergence criteria). Several other emergency department studies looking at O_3 are more difficult to interpret due to inadequate control for seasonal patterns, very low O_3 levels, or because no quantitative results were shown for O_3 (Buchdahl et al., 1996, 2000; Garty et al., 1998; Holmén et al., 1997; Lierl and Hornung, 2003; Lipsett et al., 1997; Nutman et al., 1998).

Although several studies found a significant association between O_3 concentrations and emergency department visits for respiratory causes, some inconsistencies were observed. The inconsistencies may be attributable, at least partially, to differences in model specifications and analysis approach among the various studies. For example, ambient O_3 concentrations, length of the study period, and statistical methods used to control confounding by seasonal patterns and copollutants appear to affect the observed O_3 effect on emergency department visits. The body of evidence remains inconclusive regarding effects of O_3 on the risk of emergency department visits.

7.3.3 Studies of Hospital Admissions for Respiratory Diseases

Hospital admissions represent a medical response to a serious degree of morbidity for a particular disease. Scheduled hospitalizations are planned in advance when a particular clinical treatment is needed. However, unscheduled admissions are ones that occur in response to unanticipated disease exacerbations and are more likely to be affected by environmental factors, such as air pollution. As such, the hospital admissions studies reviewed here focused specifically on unscheduled admissions. Study details and results from hospital admissions studies published over the past decade are summarized in Table AX7-4 (in the Chapter 7

Annex). As a group, these hospitalization studies tend to be larger in terms of geographic and temporal coverage, and indicate results that are generally more consistent than those reviewed above for emergency department visits. As in the case for all studies that examine changes in aggregate measures of acute disease outcomes over time, the following should be considered in comparing results: (1) difference in types of respiratory diseases for hospital admission; (2) age of study population; (3) mean level of O_3 during study; (4) single-city versus multicity studies; (5) length of study (e.g., <5 years versus >5 years); (6) analysis by season versus all year; (7) O_3-only versus multipollutant models; (8) number of exposure lag days; and (9) type of study (e.g., case-crossover versus time-series). These factors are considered in the sections below with further discussion on potential confounding of the O_3 effect estimate by seasonal factors and copollutants.

7.3.3.1 All Year and Seasonal Effects of Ozone on Respiratory Hospitalizations

The effect of O_3 on respiratory hospitalizations was examined in various studies conducted in the U.S. and abroad. Figures 7-9 and 7-10 present risk estimates from all total respiratory hospital admission studies. Burnett et al. (1995), which did not present quantitative results for O_3, and Yang et al. (2003), which only presented odd ratios, were not included in the figures. In cases where multiple lags were presented, the multiday lag was selected to represent the cumulative effect from all days examined. If only single-day lags were analyzed, the effect estimate of the shortest lag time, usually a lag of 0 or 1 day, was presented. Figure 7-9 plots the effect estimates and 95% CIs from 15 studies that analyzed all year data. The risk estimates are arranged by age groups. The preponderance of positive risk estimates, with some that are statistically significant, is readily apparent. Figure 7-10 presents the season-stratified effect estimates by region. For studies that reported risk estimates from all four seasons, only the summer and winter estimates are presented. It appears that the warm season estimates, collectively, tend to be larger, positive values compared to all year and cool season estimates. All of the negative estimates were from analyses using cool season data only, which might reflect the inverse correlation between O_3 and copollutants, namely PM, during that season. These studies are discussed below in further detail.

Among the respiratory hospitalization studies, the most robust and informative results were observed when a broad geographic area was examined using a consistent analytical methodology

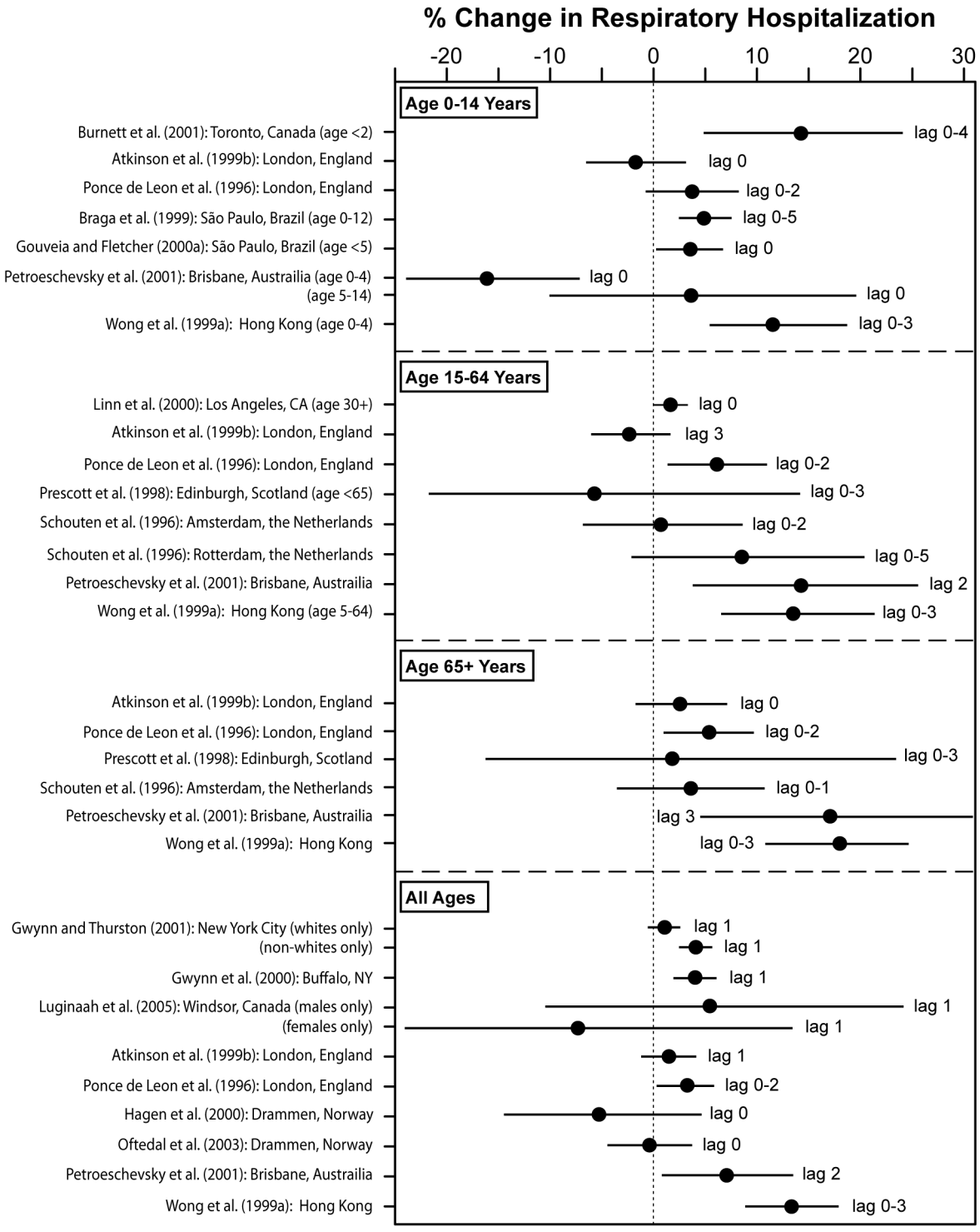


Figure 7-9. Ozone-associated percent change (95% CI) in total respiratory hospitalizations <u>for all year analyses</u> per standardized increment (see Section 7.1.3.2). Effect estimates are arranged by age groups.

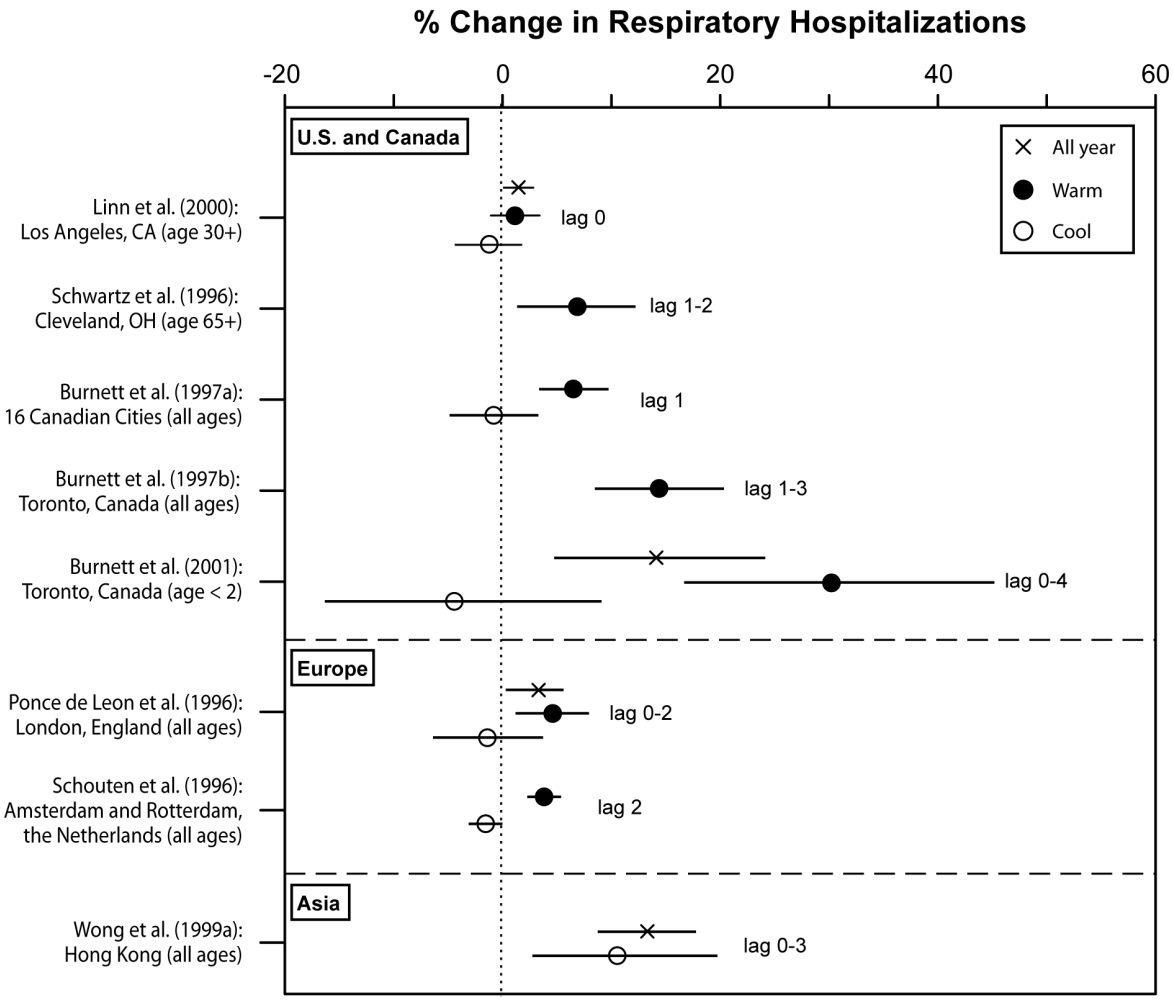


Figure 7-10. Ozone-associated percent change (95% CI) in total respiratory hospitalizations by season per standardized increment (see Section 7.1.3.2).

(Anderson et al., 1997; Burnett et al., 1995, 1997a). These studies have all reported an O_3 effect on respiratory hospital admissions. The largest such study to-date was carried out using data on all-age respiratory hospital admissions from 16 Canadian cities with populations exceeding 100,000 during the period 1981 to 1991 (Burnett et al., 1997a). In addition to O_3, the authors evaluated health effects of SO_2, NO_2, CO, and coefficient of haze (a surrogate for black carbon particle concentrations). Pooling the 16 cities, a positive association was observed between respiratory hospital admissions and the 1-day lag O_3 concentration in the spring (5.6% [95%CI:

1.6, 9.9] excess risk per 40 ppb increase in 1-h max O_3) and summer (6.7% [95%CI: 3.5, 10.0]). The results for fall were also positive, though of smaller magnitude (3.8% [95%CI: −0.2, 7.9]). There was no evidence for an O_3 effect in the winter season (−0.8% [95%CI: −4.8, 3.3]). Control outcomes related to blood, nervous system, digestive system, and genitourinary system disorders were not associated with O_3. In a previous study focused mainly on evaluating health impacts of sulfate particles, Burnett and colleagues (1995) reported results from a time-series analysis of all-age respiratory hospital admissions to 168 hospitals in Ontario, Canada over a 6-year period (1983 to 1988). The outcome data were prefiltered to remove seasonal variations using a weighted 19-day moving average. The authors reported that O_3 was associated with respiratory hospital admissions; however, no quantitative results for O_3 were presented.

Results from an analysis of five European cities indicated strong and consistent O_3 effects on unscheduled hospital admissions for COPD (Anderson et al., 1997). The five cities examined — London, Paris, Amsterdam, Rotterdam, and Barcelona — were among those included in the multicity APHEA study. The number of years of available data varied from 5 to 13 years among the cities. City-specific effect estimates were pooled across cities using weighted means. An association with O_3 was observed in full year analyses. Season-stratified analyses indicated that the O_3 effect was larger in the warm season (April-September), 4.7% (95% CI: 1.6, 7.9) excess risk per 40 ppb increase in 1-h max O_3, compared to the cool season (October-March), 1.6% (95% CI: −3.1, 7.9) excess risk. There was no significant heterogeneity in O_3 effects among the cities.

Several additional studies carried out in one or two cities over a span of five or more years provided substantial additional evidence regarding O_3 effects on respiratory hospital admissions (Anderson et al., 1998; Burnett et al., 1999, 2001; Moolgavkar et al.,1997; Petroeschevsky et al., 2001; Ponce de Leon et al., 1996; Sheppard et al., 1999 [reanalysis Sheppard, 2003]; Yang et al., 2003). Moolgavkar and colleagues (1997) reported significant and robust O_3 effects on respiratory hospital admissions in adults 65 years and older in Minneapolis and St. Paul, MN, but not in Birmingham, AL. The absence of effects in the southern city may reflect less penetration of O_3 into the indoor environment due to greater use of air conditioning, and thus less correlation between central site O_3 monitoring and actual exposures of the urban populace. Ozone effects on all-age and age-stratified asthma and total respiratory hospital admissions were observed in Brisbane, Australia (Petroeschevsky et al., 2001). Effect sizes appeared consistent

in the warm and cool seasons (data not provided). Petroeschevsky et al. commented that the year-round effect of O_3 might reflect the relatively small degree of seasonal variation in O_3 levels observed in Brisbane. Although O_3 levels were quite low year-round, they did not notably decline during the winter period. The authors also noted that given the subtropical climate in Brisbane, characterized by warm, dry winters, perhaps the proportion of the population exposed to winter O_3 concentrations was higher than in cities where inclement winter weather might force populations indoors.

Another set of studies have examined associations between O_3 and respiratory hospitalizations in single cities over shorter (<5 years) time spans. Positive and significant O_3 effects were reported in Cleveland, OH (Schwartz et al., 1996); New York City (Gwynn and Thurston, 2001); Northern New Jersey (Weisel et al., 2002); Toronto, Canada (Burnett et al., 1997b); Helsinki, Finland (Pönkä and Virtanen, 1996); São Paulo, Brazil (Braga et al., 1999; Gouveia and Fletcher, 2000a); and Hong Kong (Wong et al., 1999a). The Helsinki study reported significant effects of O_3 on both asthma and on digestive disorders in a setting of very low O_3 concentrations (Pönkä and Virtanen, 1996), which raises questions of plausibility.

Less consistent effects of O_3 were seen in other respiratory hospitalization studies (Schouten et al., 1996; Lin et al., 2003; Lin et al., 2004; Morgan et al., 1998a; Oftedal et al., 2003). In a study conducted in Amsterdam and Rotterdam, the Netherlands, associations between O_3 and respiratory admissions were observed; however, results were difficult to interpret due to the large number of statistical tests performed (Schouten et al., 1996). In a California study by Neidell (2004), a negative association was observed between hospitalizations for asthma and naturally occurring seasonal variations in O_3 within zip codes in children aged 0 to 18 years. However, the O_3 effect was found to be influenced by socioeconomic status. Among children of low socioeconomic status, O_3 generally was associated with increased hospitalizations, with statistical significance reached in certain age groups. Neidell further stated that avoidance behavior on high O_3 days may have attributed to the negative relationship observed in children of higher socioeconomic status.

No associations between respiratory hospital admissions and O_3 were seen in studies from Los Angeles, CA (Linn et al., 2000; Mann et al., 2002; Nauenberg and Basu, 1999); Vancouver, Canada (Lin et al., 2004); London, England (Atkinson et al., 1999b); Edinburgh, Scotland (Prescott et al., 1998); and Drammen, Norway (Hagen et al., 2000). Several of these studies

were carried out in locations with low O_3 levels, suggestive of a nonlinear concentration-response relationship (Lin et al., 2004; Prescott et al., 1998). The nonsignificant findings in the South Coast air basin, CA area are surprising given the elevated O_3 concentrations observed there (Mann et al., 2002). Inadequate control of seasonal confounding may underlie some of the nonsignificant and negative findings. An additional factor likely contributing to the variability of results is the relatively small sample sizes included in some of these studies.

For respiratory hospitalization outcomes, the largest, most significant associations with O_3 concentrations were observed when using short lag periods, in particular a 0-day lag (exposure on same day) and a 1-day lag (exposure on previous day). In a study of 16 Canadian cities by Burnett et al. (1997a), the strongest association between O_3 and respiratory hospitalizations was found at a 1-day lag. A decline in the magnitude and significance of the effect was seen with increasing days lagged for O_3. Anderson et al. (1997) investigated the association between O_3 and daily hospital admissions for COPD in five European cities. Lags up to 5 days were examined, and the largest risk estimates were found using 0- and 1-day lags. These results suggest that O_3 has a short-term effect on respiratory hospitalizations.

Burnett et al. (2001) investigated the association between respiratory hospitalizations and O_3 in children less than 2 years of age. Lags up to five days were examined after stratifying by season (Figure 7-11). In the summer season, significant associations between O_3 and daily admissions were found in several of the lags, with the largest risk estimate of 12.5% (95% CI: 5.7, 19.7) excess risk per 40 ppb increase in 1-h max O_3 at a 1-day lag. In comparison, the O_3-related risk estimate was 30.2% (95% CI: 18.0, 42.4) using a cumulative lag period of 5 days. The large effect estimate for the cumulative lag period indicated that O_3 exposure likely had an immediate effect that persisted over several days.

Weisel et al. (2002) stated that a lag period of 1 to 3 days between exposure to O_3 and hospital admissions or emergency department visits for asthma was plausible because it might take time for the disease to progress to the most serious responses following exposure. In addition, taking medication could delay further the progression of the adverse effect. Thus, although strongest associations are found at lags of 0 and 1 days, examining longer single-day lag periods or multiday lag periods may further enhance understanding of the effect of O_3 on hospitalizations.

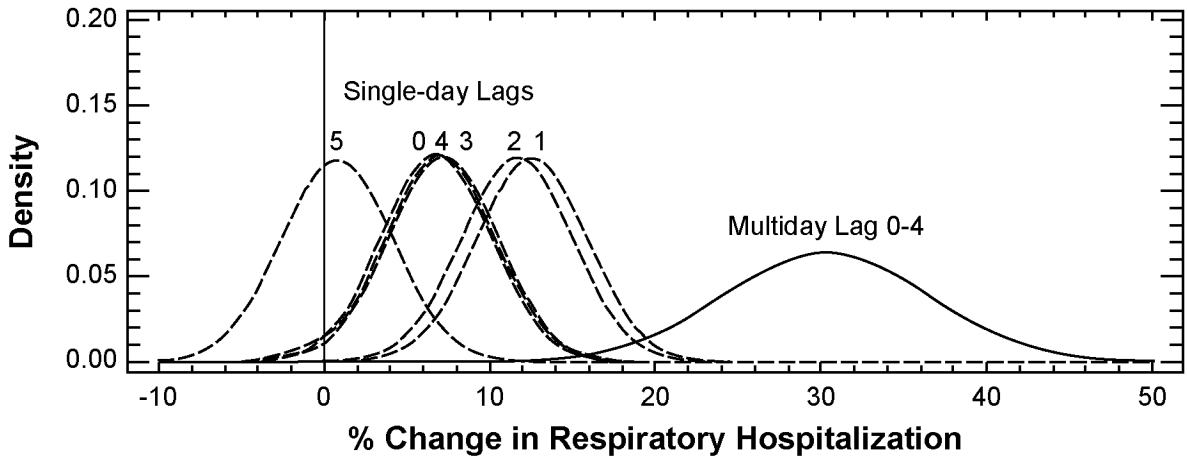


Figure 7-11. Comparison of single-day lags (0-, 1-, 2-, 3-, 4-, and 5-day) to a cumulative multiday lag (0- to 4-day) for percent changes in total respiratory hospitalizations per 40 ppb increase in 1-h max O_3 in children less than two years of age.

Source: Derived from Burnett et al. (2001).

In conclusion, while some inconsistencies are noted across studies, the evidence supports the findings of significant and robust effects of O_3 on various respiratory disease hospitalization outcomes. Large multicity studies, as well as many studies from individual cities have reported significant O_3 associations with total respiratory hospitalizations, asthma, and COPD, especially in studies analyzing the O_3 effect during the summer or warm season.

7.3.3.1 Potential Confounding of the Ozone Effect on Respiratory Hospitalizations by Copollutants

As in the case for most air pollution studies, potential confounding of the association between O_3 and respiratory hospitalizations by copollutants generally was examined using multipollutant regression models. Figure 7-12 compares the risk estimates from models with and without adjustment for PM indices. This figure indicates that O_3 risk estimates are fairly robust to PM adjustment in all year and warm season only data. None of the studies examined PM-adjusted O_3 risk estimates in cool season only data.

Several analyses of a large data set from Toronto, Canada spanning the years 1980 to 1994 reported O_3 effects on respiratory hospitalizations for all ages (Burnett et al., 1997b, 1999) and

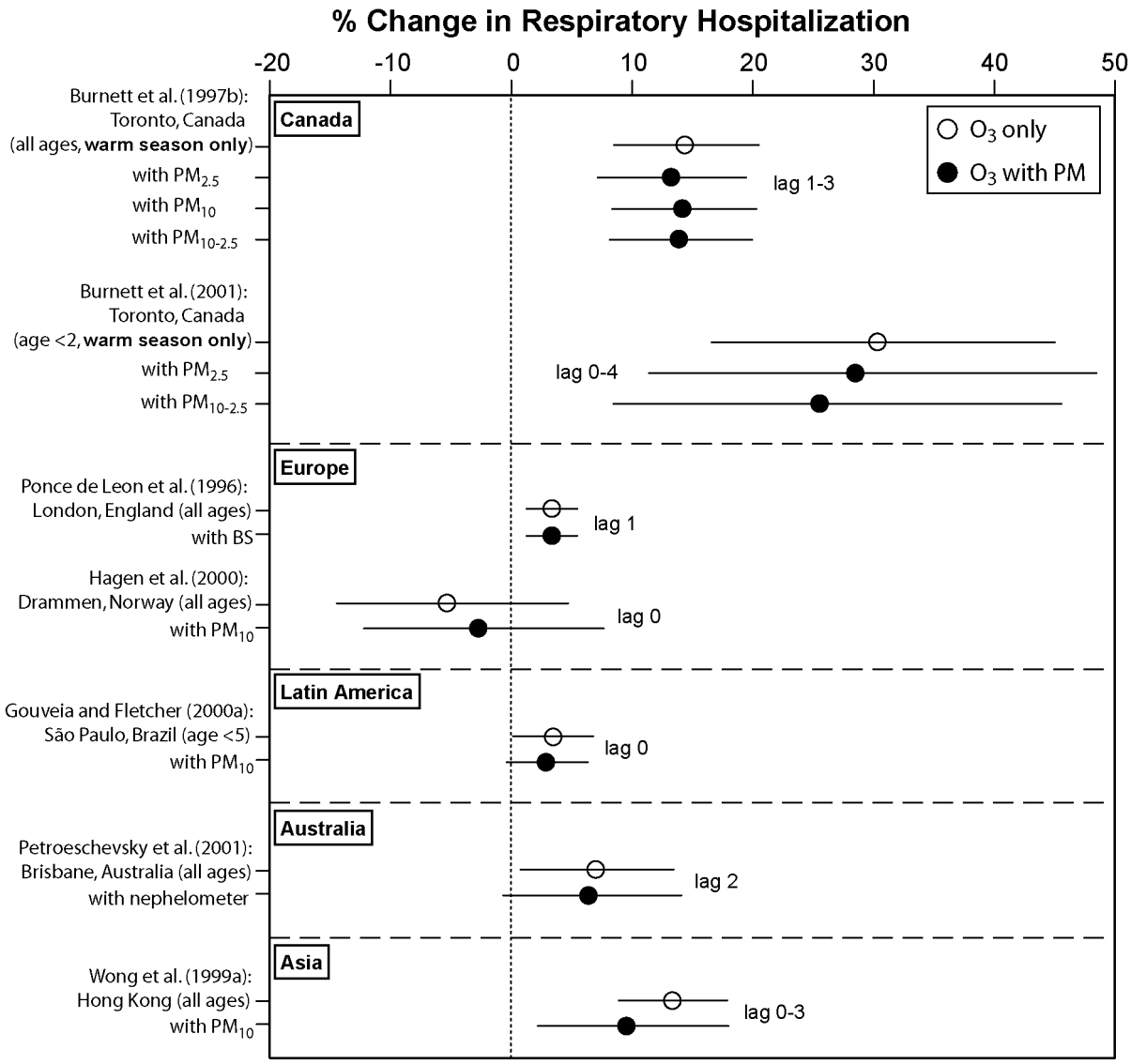


Figure 7-12. Ozone-associated percent change (95% CI) in total respiratory hospitalizations with adjustment for PM indices per standardized increment (see Section 7.1.3.2). Analyses performed using all year data unless noted otherwise.

for persons under the age of 2 years (Burnett et al., 2001). In the 1999 and 2001 studies, analyses were performed using Poisson GAM (default convergence criteria) with a nonparametric LOESS prefilter applied to the pollution and hospitalization data. All studies demonstrated that O_3 effects were robust when adjusting for PM indices, whereas PM effects

from single-pollutant models were markedly attenuated when O_3 was added to the regression. These results imply more robust associations with respiratory hospitalizations for O_3 than PM.

Results from the APHEA study indicated strong and consistent O_3 effects on unscheduled hospital admissions for COPD (Anderson et al., 1997). Significant effects also were seen for BS, TSP, and NO_2. The authors reported that among all pollutants examined, the most consistent and significant findings were for O_3. No two-pollutant model results were reported. Several additional studies also observed that there was no substantial difference in the O_3 effect after adjusting for PM in the regression model (Gouveia and Fletcher, 2000a; Petroeschevsky et al., 2001; Ponce de Leon et al., 1996).

Collectively, these results suggest that copollutants generally do not confound the association between O_3 and respiratory hospitalizations. Ozone risk estimates were robust to PM adjustment in all year and warm season only data.

7.3.4 Association of Ozone with Hospital Admissions for Cardiovascular Disease

A subset of hospital admissions studies have examined the association of O_3 with cardiovascular outcomes (see Figure 7-13). Several have found negative or inconsistent associations (Ballester et al., 2001; Burnett et al., 1999; Fung et al., 2005; Koken et al., 2003; Linn et al., 2000; Mann et al., 2002; Morgan et al., 1998a; Petroeschevsky et al., 2001; Poloniecki et al., 1997; Prescott et al., 1998). Other studies, especially those that examined the relationship when O_3 exposures were higher, have observed robust positive associations between O_3 and cardiovascular hospitalizations (Atkinson et al., 1999b; Burnett et al., 1997b; Chang et al., 2005; Tsai et al., 2003a; Wong et al., 1999a,b; Yang et al., 2004a). In Toronto, Canada, Burnett et al. (1997b) reported a positive association between O_3 and cardiovascular hospital admissions in a summer-only analysis. The results were robust to adjustment for various PM indices, while the PM effects diminished when adjusting for gaseous pollutants. Other studies stratified their analysis by temperature, warms days ($\geq$20 °C) versus cool days (<20 °C). The analysis using warms days consistently produced positive associations (Chang et al., 2005; Tsai et al., 2003a; Yang et al., 2004a). In two studies conducted in Hong Kong, total cardiovascular as well as circulatory, ischemic heart disease, and heart failure were all significantly associated with O_3 in the cool but not the warm season (Wong et al., 1999a,b).

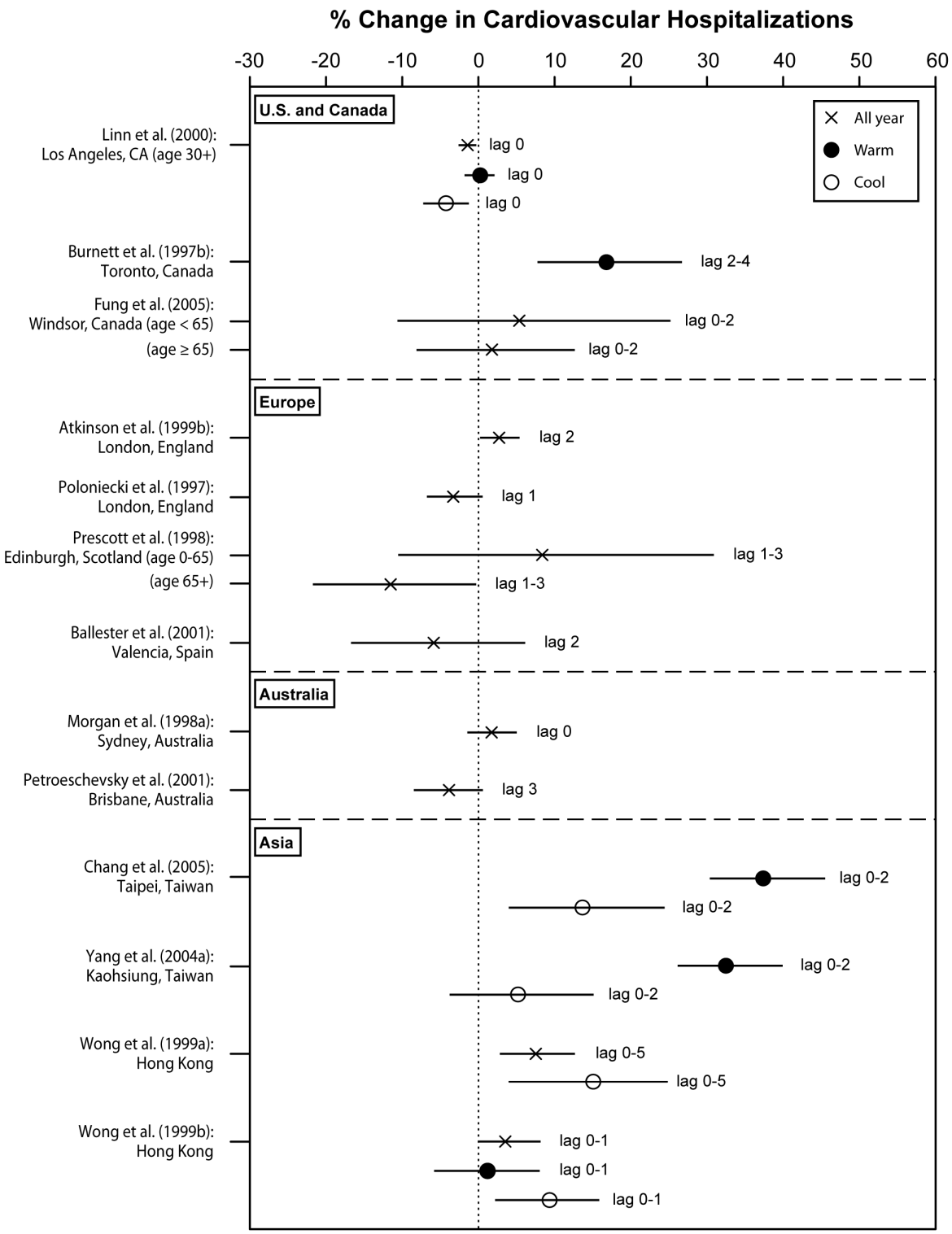


Figure 7-13. Ozone-associated percent change (95% CI) in total cardiovascular hospitalizations per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted.

In Wong et al. (1999b), O_3 concentrations were similar in both seasons, with warm season levels slightly lower, mean 8-h avg O_3 concentrations of 31.2 μg/m^3 (or 16.1 ppb), compared to the cool season, mean 34.8 μg/m^3 (or 18.0 ppb). The authors speculated that differing activity patterns and home ventilation factors may have contributed to the seasonal differences in O_3 effects. Weather in Hong Kong is mild throughout the year, but less humid and cloudy in the cool season. Thus, during the cool season people are more likely to open windows or stay outdoors, resulting in higher personal exposures even with similar ambient concentrations.

Among the growing group of hospitalization studies that examined the effect of O_3 on cardiovascular admissions, several have found inconsistent associations, especially for all year analyses. However, in studies that stratified analyses by seasonal or meteorological factors, evidence is suggestive of an association between O_3 and cardiovascular hospitalizations.

7.3.5 Summary of Acute Ozone Effects on Daily Emergency Department Visits and Hospital Admissions

- The vast majority of emergency room visits and hospitalization studies conducted over the past decade have looked at effects of O_3 on either total respiratory diseases and/or asthma. Among the hospitalization studies, O_3 was found to be associated with both outcomes in many cases. Studies of emergency department visits for respiratory conditions also reported O_3 effects, but the results tended to be less consistent across studies.

- Many of the daily emergency department visits and hospitalization studies analyzed O_3 risk estimates using year-round data. Given the strong seasonal variations in O_3 concentrations and the changing relationship between O_3 and other copollutants by seasons, inadequate adjustment for seasonal effects might have masked or underestimated the association between O_3 and the respiratory disease outcomes. Season-stratified analyses typically yielded more reliable O_3 effect estimates.

- Several studies have examined the association between O_3 and respiratory hospitalizations while controlling for other pollutants in the analytical model. In most cases, O_3 effects have been reported to be robust to adjustment for copollutants, particularly PM. Therefore, the evidence is supportive of independent O_3 effects on respiratory hospital admissions.

- A subset of hospital admission studies examined the effect of O_3 on cardiovascular outcomes. The evidence is inconclusive regarding the association between O_3 exposure and cardiovascular hospitalizations in year-round data analyses. However, in the limited number of studies that accounted for seasonal or meteorological factors, results suggested

that O_3 was associated with increased risk of cardiovascular hospital admissions during the warm season.

7.4 ACUTE EFFECTS OF OZONE ON MORTALITY

7.4.1 Summary of Key Findings on Acute Effects of Ozone on Mortality From the 1996 O_3 AQCD

A limited number of studies examined O_3-mortality associations at the time of the previous O_3 AQCD, most of which were from the 1950s and 1960s. The 1996 O_3 AQCD considered these historical studies to be flawed because of inadequate adjustment for seasonal trends or temperature and the use of questionable exposure indices. There were only a few time-series studies that examined O_3-mortality associations between the 1980s and mid-1990s. These studies used more sophisticated approaches in addressing seasonal confounding and weather models. One of these studies (Shumway et al., 1988) focused on the associations with long-term fluctuations in Los Angeles, CA but did not examine short-term associations. A study that reanalyzed the Los Angeles, CA data with a focus on the short-term associations (Kinney and Özkaynak, 1991) did find that, of the PM and gaseous criteria pollutants, O_3 (reported as total oxidants) was most strongly associated with total nonaccidental mortality. Then two studies, one using Detroit, MI data (Schwartz, 1991) and the other using St. Louis, MO and Kingston-Harriman, TN data (Dockery et al., 1992), reported that PM but not O_3 was significantly associated with mortality. However, the 1996 O_3 AQCD discussed that without sufficient presentation of model specifications, it was difficult to evaluate whether the lack of O_3-mortality associations was possibly due to mis-specification of the weather model. In summary, due to the insufficient number of studies that examined O_3-mortality associations and the uncertainties regarding weather model specifications, the 1996 O_3 AQCD was unable to quantitatively assess O_3-mortality excess risk estimates, or even provide qualitative assessment of the likelihood of O_3-mortality associations.

7.4.2 Introduction to Assessment of Current Ozone-Mortality Studies

Introductory discussions of the PM-mortality effects often cite historical air pollution incidents such as the 1952 London, England smog episode in which thousands of deaths were attributed to the air pollution from coal burning. There is no counterpart "historical episode" for

O_3-mortality effects. Instead, the early recognition of the adverse health effects of summer oxidant air pollution, mainly from Los Angeles and other major cities with a high density of automobiles, were based on symptoms such as eye and throat irritations. Thus, the focus of PM epidemiology and that of O_3 epidemiology have been historically different.

As shown in Table AX7-5 in the Chapter 7 Annex, the number of short-term mortality studies that analyzed O_3 has increased markedly since the last publication of the O_3 AQCD in 1996. The increased attention to PM-mortality associations in the early 1990s lead to the increase in studies that also examined O_3, most often as a potential confounder for PM. Although many of these PM studies also reported O_3 estimates, they often lacked specific hypotheses regarding mortality effects of O_3 as the focus of these studies was to examine the PM-mortality effect. This is in contrast to the O_3-morbidity studies, most of which were specifically designed to examine effects of "summer haze" and O_3 (or oxidants) on respiratory and other symptoms, lung functions, and emergency department visits, etc. However, new studies with hypotheses developed specifically for O_3 effects on mortality have become available, such as the large U.S. 95 communities study by Bell et al. (2004), the U.S. 14 cities study by Schwartz (2005), and the 23 European cities study by Gryparis et al. (2004) discussed in the next section.

7.4.3 Single-Pollutant Model Ozone-Mortality Risk Estimates

To facilitate a quantitative overview of the O_3-mortality effect estimates and their corresponding uncertainties, the percent excess risks of total nonaccidental mortality calculated using all year data are plotted in Figures 7-14 and 7-15. Studies that only conducted seasonal analyses will be presented in the next section. These figures do not include studies that only examined cause-specific mortality. Figure 7-14 only presents the results from single-day lag models. Results from multiday lag models are shown in Figure 7-15. All effect estimates are from single-pollutant models and include all age groups unless noted otherwise. The majority of the estimates are positive with a few exceptions. Five multicity studies, three from the U.S. (Bell et al., 2004; Samet et al., 2000 [reanalysis Dominici et al., 2003]; Schwartz, 2005) and two from Europe (Gryparis et al., 2004; Touloumi et al., 1997), also showed generally positive associations.

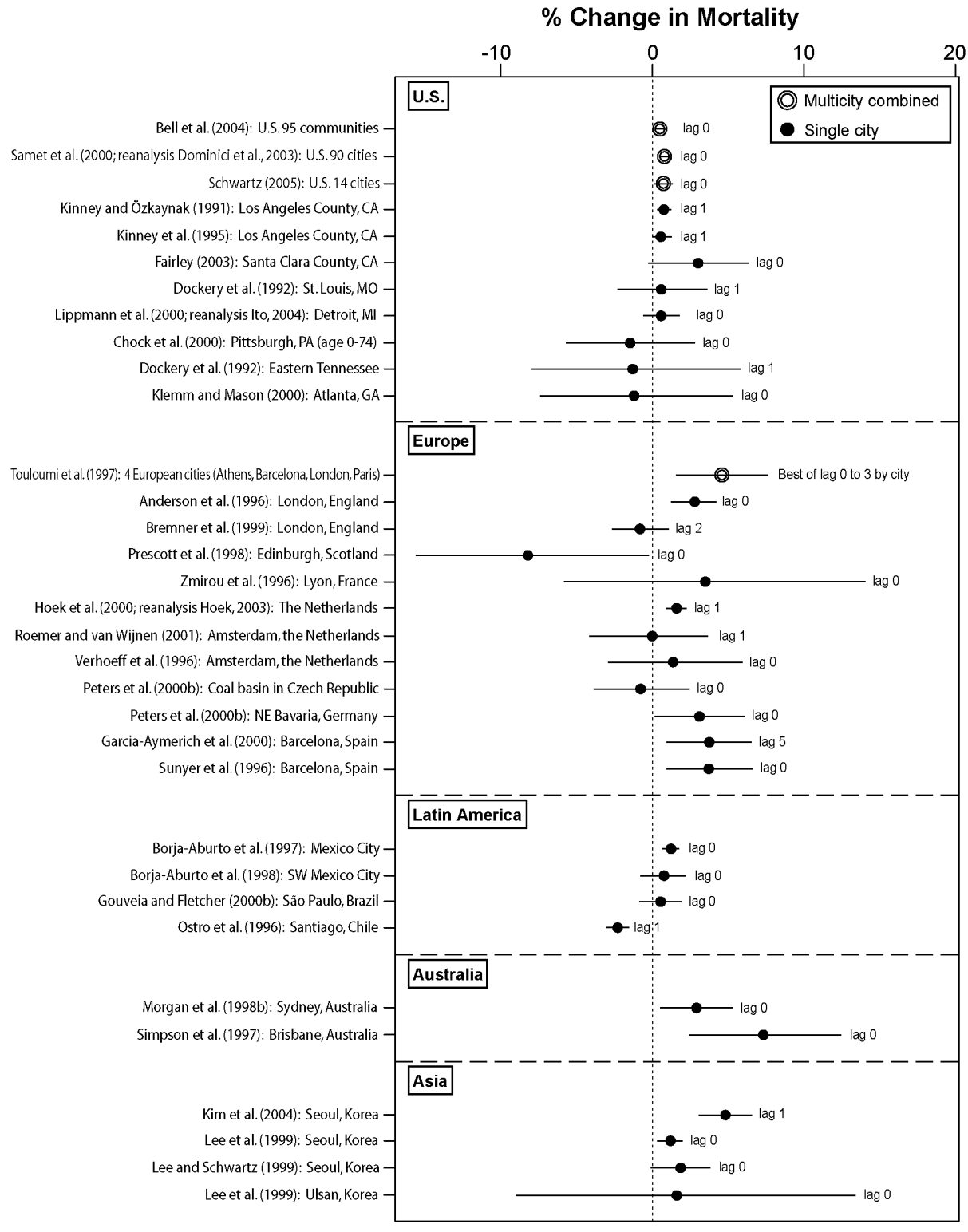


Figure 7-14. All cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) <u>for all year analyses</u> per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted. Only results from <u>single-day lag models</u> are presented.

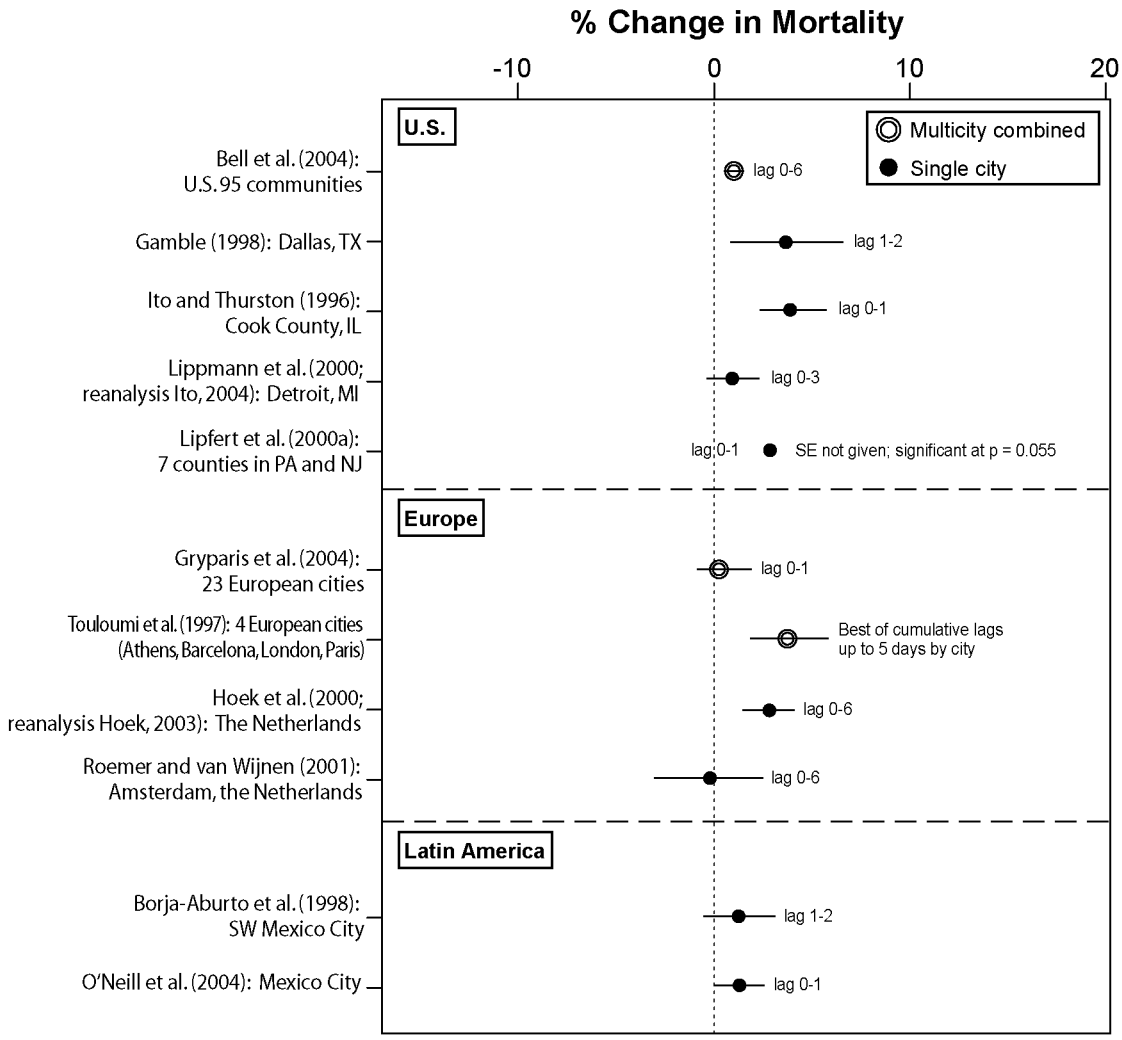


Figure 7-15. All cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) for all year analyses per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted. Only results from multiday lag models are presented.

The initial primary objective of the original NMMAPS (Samet et al., 2000; reanalysis Dominici et al., 2003) was to investigate the effects of PM, but the study also comprehensively examined mortality risk estimates from gaseous pollutants in 90 U.S. cities over the period of 1987 to 1994. Among the 90 cities, 80 monitored O_3 either year-round or during the warm season. The study illustrated that the mortality risk estimates for O_3 varied by season. The estimate using all available data was about half of that for summer-only data at a lag of 1-day

(see Section 7.6.3.2 for further discussion). Bell et al. (2004) extended the original NMMAPS by adding six more years (from 1987 to 2000) and 15 more communities (a total of 95 communities), and examined the effects of O_3 on mortality. The results of this study are discussed in detail here because of the study's emphasis on U.S. data and the inclusion of 95 large communities across the country, making this mortality study most representative of the U.S. population. In addition, this study is one of the few that have focused specifically on O_3 hypotheses testing and investigated several important issues. Among the 95 communities examined in this study, 55 monitored O_3 throughout the year and 32 only monitored during the warm season. Eight additional cities switched from warm season only to year-round monitoring or year-round to warm season only monitoring at some point during the study period. The mean 24-h avg O_3 concentration was approximately 26 ppb for the 95 communities.

Within-community results first were calculated using single-day lags of 0, 1, 2, and 3 days, and a 7-day distributed lag in O_3 exposure. A two-stage Bayesian hierarchical model was used to determine a national average effect estimate, taking into consideration city-to-city variation. Figure 7-16 presents the Bayesian community-specific and national average O_3 risk estimates for total mortality per 20 ppb increase in 24-h avg O_3 from a constrained 7-day distributed lag model. The Bayesian community-specific estimates were shrunk to the national average estimate by a factor that was inversely proportional to the heterogeneity of the community-specific relative rates. The heterogeneity of the effect estimates from the individual cities is partially attributable to differences in pollution characteristics, the use of air conditioning, time-activity patterns, and socioeconomic factors. Due to the random variation as well as the smaller sample sizes within each city, emphasis is given to the national average effect estimate.

In the U.S. 95 communities study, the largest risk estimate for O_3-mortality was obtained with a 0-day lag, followed by diminishing risk estimates with 1-, 2-, and 3-day lags (Figure 7-17). Ozone exposure at a 0-day lag was associated with a 0.50% (95% PI: 0.24, 0.78) excess risk in mortality per 20 ppb increase in 24-h avg O_3. The 7-day distributed lag model, which examined the cumulative effect from the same day and six previous days, also is shown in Figure 7-17. A cumulative excess mortality risk of 1.04% (95% PI: 0.54, 1.55) per 20 ppb increase in 24-h avg O_3 during the previous week was observed. In a related U.S. study of the 19 largest cities by Huang et al. (2005), the O_3 estimate for the summer season was 1.47% (95% PI: 0.54, 2.39) excess risk of cardiopulmonary mortality with current-day exposure. Smaller

Figure 7-16. Community-specific Bayesian estimates and national average for the percent change (95% PI) in daily mortality per 20 ppb increase in 24-h avg O_3 in the previous week using a constrained distributed lag model for 95 U.S. communities (NMMAPS), arranged by size of the effect estimate. Results from all available data are presented (32 of the 95 communities only had warm season data).

Source: Derived from Bell et al. (2004).

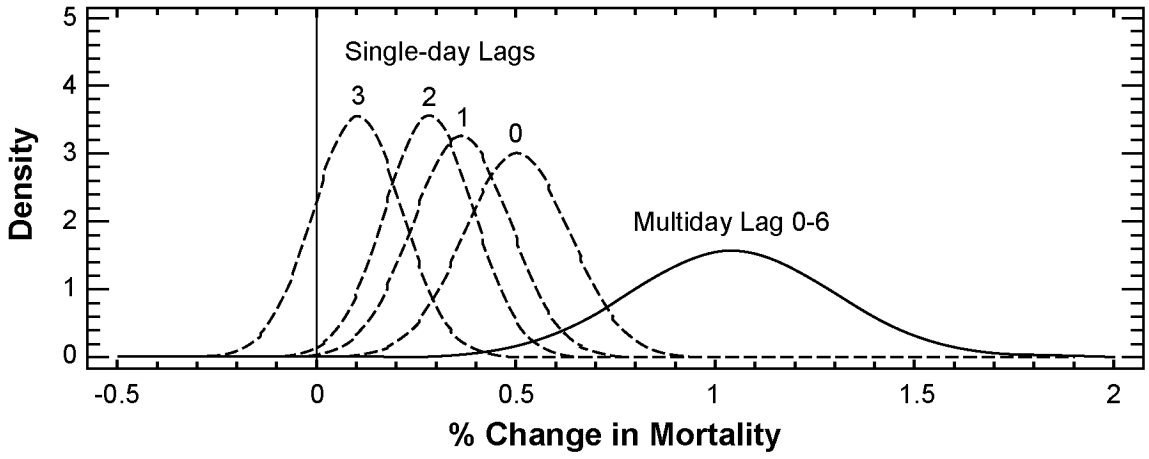


Figure 7-17. Comparison of single-day lags (0-, 1-, 2-, and 3-day) to a cumulative multiday lag (0- to 6-day) for percent changes in all cause mortality per 20 ppb increase in 24-h avg O_3 in all ages.

Source: Derived from Bell et al. (2004).

effects also were observed with 1- and 2-day lags of exposure. The effect estimate for the 7-day distributed lag was 2.52% (95% PI: 0.94, 4.10) excess risk of cardiopulmonary mortality. These findings suggest that the effect of O_3 on mortality is immediate, but also may persist over multiple days.

The influence of higher O_3 levels on the risk estimate also was evaluated in the U.S. 95 communities study. When the data were restricted to days with 24-h avg O_3 levels less than 60 ppb for the 1-day lag analysis, the national estimate did not substantially change (0.30% [95% PI: 0.06, 0.54] per 20 ppb increase for days with levels below 60 ppb versus 0.36% [95% PI: 0.12, 0.61] for all days). These results suggest that the O_3-mortality associations occur at 24-h avg O_3 levels below 60 ppb.

Schwartz (2005) examined O_3-mortality associations using data from 14 U.S. cities. A case-crossover study design was used to compare the influence of adjustment methods for temperature (regression splines of temperature versus matching case and control periods by temperature). The risk estimate obtained by matching (0.92% [95% CI: 0.06, 1.80] per 40 ppb increase in 1-h max O_3) was similar to that obtained with regression splines (0.76% [95% CI:

0.13, 1.40]), suggesting that the O_3-mortality risk estimates were not sensitive to these adjustment methods for temperature.

The APHEA 1 project (Touloumi et al., 1997) reported a pooled random effects estimate of 4.5% (95% CI: 1.6, 7.7) per 40 ppb increase in 1-h max O_3 using the best single-day lag model results from four European cities (London, Athens, Barcelona, and Paris). As an extension of the four European cities study, researchers of the APHEA 2 project investigated the effect of O_3 on total, cardiovascular, and respiratory mortality in 23 cities throughout Europe (Gryparis et al., 2004). Ozone data was available year-round in all 23 cities. A cumulative lag of 0 to 1 days was hypothesized a priori. A two-stage hierarchical model, which accounted for statistical variance and heterogeneity among cities, was used to estimate the pooled regression coefficients. Due to substantial heterogeneity among cities, random effects regression models were applied. The pooled effect estimate for the 23 European cities (0.23% [95% CI: −0.85, 1.95] per 40 ppb increase in 1-h max O_3 for all seasons) was positive but considerably smaller compared to that obtained in the APHEA 1 study. The researchers noted that there was a considerable seasonal difference in the O_3 effect on mortality, thus the small effect for the all year data might be attributable to inadequate adjustment for confounding by season. This seasonal effect will be discussed further in the next section.

Collectively, the single-pollutant model estimates from the single- and multiple-city studies shown in Figures 7-14 and 7-15 suggest an excess risk of total nonaccidental mortality associated with acute O_3 exposure. Despite the different analytical approaches and alternative model specifications used in the various studies, overall, the range of estimates were relatively narrow, with most of the positive estimates falling in the range from 0.5 to 5% excess risk in mortality per standardized increment.

7.4.4 Meta-analyses of O_3-Mortality Risk Estimates

Several studies in recent years conducted meta-analyses of O_3-mortality associations (Levy et al., 2001; Stieb et al., 2002, 2003; Thurston and Ito, 2001; World Health Organization, 2004). Figure 7-18 presents the combined O_3 risk estimates from the various meta-analyses. Most of these studies included GAM studies using default convergence criteria except Stieb et al. (2003), which compared effect estimates from GAM-affected studies to non-GAM studies. All of these meta-analyses reported fairly consistent and positive combined estimates, approximately 2%

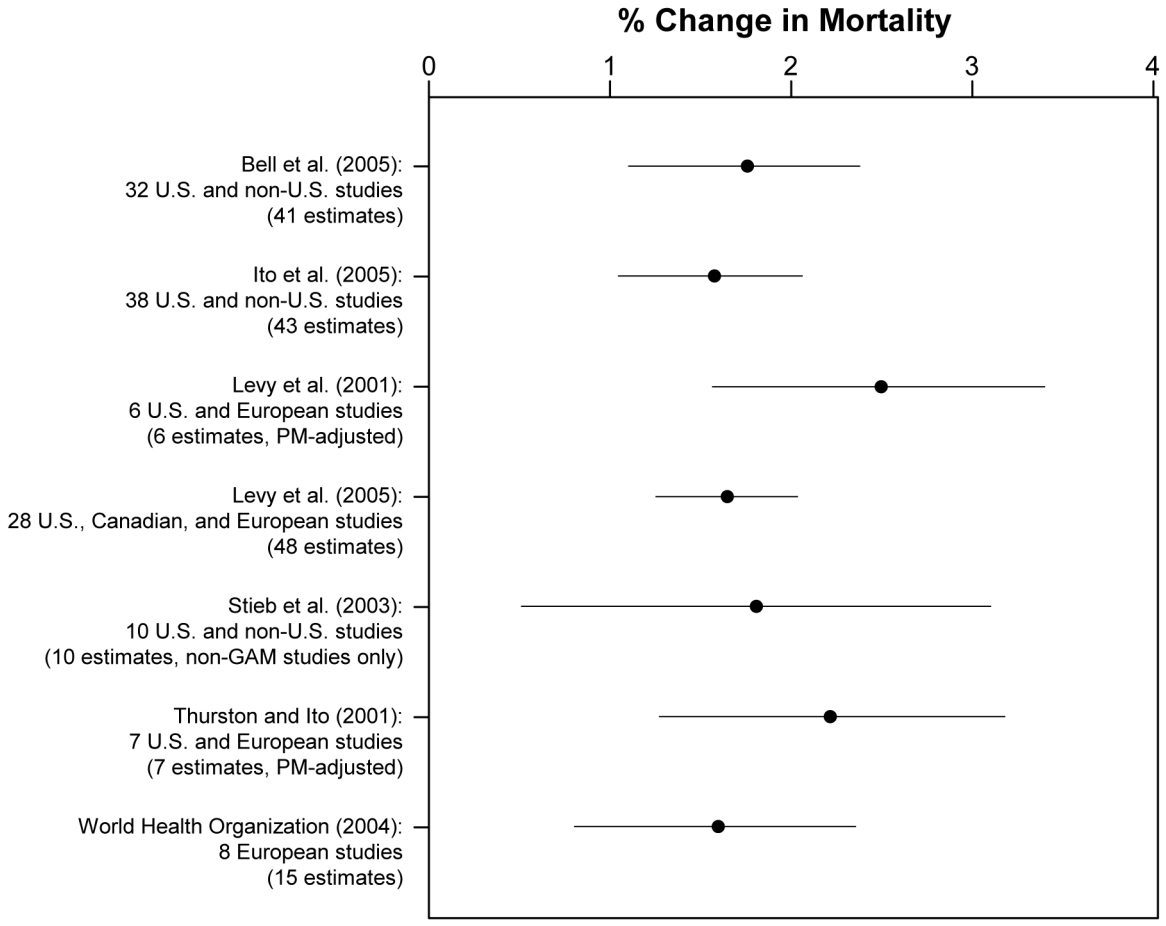


Figure 7-18. Combined all cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) from recent meta-analyses per standardized increment (see Section 7.1.3.2). Note that all meta-analyses, except Stieb et al. (2003), included studies which used Poisson GAM with default convergence criteria.

excess total nonaccidental mortality per standardized increment (see Section 7.1.3.2). However, most of these studies were not analytical in design in that they did not attempt to examine the source of heterogeneity, although one suggested an influence of weather model specification (Thurston and Ito, 2001) and another reported evidence of publication bias (World Health Organization, 2004) in the past literature. None of these studies address the issue of season-specific estimates, therefore, interpreting these combined estimates requires caution.

Most recently, three research groups conducted independent meta-analyses of O_3-mortality associations (Bell et al., 2005; Ito et al., 2005; Levy et al., 2005). These analyses attempted to evaluate the source of heterogeneity using the most up-to-date literature database. These

analyses were also systematically compared and discussed (Bates, 2005; Goodman, 2005). The all-season combined point estimates per standardized increment from these three meta-analyses were remarkably consistent: 1.75% (95% PI: 1.10, 2.37), 1.6% (95% CI: 1.1, 2.0), and 1.64% (95% CI: 1.25, 2.03), for the Bell et al., Ito et al., and Levy et al. studies, respectively. All three studies also indicated that the estimates were higher in warm seasons. Each of these studies is briefly summarized below. Their findings related to specific issues are discussed later in the corresponding sections.

Bell et al. (2005) conducted a meta-analysis of 144 effect estimates from 39 U.S. and non-U.S. studies and estimated pooled effects by lags, age groups, specific causes, and exposure metrics. The results were also compared with their NMMAPS results (Bell et al., 2004). A two-stage Bayesian hierarchical model was used to estimate the combined estimate by taking into account the within-city variance (the statistical uncertainty) and between-study variance (the heterogeneity across cities). They concluded that the results provided strong evidence of a short-term association between O_3 and mortality that was not sensitive to adjustment for PM or model specifications (discussed in Section 7.4.6). However, they suggested that, based on comparisons between the meta-analysis results and NMMAPS results, there was evidence of publication bias (1.75% [95% CI: 1.10, 2.37] per 20 ppb increase in 24-h avg O_3 for meta-analysis versus 0.50% [95% CI: 0.24, 0.78] for NMMAPS 0-day lag results).

Ito et al. (2005) conducted a meta-analysis of 43 U.S. and non-U.S. studies but also analyzed data from 7 U.S. cities to further examine the issues identified in their meta-analysis. Adjusting for PM did not substantially influence the O_3-mortality effect estimates in either the meta-analysis or 7 U.S. cities analysis. The multicity analysis further indicated that the difference in the weather adjustment model could result in a twofold difference in risk estimates (e.g., 1.96% versus 0.96% in multicity combined estimates across alternative weather models for the O_3-only, all year case). In the meta-analysis, they found suggestive evidence of publication bias (a significant asymmetry in the funnel plot), but adjusting for the asymmetry reduced the combined estimate only slightly (from 1.6% [95% CI: 1.1, 2.0] to 1.4% [95% CI: 0.9, 1.9] per 20 ppb increase in 24-h avg O_3). The extent of potential bias implicated in this study differed compared to that in Bell et al. (2005). The source of this difference is not clear, but Ito et al. state that sensitivity analyses comparing estimates from commonly used weather model

specifications suggest that the stringent weather model used in NMMAPS may tend to yield smaller risk estimates than those used in other studies.

Levy et al. (2005) analyzed 48 estimates from 28 studies from the U.S., Canada, and Europe using an empiric Bayesian meta-regression with covariates including the relationship between O_3 and other pollutants, proxies for the relationship between personal exposure and ambient concentration such as air conditioning prevalence, and statistical methods used. They found that the air conditioning prevalence (a greater effect in cities with less air conditioning) and lag time (same-day effects larger than lagged effects) were the strongest predictors of between-study variability. The warm season estimates were larger than the cool season estimates. The influences of copollutants were inconsistent, but they found a potential influence of summertime $PM_{2.5}$.

As stated earlier, the combined O_3 excess mortality risk estimates from the meta-analyses by Bell et al., Ito et al., and Levy et al. were very consistent. Although the analyses were conducted independently, there was considerable overlap among the estimates used in the three meta-analyses; thus, the agreement in the combined risk estimates was not unexpected. The common findings among these three meta-analyses, aside from the consistency in their combined estimates, include: (1) no difference in estimates between GAM studies using default versus stringent convergence criteria; (2) estimates were larger in warm seasons; and (3) no strong indication of PM confounding. Both Bell et al. and Levy et al. studies found that the estimates at lag 0-day were larger than longer lags. Both the Bell et al. and Ito et al. studies suggested evidence of publication bias. These three studies, along with the earlier meta-analyses, provide strong evidence that O_3 is associated with mortality. The combined effect estimates from the various meta-analyses ranged from 1.5 to 2.5% excess risk in all cause mortality.

7.4.5 Seasonal Variation in Ozone-Mortality Risk Estimates

Since the seasonal cycle of O_3 follows the seasonal cycle of temperature (which is inversely related to the mortality seasonal cycle), inadequate adjustment of temporal trends in the regression model may lead to negative O_3-mortality risk estimates. In addition, as discussed in Section 7.1.3.5, in some cities low-level O_3 during the winter may be negatively correlated with PM and other primary pollutants, resulting in negative correlations between O_3 and

mortality even in short-term relationships. The confounding effect by season could be substantially reduced by conducting season-stratified analyses.

A fewer number of O_3-mortality studies performed seasonal analyses. Figure 7-19 presents the studies that reported O_3 risk estimates for all cause mortality by season. For those studies that obtained O_3 risk estimates for each of the four seasons, only summer and winter results are shown. The estimates for year-round data analyses, when available, also are shown for comparisons. In all the studies, the O_3 risk estimates are larger during the warm season than the cool season, with the all year estimates generally in between the two seasonal estimates.

In three U.S. and European multicity studies (Gryparis et al., 2004; Samet et al., 2000 [reanalysis Dominici et al., 2003]; Schwartz, 2005), season-stratified analyses indicated that the O_3-mortality effect estimates were significant and positive in the warm season, with larger effects observed compared to the year-round analyses. The effect estimates from the cool season were notably smaller and less significant. In the case of the U.S. 90 cities study (of which 80 cities had O_3 data available) the winter (December, January, and February) mortality estimate was negative, which was most likely attributable to the inverse relationship between O_3 and PM in the winter.

In the U.S. 95 communities study by Bell et al. (2004), no significant difference was observed between the estimates from all available data and warm season only data (April-October); cool season only analyses were not performed. The warm season effect estimate using the 7-day constrained distributed lag model was 0.78% (95% PI: 0.26, 1.30) excess risk per 20 ppb increase in 24-h avg O_3, compared to 1.04% (95% PI: 0.54, 1.55) calculated using all available data. In the 55 communities with year-round O_3 data, the all year effect estimate was 0.96% (95% PI: 0.32, 1.57).

All three recent meta-analyses (Bell et al., 2005; Ito et al., 2005; Levy et al. 2005), found that the estimates for warm seasons were larger than all year estimates. In Bell et al., the warm season estimate was 3.02% [95% PI: 1.45, 4.63], compared to the all year estimate of 1.75% [95% PI: 1.10, 2.37]. In the subset of 10 cities examined in Ito et al., the warm season and all year estimates were 3.5% [95% CI: 2.1, 4.9] and 2.2% [95% CI: 0.8, 3.6], respectively. Likewise, Levy et al. observed a 3.38% [95% CI: 2.27, 4.42] excess risk in the warm season compared to a 1.64% [95% CI: 1.25, 2.03] excess risk using all year data. All results presented are percent excess risk in mortality per standardized increment.

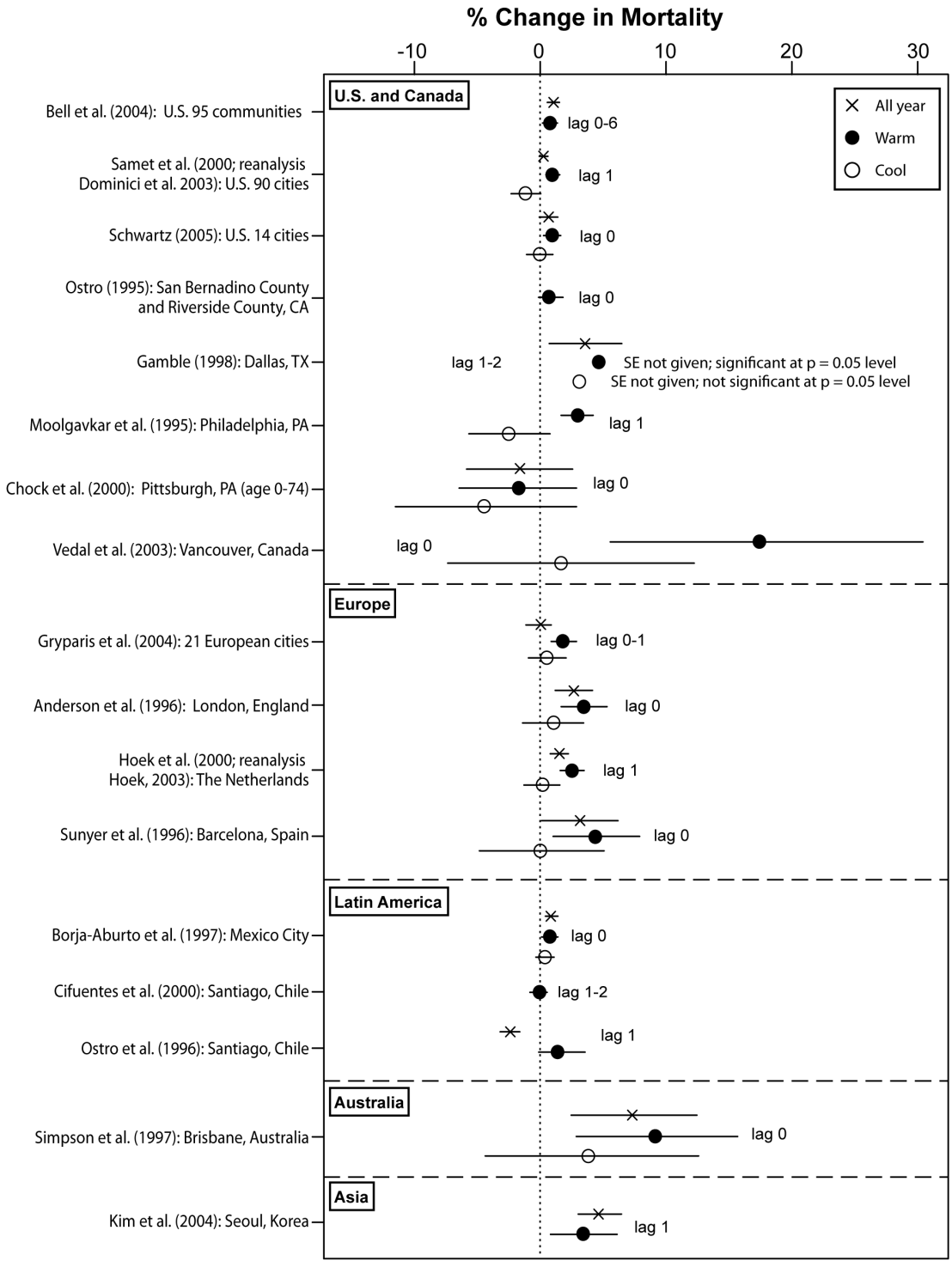


Figure 7-19. All cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) by season per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted.

Studies that conducted analysis by season indicate that O_3-mortality risk estimates are often larger in the warm season compared to the colder season. The seasonal dependence of O_3-mortality effects complicates interpretation of O_3 risk estimates calculated from year-round data without adequate adjustment of temporal trends.

7.4.6 Ozone-Mortality Risk Estimates Adjusting for PM Exposure

The confounding between "winter type" pollution (e.g., CO, SO_2, and NO_2) and O_3 is not of great concern because the peaks of these pollutants do not strongly coincide. The main confounders of interest for O_3, especially for the northeast U.S., are "summer haze" type pollutants such as acid aerosols and sulfates. Since very few studies had these chemical measurements, PM (especially $PM_{2.5}$), may serve as surrogates. However, due to the expected high correlation among the constituents of the "summer haze mix," multipollutant models including these pollutants may result in unstable coefficients, and therefore, an interpretation of such results requires some caution.

Figure 7-20 shows the O_3 risk estimates with and without adjustment for PM indices using all year data in studies that conducted two-pollutant analyses. Approximately half of the O_3 risk estimates slightly increased while the other half slightly decreased in value with the inclusion of PM in the models. In general, the O_3-mortality risk estimates were robust to adjustment for PM in the models, with the exception of Los Angeles, CA data with PM_{10} (Kinney et al., 1995) and Mexico City data with TSP (Borja-Aburto et al., 1997).

The U.S. 95 communities study by Bell et al. (2004) examined the sensitivity of acute O_3-mortality effects to potential confounding by PM_{10}. Restricting analysis to days when both O_3 and PM_{10} data were available, the community-specific O_3-mortality effect estimates as well as the national average results indicated that O_3 was robust to adjustment for PM_{10} (Bell et al., 2004). There was insufficient data available to examine potential confounding by $PM_{2.5}$.
One study (Lipfert et al., 2000a) reported O_3 risk estimates with and without sulfate adjustment. Lipfert et al. (2000a) calculated O_3 risk estimates based on mean (45 ppb) less background (not stated) levels of 1-h max O_3 in seven counties in Pennsylvania and New Jersey. The O_3 risk estimate was not substantially affected by the addition of sulfate in the model (3.2% versus 3.0% with sulfate) and remained statistically significant.

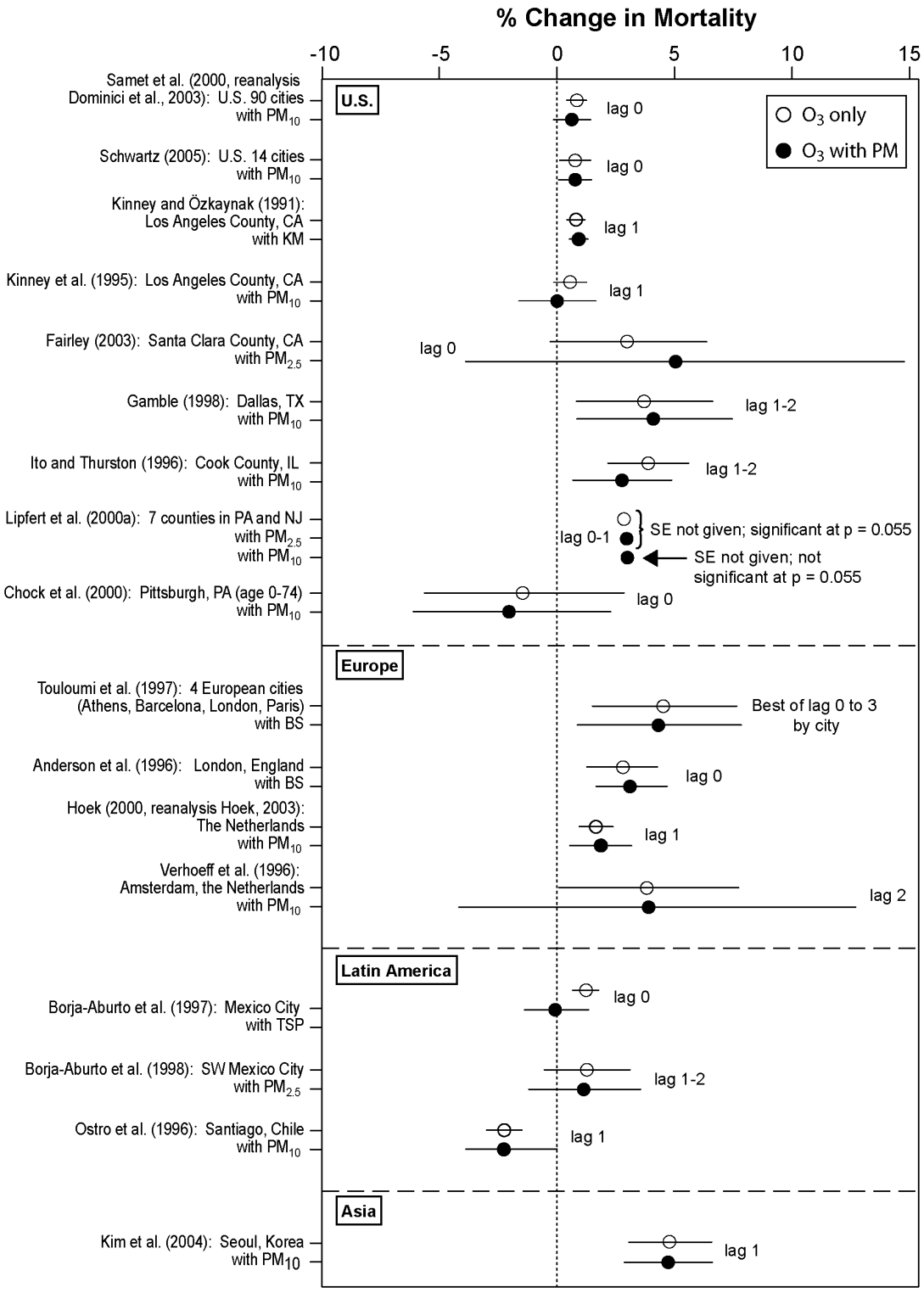


Figure 7-20. All cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) with adjustment for PM indices for all year analyses per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted.

Several O_3-mortality studies examined the effect of confounding by PM indices in different seasons (Figure 7-21). In analyses using all year data and warm season only data, O_3 risk estimates were once again fairly robust to adjustment for PM indices, with values showing both slight increases and decreases with the inclusion of PM in the model. In the analyses using cool season data only, the O_3 risk estimates all increased slightly with the adjustment of PM indices, although none reached statistical significance.

The three recent meta-analyses (Bell et al., 2005; Ito et al., 2005; Levy et al. 2005) all examined the influence of PM on O_3 risk estimates. No substantial influence was observed in any of these studies. In the analysis by Bell et al., the combined estimate without PM adjustment was 1.75% (95% PI: 1.10, 2.37) from 41 estimates, and the combined estimate with PM adjustment was 1.95% (95% PI: −0.06, 4.00) from 11 estimates per 20 ppb increase in 24-h avg O_3. In the meta-analysis of 15 cities by Ito et al., the combined estimate was 1.6% (95% CI: 1.1, 2.2) and 1.5% (95% CI: 0.8, 2.2) per 20 ppb in 24-h avg O_3 without and with PM adjustment, respectively. The additional time-series analysis of six cities by Ito et al. found that the influence of PM by season varied across alternative weather models but was never substantial. Levy et al. examined the regression relationships between O_3 and PM indices (PM_{10} and $PM_{2.5}$) with O_3-mortality effect estimates for all year and by season. Positive slopes, which might indicate potential confounding, were observed for $PM_{2.5}$ on O_3 risk estimates in the summer and all year periods, but the relationships were weak. The effect of one causal variable (i.e., O_3) is expected to be overestimated when a second causal variable (e.g., PM) is excluded from the analysis if the two variables are positively correlated and act in the same direction. However, the results from these meta-analyses as well as several single- and multiple-city studies indicate that copollutants generally do not appear to substantially confound the association between O_3 and mortality.

7.4.7 Ozone Risk Estimates for Specific Causes of Mortality

In addition to all cause mortality, several studies examined broad underlying causes of mortality, such as cardiovascular and respiratory causes. The U.S. 95 communities study (Bell et al., 2004) analyzed O_3 effect estimates from cardiovascular and respiratory mortality. Significant effects were seen at 0- and 2-day lags with results similar to total mortality. The national average estimate from the constrained distributed lag model was slightly greater for

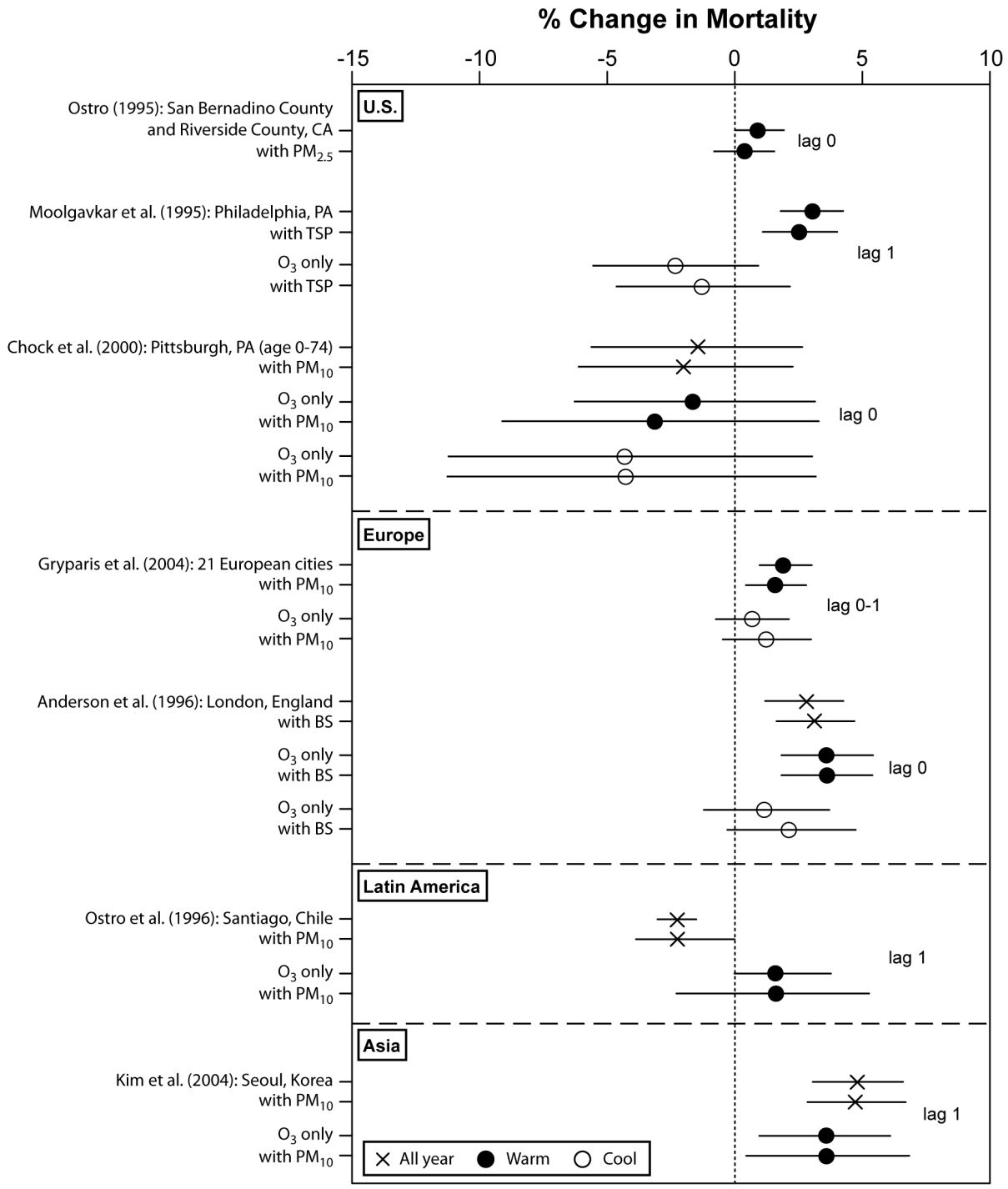


Figure 7-21. All cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) with adjustment for PM indices by season per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted.

cardiopulmonary deaths than deaths from all causes, with an excess risk of 1.28% (95% PI: 0.62, 1.97) compared to 1.04% (95% PI: 0.54, 1.55) per 20 ppb increase in 24-h avg O_3 in the preceding week. In a related study, Huang et al. (2005) examined O_3 effects on cardiopulmonary mortality during the summers (June to September) of 1987 to 1994 in 19 large U.S. cities from the NMMAPS database. In the 7-day distributed lag model, the O_3 effect estimate was 2.52% (95% PI: 0.94, 4.10) excess risk in cardiopulmonary mortality per 20 ppb increase in 24-h avg O_3.

Figure 7-22 presents the effect estimates of the association between O_3 and cardiovascular mortality for all year and warm season analyses. All studies, with the exception of Pönkä et al. (1998), showed positive associations between O_3 and cardiovascular mortality. However, as with all cause mortality, there appears to be heterogeneity in the effect estimates across studies. The cardiovascular mortality estimate from the meta-analysis by Bell et al. (2005) appears to be close to the mode of the effect estimates from the various studies, as shown in Figure 7-22. This is expected as many of these studies are included in the meta-analysis. Bell et al. observed that the posterior mean estimate for cardiovascular causes (2.23% [95% PI: 1.36, 3.08] excess risk per 20 ppb increase in 24-h avg O_3 from 25 estimates) was slightly larger than that for total mortality (1.75% [95% PI: 1.10, 2.37] excess risk from 41 estimates). However, since cardiovascular deaths account for the largest fraction (over 40%) of total deaths, it is not surprising that the risk estimates for cardiovascular mortality are somewhat similar to those from all cause mortality. Overall, the cardiovascular mortality risk estimates in the current literature show consistently positive associations with some heterogeneity (most estimates fall within the range of 1 to 8% per 40 ppb increase in 1-h avg O_3).

Several studies observed that the risk estimates for the respiratory category were larger than the cardiovascular and total nonaccidental categories (e.g., Anderson et al., 1996; Gouveia and Fletcher, 2000b; Gryparis et al., 2004; Zmirou et al., 1998). In the European 21 multicities study (Gryparis et al., 2004), the warm season effect estimate for respiratory mortality was 6.75% (95% CI: 4.38, 9.10) excess risk per 30 ppb increase in 8-h max O_3, compared to 2.70% (95% CI: 1.29, 4.32) for cardiovascular mortality and 1.82% (95% CI: 0.99, 3.06) for total mortality. In contrast, other studies have found that the risk estimates for the respiratory category were smaller or even negative while the risk estimates for total or cardiovascular categories were positive (e.g., Borja-Aburto et al., 1998; Bremner et al., 1999; Lipfert et al.,

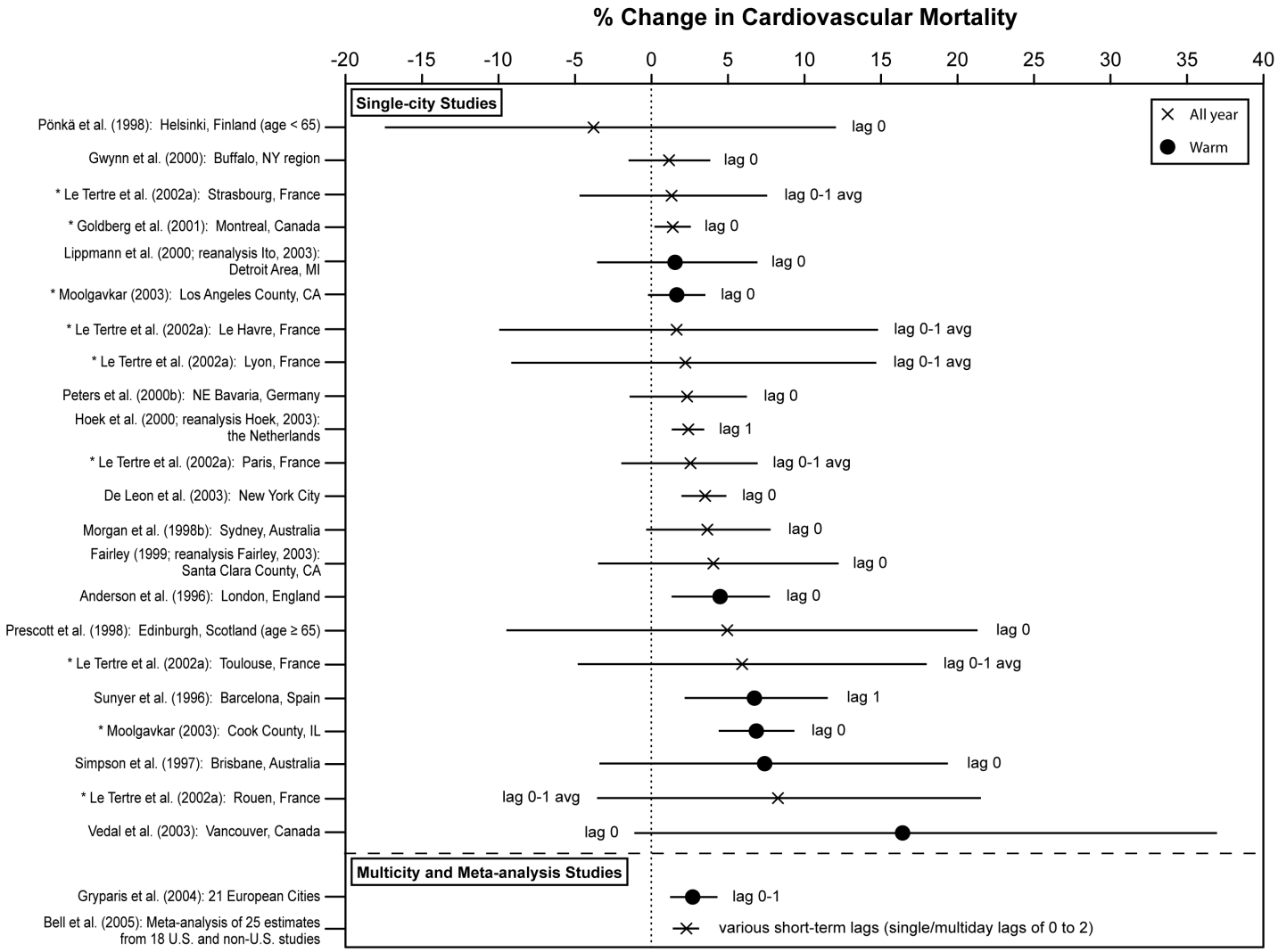


Figure 7-22. Ozone-associated cardiovascular mortality risk estimates (95% CI) per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted. *Note that Goldberg et al. (2001), Le Tertre et al. (2002a), and Moolgavkar (2003) performed analyses using Poisson GAM with default convergence criteria.

2000a; Morgan et al., 1998b). The apparent inconsistencies across studies may be due in part to the differences in model specifications, but they may also reflect the lower statistical power associated with the smaller daily counts of the respiratory category (usually accounting for less than 10% of total deaths) compared to the larger daily counts for the cardiovascular category (approximately 40 to 50% of total deaths). Thus, an examination of the differences in risk estimates across specific causes requires a large population and/or a long period of data collection. In the meta-analysis by Bell et al. (2005), which combined 23 estimates from 17 studies for respiratory mortality, the effect estimate for respiratory causes was smaller (0.94% [95% PI: −1.02, 2.96] excess risk per 20 ppb increase in 24-h avg O_3) compared to the estimates for total mortality (1.75% excess risk) and cardiovascular mortality (2.23% excess risk).

The analyses of a 9-year data set for the whole population of the Netherlands (population = 14.8 million) provided risk estimates for more specific causes of mortality, including COPD, pneumonia, and subcategories of cardiovascular causes (Hoek et al., 2000, 2001; reanalysis Hoek, 2003). The effect estimate for total nonaccidental mortality was 1.6% (95% CI: 0.9, 2.4) excess risk per 30 ppb increase in 8-h avg O_3. In comparison, the excess risk estimates for pneumonia and COPD were 5.6% (95% CI: 1.8, 9.5) and 0.8% (95% CI: −2.4, 4.2), respectively. The effect estimates for some of the cardiovascular subcategories, including heart failure (3.8% [95% CI: 0.5, 7.3]) and thrombosis-related disease (6.0% [95% CI: 1.1, 10.8]), showed greater risk estimates than that for total mortality. However, these associations were not specific to O_3. For example, most of the pollutants examined, including PM_{10}, BS, SO_2, NO_2, CO and NO_3^-, were associated with pneumonia. Therefore, it is difficult to make a causal inference specific to O_3 based on these results.

De Leon et al. (2003) examined the role of contributing respiratory causes in the association between air pollution and nonrespiratory mortality (circulatory and cancer) in New York City during the period of 1985 to 1994. The main finding of this study was that the estimated excess mortality risks for PM_{10} were higher for nonrespiratory deaths that had contributing respiratory causes compared to deaths without contributing respiratory causes in the older age (75+ years) group. This pattern was also seen for CO and SO_2, but not for O_3. Therefore, this study did not suggest a role of contributing respiratory causes in the association between O_3 and nonrespiratory causes of deaths.

In summary, several single-city studies observed positive associations between ambient O_3 concentrations and cardiovascular mortality. In addition, a meta-analysis that examined specific causes of mortality found that the cardiovascular mortality risk estimates were higher than those for total mortality. The findings regarding the effect size for respiratory mortality have been less consistent, possibly due to lower statistical power in this subcategory of mortality.

7.4.8 Ozone-Mortality Risk Estimates for Specific Subpopulations

Some studies examined O_3-mortality risk estimates in potentially susceptible subpopulations, such as those with underlying cardiopulmonary disease. Sunyer et al. (2002) examined the association between air pollution and mortality in a cohort of patients (467 men and 611 women) with severe asthma in Barcelona, Spain during the period of 1986 to 1995. A case-crossover study design was used to estimate excess odds of mortality adjusting for weather and epidemics in three groups: (1) those who had only one asthma emergency department visit; (2) those who had more than one asthma emergency department visit; and (3) those who had more than one asthma and COPD emergency department visit. Those with more than one asthma emergency department visit showed the strongest associations with the examined air pollutants, with NO_2 being the most significant predictor, followed by O_3. Sunyer et al. reported a significant association between O_3 and all cause deaths for this group during the warm season, with an odds ratio of 2.82 (95% CI: 1.15, 6.87) per 40 ppb increase in 1-h max O_3, compared to an odds ratio of 1.03 (95% CI: 0.60, 1.78) for those with only one asthma emergency department visit and 1.08 (95% CI: 0.60, 1.92) for the group with a concomitant diagnosis of COPD. In another Barcelona study, Saez et al. (1999) examined asthma mortality death among persons aged 2 to 45 years. Once again, O_3 and NO_2 were the only air pollutants that were significantly associated with asthma mortality death. While the similarity of the patterns of associations between O_3 and NO_2 makes it difficult to speculate on the specific causal role of O_3, the results of these studies suggest that individuals with severe asthma may make up a subpopulation that is sensitive to these pollutants.

Sunyer and Basagna (2001) also performed an analysis of emergency department visits by a cohort with COPD. The results from this study suggested that PM_{10}, but not gases were associated with mortality risks for the COPD cohort. However, a Mexico City study by Téllez-

Rojo et al. (2000) observed a significant association between COPD mortality and O_3, as well as PM_{10}, among patients living outside a medical unit. For a cumulative 5-day lag, an excess risk of 15.6% (95% CI: 4.0, 28.4) per 40 ppb increase in 1-h max O_3 was observed for COPD mortality.

Goldberg et al. (2003) investigated the association between air pollution and daily mortality with congestive heart failure as the underlying cause of death in patients aged 65 years or more in Montreal, Quebec, Canada during the period of 1984 to 1993. Analysis was stratified into two groups, those whose underlying cause of death was congestive heart failure and those with a diagnosis of congestive heart failure one year before their death. They found no association between daily mortality for congestive heart failure and any pollutants. However, they did find associations between daily mortality and coefficient of haze, SO_2, and NO_2 among those who were classified as having congestive heart failure before death. In the case of O_3, positive risk estimates were observed for year-round and warm season data; however, results were not significant. While the 10-year study period for this data was long, the daily mean death counts for the specific subcategory chosen was relatively small (0.7/day for mortality with congestive heart failure as underlying cause of death and 4.0/day for total mortality in patients previously diagnosed with congestive heart failure), limiting the power of the study.

In the meta-analysis by Bell et al. (2005), a combined estimate was obtained for the elderly population (age 64 years and older or 65 years and older) using 10 estimates from 9 studies. The posterior mean estimate for the elderly category (2.92% [95% PI: 1.34, 4.51] per 20 ppb increase in 24-h avg O_3) was larger than that from all ages (1.75% [95% PI: 1.10, 2.37] from 41 estimates). The results from this meta-analysis suggest that the elderly population may be particularly susceptible to O_3-related mortality.

Few studies have examined O_3-mortality effects for specific subpopulations. Among those that investigated the effect of air pollution in populations with underlying cardiopulmonary diseases, associations were not unique to O_3 but were shared with other pollutants. There is suggestive evidence that severe asthmatics may be susceptible to the mortality effects associated with NO_2 and O_3. In addition, the meta-analysis by Bell et al. (2005) suggests that the elderly population may be more affected by O_3.

7.4.9 Summary of Acute Ozone Effects on Mortality

- A substantial body of new data on acute mortality effects of O_3 has emerged since the previous O_3 AQCD. While uncertainties remain in some areas, it can be concluded that robust associations have been identified between various measures of daily O_3 concentrations and increased risk of mortality. Most of the single-pollutant model estimates from single-city studies fell in the range between 0.5 to 5% excess deaths per standardized increment. The corresponding summary estimates in large multicity studies and meta-analyses ranged between 0.5 to 2.5%, with some studies noting heterogeneity across cities and studies. These associations could not be readily explained by confounding due to time, weather, nor copollutants, but model specifications likely contributed to some of the observed heterogeneity in risk estimates across studies.

- The majority of the available O_3-mortality risk estimates were computed using all year data. The results from the studies that conducted analysis by season suggest that the O_3 risk estimates were larger in the warm season. Some of the risk estimates in the cool season were negative, possibly reflecting the negative correlation between low-level O_3 and PM (and other primary pollutants) during that season. Thus, even with adjustment for temporal trends, the O_3 risk estimates obtained for year-round data may be misleading. In locations with considerable seasonal variation, season-specific analyses may better elucidate the effect of O_3 on mortality.

- The majority of the available O_3-mortality risk estimates were computed for a single-day lag. Choosing the optimal lag out of several lags examined may bias the single-day risk estimate upward. However, recent findings from the largest U.S. 95 communities study indicated that a strong association between O_3 and mortality was observed with a 7-day distributed lag model. Thus, it is possible that the effect of acute O_3 exposure on mortality persists over several days. Further research is needed to understand the nature of cumulative effects.

- Some studies examined specific subcategories of mortality, but most of these studies had limited statistical power to detect associations due to the small daily mortality counts. A recent meta-analysis indicated that there was a slightly greater risk of cardiovascular mortality compared to total mortality.

- Few studies examined the effect of O_3 on mortality in subpopulations with underlying cardiopulmonary diseases. Similar to cause-specific mortality, these population-specific studies had limited statistical power to detect associations. The evidence suggests that individuals with severe asthma may be at increased risk of O_3-related mortality; however, similar results were seen with other pollutants.

7.5 EFFECTS OF CHRONIC OZONE EXPOSURE

7.5.1 Summary of Key Findings on Studies of Health Effects and Chronic Ozone Exposure from the 1996 O_3 AQCD

The 1996 O_3 AQCD concluded that there was insufficient evidence from the limited number of studies to determine whether long-term ambient O_3 exposures resulted in chronic health effects. However, the aggregate evidence suggested that chronic O_3 exposure, along with other environmental factors, could be responsible for health effects in exposed populations.

7.5.2 Introduction to Morbidity Effects of Chronic Ozone Exposure

Several new longitudinal epidemiologic investigations have yielded information on health effects of long-term O_3 exposures. Epidemiologic interest in investigating long-term effects has been motivated by several considerations. Animal toxicology studies carried out from the late 1980s onward demonstrated that long-term exposures can result in permanent changes in the small airways of the lung, including remodeling of the airway architecture (specifically the distal airways and centriacinar region) and deposition of collagen, as discussed earlier in Chapter 5. These changes result from the damage and repair processes that occur with repeated exposure. Indices of fibrosis also were found to persist after exposure in some of the studies. Collectively, these findings provide a potential pathophysiologic basis for the changes in airway function observed in children in longitudinal studies. Seasonal ambient patterns of exposure may be of greater concern than continuous daily exposure. In the classical study by Tyler et al. (1988), young monkeys with seasonal exposure to O_3, but not those with daily exposure, experienced increases in total lung collagen content, chest wall compliance, and inspiratory capacity, suggesting a delay in lung maturation in seasonally-exposed animals.

Controlled human exposure studies clearly demonstrated acute inflammation in the lung at ambient exposure levels. Epidemiologic studies could examine whether repeated exposures over multiple episode periods and/or multiple years would lead to persistent inflammation and result in damage to the human lung, especially in the small, terminal bronchiolar regions where vulnerability is greatest. However, the challenges to addressing these issues in epidemiologic studies are formidable, and as a result there exists relatively limited literature in this area. Long-term O_3 concentrations tend to be correlated with long-term concentrations of other pollutants, making specific attribution difficult. Subtle pulmonary effects require health outcome measures

that are sensitive, and must usually be directly collected from individual human subjects, rather than from administrative data bases. Although these factors make chronic studies difficult and expensive to conduct, efforts must be made to design studies with adequate power to examine the hypothesis being tested. Epidemiologic studies have the potential to provide important new insights on the links between chronic exposure to O_3 and the occurrence of human health effects.

This section reviews studies published from 1996 onward in which health effects were tested in relation to O_3 exposures extending from several weeks to many years (Table AX7-6 in the Chapter 7 Annex). The available literature falls into four general categories: (1) studies examining seasonal changes in lung function as related to O_3 exposures in peak season; (2) studies addressing smaller increases in lung function during childhood or decline of lung function beyond childhood in relation to long-term O_3 exposures; (3) studies addressing respiratory inflammation in high versus low exposure groups or time periods; and (4) studies addressing longitudinal and cross-sectional associations between long-term O_3 exposures and asthma development and prevalence.

7.5.3 Seasonal Ozone Effects on Lung Function

While it has been well-documented in both chamber and field studies that daily, multihour exposures to O_3 result in transient declines in lung function, much less is known about the effects of repeated exposures to O_3 over extended periods on lung function. Several new studies reported over the past decade have examined lung function changes over seasonal time periods with differing levels of O_3 exposures (Frischer et al., 1999; Horak et al., 2002a,b; Ihorst et al., 2004; Kinney and Lippmann, 2000; Kopp et al., 2000). The seasonal effects of O_3 are examined first in this section. In the next section is a discussion of effects over years, as opposed to over seasons, in addition to multiyear analyses of seasonal studies.

In a large Austrian study, Frischer et al. (1999) collected repeated lung function measurements in 1,150 school children (mean age 7.8 years) from nine towns that differed in mean O_3 levels. Lung function was measured in the spring and fall over a three-year period from 1994 to 1996, yielding six measurements per child. Mean summertime O_3 exposure ranged from 32.4 to 37.3 ppb during the three summers. Growth-related increases in lung function over the summer season were reduced in relation to seasonal mean O_3 levels. Ozone was associated with a change of −156.6 mL (95% CI: −209.5, −103.7) (central estimate: −0.029 mL/day/ppb × 90

days/year × 3 years × 20 ppb) in FEV_1 increase for each 20 ppb increase in mean 24-h avg O_3 concentrations over the three summers and −129.6 mL (95% CI: −193.1, −66.1) over the three winters. When analyses were restricted to children who had spent the whole summer period in their community, the changes were greater, with an O_3-related −183.6 mL (95% CI: −278.9, −88.3) change in FEV_1 increase over three summers. Other pollutants (PM_{10}, SO_2, and NO_2) had less consistent associations with changes in lung function. Horak et al. (2002a,b) extended the study of Frischer et al. (1999) with an additional year of data and stated that seasonal mean O_3 was associated with a negative effect on increases in lung function, confirming results from the previous three-year study. In an editorial, Tager (1999) stated that the Frischer et al. (1999) data provided the first prospective evidence of an association between exposure to ambient air pollution and alterations in lung function in children. Tager further noted that the prospective study design represented a substantial improvement over data derived from cross-sectional studies and should be emulated. However, Tager also cautioned that it was difficult to attribute the reported effects to O_3 alone independently of copollutants.

Kopp et al. (2000), in a cohort of 797 children in Austria and southwestern Germany, reported smaller increases in lung function in children exposed to high (44 to 52 ppb O_3) levels of ambient O_3. Children residing in low O_3 (24 to 33 ppb) areas experienced a 43 mL increase in FEV_1 whereas those in high O_3 areas only experienced a 16 mL increase during the summer of 1994. Similar results were found in data from the summer of 1995. In another Austrian study, Ihorst et al. (2004) examined 2,153 children with a median age of 7.6 years and reported summer pulmonary function results revealing a significantly lower FVC and FEV_1 increase associated with higher O_3 exposures in the summer, but not in the winter.

In a pilot study (Kinney and Lippmann, 2000), 72 nonsmoking adults (mean age 20 years) from the second year class of students at the U.S. Military Academy at West Point, NY provided two lung function measurements, one before and one after a five-week long summer training program at four locations. There was a greater decline in FEV_1 among students at the Fort Dix location (78 mL) as compared to students at the other locations (31 mL). Ozone levels at Fort Dix averaged 71 ppb (mean of daily 1-h max O_3) over the summer training period versus mean values of 55 to 62 ppb at the other three locations. In addition to the higher mean O_3 level, Fort Dix had greater peak O_3 values (23 hours >100 ppb) compared to the other locations (1 hour >100 ppb). Ambient levels of other pollutants, PM_{10} and SO_2, were relatively low during the

study and did not vary across the four sites. Though conclusions are limited by the small size of the study, results are consistent with a seasonal decline in lung function that may be due, in part, to O_3 exposures. An exploratory observation from this study was that there appeared to be a larger decline for those subjects who completed their post-summer lung function measurements in the first two weeks after returning from training compared to those measured three to four weeks after training, which is consistent with some degree of rebound of function following the summer exposure period.

Collectively, the above studies indicate that seasonal O_3 exposure is associated with smaller increases in lung function in children. The study by Kinney and Lippman (2000) provide limited evidence that seasonal O_3 also may affect lung function in adults, though the effect may be somewhat transient.

7.5.4 Chronic Ozone Exposure Effects on Lung Function and Respiratory Symptoms

Lung capacity grows during childhood and adolescence as body size increases, reaches a maximum during the 20s, and then begins to decline steadily and progressively with age. There has long been concern that long-term exposure to air pollution might lead to slower growth in lung capacity, diminished maximally attained capacity, and/or more rapid decline in capacity with age. The concern arises by analogy with cigarette smoking, where it is well-documented that lung function declines more rapidly with age in a dose-dependent manner among adults who smoke cigarettes. Adults who stop smoking return to a normal rate of decline in capacity, although there is no evidence that they regain the capacity previously lost due to smoking (Burchfiel et al., 1995). Because O_3 is a strong respiratory irritant and is associated with acute lung function declines as well as inflammation and re-structuring of the respiratory airways, it seems plausible that there might be a negative impact of long-term O_3 exposures on lung function. Exposures that negatively affect increases in lung function during childhood, in particular, might have greater long-term risks. Thus, studies of effects on the rates of increases in lung function in children are especially important.

Several studies published over the past decade have examined the relationship between lung function and long-term O_3 exposure. The most extensive and robust study of respiratory effects in relation to long-term air pollution exposures among children in the U.S. is the

Children's Health Study carried out in 12 communities of southern California starting in 1993 (Avol et al., 2001; Gauderman et al., 2000, 2002, 2004a,b; Peters et al., 1999a,b). The first cohort included children from the fourth, seventh, and tenth grades. A total of 3,676 students completed questionnaires regarding their lifetime residential histories, historic and current health status, residential characteristics, and physical activity. Among those students, 3,293 also performed pulmonary function tests at the time of enrollment. Peters et al. (1999a) examined the relationship between long-term (1986-1990) O_3 exposures and self-reports of respiratory symptoms and asthma in a cross-sectional analysis. For outcomes of current asthma, bronchitis, cough, and wheeze, the reported odds ratios were 0.95 (95% CI: 0.70,1.29), 1.14 (95% CI: 0.84, 1.55), 0.98 (95% CI: 0.82, 1.17), and 1.08 (95% CI: 0.87, 1.35), respectively, per 40 ppb increase in 1-h max O_3. In another cross-sectional analysis examining the relationship between lung function at baseline and levels of air pollution in the community, there was evidence that annual mean O_3 levels were associated with decreased FVC, FEV_1, PEF, and FEF_{25-75} (the latter two being statistically significant) among females but not males (Peters et al., 1999b).

Avol et al. (2001) examined 110 children from the first cohort who had moved from the participating communities in southern California to other states to determine whether changes in air quality caused by relocation were associated with changes in annual increases in lung function. With the exception of FEV_1, the O_3 effect estimates for all other spirometric parameters were negative, but the associations were not as strong as those observed for PM_{10}.

A second cohort of fourth graders (n = 1,678) were recruited in 1996 and followed over four years to examine the association between long-term exposure to air pollution and changes in lung function (Gaunderman et al., 2002). In general, smaller increases in various lung function parameters were observed in communities with higher 4-year average O_3 levels (for examples, see Figure 7-23). The strongest effect of O_3 was on PEF — children from the least-polluted community had a 1.21% (95% CI: 0.36, 2.06) greater increase in PEF compared to those from the most-polluted communities. However, in the 4-year and 8-year longitudinal analysis of the first cohort, Gauderman et al. (2000, 2004) stated that the results provided little evidence that long-term exposure to ambient O_3 was associated with significant deficits in the growth rate of lung function in children.

In both cohorts of fourth graders, stratified analyses by time spent outdoors indicated a stronger association between long-term O_3 exposure and smaller increases in lung function in

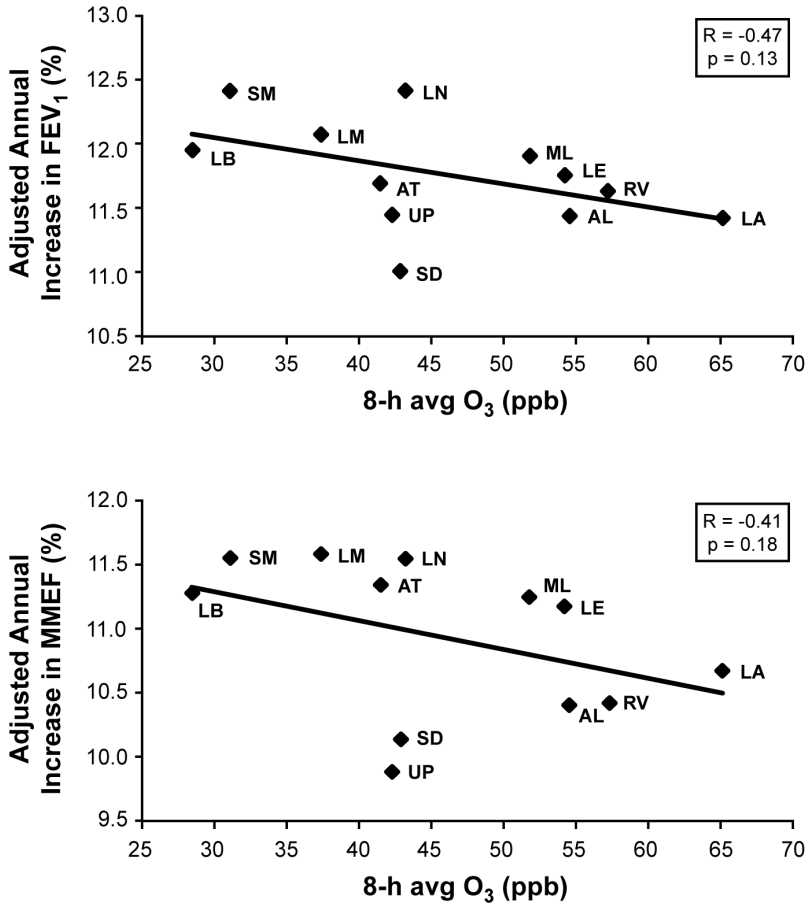


Figure 7-23. Adjusted average annual increases in FEV_1 and maximal midexpiratory flow (MMEF) versus the mean 8-h avg O_3 (10 a.m. to 6 p.m.) concentration over a 4-year period in the 12 southern California communities of the Children's Health Study.

AL = Alpine; AT = Atascadero; LA = Lake Arrowhead; LB = Long Beach; LE = Lake Elsinore; LM = Lompoc; LN = Lancaster; ML = Mira Loma; RV = Riverside; SD = San Dimas; SM = Santa Maria; UP = Upland

Source: Gauderman et al. (2002).

children who spent more time outdoors, as shown in Table 7-3 (Gauderman et al., 2002). A study by Jedrychowski et al. (2001) found a link between repeated respiratory symptoms and smaller lung function increases. Gauderman et al., therefore, suggested that the observation of reduced increases in lung function with increasing annual average air pollution might be a consequence of repeated acute respiratory events after short-term increases in pollutant levels.

Table 7-3. Difference in Annual Percent Increases in Lung Function from the Least to the Most Polluted Community in the Children's Health Study by Time Spent Outdoors [a]

Lung Function Parameter	Cohort [b]	More Time Outdoors [c] % Change (95% CI) [d]	Less Time Outdoors [c] % Change (95% CI) [d]
FVC	Cohort 1	−0.02% (−0.57, 0.54)	−0.04% (−0.45, 0.37)
	Cohort 2	−0.57% (−1.03, −0.09)	−0.06% (−0.76, 0.66)
FEV_1	Cohort 1	−0.25% (−1.18, 0.68)	−0.05% (−0.58, 0.49)
	Cohort 2	−0.68% (−1.36, 0.00)	−0.29% (−1.02, 0.46)
MMEF	Cohort 1	−0.55% (−2.08, 1.01)	0.23% (0.89, 1.36)
	Cohort 2	−0.48% (−1.71, 0.78)	−0.80% (−2.07, 0.50)
PEF	Cohort 1	−0.77% (−2.03, 0.52)	0.25% (−0.65, 1.16)
	Cohort 2	−1.33% (−2.43, −0.24)	−0.71% (−1.71, 0.30)

[a] Results are derived from Gauderman et al. (2002).
[b] Cohort 1 includes children enrolled in 1993 as 4th graders and followed through 1997 (n = 1,457). Cohort 2 includes children enrolled in 1996 as 4th graders and followed through 2000 (n = 1,678).
[c] More or less time outdoors is based on reported time spent outdoors during weekday afternoons. Subjects were split into the two groups on the basis of the median reported time outdoors within each cohort.
[d] Percent change in lung function is per 30 ppb increase in 8-h avg O_3 (10 a.m.-6 p.m.).

The findings of larger deficits in children who spend more time outdoors in the afternoon adds some support to the possibility. Results from this study also indicate the importance of reducing exposure misclassification. Most long-term epidemiologic studies, including the Children's Health Study, estimated O_3 exposure using centrally-located ambient monitors. Gonzales et al. (2003) and Künzli et al. (1997) evaluated the use of retrospective questionnaires to reconstruct past time-activity and location pattern information. Both studies found that questionnaires or activity diaries might improve the assessment of chronic exposure in epidemiologic studies.

In a study conducted in Austria and Germany, Ihorst et al. (2004) found that there were no associations between increases in lung function and mean summer O_3 levels for FVC and FEV_1 over a 3.5-year period, in contrast to the significant seasonal effects discussed in the earlier

section. Unlike the reduced increases in lung function parameters over the first two summers among children in high O_3 areas, a greater increase was observed during the third summer and no difference was observed during the fourth summer. The authors then concluded that medium-term effects on school children lung function were possibly present but were not detected over a 3- to 5-year period due to partial reversibility. The study by Frischer et al. (1999) showed results similar to the Ihorst et al. (2004) study. Although O_3 was related to smaller increases in lung function when three years of data were analyzed collectively, the magnitude and direction of the effect changed throughout the years. Ozone was associated with a change of −34.0 mL (95% CI: −58.7, −9.3) in FEV_1 increase in the first year, compared to +7.3 mL (95% CI: −20.8, 35.6) in the third year for each 20 ppb increase in mean 24-h avg O_3 (Frischer et al., 1999).

Calderón-Garcidueñas et al. (2003) examined chest X-rays and lung function in 174 children from Mexico City and 27 control children from low pollution areas (Tuxpam and Tlaxcala, Mexico). The Mexico City children exhibited lung hyperinflation (67%), interstitial markings (49%), and a mild restrictive pattern by spirometry (10%). In children with increased interstitial markings, FEF_{75} values were significantly declined ($r = 0.42$, $p < 0.003$). No significant abnormalities were observed in the control children. In another study of similar design, the prevalence of respiratory symptoms was higher in Mexico City children compared to children from the clean coastal town of Manzanillo, Mexico (Calderón-Garcidueñas et al., 1999).

A study by Gong et al. (1998b) examined lung function changes in 164 nonsmoking adults (mean age 45 years) from a high O_3 community in southern California, recruited from a cohort of 208 who had been tested on two previous occasions. In the earlier analysis by Detels et al. (1987), a significant decline in lung function was observed from 1977/1978 to 1982/1983. In contrast, Gong et al. observed a slight increase in FVC and FEV_1 from 1982/1983 to 1986/1987. A consistent decline in FEV_1/FVC ratio was observed at all three time points ($p < 0.0001$). Among the 45 subjects who further participated in the controlled exposure study (0.40 ppm O_3 over 2 hours with intermittent exercise), acute changes in lung function were not associated with long-term changes in lung function over a decade.

Evidence for a relationship between long-term O_3 exposures and decrements in maximally attained lung function was observed in a nationwide cohort of 520 first year students at Yale College in New Haven, CT (Galizia and Kinney 1999; Kinney et al., 1998). Each student performed one lung function test in the spring of their first year at college. Ozone exposures

were estimated by linking 10-year mean summer season 1-h max O_3 levels at the nearest monitoring station to the residential locations reported each year from birth to the time of measurement. Students who had lived four or more years in areas with long-term mean O_3 levels above 80 ppb had significantly lower FEV_1 (−3.07% [95% CI: −0.22, −5.92]) and FEF_{25-75} (−8.11% [95% CI: −2.32, −13.90]) compared to their classmates with lower long-term O_3 exposures. Stratification by gender indicated that males had much larger effect estimates than females, which might reflect higher outdoor activity levels and corresponding higher O_3 exposure during childhood.

A similar study of 130 first year college freshmen at the University of California at Berkeley also reported significant effects of O_3 on lung function (Künzli et al., 1997; Tager et al., 1998). Enrollment was limited to students from either the San Francisco or Los Angeles, CA metropolitan areas. After controlling for city of origin, long-term O_3 exposures were associated with declines in FEF_{25-75} and FEF_{75} (forced expiratory flow after 75% of FVC has been exhaled). No effects were seen for PM_{10} and NO_2. Künzli and colleagues noted that significant changes in these mid- and end-expiratory flow measures could be considered early indicators for pathologic changes that might ultimately progress to COPD, as evidenced by animal studies that showed that the primary site of O_3 injury in the lung was the centriacinar region (Chapter 5).

Sherwin et al. (2000) examined lungs from autopsies of young residents in Miami, FL and Los Angeles, CA for centriacinar region inflammatory diseases. A trend towards greater degrees of centriacinar region alterations was observed in the lungs of Los Angeles residents compared to Miami residents, independent of a smoking effect. The results suggest that the greater extent and severity of centriacinar region alterations might be related to the higher O_3 levels in Los Angeles. Beyond the challenge of differentiating the lifetime of exposure for subjects in the two cities, various confounding factors also can impact this study. The pathogenesis of centriacinar region alteration is undoubtedly multifactorial with respiratory infection and adverse environmental influences being two major considerations. In addition, Sherwin et al. (2000) noted that the study was limited due to the relatively small number of cases available. Nonetheless, as observed by Tager (1993), the use of human postmortem specimens is of interest in future epidemiologic studies.

The results of the southern California Children's Health Study, as well as those from the European studies, provide little evidence for impacts of long-term O_3 exposures on lung function in children. However, further study is needed to better address this difficult question. There is limited evidence that young adults who grew up in high O_3 communities may have reduced lung function compared to those from low O_3 communities.

7.5.5 Chronic Ozone Exposure and Respiratory Inflammation

As noted in Chapter 6, human chamber studies have demonstrated that brief (2 to 6.6 hours) exposures to O_3 while exercising result in inflammation in the lung, including the alveolar region where gas exchange takes place. This acute exposure effect is potentially important for effects of chronic exposure because repeated inflammation can result in the release of substances from inflammatory cells that can damage the sensitive cells lining the lung. Over extended periods, repeated insults of this kind can lead to permanent damage to and restructuring of the small airways and alveoli. In addition, since inflammation is a fundamental feature of asthma, there is concern that O_3-induced inflammation can exacerbate existing asthma or perhaps promote the development of asthma among genetically pre-disposed individuals. Several studies are discussed next, examining different outcomes related to inflammation.

In a study by Kinney et al. (1996b), bronchoalveolar lavage fluids were collected in the summer and winter from a group of 19 adult joggers living and working on an island in New York harbor. The mean 1-h max O_3 concentrations for a 3-month period were 58 ppb (maximum 110) in the summer and 32 ppb (maximum 64) in the winter. PM_{10} and NO_2 concentrations were similar across the two seasons. There was little evidence for acute inflammation in bronchoalveolar lavage fluids collected during the summer as compared to that collected from the same subjects in the winter. However, there was evidence of enhanced cell damage, as measured by lactate dehydrogenase, in the summer lavage fluids. These results indicate that acute inflammatory responses may diminish with repeated exposures over the course of a summer (which have been demonstrated in multiday chamber exposures, Chapter 6, Section 6.9) but cell damage may be ongoing.

Pollution effects in the nose can be viewed as a potential surrogate measure for effects that may occur in the lungs, though doses to nasal tissues are usually higher for a given pollutant concentration. In Chapter 5, morphological effects of O_3 on the upper respiratory tract indicated

quantitative changes in the nasal transitional respiratory epithelium. The persistent nature of the O_3-induced mucous cell metaplasia in rats, as discussed in Chapter 5, suggests that O_3 exposure may have the potential to induce similar long-lasting alterations in the airways of humans. A series of interesting studies in Mexico City have demonstrated inflammation and genetic damage to cells in the nasal passages of children chronically exposed to O_3 and other air pollutants (Calderón-Garcidueñas et al., 1995, 1997, 1999, 2001, 2003). Nasal lavage samples and nasal biopsies from children living in Mexico City were compared to those from children living in a clean coastal town with no detectable air pollutants. In the first study, urban children (n = 38) from Mexico City were found to have significantly higher polymorphonuclear leukocyte counts and abnormal nasal cytologies compared to nonurban children (n = 28) (Calderón-Garcidueñas et al., 1995). A more recent study of similar design examined nasal abnormalities and serum cytokines in both urban and nonurban children (Calderón-Garcidueñas et al., 2003). Twenty-two percent of the 112 Mexico City children showed a grossly abnormal nasal mucosa. No significant abnormalities were observed in the control children. In addition, the Mexico City children had more serum interleukin-10 and interleukin-6, and less serum interleukin-8 than controls. Twenty-five children with whitish-gray nasal lesions showed a significant association between tumor necrosis factor α and interleukin-8 ($r = 0.89$, $p < 0.0001$), which suggested the potential importance of the nose in the production of proinflammatory cytokines.

Calderón-Garcidueñas et al. (1997) also observed that cells collected from the lining of the nose had significantly higher amounts of DNA damage in the urban children in Mexico City (n = 129) versus nonurban children (n = 19). Among exposed children, the extent of DNA damage was greater in older children, who had spent more time outdoors and were more engaged in physical activities compared to the younger children. Another study of 86 urban and 12 nonurban children reported similar findings, and also noted increased levels of specific DNA mutations (Calderón-Garcidueñas et al., 1999). Fortoul et al. (2003) examined DNA strand breaks in nasal epithelial cells from asthmatic and nonasthmatic medical students in Mexico City and noted greater genotoxic damage in asthmatics. These results indicate that asthmatics may have a greater susceptibility for DNA damage, or a decreased ability to repair it, compared to nonasthmatic subjects. However, because of the complex mixture of pollutants present in Mexico City, it is not possible to uniquely attribute these observed changes to O_3 concentrations.

Another outcome of inflammation was examined in a study by Frischer et al. (2001). In this cross-sectional study, urinary eosinophil protein was analyzed as a marker of eosinophil activation in 877 school children living in nine Austrian communities with varying O_3 exposure. The results indicated that O_3 exposure was significantly associated with eosinophil inflammation.

In the Mexico City studies, specific attribution of these adverse respiratory and genotoxic effects to O_3 is difficult given the complex pollutant mixture present in the ambient air. In particular, the DNA effects seem more plausibly related to other components of urban air, such as semi-volatile organic compounds. However, the inflammatory changes such as increased eosinophil levels observed in the Austrian study would be consistent with known effects of O_3.

7.5.6 Risk of Asthma Development

Recent longitudinal cohort studies have reported associations between the onset of asthma and long-term O_3 exposures (McConnell et al., 2002; McDonnell et al., 1999). Significant associations between new cases of asthma among adult males and long-term O_3 exposure were observed in a cohort of nonsmoking adults in California (Greer et al., 1993; McDonnell et al., 1999). The Adventist Health and Smog (AHSMOG) study cohort of 3,914 (age 27-87 years, 36% male) was drawn from nonsmoking, non-Hispanic white California Seventh Day Adventists. Subjects were surveyed in 1977, 1987, and 1992. To be eligible, subjects had to have lived 10 or more years within 5 miles of their current residence in 1977. Residences from 1977 onward were followed and linked in time and space to interpolate concentrations of O_3, PM_{10}, SO_2, and NO_2. New asthma cases were defined as self-reported doctor-diagnosed asthma at either the 1987 or 1992 follow-up questionnaire among those who had not reported having asthma upon enrollment in 1977. During the 10-year follow-up (1977-1987), the incidence of new asthma was 2.1% for males and 2.2% for females (Greer et al., 1993). A relative risk of 3.12 (95% CI: 1.16, 5.85) per 10 ppb increase in annual mean O_3 (exposure metric not stated) was observed in males, compared to a relative risk of 0.94 (95% CI: 0.65, 1.34) in females. In the 15-year follow-up study (1977-1992), 3.2% of the eligible males and a slightly greater 4.3% of the eligible females developed adult asthma (McDonnell et al., 1999). For males, the relative risk of developing asthma was 2.27 (95% CI: 1.03, 4.87) per 30 ppb increase in 8-h avg

O_3 (9 a.m.-5 p.m.). Once again, there was no evidence of an association between O_3 and new-onset asthma in females (relative risk of 0.85 [95% CI: 0.55, 1.29]). The lack of an association does not necessarily indicate no effect of O_3 on the development of asthma among females. For example, differences in time-activity patterns in females and males may influence relative exposures to O_3, leading to greater misclassification of exposure in females. The consistency of the results in the two studies with different follow-up times and indices of O_3 exposure provides supportive evidence that long-term O_3 exposure may be associated with asthma incidence in adult males. However, as the AHSMOG cohort was drawn from a narrow subject definition, the representativeness of this cohort to the general U.S. population may be limited.

A similar study of incident asthma cases in relation to O_3 among children was carried out in the Children's Health Study (McConnell et al., 2002). 3,535 initially nonasthmatic children (ages 9 to 16 years at enrollment) were followed for up to 5 years to identify new-onset asthma cases. Communities were stratified by pollution levels, with six high-O_3 communities (mean 1-h max O_3 of 75.4 ppb [SD 6.8] over four years) and six low-O_3 communities (mean 50.1 ppb [SD 11.0]). A total of 265 children reported a new diagnosis of asthma during the follow-up period. Asthma risk was not higher for residents of the six high-O_3 communities versus residents of the six low-O_3 communities. However, within the high-O_3 communities, asthma risk was 3.3 (95% CI: 1.9, 5.8) times greater for children who played three or more sports as compared with children who played no sports. This association was absent in the low-O_3 communities (relative risk of 0.8 [95% CI: 0.4, 1.6]). No associations with asthma were seen for PM_{10}, $PM_{2.5}$, NO_2, or inorganic acid vapors. These results suggest effect modification of the impacts of O_3 on asthma risk by physical activity. Playing sports may indicate outdoor activity when O_3 levels are higher and an increased ventilation rate, which may lead to increased O_3 exposure. It should be noted, however, that these findings were based on a small number of new asthma cases (n = 29 among children who played three or more sports) and it is not clear to what extent the key findings were based on a priori hypotheses. Replication of these findings in other cohorts would lend greater weight to a causal interpretation.

Recent cross-sectional surveys have detected no associations between long-term O_3 exposures and asthma prevalence, asthma-related symptoms, or allergy to common aeroallergens in children after controlling for covariates (Charpin et al., 1999; Kuo et al., 2002; Ramadour et al., 2000). It should be noted that O_3 levels were quite low in all cases, with a range of 16 to

27 ppb for 8-h max O_3. The longitudinal study design, which observes new onset of asthma prospectively, provides stronger evidence on the question of asthma development.

7.5.7 Respiratory Effects of Chronic Ozone Exposure on Susceptible Populations

Studies on the effect of long-term O_3 exposure on respiratory health has focused mostly on children, a potentially susceptible population. Ozone exposure was associated with smaller increases in lung function and respiratory inflammation in children. Other studies have investigated additional groups of potentially susceptible individuals. McConnell et al. (1999) examined the association between O_3 levels and the prevalence of chronic lower respiratory tract symptoms in southern California children with asthma (n = 3,676). In this cross-sectional study, bronchitis, phlegm, and cough were not associated with annual mean O_3 concentrations in children with asthma or wheeze. All other pollutants examined, PM_{10}, $PM_{2.5}$, NO_2, and gaseous acid, was associated with an increase in phlegm, but not cough.

In another analysis from the Children's Health Study, McConnell et al. (2003) investigated the relationship between air pollutants and bronchitic symptoms among 475 children with asthma. For a 1 ppb increase in 8-h avg O_3 concentrations averaged over 4 years, the between-community odds ratio was 0.99 (95% CI: 0.98, 1.01) compared to the within-community odds ratio of 1.06 (95% CI: 1.00, 1.12). The authors commented that if the larger within-community effect estimates were correct, then other cross-sectional (between-community) studies might have underestimated the true effect of air pollution on bronchitic symptoms in children. These differences might be attributable to confounding by poorly measured or unmeasured risk factors that vary between communities. In two-pollutant models, the within-community effect estimates for O_3 were markedly reduced and in some cases no longer significant (odds ratios not provided). However, given the high correlation between O_3 and the other pollutants, a causal role for O_3 should not be excluded.

One recent study examined a susceptible group not examined before. Goss et al. (2004) investigated the effect of O_3 on pulmonary exacerbations and lung function in individuals with cystic fibrosis over the age of 6 years (n = 11,484). The study included patients enrolled in the Cystic Fibrosis Foundation National Patient Registry. The registry contained demographic and clinical data collected annually at accredited centers for cystic fibrosis. In 1999 and 2000, the

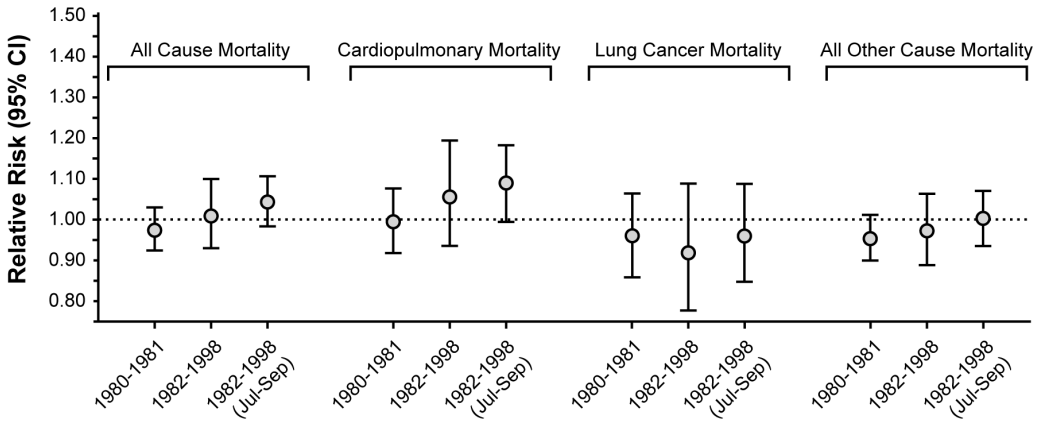
annual mean O_3 concentration from 616 monitors in the U.S. EPA Aerometric Information Retrieval System (AIRS) was 51.0 ppb (SD 7.3). Exposure was assessed by linking air pollution values from AIRS with the patient's home zip code. No clear association was found between annual mean O_3 and lung function parameters. However, a 40 ppb increase in annual mean 1-h max O_3 was associated with a 46% (95% CI: 13, 87) increase in the odds of two or more pulmonary exacerbations. Significant excess odds of pulmonary exacerbations also were observed with increased annual mean PM_{10} and $PM_{2.5}$ concentrations.

In summary, some studies have identified and investigated potentially susceptible populations. Although effects are not specific to O_3 exposure, the results suggest that O_3 may contribute to the adverse respiratory health responses observed in individuals with asthma and cystic fibrosis.

7.5.8 Effects of Chronic Ozone Exposure on Mortality and Cancer Incidence

There is inconsistent and inconclusive evidence for a relationship between long-term O_3 exposure and increased mortality and cancer risk (see Table AX7-7 in the Chapter 7 Annex). In a large prospective cohort study of approximately 500,000 U.S. adults, Pope et al. (2002) examined the effects of long-term exposure to air pollutants on mortality. All cause, cardiopulmonary, lung cancer, and all other cause mortality risk estimates for long-term O_3 exposure are shown in Figure 7-24. Consistent positive associations were not observed between O_3 and mortality. The mortality risk estimates were larger when analyses were restricted to the summer months (July to September) when O_3 levels were generally higher. The O_3-mortality risk estimates were positive for all cause and cardiopulmonary mortality, with a marginally significant estimate for cardiopulmonary mortality in the summer months. A negative, nonsignificant O_3 risk estimate was observed for lung cancer mortality. Consistent positive and significant effects of $PM_{2.5}$ were observed for both lung cancer and cardiopulmonary mortality.

Lipfert et al. (2000b, 2003) reported positive effects on all cause mortality for peak O_3 exposures (95th percentile levels) in the U.S. Veterans Cohort study of approximately 50,000 male middle-aged men recruited with a diagnosis of hypertension. The actual analysis involved smaller subcohorts based on exposure and mortality follow-up periods. Four separate exposure periods were associated with three mortality follow-up periods. In a preliminary screening of regression results, Lipfert et al. (2000b) observed a negative association for mean O_3 and a



Years of Data Collection	No. of Metropolitan Areas	No. of Participants in Thousands	1-h Max O_3 Mean (SD)
1980	134	569	47.9 ppb (11.0)
1982-1998	119	525	45.5 ppb (7.3)
1982-1998 (Jul-Sep)	134	557	59.7 ppb (12.8)

Figure 7-24. Adjusted O_3-mortality relative risk estimates (95% CI) by cause of mortality and time period of analysis per subject-weighted mean O_3 concentration in the Cancer Prevention Study II by the American Cancer Society.

Source: Derived from Pope et al. (2002).

positive relationship for peak O_3; thus, peak O_3 was used in subsequent analyses. The mean of the peak values ranged from 85 to 140 ppb over the four exposure periods. For concurrent exposure periods, peak O_3 was positively associated with all cause mortality, with a 9.4% (95% CI: 0.4, 18.4) excess risk per mean 95th percentile O_3 less estimated background level (not stated). When exposure periods preceding death were considered, no association between O_3 and mortality was observed (−0.2% [95% CI: −12.5, 12.1]). In a further analysis, Lipfert et al. (2003) reported the strongest positive association for concurrent exposure to peak O_3 for the subset with low diastolic blood pressure during the period of 1982-1988. Once again, the O_3 effect was diminished when exposure (1982-1988) preceded mortality (1989-1996).

A long-term prospective cohort study (AHSMOG) of 6,338 nonsmoking, non-Hispanic white individuals living in California examined the association between air pollutants and lung cancer incidence (Beeson et al., 1998). Over the follow-up period of 1977 to 1992, 20 females

(35% smokers, n = 7) and 16 males (37.5% smokers, n = 6) developed lung cancer. An association was observed between long-term O_3 exposure and increased incidence of lung cancer in males only. The relative risk for incident lung cancer among males was 3.56 (95% CI: 1.35, 9.42) for an interquartile range increase in hours per year (556 hours/year) when O_3 levels exceeded 100 ppb (Beeson et al., 1998). A stronger association was observed in males who never smoked (4.48 [95% CI: 1.25, 16.04]) compared to those who smoked in the past (2.15 [95% CI: 0.42, 10.89]) (Beeson et al., 1998).

A related study by Abbey et al. (1999) examined the effects of long-term O_3 exposure on all cause (n = 1,575), cardiopulmonary (n = 1,029), nonmalignant respiratory (n = 410), and lung cancer (n = 30) mortality in the same AHSMOG study population. A particular strength of this study was the extensive effort devoted to assessing long-term air pollution exposures, including interpolation to residential and work locations from monitoring sites over time and space. No associations with long-term O_3 exposure were observed for all cause, cardiopulmonary, and nonmalignant respiratory mortality. However, effects of O_3 on lung cancer mortality confirmed the results of the previous study by Beeson and colleagues. An association between lung cancer mortality and chronic O_3 exposure was observed in males only, with a relative risk of 4.19 (95% CI: 1.81, 9.69) (Abbey et al., 1999). The gender-specific O_3 effects may be partially attributable to the differences in activity and time spent outdoors. The questionnaires indicated that males spent approximately twice as much time outdoors and performed more vigorous exercises outdoors, especially during the summer, compared to the females. However, the very small numbers of lung cancer deaths (n = 12 for males) raise concerns with regard to the precision of the effect estimate, as evidenced by the wide confidence intervals. The lack of an association of chronic O_3 exposure with other mortality outcomes, which had much larger samples sizes, also is of concern. A study by Pereira et al. (2005) provides supportive evidence of an association between O_3 and increase risk of cancer. The correlation between average air pollution data from 1981 to 1990 and cases of larynx and lung cancer in 1997 were assessed in communities of São Paulo, Brazil. Of all the pollutants examined (PM_{10}, NO_2, NO_x, SO_2, CO, and O_3), O_3 was best correlated with cases of larynx ($r = 0.9929$, $p = 0.007$) and lung cancer ($r = 0.7234$, $p = 0.277$).

Few studies have examined the effect of chronic O_3 exposure on mortality outcomes and incidence of cancer. Consistent associations with long-term O_3 exposure were not observed for

all cause and cardiopulmonary mortality. There is limited evidence supportive of an association between O_3 exposure and lung cancer incidence and mortality; however, the small number of lung cancer cases, differential effects by gender, and the lack of O_3 effects on other mortality outcomes raise concerns regarding plausibility.

7.5.9 Effects of Ozone on Birth-Related Health Outcomes

In recent years, air pollution epidemiologic studies have examined impacts on birth-related endpoints including intrauterine, perinatal, postneonatal, and infant deaths; premature births; intrauterine growth retardation; very low birth weight (weight <1500 grams) and low birth weight (weight <2500 grams); and birth defects. However, the majority of these studies did not examine the effect of O_3. In the limited studies that investigated O_3, no associations were observed between O_3 and birth outcomes, with the exception of birth defects. The following is a synopsis of the literature on this topic.

Pereira et al. (1998) investigated impacts of air pollution on intrauterine mortality in São Paulo, Brazil during 1991 and 1992. NO_2, SO_2, CO, O_3, and PM_{10} were examined. Intrauterine mortality was most significantly associated with NO_2, and less for SO_2 and CO. No association was found for O_3 or PM_{10}. Pereira et al. also sampled blood from the umbilical cord of healthy non-smoking pregnant women soon after delivery in 1995 and analyzed for levels of carboxyhemoglobin. They found an association between carboxyhemoglobin and ambient CO after adjusting for passive smoking and weight, suggesting an impact of CO on the fetus. Loomis et al. (1999) examined the association between air pollutants and infant mortality in Mexico City in the years 1993 to 1995. NO_2, SO_2, CO, O_3, and $PM_{2.5}$ were examined. They reported that the strongest association was found for $PM_{2.5}$ with a 3- to 5-day cumulative lag. They noted that infant mortality also was associated with NO_2 and O_3 at a 3- to 5-day lag, but not as consistently as $PM_{2.5}$. There have been air pollution studies that examined postneonatal mortality (Bobak and Leon, 1992; Bobak and Leon, 1999; Kaiser et al., 2004; Woodruff et al., 1997), but these studies did not examine O_3.

Ritz and Yu (1999) investigated the effects of ambient CO on low birth weight among children born in southern California between 1989 and 1993. They focused on CO because "a biologic mechanism for fetal effects has been proposed for CO, but not for other air

pollutants." They found that exposure to higher levels of ambient CO during the last trimester was associated with an increased risk for low birth weight. Using available data, they also estimated the last-trimester exposures for NO_2, O_3, and PM_{10}. NO_2 and PM_{10} were positively associated with CO, but O_3 was negatively associated with CO ($r = -0.65$). Ha et al. (2001; a GAM analysis using default convergence parameters) examined CO, NO_2, SO_2, O_3, and TSP for their associations with low birth weight in Seoul, Korea during the years 1996 and 1997. They estimated first and third trimester exposures by averaging daily air pollution levels during the corresponding days for the registered births. Ha et al. found that first trimester exposures of CO, NO_2, SO_2, and TSP were associated with increased risk of low birth weight, whereas O_3 was associated with a decreased risk. The opposite pattern was observed for third trimester exposures, with an increased risk of low birth weight found only for O_3. When exposures from both trimesters were examined simultaneously, the associations of first trimester exposures of CO, NO_2, SO_2, and TSP with increased risk of low birth weight remained; however, the association between third trimester O_3 exposure and low birth weight was diminished. Based on these results, Ha et al. concluded that exposures to CO, NO_2, SO_2, and TSP in the first trimester were risk factors for low birth weight. Note that neither of these studies examined the air pollution effect by season. Other studies that examined the associations between air pollution and low birth weight (Bobak, 2000; Bobak and Leon, 1999; Lin et al., 2001; Maisonet et al., 2001; Wang et al., 1997) did not examine O_3 data, and found associations between low birth weight and either one or more of CO, SO_2, NO_2 and PM indices. Collectively, these results do not indicate strong evidence of the role of O_3 in low birth weight.

Two studies by Dejmek et al. (1999; 2000) examined the relationship between ambient air pollution and risk of intrauterine growth retardation in a highly polluted area of Northern Bohemia (Teplice District). Both studies, however, focused on PM indices and did not analyze gaseous pollutants.

A few studies have examined the association between air pollution and premature births (Bobak, 2000; Ritz et al., 2000; Xu et al., 1995), but only Ritz et al. (2000) included O_3 in their analysis. Ritz et al. evaluated the effect of air pollution exposure during pregnancy on the occurrence of preterm birth in a cohort of 97,518 neonates born in southern California. CO, NO_2 SO_2, O_3, and PM_{10} data measured at 17 air quality monitoring stations were used to estimate the

average exposures for the first month and the last 6 weeks of pregnancy. They found associations between PM_{10} levels averaged for the last 6 weeks of pregnancy as well as PM_{10} levels averaged over the first month of pregnancy. Similar but weaker associations were found for CO. No association was found for O_3. The reported correlation matrix indicated that O_3 was negatively correlated with CO ($r = -0.45$) and only weakly correlated with PM_{10} ($r = 0.2$). The results from Beijing, China (Xu et al., 1995) and the Czech Republic (Bobak, 2000) suggested that SO_2 and TSP were associated with preterm births. Considering that O_3 tends to be negatively correlated with winter-type pollutants, O_3 is unlikely to be an important risk factor for preterm births.

Ritz et al. (2002) evaluated the effect of air pollution on the occurrence of birth defects in neonates and fetuses delivered in southern California from 1987 to 1993 as ascertained by the California Birth Defects Monitoring Program. They averaged air pollution (CO, O_3, PM_{10}, and NO_2) levels measured at the assigned ambient station over the first, second, and third month of gestation. Conventional, polytomous, and hierarchical logistic regressions were used to estimate odds ratios for subgroups of cardiac and orofacial defects. Concentration-response relationships were observed for second month CO exposure on ventricular septal defects, and second month O_3 exposure on aortic artery and valve defects, pulmonary artery and valve anomalies, and conotruncal defects. The odds ratios observed for these outcomes were similar and quite large (e.g., the odds ratios comparing the highest [monthly 24-h avg mean 34.9 ppb] to lowest [monthly mean 6.4 ppb] O_3 quartiles ranged from 2.0 to 2.7), and were not sensitive in multipollutant models. Ritz et al. reported that they did not observe consistently increased risks and concentration-response patterns for NO_2 and PM_{10} after controlling for the effects of CO and O_3. Results from this study were in contrast to those with other birth-related outcomes in that both CO and O_3, presumably negatively correlated pollutants, were associated with birth defects. Further, O_3 showed associations with more birth defect outcomes compared to CO. It should be noted, however, that the concentration-response relationships were quite specific to exposures during the second month. Associations with third month exposures were often negative (though not significantly). Since both CO and O_3 show strong seasonal peaks, it is possible that seasonal confounding could have played some role in these associations. This is the only study to date that examined the relationship between air pollution and birth defects.

In summary, O_3 was not an important predictor of birth-related outcomes including intrauterine and infant mortality, premature births, and low birth weight. Birth-related outcomes generally appear to be associated with air pollutants that tend to peak in the winter and are possibly traffic-related, most notably CO. The strong results for CO are consistent with its ability to cross the placental barrier and the high affinity that hemoglobin in fetal blood has for binding with it. However, most of these studies did not analyze the data by season, and therefore seasonal confounding may have influenced the reported associations. One study reported associations between exposures to O_3 in the second month of pregnancy and birth defects. Since the O_3 effect estimates were relatively large (odds ratios $\geq$2.0 at the highest O_3 quartile), the potential role of O_3 on birth defects should be further investigated.

7.5.10 Summary of Chronic Ozone Exposure Effects on Morbidity and Mortality

- In the past decade, important new longitudinal studies have examined the effect of chronic O_3 exposure on respiratory health outcomes, including seasonal declines in lung function, increases in inflammation, and development of asthma in children and adults. Seasonal O_3 effects on lung function have been reported in several studies; however, it remains uncertain to what extent these changes are transient. There is suggestive evidence that chronic exposure to O_3 also may be associated with airway inflammation. In contrast to the supportive evidence from chronic animal studies, epidemiologic studies of new asthma development and longer-term lung function declines remain inconclusive at present.

- Few studies have investigated the effect of long-term O_3 exposure on mortality and cancer incidence. Uncertainties regarding the exposure period of relevance, differential effects by gender, and inconsistencies across outcomes raise concerns regarding plausibility. There is currently little evidence for a relationship between chronic O_3 exposure and increased risk of mortality.

- A limited number of studies have examined the relationship between air pollution and birth-related health outcomes, including mortality, premature births, low birth weights, and birth defects. The most consistent associations with various birth outcomes were observed for CO. One study reported a large effect of O_3 on cardiac defects. The potential role of O_3 on birth defects needs to be further examined.

7.6 INTERPRETIVE ASSESSMENT OF THE EVIDENCE IN EPIDEMIOLOGIC STUDIES OF OZONE HEALTH EFFECTS

7.6.1 Introduction

In the 1996 O_3 AQCD, the epidemiologic section focused primarily on individual-level camp and exercise studies, and studies of hospital admissions and emergency room visits. The field studies indicated concentration-response relationships of O_3 exposure from the ambient air with declines in pulmonary function, increases in respiratory symptoms, and exacerbation of asthma, especially in children. Numerous new studies provide additional evidence for evaluating associations between O_3 exposure and the above respiratory health outcomes. The 1996 O_3 AQCD review of aggregate population time-series studies indicated an association between ambient O_3 concentrations and increased hospitalizations. Limited studies examined the O_3-mortality relationship. The current O_3 AQCD further presents results from time-series studies that have addressed previously unresolved issues regarding potential linkages between ambient O_3 concentrations and health outcomes, particularly mortality. Daily time-series studies minimize confounding by population characteristics (e.g., cigarette smoking, diet, occupation) by following the same population from day to day. However, confounders operating over shorter time scales can affect O_3 risk estimates in these studies.

In this section, the issues and attendant uncertainties that affect the interpretation of O_3 health effects will be discussed. The use of various indices to represent O_3 exposure in epidemiologic studies is discussed first. Also, of interest is the issue of confounding by temporal factors, meteorological factors, and copollutants. The shape of the concentration-response function and heterogeneity of O_3 effects also will be discussed briefly. All of these topics are of much importance for characterizing and interpreting ambient O_3-health effects associations.

7.6.2 Ozone Exposure Indices

Three O_3 indices were used most often to indicate daily O_3 exposure: maximum 1-h average (1-h max); maximum 8-h average (8-h max); and 24-h average (24-h avg). The 8-h max O_3 is a frequently used index in newer epidemiologic studies, as it best reflects the current U.S. EPA NAAQS. The O_3 exposure indices are highly correlated as indicated in several studies. In the 21 European multicities acute mortality study (Gryparis et al., 2004), 1-h max O_3 was found to be highly correlated with 8-h max O_3, with a median correlation coefficient of 0.98

(range 0.91–0.99). Among single-city studies, the 1-h max O_3 and 8-h max O_3 also were found to have correlation coefficients ranging from 0.91 to 0.99 in various cities such as Atlanta, GA (Tolbert et al., 2000; White et al., 1994); southern New England (Gent et al., 2003); Ontario, Canada (Burnett et al., 1994); and Mexico City (Loomis et al., 1996; Romieu et al., 1995). In addition, 1-h max O_3 was highly correlated with 24-h avg O_3, as observed in the Mexico City study by Loomis et al. (1996) ($r = 0.77$) and in the Ontario, Canada study by Burnett et al. (1994) ($r = 0.87$).

All studies discussed in Sections 7.2 to 7.5 were examined for presentation of the three O_3 exposure indices. Several presented the concentration data and correlations among 1-h max, 8-h max, and 24-h avg O_3 ambient measures. Some presented the associated risk estimates of comparable analyses for the three exposure indices. No papers provided a statistical analysis comparing results from the different indices. Summary of the available data is provided below starting with two multicity mortality studies.

In the large U.S. 95 communities study by Bell et al. (2004), increases in O_3-associated daily mortality were estimated using all three O_3 indices. The increments used in this document to standardize expressions of excess risks are 40 ppb for 1-h max O_3, 30 ppb for 8-h max O_3, and 20 ppb for 24-h avg O_3, as discussed in Section 7.1.3.2. For these increments, the effect estimates calculated by Bell et al. (2004) using all available data were 1.34% (95% PI: 0.84, 1.85), 1.28% (95% PI: 0.88, 1.73), and 1.04% (95% PI: 0.54, 1.55) excess risk in mortality for 1-h max O_3, 8-h max O_3, and 24-avg O_3, respectively. A statistical test examining differences among these risk estimates indicated that there were no significant differences by exposure index. In the European study of 21 cities (of the 23 cities, two did not have 8-h max O_3 data), the O_3-mortality effect estimate for the summer season was slightly smaller for 8-h max O_3, 1.82% (95% CI: 0.99, 3.06) excess risk, compared to 1-h max O_3, 2.59% (95% CI: 1.32, 4.10) excess risk; however, the two risk estimates were not significantly different (Gryparis et al., 2004).

Several single-city mortality studies examined multiple O_3 exposure indices (Anderson et al., 1996; Dab et al., 1996; Sunyer et al., 2002; Zmirou et al., 1996; Borja-Aburto et al., 1997). These studies did not differentiate risk estimates by exposure index as the results were considered similar. Hospital admission studies also provided limited data for O_3 index comparisons. Schouten et al. (1996) found similar O_3 effects on total respiratory hospitalizations

from 8-h max O_3 and 1-h max O_3 in the summer. Both indices resulted in a 4.0% excess risk per standardized increment. For emergency department visits, the examples of Delfino et al. (1998b) and Weisel et al. (2002) indicated no difference in effect estimate when using various O_3 indices. Tolbert et al. (2000) noted an increase in emergency room visits of 4.0% per standard deviation increase (approximately 20 ppb) for both 1-h max O_3 and 8-h max O_3 as being expected since the correlation between the indices was 0.99.

Peak flow asthma panel studies generally used only one index; thus, there were limited data available for comparison. One respiratory symptom study (Gent et al., 2003) examined both 1-h max O_3 and 8-h max O_3 but noted no differences in the results. Only one FEV_1 panel study examined more than one O_3 exposure index. Chen et al. (1999) examined 1-h max O_3 and 24-h avg O_3 and reported a decrement in FEV_1 of −25.6 mL (95% CI: −49.1, −2.1) for 1-h max O_3 and −13.6 mL (95% CI: −33.2, 6.0) for 24-h avg O_3 in children at a 1-day lag. For 2- and 7-day lags, smaller differences were observed between the two indices. Despite the apparent differences, the effect estimates calculated using 1-h max O_3 and 24-h avg O_3 concentrations were not found to be significantly different for any of the lags examined.

Limited information is available to reach conclusions for comparison of the three indices 1-h max O_3, 8-h max O_3, and 24-h avg O_3. Studies conducted in various cities have observed very high correlations among the daily O_3 indices. For the same distributional increment, the excess health risk estimates and significance of associations were generally comparable for the three O_3 indices across all outcomes. The high correlation among the indices presents a challenge in distinguishing the most appropriate measure for epidemiologic studies. Exploratory analyses using various O_3 exposure indices are valuable in understanding relationships. However, to address the issue of multiple hypothesis testing, hypotheses that are confirmatory and exploratory should be decided a priori and reported accordingly.

7.6.3 Confounding by Temporal Trends and Meteorologic Effects in Time-Series Studies

The challenge of analyzing acute O_3 effects in time-series studies is to avoid bias due to confounding by daily to seasonal temporal factors. On a seasonal scale, the analysis must remove the influence of the strong seasonal cycles that usually exist in both health outcomes and O_3. On a daily scale, weather factors and other air pollutants also may confound the association

of interest. This section discusses the interpretation of effect estimates after adjusting for temporal trends and meteorologic effects.

7.6.3.1 Assessment of Ozone Effects after Adjusting for Temporal Trends and Meteorologic Effects

The relationship between O_3 and health outcomes are significantly affected by temporal trends and meteorological factors, namely temperature. Analyses of the association between health outcomes and O_3 concentrations using raw data, therefore, can be misleading. In Díaz et al. (1999), a U-shaped relationship was observed between mortality and O_3 concentrations, in which the negative portion of the slope was likely due to the opposing seasonal cycles in mortality (high in winter) and temperature (low in winter). Goldberg and Burnett (2003) report a positive slope for the temperature-mortality relationship being fitted most tightly in the mild temperature range where mortality effects of temperature are not expected. It is possible that temperature has mortality effects in the mild temperature range, however because daily fluctuations of air pollution, especially O_3, are strongly influenced by weather conditions, ascribing the association between temperature and mortality entirely to effects of temperature may underestimate the effects of air pollution.

Sensitivity analyses specifically for O_3 effects were performed in the U.S. 95 communities data by Bell et al. (2004). They found that varying the degrees of freedom from 7 to 21 per year did not significantly affect the O_3-mortality estimates, with effect estimates ranging from 0.82 to 1.08% excess risk per 20 ppb increase in 24-h avg O_3 during the previous week. Using more degrees of freedom in temporal trend fitting (i.e., controlling shorter temporal fluctuations) means ascribing more details of daily health outcomes to unmeasured potential confounders and possibly taking away real weather and air pollution effects. However, results from this large multicity study indicated that O_3 effects were robust to aggressive smoothing of temporal trends. In a related analysis of 19 U.S. cities by Huang et al. (2005), sensitivity of summertime O_3 risk estimates to varying degrees of freedom (4 to 16 per year) for temporal trend adjustment was examined. The extent of change in the risk estimates, while varied from city to city (graphically presented), was not substantial. Huang et al. concluded that the risk estimates were robust to the adjustment for long-term trends.

Ito et al. (2005) examined sensitivity of O_3-mortality risk estimates to the extent of temporal trend adjustment and to alternative weather model specifications using data from seven

U.S. cities (Cook County, IL; Detroit, MI; Houston, TX; Minneapolis, MN; New York City; Philadelphia, PA; and St. Louis, MO). They found that varying the degrees of freedom from 4 to 26 per year did not substantially or systematically affect the O_3-mortality estimates, except for Cook County where the percent excess O_3-mortality risk estimates were considerably reduced when the temporal adjustment term with 26 degrees of freedom was applied. Ito et al. noted that the O_3 risk estimates were generally more sensitive to alternative weather models than to the degrees of freedom for temporal adjustment.

Schwartz (2005) examined the sensitivity of the O_3-mortality relationship to methods used to control for temperature. Initially, temperature lagged 0 and 1 day was controlled using nonlinear regression splines with 3 degrees of freedom each. In a comparison analysis, control days were restricted to a subset that was matched on temperature. The effect estimates for all year data using nonlinear regression splines (0.8% [95% CI: 0.1, 1.4] excess risk per 40 ppb increase in 1-h max O_3) and temperature matched controls (0.9% [95% CI: 0.04, 1.8] excess risk) were not significantly different. Results were similar when restricting analysis to warm season only data.

Temporal cycles in daily hospital admissions or emergency department visits are often considerably more episodic and variable than is usually the case for daily mortality. As a result, smoothing functions that have been developed and tuned for analyses of daily mortality data may not work as well at removing cyclic patterns from morbidity counts. Two methods are commonly used to adjust for temporal trends. The pre-adjustment method involves applying the adjustment to both outcome and air pollution variables prior to the regression analysis. The co-adjustment method involves applying the adjustment as part of the regression analysis, by fitting a function of time while simultaneously fitting the regression effect of air pollution and weather factors. As shown in a hospital admissions study by Burnett et al. (2001; used Poisson GAM with default convergence criteria), the co-adjustment approach may lead to biased air pollution effect estimates in cases where both outcome and pollution variables exhibit strong seasonal cycles. Using year-round data, pre-adjustment followed by regression analysis yielded a 14% (95% CI: 5, 24) increase in admissions per 40 ppb increase in 1-h max O_3 with a multiday lag of 0 to 4 days. The co-adjustment method resulted in a 7% (95% CI: 3, 11) decrease in admissions. When the authors limited the analysis to the warm season (May-August), both methods yielded similar results (32% [95% CI: 21, 44] versus 30% [95% CI: 17,

45] increase for co-adjustment and pre-adjustment, respectively) implying that stratification by season can remove a significant amount of the confounding seasonality (which also may include seasonally-varying population behavior and ventilation conditions). This finding may be important to consider in reviewing the acute O_3 mortality and morbidity literature since the vast majority of studies published over the past decade have used the co-adjustment method. However, the use of pre-adjustment versus co-adjustment in time-series studies is an unresolved issue. More empirical research in different locales is needed to evaluate the merits of these two methods as far as O_3 is concerned, and to determine what endpoints may be affected.

More sensitivity analysis of O_3 effect estimates to the extent of adjustment for temporal trends and meteorological factors is needed, but perhaps it is equally as important to evaluate the epidemiologic adequacy of a given adjustment. For example, do the fitted mortality series sufficiently depict influenza epidemics? Or, when larger degrees of freedom (e.g., 12 degrees of freedom per year) are used, what "unmeasured" confounders, other than weather and pollution, are the investigators trying to adjust? Even in PM studies that conducted sensitivity analyses, investigators rarely stated assumptions clearly and not enough discussions were provided as to potential reasons for the sensitivity of results.

Given their relationship to health outcomes and O_3 exposure, adjusting for temporal trends and meteorologic factors is critical to obtain meaningful O_3 effect estimates. While the prevailing analytical approaches fit the data flexibly, the estimated effects of meteorologic variables and their impact on the adjusted O_3 effects are not adequately discussed. More work is needed in this area to reduce the uncertainty involved in the epidemiologic interpretation of O_3 effect estimates.

7.6.3.2 Importance of Season-Specific Estimates of Ozone Health Effects

Analysis of O_3 health effects is further complicated as relationships of O_3 with other pollutants and with temperature appear to change across seasons. Moolgavkar et al. (1995) examined the relationship between daily mortality and air pollution by season in Philadelphia, PA for the period of 1973 to 1988. During the summer, there was a positive relationship between O_3 and TSP, as well as O_3 and SO_2. In contrast, the relationship of O_3 with TSP and SO_2 inversed during the winter. Ozone showed positive associations only in the summer when the mean O_3 concentration was the highest. The effect of O_3 on mortality was negative (though

not significantly) in the winter when the mean O_3 concentration was low. In the summer multipollutant model, O_3 was the only pollutant that remained significant. Similar results were found in another Philadelphia study by Moolgavkar and Luebeck (1996). Both studies did not analyze year-round data, therefore the relationship between the excess risk estimates for all year and each season could not be compared. The results from these studies, however, suggest that year-round analyses may mask the positive (or negative) associations that may exist in particular seasons.

Ito et al. (2005) examined O_3-mortality associations in seven U.S. cities, but also described the relationship between O_3 and PM for summer months (June-August) and winter months (December-February) in these cities (see Figure 7-25). The O_3-PM relationships were positive in the summer and negative in the winter in all of these cities, except in Houston, where the O_3-PM association was not clearly positive in the warmer months but positive in colder months. Ito et al. found that O_3-mortality associations were mostly weaker, null, or even negative in the winter compared to the summer in most of these cities. Once again, the exception was Houston where the cold season O_3-mortality association was positive and larger than those for year-round or warmer months. Findings from this study suggest the influence of seasonal O_3-PM relationships on O_3-mortality associations.

In the analyses of the U.S. 90 cities data (of which 80 cities had O_3 data available) by Samet et al. (2000; reanalysis Dominici et al., 2003), the focus of the study was PM_{10}, but O_3 and other gaseous pollutants also were analyzed in single- and multiple-pollutant models. In the reanalysis (Dominici et al., 2003), O_3 was associated with an excess risk of mortality in analyses of all available data (0.4% [95% PI: 0.1, 0.7]) and summer only data (1.0% [95% PI: 0.5, 1.6]; however, a negative association was observed for the winter only analysis (−1.1% [95% PI: −2.2, 0.1]). A twofold greater effect was estimated using summer data compared to all available data. It should be noted that the analyses by Samet et al. and Dominici et al. used a weather model specification that is more detailed than other studies in that it had multiple terms for temperature and dewpoint (these two variables are generally highly correlated). Thus, it is possible that the high concurvity of O_3 with these weather covariates may have produced these conflicting results. Another possibility is that the apparent negative relationship between O_3 and mortality in the winter may have been due to confounding by PM. In the larger U.S. 95 communities study by Bell et al. (2004), the all available data and summer only analyses also

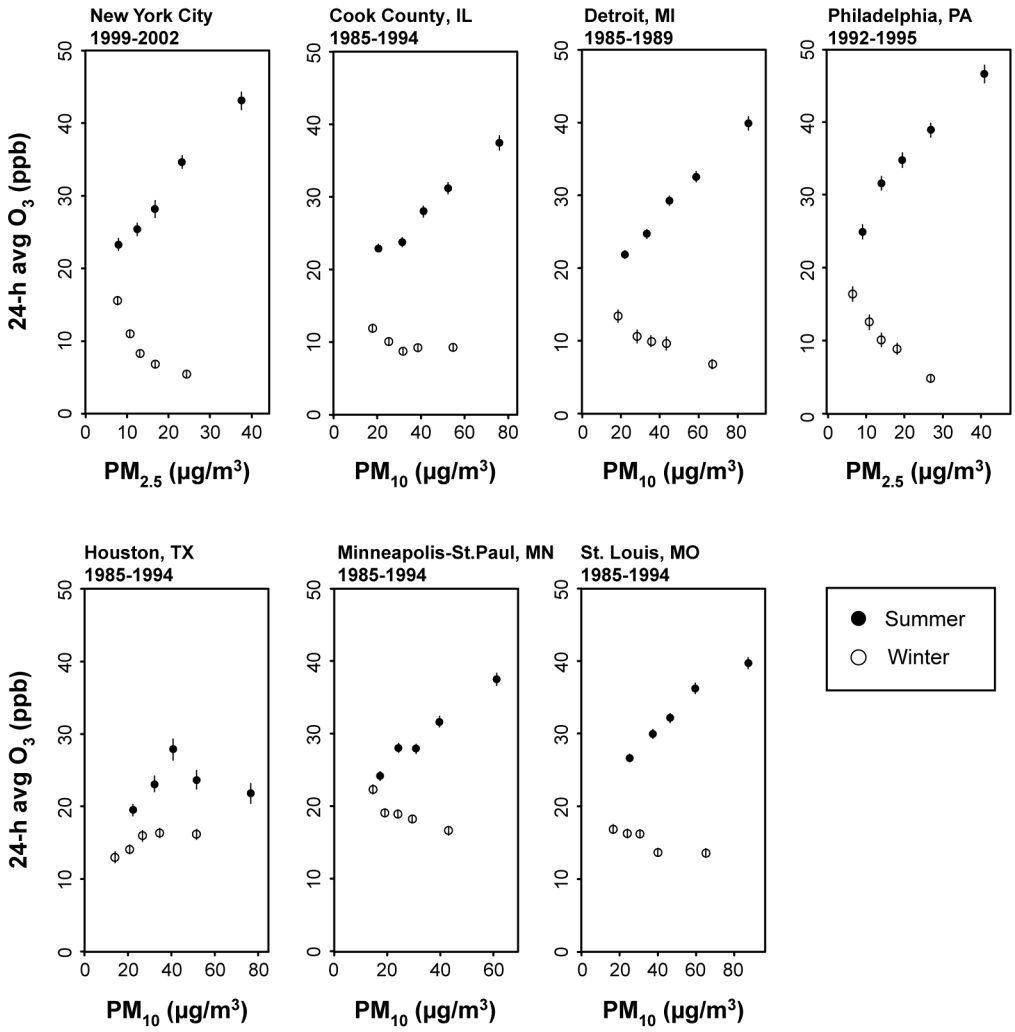


Figure 7-25. The relationship between PM and O_3 in the summer (June through August) and the winter (December through February) as sorted and averaged by quintiles of PM.

Source: Derived from Ito et al. (2005).

indicated positive risk estimates (1.04% [95% PI: 0.54, 1.55] and 0.78% [95% PI: 0.26, 1.30] excess risk per 20 ppb increase in 24-h avg O_3, respectively, using a constrained distributed 7-day lag model), but the two estimates were similar in magnitude. Winter only analyses were not performed. Note that 32 of the 95 communities only had warm season data available. Results from the U.S. 95 communities study appear to conflict with the strong seasonal variation

observed in the U.S. 90 cities study. However, there are several differences between the two studies that might account for these results. First, the U.S. 95 communities study nearly doubled the study period by extending the analysis by six additional years (1987 to 2000 versus 1987 to 1994) and included 15 additional cities to the original 80. Also note that the warm seasons are characterized differently in the two studies. The U.S. 90 cities study defined summer as a three-month period of June through August, while the 95 communities study defined warm season as a seven-month period of April through October. In addition, the results presented in the U.S. 90 cities study were from a single-day lag model (lag 1-day) while the estimates from the 95 communities study were calculated using a constrained distributed 7-day lag model. The difference in seasonal O_3 effects observed in the two related studies might be attributable to some of these factors.

Many studies reported larger excess mortality risks in the warm (or summer) season than in the cool (or winter) season (see Figure 7-19 in Section 7.4.5). These studies showed cool season risk estimates that were either smaller compared to warm season estimates or slightly negative. Of the studies that analyzed data by season, only one study in Pittsburgh, PA (Chock et al., 2000) showed negative risk estimates in the summer. The studies that observed larger, positive associations between O_3 and mortality in warm seasons are consistent with the expectation that O_3, if harmful, should have a stronger association with health outcomes in the summer when exposure to O_3 is higher. However, the negative O_3-mortality associations seen in the winter suggest that further examination of this issue is required. Specifically, if the O_3 level in the winter is shown to be negatively associated with factors (e.g., PM) that are positively associated with mortality, then these potentially spurious negative O_3-mortality associations can be explained. Several examples of this phenomenon also exist in morbidity studies investigating the effect of O_3 on daily hospital admissions and emergency department visits (Anderson et al., 1998; Burnett et al., 2001; Prescott et al., 1998; Thompson et al., 2001).

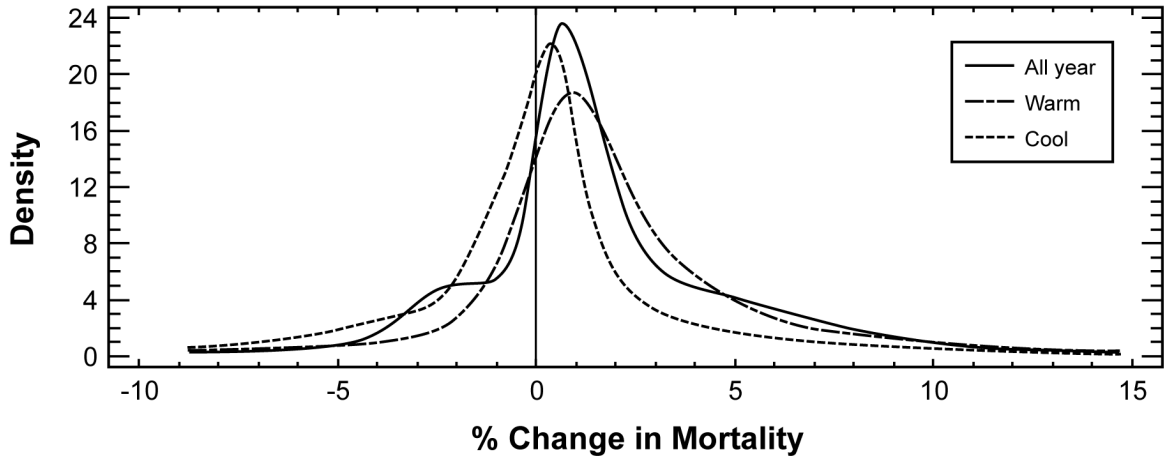
Unlike the time-series studies examining outcomes of mortality, hospital admissions, and emergency department visits, most acute field studies did not perform year-round analyses. These acute field studies that examined the relationship between O_3 and lung function, respiratory symptoms, and inflammation focused primarily on the O_3 effect during the warm season when O_3 levels were expected to be high and subjects spent more time outdoors and were physically active.

There are seasonal (e.g., air conditioning use) or seasonally-modified (e.g., time spent outdoors, air exchange rates) factors that affect the relationship between ambient concentrations and personal exposures to O_3, as discussed in Section 3.9. The influence of combinations of these factors across seasons on air pollution health effects can become quite complex. For example, longer time spent outdoors in the summer may increase personal exposure to O_3 for some segment of the population, but the increased use of air conditioners may reduce exposures to ambient O_3 for those who spend much of their time indoors. In the meta-analysis by Levy et al. (2005), the combined risk estimate from the warm season was greater (3.38% [95% CI: 2.27, 4.42] per 40 ppb increase in 1-h max O_3) compared to the estimate from all year data (1.64% [95% CI: 1.25, 2.03]). However, further analysis suggested that the O_3-mortality risk estimates were smaller in cities with high air conditioning prevalence. These seasonal factors that influence the relationship between ambient concentrations and personal exposures make the interpretation of the concentration-response relationships obtained from analyses of year-round data less straightforward.

In some cities, O_3 is only monitored during the warm season. For example, 34% of the communities in the U.S. 95 communities study only collected O_3 data during the warm season (Bell et al., 2004). The cities with larger populations and/or higher O_3 concentrations generally collected year-round data. There is some concern that differential data availability may contribute to the seasonal differences in O_3 health effects observed.

The potential influence of season on O_3 effect estimates was examined using summary density curves. The O_3 effect observed in all year data was compared to effects from warm season and cool season only data (Figures 7-26 and 7-27). Summary probability density curves were calculated to review the effect estimates from the various studies (see Annex Section AX7-2 for further explanation of summary density curves). The summary density curves shown in Figures 7-26 and 7-27 were smoothed by multiplying a constant to the standard error of each effect estimate in the calculation of the individual distribution functions. Since the normal distribution is unimodal, this constant will oversmooth when the density is multimodal.

In Figure 7-26, the summary density curves of O_3-associated all cause (nonaccidental) mortality are presented (see Figure 7-19 in Section 7.4.5 for the effect estimates). The summary density curves are calculated using results from 14 studies that reported at least two of the three estimates. This figure indicates that 75% of the area under the density curve has a value greater

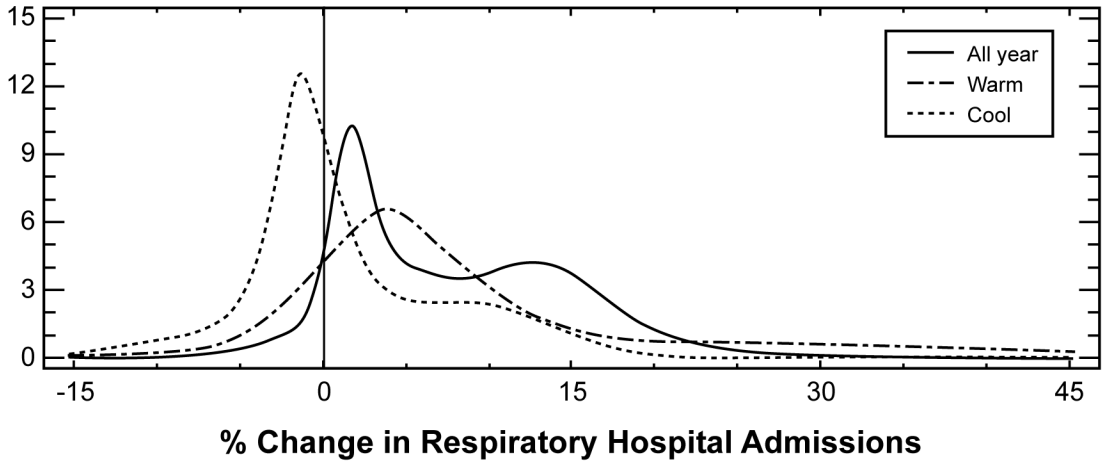


	All year	Warm season	Cool season
% area under the density curve and >0	75%	78%	62%
Mean (SD) effect estimates	1.2% (2.8%)	1.3% (2.6%)	0.1% (3.1%)
Mode effect estimates	0.7%	0.9%	0.4%

Figure 7-26. Summary density curves of the percent change in all cause mortality for all year data and by season per standardized increment (see Section 7.1.3.2). Effect estimates from 14 studies have been included in the summary density curves (see Figure 7-19 in Section 7.4.5 for the effect estimates).

than zero for all year data compared to 78% for warm season data and 62% for cool season data. Therefore, both all year and warm season data generally indicate a positive O_3 effect on mortality. The mean effect estimates for all year data and warm season only data are 1.2%(SD 2.8) and 1.3% (SD 2.6) excess risk in mortality per 40 ppb increase in 1-h max O_3, respectively. A slightly larger mode of effects is observed for warm season data (0.9% excess risk) compared to all year data (0.7%). The cool season only data indicate that there is no excess risk (mean 0.1% [SD 3.1]) associated with O_3 concentrations.

Similar observations are made when examining the O_3 effect on total respiratory hospital admissions (Figure 7-27). Six studies provided season-specific estimates as well as all year results (see Figure 7-10 in Section 7.3.3 for the effect estimates). Once again, a large percent of the area under the summary density curve is greater than zero when using all year and warm season data, 92% and 84%, respectively, compared to cool season data, 49%. The mean O_3



	All year	Warm season	Cool season
% area under the density curve and >0	92%	84%	49%
Mean (SD) effect estimates	6.5% (6.4%)	6.3% (9.1%)	0.8% (6.1%)
Mode effect estimates	1.8%	4.0%	−1.3%

Figure 7-27. Summary density curves of the percent change in total respiratory hospital admissions for all year data and by season per standardized increment (see Section 7.1.3.2). Effect estimates from six studies have been included in the summary density curves (see Figure 7-10 in Section 7.3.3 for the effect estimates).

effect estimates for warm season data only, 6.3% (SD 9.1) excess risk per 40 ppb increase in 1-h max O_3, and all year analyses, 6.5% (SD 6.4) excess risk, are similar. A larger mode of effects is observed for warm season data (3.9% excess risk) compared to all year data (1.8% excess risk). A small O_3 effect (0.8% [SD 6.1] excess risk) is observed when using cool season data only.

Integrating seasonal influences across the various health outcomes supports the view that O_3 effects are different in the cool and warm seasons, with greater effects observed during the warm season. As this relates to potentially higher O_3 exposures during the warm season, the larger effects are consistent with causal inference. Therefore, these results indicate that the focus should be on warm season data to derive quantitative relationships for the effect of O_3 on health outcomes. This conclusion is supported by epidemiologic researchers who mainly examine warm season as an a priori hypothesis. However, studying summer data only when all year data

are available weakens the power of the study since less data are analyzed. In addition, increased adverse health outcomes are observed in the winter, some of which may be attributable to O_3. The O_3 effect in the wintertime may be masked by the effects of PM due to the negative correlation between these variables (see Section 7.6.4.2 for further discussion). Therefore, analysis of all year data may be improved by adjusting for PM indices in addition to adequate adjustment of meteorological factors and temporal trends.

Seasonality influences the relationship between O_3 and health outcomes as it may serve as an indicator for time-varying factors, including temperature, copollutant concentrations, infiltration, and human activity patterns. Given the potentially significant effect of season, O_3 effect estimates computed for year-round data need to be interpreted with caution. Small or no effects may simply reflect the cancellation of positive associations in the summer and negative associations in the winter, or the presence of confounding due to the strong seasonal character of O_3 concentrations.

7.6.4 Assessment of Confounding by Copollutants

Potential confounding by daily variations in copollutants is another analytical issue to be considered. With respect to copollutants, daily variations in O_3 tend to not correlate highly with most other criteria pollutants (e.g., CO, NO_2, SO_2, PM_{10}), but may be more correlated with secondary fine PM (e.g., $PM_{2.5}$, sulfates) measured during the summer months. Assessing the independent health effects of two pollutants that are correlated over time is not straightforward. If high correlations between O_3 and PM or other gaseous pollutants exist in a given area, then disentangling their relative individual contributions to observed health effects associations becomes very difficult. The changing relationship between O_3 and other copollutants also is of issue. In some urban locations, the correlation between PM indices and O_3 is positive in the summer and negative in the winter. This section will further discuss the correlation between O_3 and copollutants and confounding of the O_3 effect by copollutants.

7.6.4.1 Relationship between Personal Exposure to Ozone and Copollutants

Ambient levels of PM, NO_2, SO_2, and CO, measured at central monitoring sites, have been found to be highly correlated to ambient O_3 concentrations. A very limited number of studies have examined the association between personal O_3 concentrations and personal exposures to

other copollutants. An issue of particular interest is the correlation between personal exposure to O_3 and personal exposure to the ambient component of $PM_{2.5}$. Only one study examined personal exposure to PM of ambient origin. In a Baltimore, MD study of susceptible populations (older adults, individuals with COPD, and children), Sarnat et al. (2001) found that ambient 24-h avg O_3 concentrations and ambient 24-h avg $PM_{2.5}$ levels were positively associated ($\beta = 0.84$, $r = 0.67$) in the summer and negatively associated ($\beta = -0.67$, $r = -0.67$) in the winter. A significant association also was observed between ambient O_3 concentrations and personal $PM_{2.5}$ of ambient origin, with a mixed regression effect estimate of $\beta = 0.37$ (95% CI: 0.25, 0.49) in the summer and $\beta = -0.36$ (95% CI: −0.31, −0.41) in the winter. However, no relationship was found between 24-h avg personal O_3 exposure and personal exposure to $PM_{2.5}$ of ambient origin. While the results from this study provide limited evidence for a lack of an association between personal O_3 levels and personal exposure to $PM_{2.5}$ of ambient origin, additional research is necessary to address this issue.

7.6.4.2 Assessment of Confounding Using Multipollutant Regression Models

Multipollutant regression models are generally used to determine whether the pollutant-specific effect is robust. However, due to the multicollinearity among O_3 and pollutants, and the changing correlations by seasons, multipollutant models may not adjust for potential confounding adequately, especially when using year-round data. These limitations need to be considered when evaluating results from multipollutant models. Results from the U.S. 90 cities study, which included 80 cities with O_3 data, indicated that while the addition of PM_{10} in the model did not substantially change the O_3-mortality risk estimates, slight declines in the O_3 effect were observed, as shown in Figure 7-28 (Samet et al., 2000; reanalysis Dominici et al., 2003). In the extended U.S. 95 communities study (Bell et al., 2004), the city-specific O_3-mortality effects were robust to adjustment for PM_{10}, as indicated by the nearly 1:1 ratio between estimates with and without PM_{10} adjustment shown in Figure 7-29. This finding suggested that PM_{10} generally did not confound the association between O_3 and mortality. Limited data were available to examine the potential confounding effect of $PM_{2.5}$ on the O_3-mortality relationship. A weighted second-stage linear regression indicated that there was no association between long-term $PM_{2.5}$ average and the community-specific O_3-mortality effect estimate. Several other mortality and morbidity studies have investigated confounding of O_3 risk estimates using

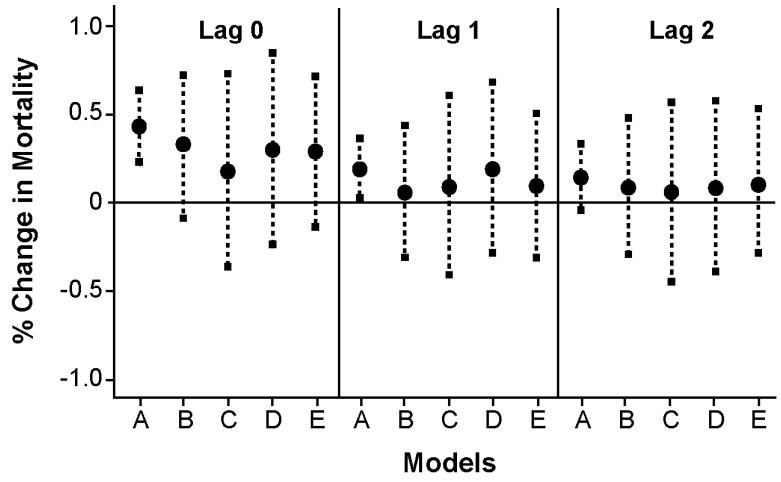


Figure 7-28. Posterior means and 95% PIs of the national average estimate of O_3 effects on total mortality from non-external causes per 10 ppb increase in 24-h avg O_3 at 0-, 1-, and 2-day lags within sets of 80 U.S. cities with pollutant data available. Models A = O_3 only; B = O_3 + PM_{10}; C = O_3 + PM_{10} + NO_2; D = O_3 +PM_{10} +SO_2; E = O_3 + PM_{10} + CO.

Source: Derived from Dominici et al. (2003).

multipollutant models with year-round data, and most have reported that O_3 effects were robust to adjustment for copollutants (see Figures 7-11 and 7-20 in Sections 7.3.3 and 7.4.6, respectively).

The pollutant most correlated with O_3 in the summer is sulfate (which is in the fine particle size range), especially in the eastern U.S. Therefore, the main potential confounders of interest for O_3 are $PM_{2.5}$ and sulfate in the summer. Once again, the results from two-pollutant regression models with O_3 and sulfate (or $PM_{2.5}$) should be interpreted with caution because both of these pollutants are formed under the same atmospheric condition and are both part of the "summer haze" pollution mix. A simple two-pollutant regression model does not address their possible synergistic effects, and the high correlation between the two pollutants may lead to unstable and possibly misleading results. In any case, most studies that analyzed O_3 with PM indices did not have $PM_{2.5}$ data and very few examined sulfate data. The studies that did have $PM_{2.5}$ data, including Santa Clara County, CA (Fairley, 1999; reanalysis Fairley, 2003), Philadelphia, PA (Lipfert et al., 2000a), and Detroit, MI (Lippmann et al., 2000; reanalysis Ito,

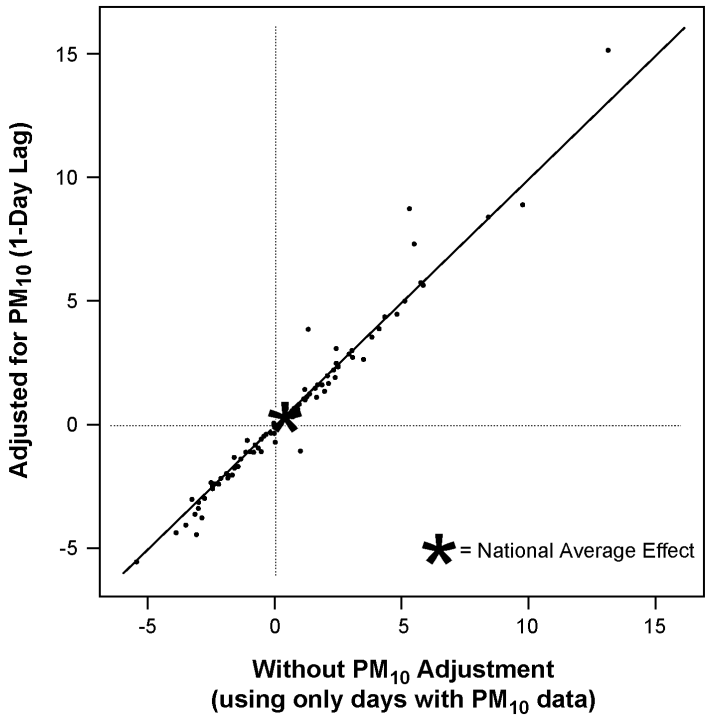


Figure 7-29. Maximum likelihood estimates of O_3-mortality for 95 U.S. communities, determined using a constrained distributed lag model for lags 0 through 6 days. Same data set was used for O_3 estimates with and without adjustment for PM_{10}.

Source: Derived from Bell et al. (2004).

2003), examined copollutant models for year-round data only, but O_3-mortality risk estimates were not substantially affected by the addition of $PM_{2.5}$. The updated analysis of Philadelphia and Detroit data by season suggested that O_3-mortality risk estimates were not sensitive to adjustment for $PM_{2.5}$ in all year or seasonal analyses (Ito et al., 2005). A mortality study by Lipfert et al. (2000a) also found that all year O_3 risk estimates were not affected by the addition of sulfate.

Other studies have estimated O_3 health risks with copollutants in the model by season. Respiratory hospitalization studies conducted during the warm season in Canada observed consistent O_3 risk estimates with the inclusion of $PM_{2.5}$ in the model (Burnett et al., 1997b, 2001). In one of these studies (Burnett et al., 1997b), the effect of O_3 also was adjusted for sulfate. With the addition of sulfate in the model, the risk estimate for O_3 on respiratory

hospitalizations remained relatively stable, from an 14.4% (95% CI: 8.7, 20.5) excess risk to a 11.7% (95% CI: 5.6, 18.0) excess risk per 25 ppb increase in 12-h avg O_3 at a 1-day lag. In contrast, the effects for sulfate were reduced in half after adjusting for O_3. Amongst the mortality studies (see Figure 7-21 in Section 7.4.6), adjusting for copollutants, in particular PM indices, did not substantially change the warm season O_3-mortality effect estimates, with both slight reductions and increases observed in the adjusted estimates. In the analysis using cool season data only, the O_3 effect estimates were generally negative, but none were statistically significant. The O_3 risk estimates all increased slightly with the adjustment of PM indices. The inverse relationship between O_3 and PM during the cool season most likely influenced the O_3-mortality effect estimates in the single-pollutant models.

In field studies, power to assess independent O_3 effects may be limited by small sample sizes and short follow-up times. Yet, the O_3 effect also was robust to the addition of copollutants in multipollutant models, with a few exceptions. For example, the effect of O_3 on PEF was not robust to adjustments for $PM_{2.5}$ and sulfate, in studies by Romieu et al. (1996) and Neas et al. (1999). In general, however, O_3 effects on respiratory symptoms (Romieu et al., 1996), lung function parameters (Brauer et al., 1996, Gold et al., 1999), and asthma medication use (Gent et al., 2003) were robust to inclusion of $PM_{2.5}$. Further, the effects for O_3 were observed to be stronger than those for PM.

Multipollutant regression analyses indicated that O_3 risk estimates, in general, were not sensitive to the inclusion of copollutants, including $PM_{2.5}$ and sulfate. These results suggest that the effect of O_3 on respiratory health outcomes appears to be robust and independent of the effects of other copollutants.

7.6.5 Concentration-Response Function and Threshold

An important consideration in characterizing the public health impacts associated with O_3 exposure is whether the concentration-response relationship is linear across the full concentration range or instead shows evidence of a population threshold. Of particular interest is the shape of the concentration-response curve at and below the level of the current 8-h standard of 80 ppb. The slope of the O_3 concentration-response relationship has been explored in several studies.

To examine the shape of the concentration-response relationship between O_3 and mortality, Gryparis et al. (2004) used meta-smoothing to combine smooth curves across the 23 European cities in a hierarchical model. For the summer period, while the estimated concentration-response curve did not appear to deviate significantly from linearity, there were indications of decreasing effectiveness at lower exposures.

In the U.S. 95 communities study (Bell et al., 2004), effect estimates calculated using only days with 24-h avg O_3 levels less than 60 ppb were compared to those using all data. At a lag of 1 day, O_3 was associated with an excess risk of 0.36% (95% PI: 0.12, 0.60) per 20 ppb increase in 24-h avg O_3 using data from all days and only a slightly smaller risk of 0.30% (95% PI: 0.08, 0.54) when data were limited to days less than 60 ppb. These results suggest that if there is a threshold, it must be notably lower than a 24-h avg O_3 of 60 ppb.

Fairley (2003) reanalyzed the Santa Clara County mortality data using GAM with stringent convergence criteria and examined a new exposure index for O_3. He noted O_3 concentrations exceeding 60 ppb each hour and calculated a daily sum of these exceedances. Fairley's index incorporates measures of concentration and exposure duration; this index represents a linear time-integrated concentration, also known as dosage. The O_3 index with the 60 ppb "threshold level" was found to be significantly associated with mortality in single-pollutant models as well as in multi-pollutant models. Two other "threshold levels" were examined, 40 ppb and 80 ppb. Both produced statistically significant results in single-pollutant models. These results suggest that the threshold for O_3-mortality effects, if it exists, is likely less than 40 ppb. The implication for thresholds in terms of the three standard indices (i.e., 1-h max, 8-h max, and 24-h avg) is unclear, but there may be an empirical relationship.

Vedal et al. (2003) observed that the annual mean 1-h daily max O_3 concentration of 27.3 ppb in Vancouver, Canada, was lower than that in any of the 80 NMMAPS cities (Samet et al., 2000); thus, a study in this city might focus better on the shape of the concentration-response curve at lower levels. In this study, an O_3 effect was observed on total mortality at a 0-day lag during the summer. Ozone effects on respiratory mortality at a 2-day lag and cardiovascular mortality at a 0-day lag also were observed in the summer. The effect of O_3 on mortality was robust in two-pollutant models. Vedal et al. questioned if O_3, other gaseous pollutants, and PM were acting as surrogate markers of pollutant sources that contain more toxic compounds, since the low measured concentrations were unlikely, in their opinion, to cause the

observed effects. They further stated that measurement error and interference by meteorological factors might have contributed to the inability to detect a threshold. Vedal et al. (2003) concluded that O_3 concentrations were associated with adverse effects on mortality even at low levels. Although this study supports the argument that there are no threshold concentrations below which adverse effects cannot be detected, the results must be interpreted with caution as concerns remain.

Kim et al. (2004) investigated the presence of a threshold in O_3-mortality effects in Seoul, Korea by analyzing data using a log linear GAM (linear model), a cubic natural spline model (nonlinear model), and a B-mode splined model (threshold model). Models were stratified by season and adjusted for PM_{10}, long-term time trend, and meteorological variables. An estimated threshold value of 47 ppb was observed for 1-h daily max O_3. None of the other pollutants examined, including PM_{10}, SO_2, NO_2, and CO, had a nonlinear association with mortality. Using summer data only, the B-spline model resulted in an excess mortality risk of 7.1% (95% CI: 3.1, 11.2) per 40 ppb increase in 1-h max O_3, compared to an excess risk of 3.6% (95% CI, 0.5, 6.8) calculated using the log linear model. If a threshold truly exists, results from the Kim et al. study suggest that the use of log-linear models may underestimate the O_3 effect on mortality at levels above the threshold.

Other studies examining the effect of O_3 on mortality also have found suggestive evidence for a possible threshold level. In a London, England study (Anderson et al., 1996), an adjusted O_3-mortality bubble plot suggested that a threshold might exist around 50 ppb for 8-h avg O_3. A study by Simpson et al. (1997) in Brisbane, Australia observed a significant excess risk in mortality only in the highest quintile of O_3 exposure, which had a mean concentration of 42 ppb for 1-h max O_3.

Among several studies with morbidity outcomes, examination of the shape of the concentration-response function indicated evidence of an effect threshold. In a study of all-age respiratory hospital admissions in Toronto, Canada, effects of O_3 appeared to become apparent only above approximately 30 ppb daily 1-h max O_3 (Burnett et al., 1997b). In London, England, Ponce de Leon et al. (1996) observed an indication of a threshold in the O_3 effect on hospitalizations at 40 to 50 ppb for 8-h max O_3 and 50 to 60 ppb for 1-h max O_3. In a study of emergency department visits for asthma in St. John, Canada, effects observed in the over 15 years age group were apparent only when data above the 95th percentile (75 ppb daily 1-h

max O_3) were included (Stieb et al., 1996). However, other morbidity studies observed a monotonic increase in the concentration-response function, suggesting that there was no threshold in O_3 effects on hospitalizations and emergency department visits (Burnett et al., 1997a; Jaffe et al., 2003; Petroeschevsky et al., 2001; Tenías et al., 1998).

In a field study by Mortimer et al. (2002), the associations of ambient O_3 levels with PEF and asthma symptoms were investigated in eight urban cities in the U.S. The mean 8-h avg O_3 was 48 ppb, with less than 5% of days exceeding 80 ppb. Analysis performed using all data indicated that a 15 ppb change in 8-h avg O_3 was associated with decrements in PEF (−0.59% [95% CI: −1.05, −0.13]) and increased incidence of respiratory symptoms (odds ratio of 1.16 [95% CI: 1.02, 1.30]) over multiday lag periods. When data were restricted to days when ambient O_3 concentrations were less than 80 ppb, the O_3 effects persisted, with a significant PEF decline (−0.70% [95% CI: −1.29, −0.12]) and incidence of morning symptoms (odds ratio of 1.17 [95% CI: 1.01, 1.35]). A study by Chen et al. (1999) also found that there was no clear threshold in the O_3 effect on FEV_1 and FVC in Taiwanese school children.

The studies of both Brauer et al. (1996) and Korrick et al. (1998) demonstrate that exposure duration and exercise level, in addition to O_3 concentration, must be considered when evaluating thresholds. In the study by Brauer et al., the mean O_3 concentration during the 11-hour work shift was 26.0 ppb (SD 11.8). Workers experienced a change of −180.0 mL (95% CI: −227.0, −133.0) in FEV_1 levels the next morning per 40 ppb increase in 1-h max O_3. The hikers in the study by Korrick et al. (1998) were exposed to mean O_3 levels of 40 ppb (SD 12) over the duration of their hike (mean 8 hours). Korrick et al. observed a mean change of −62.5 mL (95% CI: −115.3, −9.7) in pre-hike to post-hike FEV_1 per 30 ppb increase in 8-h avg O_3 when all hikers were included in the analysis; however, when analysis was restricted to hikers with wheeze or asthma, a larger change of −182.5 mL (95% CI: −312.2, −52.9) was observed. In both studies, large reductions in lung function were observed in subjects exposed to relatively low levels of O_3 over multiple hours while active outdoors.

Note that adjusting for seasonal cycles does not address the issue of the changing relationship between O_3 concentrations and personal exposure across seasons. The ambient O_3 levels are lower in the cold season, but people are likely to be exposed to even lower levels of O_3 during this season due to the shorter time spent outdoors and the longer time spent indoors with closed windows. This is in contrast to what occurs with fine particles, which can

effectively penetrate the indoors. Thus, a more "accurate" concentration-response relationship may need to be examined in a summer-only data set. Even for summer data, however, an interpretation of the relationship is not straightforward because of the possible influence of the use of air conditioning (an effective remover of O_3). Greater use of air conditioning is expected on hot days when the O_3 level is higher, but the use of air conditioning may also vary from city to city and across social class within a city. Using $PM_{2.5}$ and sulfate as an example, Brauer et al. (2002) observed that surrogate measures of exposure (i.e., those from centrally-located ambient monitors) that were not highly correlated with personal exposures obscured the presence of thresholds in epidemiologic studies of larger populations. Likewise, exposure measurement error may reduce the ability to detect a threshold in O_3 population studies that used ambient O_3 concentrations as an indicator of personal exposure.

Limited studies have examined the issue of thresholds in O_3 health effects studies. Some studies have found a low level threshold while others have found no threshold in O_3 effects. Levy et al. (2001) states that the molecular effects of O_3 are mediated by antioxidants in the lung lining fluid, which raises the possibility that there may be a threshold levels below which O_3 would have few or no adverse effects. However, due to the variability in individual sensitivities and antioxidant levels, this threshold may not be seen at the population level.

From 1990 to 2004, the 10th percentile values (which represent the lower concentration range) of the warm season (May to September) 8-h max O_3 concentrations averaged for all available monitors throughout the U.S. were approximately 40 ppb (see discussion in Section 3.2). While no conclusion can be made regarding the threshold issue, the limited evidence suggests that if there is a threshold level in O_3 health effects, it is likely near the lower limit of ambient O_3 concentrations in the U.S.

7.6.6 Heterogeneity of Ozone Health Effects

As described in Chapter 3 of this AQCD, O_3 concentrations tend to be more spatially variable than $PM_{2.5}$ concentrations in urban areas. In addition, relative personal exposures to O_3 likely vary by region. The geographic variability in O_3 concentrations and personal exposures may contribute to the heterogeneity in observed O_3 health effects. The degree of influence of the geographic variability on heterogeneity in effects will vary by study as study design affects different aspects of exposure (e.g., time period and duration of exposure).

More than 80% of the O_3-mortality estimates from the various studies conducted in North America, South America, Europe, and Australia were between 0.5 and 5% excess risk per 40 ppb increase in 1-h max O_3 using year-round data. In general, the O_3-mortality estimates were greater when using summer only data compared to year-round data. Though not all statistically significant, most of the O_3-mortality estimates were greater than zero, indicating a positive relationship between O_3 exposure and mortality. The O_3 risk estimates from the numerous hospitalization and emergency department visit studies were generally larger in magnitude and more variable from study to study compared to the mortality studies. These differences in the O_3 effect estimates may be attributable to the greater variability in the outcome measure in hospitalization studies compared to mortality studies, such as more subcategories of outcome and varying degrees of severity.

Three recent meta-analyses that included both U.S. and non-U.S. studies found consistent all-year combined point estimates: 1.75% (95% PI: 1.10, 2.40), 1.6% (95% CI: 1.1, 2.0), and 1.64% (95% CI: 1.25, 2.03) per 20 ppb increase in 24-h average O_3, for Bell et al. (2005), Ito et al. (2005), and Levy et al. (2005), respectively. Bell et al. further observed that the pooled estimate for U.S. studies (11 estimates), 1.69% (95% PI: 0.94, 2.78), was similar to the pooled estimate for the non-U.S. studies (30 estimates), 1.85% (95% PI: 0.94, 2.78). Levy et al. compared North American studies to European studies and also found nearly identical effect estimates.

As differences in study design, population, and data analysis may affect risk estimates, studies that were conducted in multiple cities using standardized methods were further examined to investigate the geographic heterogeneity of O_3 effects. Bell et al. (2004) conducted a time-series analysis of O_3 and mortality in 95 U.S. communities from 1987 to 2000. A 20 ppb increase in 24-h avg O_3 levels in the previous week was associated with an increase of 1.04% (95% PI: 0.54, 1.55) excess risk of mortality in the pooled analysis of 95 communities using all available data. Intercity heterogeneity was observed among the 95 communities, which the authors noted as plausible given the city-specific differences in pollution characteristics, the use of air conditioning, time-activity patterns, and socioeconomic factors. Although some heterogeneity was observed among the communities (see Figure 7-16 of Section 7.4.3), the range of the community-specific Bayesian estimates was fairly narrow. Note that the community-specific Bayesian estimates are shrunken estimates of the percent changes in daily

mortality. The larger the heterogeneity (across-community variance relative to within-community variance), the less the Bayesian estimates shrink toward the national average. Of the 95 U.S. communities, 93 had positive O_3-mortality risk estimates. Only 5 had risk estimates greater than 2.0% per 20 ppb increase in 24-h avg O_3 during the previous week, with all communities indicating an excess mortality risk less than 3.5%.

Greater heterogeneity was observed in the European study of 23 cities in 14 countries (Gryparis et al., 2004). In the year-round analyses, only 8 of the 23 cities had positive O_3-mortality effect estimates. However, in the analyses using summer data only, the risk estimates were positive in 19 of the 23 cities, with a range of 0.8 to 8% excess risk per 40 ppb increase in 1-h max O_3. The heterogeneity may be attributable to the considerable variability among countries in factors that may influence the relationship between ambient O_3 concentrations and personal exposure to O_3, such as climate, use of air conditioning, personal activity patterns, and socioeconomic factors. In addition, the variability in the concentration and composition of copollutants by cities or countries may contribute to the heterogeneity in the O_3-mortality effects. For example, concentrations of NO_2 may vary widely by region, depending on the differences in traffic density.

Among the hospitalization studies, Burnett et al. (1997a) conducted the largest study of 16 Canadian cities. The mean daily 1-h max O_3 was 31 ppb in the 16 cities. The pooled O_3 estimate was 5.6% (95% CI: 3.4, 7.9) excess risk in respiratory hospitalization per 40 ppb increase in 1-h max O_3 using warm season data (April to December). The risk estimates were fairly homogenous across the 16 Canadian cities, ranging from 3.1% for Vancouver to 7.7% for Quebec City.

Anderson et al. (1997) investigated the association between O_3 and hospital admissions for COPD in five European cities — London, Paris, Amsterdam, Rotterdam, and Barcelona. The pooled effect estimate was 5.0% (95% CI: 2.6, 7.6) excess risk per 30 ppb increase in 8-h max O_3 for year-round data. Results from the APHEA study showed similar variability to that from the Burnett et al. (1997a) study. The year-round effects estimates were lower in the two Dutch cities (2.5% excess risk) compared to that in Paris (7.7% excess risk); however, analyses indicated that there was no significant heterogeneity in effects by city. The authors further noted that among the pollutants examined (O_3, BS, TSP, SO_2, and NO_2), O_3 had the most consistent and significant findings.

Among the field studies, various respiratory health outcomes were examined, including PEF, spirometric parameters, respiratory symptoms, and medication use. Only one field study investigated the O_3 effect in several locations (Mortimer et al., 2002). Mortimer et al. (2002) investigated the association of ambient O_3 concentrations with PEF and asthma symptoms in asthmatic children living in eight urban cities in the U.S. — St. Louis, MO; Chicago, IL; Detroit, MI; Cleveland, OH; Washington, DC; Baltimore, MD; East Harlem, NY; and Bronx, NY. In the analysis pooling data from all eight cities, a 30 ppb increase in 8-h avg O_3 was associated with a decrement of −1.18% (95%CI: −2.10, −0.26) in morning PEF for a 5-day cumulative lag period. The percent changes in PEF were negative in all cities except for Baltimore, 0.49%. Among the other seven cities, the percent changes in PEF were quite homogenous, with values ranging from −1.08% for Washington, DC to −1.71% for St. Louis. A 30 ppb increase in 8-h avg O_3 also was associated with an increased incidence of morning symptoms in the pooled analysis (odds ratio of 1.35 [95% CI: 1.04, 1.69] for a 4-day cumulative lag period). In all cities except for St. Louis, there was an increase in the incidence of morning symptoms. In these cities, the odds ratios for incidence of morning symptoms varied more compared to the PEF measurements, ranging from 1.19 for Chicago to 2.96 for Detroit. The greater variance may indicate the lack of standardization in the use of symptoms as a health outcome measure.

Most of the multicity and meta-analyses studies consistently found positive associations between O_3 and mortality. Consistent O_3 effects on hospitalizations and various respiratory health outcomes also were found. The observed heterogeneity of O_3 effects may be partially attributable to the use of centrally-located ambient monitors to assess exposure. There may be differences in relative personal exposures to O_3 due to varying factors, such as use of air conditioning and activity patterns, that affect the relationship between personal exposure and ambient concentrations. For example, Levy et al. (2005) found suggestive evidence that air conditioning prevalence was a predictor of heterogeneity in O_3 risk estimates in their meta-analysis. The variability in the concentration and composition of other pollutants present also may contribute to the heterogeneity of the effect of O_3 on health outcomes as confounding by copollutants may vary by region.

7.6.7 Health Effects of Ozone in Susceptible Populations

In this section, the effects of O_3 on morbidity and mortality in potentially susceptible populations will be examined. In epidemiologic studies of O_3 health effects, the most widely studied subpopulation was asthmatics. Also of interest were the observed health effects of O_3 on different age groups, particularly children and the elderly. This section begins with a discussion of the O_3-related health effects in asthmatics.

7.6.7.1 Health Effects Associated with Ambient Ozone Exposure in Asthmatics

Epidemiologic studies of health effects from acute O_3 exposure in asthmatics have examined a range of outcomes: pulmonary function, respiratory symptoms, inflammation, emergency room visits, hospital admissions, and mortality. Chronic O_3 exposure studies have investigated similar outcomes, with the exception of emergency room visits and hospitalizations. Both are discussed in the earlier text. This subsection draws together this information to examine whether the evidence indicates that O_3 exposure impacts asthmatics.

In Germany and Mexico City, O_3 exposure was associated with a decline in FEV_1 in asthmatic adults and children (Höppe et al., 1995a, 2003; Romieu et al., 2002). Change in FEV_1 also was examined in a group of asthmatic hikers in Mount Washington, NH (Korrick et al., 1998). Compared to the healthy subjects, the asthmatic subjects experienced a four-fold greater decline in FEV_1 with the same exposure to O_3 (mean change of −1.08% [95% CI: −2.49, 0.33] versus −4.47% [95% CI: −7.65, −1.29] per 30 ppb increase in 8-h avg O_3). The results from the hiker study are consistent with those observed in controlled human exposure studies (discussed in Chapter 6), which also indicate greater decrements in FEV_1 among mild asthmatics versus nonasthmatic subjects with heavy intermittent exercise.

PEF was examined in panels of asthmatic children in several field studies (see Figures 7-1 and 7-2). Collectively, most of the studies indicated decrements of morning PEF, though only a few estimates were statistically significant. One multicity study of eight urban areas in the U.S. observed O_3-related reductions in morning PEF that were not significant in each individual city (Mortimer et al., 2002); however, the analysis combining data from all eight cities indicated a significant decline in PEF with a cumulative lag of 1 to 5 days of O_3 exposure. The odds ratio for the incidence of ≥10% decline in morning PEF was greater than one, which was discussed by the author as an indication that O_3 exposure might be associated with clinically important

changes in PEF in asthmatic children. The study examined 846 asthmatic children, the largest asthma panel study reported.

Mortimer et al. (2000) observed that the subpopulation of asthmatic children with a history of low birth weight or premature birth had greater O_3-associated declines in PEF (mean change of −3.66% [95% CI: −5.30, −2.02] per 30 ppb increase in 8-h avg O_3) than normal birth weight children (−0.60% [95% CI: −1.58, 0.38]). Low birth weight and prematurity are associated with reduced lung function, higher levels of airway reactivity and increased susceptibility to lung damage (Barker et al., 1993; Rona et al., 1993), which may explain why these factors are found to increase susceptibility to respiratory insults of air pollution in children.

Lung function parameters have been evaluated for clinical significance. A reversible 5 to 15% decline in FEV_1 in an individual may have clinical importance to asthma morbidity (American Thoracic Society, 1991; Lebowitz et al., 1987; Lippmann, 1988). The National Institutes of Health (1997) has stated that PEF below 80% of the personal best indicates a need for additional medication use in asthmatics. At a population level the mean changes in lung function attributable to O_3 exposure do not generally exceed 10% changes in FEV_1 or PEF per standardized increment of O_3. At an individual level, a subpopulation of susceptible asthmatics are likely experiencing clinically significant declines in lung function. Höppe et al. (2003) examined effects of O_3 on the lung function of potential risk groups, performing both group and individual analyses. For the group mean values, consistent O_3 effects were not detectable. On an individual basis, a potential pattern of O_3 sensitivity was observed. About 20% of the asthmatics and children were regarded as O_3 responders (i.e., individuals with >10% change in FEV_1) compared to only 5% of the elderly and athletes. These results indicated that while the population as a whole was not reacting to O_3, susceptible individuals were experiencing clinically significant declines in lung function in response to O_3 exposure.

Respiratory symptom increases in asthma panels were examined in several field studies, some of which also examined PEF as discussed above. The outcome definition of symptoms varied among these studies. Collectively, the results are suggestive of a potential O_3 effect on respiratory symptoms, but the evidence is not strong in the available studies. Two U.S. studies that examined larger panels might be better to draw inferences from as the large sample size provided greater power to examine the effect of O_3 on respiratory symptoms. The eight U.S. urban cities study reported that morning symptoms in the 846 asthmatic children were most

strongly associated with a 4-day cumulative lag of O_3 concentrations (Mortimer et al., 2002). A New England study examined 271 asthmatic children and observed an O_3 effect on a variety of respiratory symptoms at a lag of 1 day among the 130 subjects who used maintenance asthma medications (Gent et al., 2003).

Few epidemiologic studies have examined airway inflammation in asthmatics. A Mexico City study indicated that supplementation with antioxidants may modulate the impact of O_3 exposure on the small airways of children with moderate-to-severe asthma (Romieu et al., 2002). A related study indicated that asthmatic children with GSTM1 null genotype were found to be more susceptible to the impact of O_3 exposure on small airways (Romieu et al, 2004). A chronic exposure study in Mexico City examined DNA strand breaks in nasal epithelial cells in asthmatic and nonasthmatics medical students and noted greater genotoxic damage in asthmatics (Fortoul et al., 2003).

Emergency department visits for asthmatics have been examined in several studies and range from negative to positive results (see Figure 7-8 in Section 7.3.2). Warm season studies tended to yield positive outcomes, as expected based on earlier discussions. Two studies in Atlanta, GA (Tolbert et al., 2000) and Valencia, Spain (Tenías et al., 1998) indicated positive effects in warm season analyses. Further, a Canadian study, one of the larger studies conducted in the summertime, reported a large increase in asthma emergency department visits when the daily 1-h max O_3 concentration exceeded 75 ppb (Stieb et al., 1996). A three-city study in Ohio also indicated an increased risk of asthma visits during the summer (Jaffe et al., 2003). Other studies of mostly year-long data tended to produce inconsistent results, with some finding negative estimates (Atkinson et al., 1999a; Castellsague et al., 1995; Thompson et al., 2001; Tobías et al., 1999).

Hospital admission studies that specifically examined asthmatics were fewer in number than those that examined total respiratory diseases. Associations were noted in all age groups in studies conducted in Seattle, WA (Sheppard, 2003), New Jersey (Weisel et al., 2002), Toronto, Canada (Burnett et al., 1999), London, England (Anderson et al., 1998), Brisbane, Australia (Petroeschevsky et al., 2001), and Hong Kong (Wong et al., 1999a). However, several other studies, mostly examining the effect of O_3 on asthmatic children, did not observe a significant relationship (Gouveia and Fletcher, 2000a; Lin et al., 2003; Morgan et al., 1998; Nauenberg and Basu, 1999; Schouten et al., 1996).

Acute mortality related to asthma was examined in Barcelona, Spain (Saez et al., 1999; Sunyer et al., 2002). Severe asthmatics with more than one asthma emergency visit showed the strongest mortality associations with O_3 (Sunyer et al., 2002).

Recent reports from longitudinal cohort studies in California have reported associations between the onset of asthma and long-term O_3 exposures (Greer et al., 1993; McConnell et al., 2002; McDonnell et al., 1999). In adult studies, associations were seen in males but not females (Greer et al., 1993; McDonnell et al., 1999). Among children residing in high O_3 communities, McConnell et al. (2002) observed that asthma risk was elevated for those who played three or more sports as compared with those who did not play sports. Playing sports may indicate outdoor activity and an increased ventilation rate which may lead to increased dose of O_3. These outcomes would benefit from replication in other cohorts in regards to indicating weight of a causal interpretation.

A few studies provide limited discussion of concentration-response functions and thresholds. In the eight U.S. urban cities study, the odds ratios for incidence of $\geq$10% decline in morning PEF and incidence of morning symptoms when excluding days with 8-h avg O_3 greater than 80 ppb were nearly identical to those including data from all days (Mortimer et al., 2002). In the New England asthma panel study (Gent et al., 2003), some of the associations for symptoms occurred at 1-h max O_3 levels below 60 ppb. In the St. John, Canada study (Stieb et al., 2003), an effect of O_3 on emergency department visits was reported with evidence of a threshold somewhere in the range below a 1-h max O_3 of 75 ppb in the 15 years and over age group.

Overall, asthma subjects have been examined across most health endpoints of interest. The results reported in these studies vary with some indicating a positive excess risk associated with O_3. While no endpoint in itself seems to indicate an unquestionable demonstration of an association, studies with adequate sample size and power consistently provide positive results, especially during the summer months when higher O_3 levels occur. This view is strengthened as positive results are obtained cohesively across the varied outcomes. Therefore, based on the evidence, it seems prudent to consider asthmatics as a susceptible group that requires protection from O_3 exposures.

7.6.7.2 Age-Related Differences in Ozone Effects

Several mortality studies have investigated age-related differences in O_3 effects. Among the studies that observed positive associations between O_3 and mortality, a comparison of all age or younger age (≤65 years of age) O_3-mortality risk estimates to that of the elderly population (>65 years) indicates that, in general, the elderly population is more susceptible to O_3 effects (Borja-Aburto et al. 1997; Bremner et al., 1999; Gouveia and Fletcher 2000b; O'Neill et al., 2004; Simpson et al., 1997; Sartor et al., 1995; Sunyer et al., 2002). For example, a study by Gouveia and Fletcher (2000b) examined the O_3-mortality effect by age in São Paulo, Brazil. There were 151,756 deaths for all non-violent causes over the period of 1991 to 1993, of which 49% occurred in the elderly. Among all ages, O_3 was associated with a 0.6% (95% CI: −0.8, 2.0) excess risk in all cause mortality per 40 ppb increase in 1-h max O_3. In comparison, in the elderly population, the O_3-mortality risk estimate was nearly threefold greater, 1.7% (95% CI: 0.0, 3.3). Similarly, a Mexico City study found that O_3-mortality risk estimates were 1.3% (95% CI: 0.04, 2.6) and 2.8% (95% CI: 1.0, 4.6) per 20 ppb increase in 24-h avg O_3 concentration in all ages and the elderly, respectively (O'Neill et al., 2004).

The meta-analysis by Bell et al. (2005) found a larger effect estimate for the elderly (2.92% [95% PI: 1.34, 4.51] per 20 ppb increase in 24-h avg O_3) than for all ages (1.75% [95% PI: 1.10, 2.37]). In the large U.S. 95 communities study (Bell et al., 2004), effect estimates were slightly higher for those aged 65 to 74 years, 1.40% (95% PI: 0.56, 2.25) excess risk per 20 ppb increase in 24-h avg O_3, compared to individuals less than 65 years and 75 years or greater, 1.00% (95% PI: 0.20, 1.85) and 1.04% (95% PI: 0.36, 1.75), respectively, using a constrained distributed 7-day lag model. Bell et al. (2004) notes that despite somewhat similar effect estimates, the absolute effect of O_3 is substantially greater in the elderly population due to the higher underlying mortality rates, which leads to a larger number of extra deaths for the elderly compared to the general population.

Few mortality studies examined another potentially susceptible age group, young children under the age of 5 years. The results were mixed, with one Mexico City study showing a lower risk of O_3-related all cause mortality in young children compared to all ages and the elderly (Borja-Aburto et al., 1997) and one São Paulo, Brazil study showing a greater risk in respiratory mortality in young children compared to the elderly (Gouveia and Fletcher, 2000b). It should be

noted that approximately 10% of mortality occurred in young children, thus the statistical power to study the O_3 effect in this age group was limited.

With respect to age-specificity of associations between O_3 and acute respiratory hospitalizations or emergency department visits, no clear pattern emerges from recent studies. Associations have been reported for all ages (Anderson et al., 1997; Burnett et al., 1995, 1997b, 1999; Weisel et al., 2002), adults or elderly (Burnett et al., 1997a; Delfino et al., 1997b, 1998b; Moolgavkar et al., 1997; Schwartz et al., 1996; Yang et al., 2003), and children (Burnett et al., 2001; Gouveia and Fletcher, 2000a; Lin et al., 1999; Pönkä and Virtanen, 1996; Tolbert et al., 2000; Yang et al., 2003). Interestingly, studies that have examined effects in multiple age strata often have seen effects only in non-pediatric strata (Delfino et al., 1997b, 1998b; Stieb et al., 1996; Jones et al., 1995). Several studies that focused on children did not report significant O_3 effects, though in some cases these studies are limited by small size, inadequate control of seasonal patterns, or very low O_3 levels (Lierl and Hornung, 2003; Lin et al., 2003; Thompson et al., 2001). If O_3 is causally related to exacerbations of respiratory diseases leading to hospital usage, one would expect to see effects most prominently among children, for whom asthma is more prevalent and O_3 exposures may be greater. However, once again, children only comprised of about 20% of the total hospitalizations, which limits the power to examine age-specific O_3 effects.

A few field studies compared the effect of O_3 in different age groups. Korrick et al. (1998) examined changes in FEV_1 and FVC related to O_3 exposure in a group of hikers ranging in age from 18 to 64 years, and found that there was no association between O_3 responsiveness and age. Brauer et al. (1996), in a study of berry pickers aged 10 to 69 years, also observed that subject age was not significantly associated with O_3-related changes in lung function. However, a study by Höppe et al. (1995a, 2003) observed that children, but not seniors (69 to 95 years of age), experienced a decline in lung function associated with O_3 exposure. The results by Höppe et al. are consistent with the diminishing responses to O_3 exposure with increasing age observed in clinical studies. The clinical studies by Drechsler-Parks (1995) and Bedi et al. (1989) found that subjects aged 56 to 89 years had markedly reduced responses to O_3 exposure compared to young adults.

Many field studies focused on the effect of O_3 on the respiratory health of school children. In general, children experienced decrements in pulmonary function parameters, including PEF,

FEV_1, and FVC (Castillejos et al., 1995; Chen et al., 1999; Gielen et al., 1997; Gold et al., 1999; Jalaludin et al., 2000; Mortimer et al., 2002; Romieu et al., 1996; Thurston et al., 1997). Increases in respiratory symptoms (Delfino et al., 2003; Gold et al., 1999; Neas et al., 1995; Romieu et al., 1996, 1997; Thurston et al., 1997) and asthma medication use (Delfino et al., 1996; Just et al., 2002; Ostro et al., 2001; Thuston et al., 1997) also were observed in children. These respiratory health effects were observed in both healthy and asthmatic children. In one German study (Höppe et al., 2003), juvenile asthmatics and healthy children were found to be particularly susceptible to O_3 effects on lung function. Approximately 20% of the children and asthmatics experienced a greater than 10% change in FEV_1, compared to only 5% of the elderly population and athletes.

The American Academy of Pediatrics (2004) notes that children and infants are among the most susceptible to many air pollutants, including O_3. Eighty percent of alveolar are formed postnatally and changes in the lung continue through adolescence (Dietert et al., 2000). Children spend more time outdoors, which results in increased exposure to air pollutants (Wiley et al., 1991a,b). Further, children have a high minute ventilation and high levels of physical activity which increase their dose (Plunkett et al., 1992).

Collectively, there is supporting evidence of age-related differences in susceptibility to O_3 health effects. The elderly population (>65 years of age) appear to be at increased risk of O_3-related mortality and hospitalizations, and children (<18 years of age) experience other potentially adverse respiratory health outcomes with increased O_3 exposure. One epidemiologic study also found that the lung function response to O_3 exposure may be diminished in elderly populations; this finding is further supported by evidence from clinical studies.

7.6.8 Summary of Key Findings and Conclusions Derived From Ozone Epidemiologic Studies

In the previous 1996 O_3 AQCD, there was considerable evidence of O_3-related respiratory health effects from individual-level camp and exercise studies, as well as some consistent evidence from time-series studies of emergency room visits and hospitalizations. Since the 1996 document, more field studies have been conducted, with some emphasis on additional outcome markers such as respiratory symptoms and asthma medication use. Another significant addition to the current O_3 AQCD is the substantial number of short-term O_3 mortality studies. The recent

publication of an analysis examining the relationship between O_3 and mortality in 95 U.S. communities (Bell et al., 2004) and three meta-analysis on O_3-mortality associations (Bell et al., 2005; Ito et al., 2005; Levy et al., 2005) also contribute significantly to the evidence base. Considering the wide variability in possible study designs and statistical model specification choices, the reported O_3 risk estimates for the various health outcomes are in reasonably good agreement. In the case of O_3-mortality time-series studies, combinations of choices in model specifications (the number of weather terms and degrees of freedom for smoothing of mortality-temporal trends) alone may explain the extent of the difference in O_3 risk estimates across studies. As use of time-series studies to investigate air pollution effects has become more common, there has been a great effort to evaluate the issues surrounding these studies.

In this section, conclusions regarding O_3 health effects from the epidemiologic evidence and the issues that may affect the interpretation of the effect estimates are briefly summarized. A more integrative synthesis of all relevant information will be presented in Chapter 8 of this AQCD.

(1) Field/panel studies of acute O_3 effects. Results from recent field/panel studies continue to confirm that short-term O_3 exposure is associated with acute decrements in lung function and increased respiratory symptoms, particularly in children and asthmatics. There is also suggestive evidence that O_3 is related to increased asthma medication use. Taken together with the evidence from controlled human exposure studies, O_3 is likely causally related to the various respiratory health outcomes. The current evidence is limited but supportive of a potential effect of O_3 on heart rate variability, ventricular arrhythmias, and the incidence of myocardial infarctions.

(2) Acute O_3 effects on emergency department visits and hospitalizations. Large multicity studies, as well as many studies from individual cities have reported an association of O_3 concentrations with respiratory and cardiovascular hospital admissions. Studies using year-round data noted some inconsistencies in the O_3 effect on daily hospitalizations. However, studies with data restricted to the summer or warm season, in general, indicated positive and robust associations between ambient O_3 concentrations and cardiopulmonary hospital admissions. Results for emergency department visits are less consistent.

(3) Acute O_3 effects on mortality. The majority of the studies suggest an elevated risk of all cause mortality associated with acute exposure to O_3, especially in the summer or warm season when O_3 levels are typically high. Slightly greater O_3 effects were observed for cardiovascular mortality. Results from a recent, large U.S. multicity time-series study provide the strongest evidence to-date for O_3 effects on acute mortality. Recent meta-analyses also showed consistent risk estimates that are

unlikely to be confounded by PM; however, future work is needed to better understand the influence of model specifications on the risk coefficient.

(4) Chronic O_3 exposure effects on morbidity and mortality. Fewer studies have investigated the effect of chronic O_3 exposure on morbidity and mortality. The strongest evidence is for negative seasonal effects of O_3 on lung function in adults and children. Less conclusive are longer-term studies investigating the association of chronic O_3 exposure on yearly lung function, asthma incidence, and respiratory symptoms. Chronic O_3-mortality studies observed inconsistencies across exposure periods, cause-specific mortality outcomes, and gender.

(5) Exposure assessment. Exposure misclassification may result from the use of stationary ambient monitors to determine exposure in population studies. Although central ambient monitors do not explain the variance of individual personal exposures, significant correlations are found between aggregate personal O_3 measurements and O_3 concentrations from ambient monitors. A simulation study indicated that the use of ambient monitor data will tend to underestimate the O_3 effect. A better understanding of the factors that affect the relationship between ambient concentrations and personal exposures will improve interpretation of the O_3 effect estimates.

(6) Ozone exposure indices. The three most commonly used daily O_3 exposure indices, 1-h max O_3, 8-max O_3, and 24-h avg O_3, were found to be highly correlated in studies conducted in various regions. In addition, the effect estimates and significance of associations across all health outcomes were comparable when using the standardized distributional increment of 40 ppb, 30 ppb, and 20 ppb for 1-h max O_3, 8-h max O_3, and 24-h avg O_3, respectively.

(7) Lag structures for O_3 exposure and effect. The lag time between O_3 exposure and effect may differ depending on various factors such as the specific health outcome of interest, the mechanism of effect, and preexisting health conditions. The majority of the studies found an immediate O_3 effect, with the strongest associations observed between health outcomes and O_3 exposure on the same day and/or previous day. Some studies found large cumulative effects of O_3 over longer lag periods, indicating that multiday lags also may be relevant for some health outcomes, including mortality.

(8) Sensitivity to model specifications for temporal trends. Ozone effect estimates that were reported in studies whose main focus was PM often were calculated using the same model specifications as PM. While the sensitivity of the O_3 risk estimates to alternative model specifications has not been throughly investigated, limited evidence indicates that O_3 effects may be robust to various model specifications for temporal trend adjustment.

(9) Influence of seasonal factors. An evaluation of the confounding effects of meteorologic factors and copollutants on O_3 risk estimates is complicated by their changing relationships with O_3 across seasons. In addition, seasonal or seasonally-

modified factors (e.g., air conditioning use, time spent outdoors) complicate interpretation of all year effect estimates as they affect the relationship between ambient concentrations and personal exposures. Given the potentially significant influence of season, season-specific analyses are more informative in assessing O_3 health risks.

(10) Confounding by copollutants. Multipollutant regression models often are used to adjust for confounding by copollutants. Although there is some concern regarding the use of multipollutant models given the varying concurvity across pollutants, results generally suggest that the inclusion of copollutants into the models do not substantially affect O_3 risk estimates. These findings indicate that effects of O_3 on various health outcomes are robust and independent of the effects of other copollutants.

(11) Concentration-response function. In the limited mortality and morbidity studies that have specifically examined the O_3 concentration-response relationship, the evidence is inconclusive regarding the presence of an effect threshold. Factors such as exposure measurement error may reduce the ability to detect a threshold in population studies.

(12) Heterogeneity of O_3 health effects. Consistent O_3 effect estimates generally were observed for mortality, hospitalizations, and other respiratory health outcomes in multicity studies. Some of the observed geographic heterogeneity in effects may be attributable to the differences in relative personal exposure to O_3, which is affected by factors such as air conditioning prevalence and activity patterns, and the varying concentrations and compositions of copollutants present by region.

(13) Ozone health effects in asthmatics. The effects of O_3 on asthmatics have been examined widely in both time-series studies and panel studies. Associations of O_3 with various respiratory health outcomes, including lung function declines, increased respiratory symptoms, and emergency department visits, were observed. These findings, along with the pathophysiologic understanding of asthma as a chronic inflammatory disease, indicate that asthmatics may be a susceptible population that requires protection from O_3 exposures.

(14) Age-related differences in O_3 health effects. Supporting evidence exists for heterogeneity in the effects of O_3 by age. The elderly population (>65 years of age) appear to be at greater risk of O_3-related mortality and hospitalizations compared to all age or younger populations. In addition, potentially adverse respiratory health outcomes were associated with O_3 exposure in children (<18 years of age). One epidemiologic study provided limited evidence that lung function responses to O_3 exposure is diminished in the elderly population.

REFERENCES

Abbey, D. E.; Nishino, N.; McDonnell, W. F.; Burchette, R. J.; Knutsen, S. F.; Beeson, W. L.; Yang, J. X. (1999) Long-term inhalable particles and other air pollutants related to mortality in nonsmokers. Am. J. Respir. Crit. Care Med. 159: 373-382.

American Academy of Pediatrics, Committee on Environmental Health. (2004) Ambient air pollution: health hazards to children. Pediatrics 114: 1699-1707.

American Thoracic Society. (1991) Lung function testing: selection of reference values and interpretative strategies Am. Rev. Respir. Dis. 144: 1202-1218.

Anderson, H. R.; Ponce de Leon, A.; Bland, J. M.; Bower, J. S.; Strachan, D. P. (1996) Air pollution and daily mortality in London: 1987-92. Br. Med. J. 312: 665-669.

Anderson, H. R.; Spix, C.; Medina, S.; Schouten, J. P.; Castellsague, J.; Rossi, G.; Zmirou, D.; Touloumi, G.; Wojtyniak, B.; Ponka, A.; Bacharova, L.; Schwartz, J.; Katsouyanni, K. (1997) Air pollution and daily admissions for chronic obstructive pulmonary disease in 6 European cities: results from the APHEA project. Eur. Respir. J. 10: 1064-1071.

Anderson, H. R.; Ponce de Leon, A.; Bland, J. M.; Bower, J. S.; Emberlin, J.; Strachen, D. P. (1998) Air pollution, pollens, and daily admissions for asthma in London 1987-92. Thorax 53: 842-848.

Anderson, H. R.; Bremner, S. A.; Atkinson, R. W.; Harrison, R. M.; Walters, S. (2001) Particulate matter and daily mortality and hospital admissions in the west midlands conurbation of the United Kingdom: associations with fine and coarse particles, black smoke and sulphate. Occup. Environ. Med. 58: 504-510.

Atkinson, R. W.; Anderson, H. R.; Strachan, D. P.; Bland, J. M.; Bremner, S. A.; Ponce de Leon, A. (1999a) Short-term associations between outdoor air pollution and visits to accident and emergency departments in London for respiratory complaints. Eur. Respir. J. 13: 257-265.

Atkinson, R. W.; Bremner, S. A.; Anderson, H. R.; Strachan, D. P.; Bland, J. M.; Ponce de Leon, A. (1999b) Short-term associations between emergency hospital admissions for respiratory and cardiovascular disease and outdoor air pollution in London. Arch. Environ. Health 54: 398-411.

Atkinson, R. W.; Anderson, H. R.; Sunyer, J.; Ayres, J.; Baccini, M.; Vonk, J. M.; Boumghar, A.; Forastiere, F.; Forsberg, B.; Touloumi, G.; Schwartz, J.; Katsouyanni, K. (2001) Acute effects of particulate air pollution on respiratory admissions: results from APHEA 2 project. Am. J. Respir. Crit. Care Med. 164: 1860-1866.

Avol, E. L.; Trim, S. C.; Little, D. E.; Spier, C. E.; Smith, M. N.; Peng, R.-C.; Linn, W. S.; Hackney, J. D.; Gross, K. B.; D'Arcy, J. B.; Gibbons, D.; Higgins, I. T. T. (1990) Ozone exposure and lung function in children attending a southern California summer camp. Presented at: 83rd annual meeting and exhibition of the Air & Waste Management Association; June; Pittsburgh, PA. Pittsburgh, PA: Air & Waste Management Association; paper no. 90-150.3.

Avol, E. L.; Navidi, W. C.; Rappaport, E. B.; Peters, J. M. (1998) Acute effects of ambient ozone on asthmatic, wheezy, and healthy children. Cambridge, MA: Health Effects Institute; research report no. 82.

Avol, E. L.; Gauderman, W. J.; Tan, S. M.; London, S. J.; Peters, J. M. (2001) Respiratory effects of relocating to areas of differing air pollution levels. Am. J. Respir. Crit. Care Med. 164: 2067-2072.

Ballester, F.; Tenías, J. M.; Pérez-Hoyos, S. (2001) Air pollution and emergency hospital admissions for cardiovascular diseases in Valencia, Spain. J. Epidemiol. Community Health 55: 57-65.

Barker, D. J. P.; Gluckman, P. D.; Godfrey, K. M.; Harding, J. E.; Owens, J. A.; Robinson, J. S. (1993) Fetal nutrition and cardiovascular disease in adult life. Lancet 341: 938-941.

Bates, D. V. (2005) Ambient ozone and mortality. Epidemiology 16: 427-429.

Bates, D. V.; Sizto, R. (1983) Relationship between air pollutant levels and hospital admissions in Southern Ontario. Can. J. Public Health 74: 117-122.

Bates, D. V.; Sizto, R. (1987) Air pollution and hospital admissions in southern Ontario: the acid summer haze effect. Environ. Res. 43: 317-331.

Bates, D. V.; Sizto, R. (1989) The Ontario Air Pollution study: identification of the causative agent. Environ. Health Perspect. 79: 69-72.

Bates, D. V.; Baker-Anderson, M.; Sizto, R. (1990) Asthma attack periodicity: a study of hospital emergency visits in Vancouver. Environ. Res. 51: 51-70.

Bedi, J. F.; Horvath, S. M.; Drechsler-Parks, D. M. (1989) Adaptation by older individuals repeatedly exposed to 0.45 parts per million ozone for two hours. JAPCA 39: 194-199.

Beeson, W. L.; Abbey, D. E.; Knutsen, S. F. (1998) Long-term concentrations of ambient air pollutants and incident lung cancer in California adults: results from the AHSMOG study. Environ. Health Perspect. 106: 813-823.

Bell, M. L.; McDermott, A.; Zeger, S. L.; Samet, J. M.; Dominici, F. (2004) Ozone and short-term mortality in 95 US urban communities, 1987-2000. JAMA J. Am. Med. Assoc. 292: 2372-2378.

Bell, M. L.; Dominici, F.; Samet, J. M. (2005) A meta-analysis of time-series studies of ozone and mortality with comparison to the national morbidity, mortality, and air pollution study. Epidemiology 16: 436-445.

Bobak, M. (2000) Outdoor air pollution, low birth weight, and prematurity. Environ. Health Perspect. 108: 173-176.

Bobak, M.; Leon, D. A. (1992) Air pollution and infant mortality in the Czech Republic, 1986-1988. Lancet (8826): 1010-1014.

Bobak, M.; Leon, D. A. (1999) Pregnancy outcomes and outdoor air pollution: an ecological study in districts of the Czech Republic 1986-8. Occup. Environ. Med. 56: 539-543.

Borja-Aburto, V. H.; Loomis, D. P.; Bangdiwala, S. I.; Shy, C. M.; Rascon-Pacheco, R. A. (1997) Ozone, suspended particulates, and daily mortality in Mexico City. Am. J. Epidemiol. 145: 258-268.

Borja-Aburto, V. H.; Castillejos, M.; Gold, D. R.; Bierzwinski, S.; Loomis, D. (1998) Mortality and ambient fine particles in southwest Mexico City, 1993-1995. Environ. Health Perspect. 106: 849-855.

Bourcier, T.; Viboud, C.; Cohen, J.-C.; Thomas, F.; Bury, T.; Cadiot, L.; Mestre, O.; Flahault, A.; Borderie, V.; Laroche, L. (2003) Effects of air pollution and climatic conditions on the frequency of ophthalmological emergency examinations. Br. J. Ophthalmol. 87: 809-811.

Braga, A. L. F.; Conceição, G. M. S.; Pereira, L. A. A.; Kishi, H. S.; Pereira, J. C. R.; Andrade, M. F.; Gonçalves, F. L. T.; Saldiva, P. H. N.; Latorre, M. R. D. O. (1999) Air pollution and pediatric respiratory hospital admissions in São Paulo, Brazil. J. Environ. Med. 1: 95-102.

Brauer, M.; Brook, J. R. (1997) Ozone personal exposures and health effects for selected groups residing in the Fraser Valley. In: Steyn, D. G.; Bottenheim, J. W., eds. The Lower Fraser Valley Oxidants/Pacific '93 Field Study. Atmos. Environ. 31: 2113-2121.

Brauer, M.; Blair, J.; Vedal, S. (1996) Effect of ambient ozone exposure on lung function in farm workers. Am. J. Respir. Crit. Care Med. 154: 981-987.

Bremner, S. A.; Anderson, H. R.; Atkinson, R. W.; McMichael, A. J.; Strachan, D. P.; Bland, J. M.; Bower, J. S. (1999) Short term associations between outdoor air pollution and mortality in London 1992-4. Occup. Environ. Med. 56: 237-244.

Brook, R. D.; Franklin, B.; Cascio, W.; Hong, Y.; Howard, G.; Lipsett, M.; Luepker, R.; Mittleman, M.; Samet, J.; Smith, S. C., Jr.; Tager, I. (2004) Air pollution and cardiovascular disease. A statement for healthcare professionals from the Expert Panel on Population and Prevention Science of the American Heart Association. Circulation 109: 2655-2671.

Buchdahl, R.; Parker, A.; Stebbings, T.; Babiker, A. (1996) Association between air pollution and acute childhood wheezy episodes: prospective observational study. Br. Med. J. 312: 661-664.

Buchdahl, R.; Willems, C. D.; Vander, M.; Babiker, A. (2000) Associations between ambient ozone, hydrocarbons, and childhood wheezy episodes: a prospective observational study in south east London. Occup. Environ. Med. 57: 86-93.

Burchfiel, C. M.; Marcus, E. B.; Curb, J. D.; Maclean, C. J.; Vollmer, W. M.; Johnson, L. R.; Fong, K.; Rodriguez, B. L.; Masaki, K. H.; Buist, A. S. (1995) Effects of smoking and smoking cessation on longitudinal decline in pulmonary function. Am. J. Respir. Crit. Care Med. 151: 1778-1785.

Burnett, R. T.; Dales, R. E.; Raizenne, M. E.; Krewski, D.; Summers, P. W.; Roberts, G. R.; Raad-Young, M.; Dann, T.; Brook, J. (1994) Effects of low ambient levels of ozone and sulfates on the frequency of respiratory admissions to Ontario hospitals. Environ. Res. 65: 172-194.

Burnett, R. T.; Dales, R.; Krewski, D.; Vincent, R.; Dann, T.; Brook, J. R. (1995) Associations between ambient particulate sulfate and admissions to Ontario hospitals for cardiac and respiratory diseases. Am. J. Epidemiol. 142: 15-22.

Burnett, R. T.; Brook, J. R.; Yung, W. T.; Dales, R. E.; Krewski, D. (1997a) Association between ozone and hospitalization for respiratory diseases in 16 Canadian cities. Environ. Res. 72: 24-31.

Burnett, R. T.; Cakmak, S.; Brook, J. R.; Krewski, D. (1997b) The role of particulate size and chemistry in the association between summertime ambient air pollution and hospitalization for cardiorespiratory diseases. Environ. Health Perspect. 105: 614-620.

Burnett, R. T.; Smith-Doiron, M.; Stieb, D.; Cakmak, S.; Brook, J. R. (1999) Effects of particulate and gaseous air pollution on cardiorespiratory hospitalizations. Arch. Environ. Health 54: 130-139.

Burnett, R. T.; Smith-Doiron, M.; Stieb, D.; Raizenne, M. E.; Brook, J. R.; Dales, R. E.; Leech, J. A.; Cakmak, S.; Krewski, D. (2001) Association between ozone and hospitalization for acute respiratory diseases in children less than 2 years of age. Am. J. Epidemiol. 153: 444-452.

Burr, D.; Doss, H. (2005) A Bayesian semiparametric model for random-effects meta-analysis. J. Am. Stat. Assoc. 100: 242-251.
Calderón-Garcidueñas, L.; Rodriguez-Alcaraz, A.; García, R.; Ramírez, L.; Barragan, G. (1995) Nasal inflammatory responses in children exposed to a polluted urban atmosphere. J. Toxicol. Environ. Health 45: 427-437.
Calderón-Garcidueñas, L.; Osnaya, N.; Rodríguez-Alcaraz, A.; Villarreal-Calderón, A. (1997) DNA damage in nasal respiratory epithelium from children exposed to urban pollution. Environ. Mol. Mutagen. 30: 11-20.
Calderón-Garcidueñas, L.; Wen-Wang, L.; Zhang, Y.-J.; Rodriguez-Alcaraz, A.; Osnaya, N.; Villarreal-Calderón, A.; Santella, R. M. (1999) 8-hydroxy-2'-deoxyguanosine, a major mutagenic oxidative DNA lesion, and DNA strand breaks in nasal respiratory epithelium of children exposed to urban pollution. Environ. Health Perspect. 107: 469-474.
Calderón-Garcidueñas, L.; Valencia-Salazar, G.; Rodríguez-Alcaraz, A.; Gambling, T. M.; García, R.; Osnaya, N.; Villarreal-Calderon, A.; Devlin, R. B.; Carson, J. L. (2001) Ultrastructural nasal pathology in children chronically and sequentially exposed to air pollutants. Am. J. Respir. Cell Mol. Biol. 24: 132-138.
Calderón-Garcidueñas, L.; Mora-Tiscareño, A.; Fordham, L. A.; Valencia-Salazar, G.; Chung, C. J.; Rodriguez-Alcaraz, A.; Paredes, R.; Variakojis, D.; Villarreal-Calderón, A.; Flores-Camacho, L.; Antunez-Solis, A.; Henríquez-Roldán, C.; Hazucha, M. J. (2003) Respiratory damage in children exposed to urban pollution. Pediatr. Pulmonol. 36: 148-161.
Cassino, C.; Ito, K.; Bader, I.; Ciotoli, C.; Thurston, G.; Reibman, J. (1999) Cigarette smoking and ozone-associated emergency department use for asthma by adults in New York City. Am. J. Respir. Crit. Care Med. 159: 1773-1779.
Castellsague, J.; Sunyer, J.; Sáez, M.; Antó, J. M. (1995) Short-term association between air pollution and emergency room visits for asthma in Barcelona. Thorax 50: 1051-1056.
Castillejos, M.; Gold, D. R.; Damokosh, A. I.; Serrano, P.; Allen, G.; McDonnell, W. F.; Dockery, D.; Velasco, S. R.; Hernandez, M.; Hayes, C. (1995) Acute effects of ozone on the pulmonary function of exercising schoolchildren from Mexico City. Am. J. Respir. Crit. Care Med. 152: 1501-1507.
Chan, C.-C.; Wu, T.-H. (2005) Effects of ambient ozone exposure on mail carriers' peak expiratory flow rates. Environ. Health Perspect. 113: 735-738.
Chang, C.-C.; Tsai, S.-S.; Ho, S.-C.; Yang, C.-Y. (2005) Air pollution and hospital admissions for cardiovascular disease in Taipei, Taiwan. Environ. Res. 98: 114-119.
Charpin, D.; Pascal, L.; Birnbaum, J.; Armengaud, A.; Sambuc, R.; Lanteaume, A.; Vervloet, D. (1999) Gaseous air pollution and atopy. Clin. Exp. Allergy 29: 1474-1480.
Chen, P.-C.; Lai, Y.-M.; Wang, J.-D.; Yang, C.-Y.; Hwang, J.-S.; Kuo, H.-W.; Huang, S.-L.; Chan, C.-C. (1998) Adverse effect of air pollution on respiratory health of primary school children in Taiwan. Environ. Health Perspect. 106: 331-335.
Chen, P.-C.; Lai, Y.-M.; Chan, C.-C.; Hwang, J.-S.; Yang, C.-Y.; Wang, J.-D. (1999) Short-term effect of ozone on the pulmonary function of children in primary school. Environ. Health Perspect. 107: 921-925.
Chen, L.; Jennison, B. L.; Yang, W.; Omaye, S. T. (2000) Elementary school absenteeism and air pollution. Inhalation Toxicol. 12: 997-1016.
Chen, L.; Yang, W.; Jennison, B. L.; Goodrich, A.; Omaye, S. T. (2002) Air pollution and birth weight in northern Nevada, 1991-1999. Inhalation Toxicol. 14: 141-157.
Chew, F. T.; Goh, D. Y. T.; Ooi, B. C.; Saharom, R.; Hui, J. K. S.; Lee, B. W. (1999) Association of ambient air-pollution levels with acute asthma exacerbation among children in Singapore. Allergy (Copenhagen) 54: 320-329.
Chock, D. P.; Winkler, S. L.; Chen, C. (2000) A study of the association between daily mortality and ambient air pollutant concentrations in Pittsburgh, Pennsylvania. J. Air Waste Manage. Assoc. 50: 1481-1500.
Cifuentes, L. A.; Vega, J.; Köpfer, K.; Lave, L. B. (2000) Effect of the fine fraction of particulate matter versus the coarse mass and other pollutants on daily mortality in Santiago, Chile. J. Air Waste Manage. Assoc. 50: 1287-1298.
Clyde, M. A. (1999) Bayesian model averaging and model search strategies. In: Bernardo, J. M.; Berger, J. O.; Dawid, A. P.; Smith, A. F. M., eds. Bayesian Statistics 6: proceedings of the Sixth Valencia International Meeting, June; pp. 157-185. Oxford, UK. Oxford, UK: Clarendon Press.
Clyde, M. (2000) Model uncertainty and health effect studies for particulate matter. Environmetrics 11: 745-763.
Clyde, M. A.; Guttorp, P.; Sullivan, E. (2000) Effects of ambient fine and coarse particles on mortality in Phoenix, Arizona. Seattle, WA: University of Washington, National Research Center for Statistics and the Environment; NRCSE technical report series, NRCSE-TRS no. 040. Available: http://www.nrcse.washington.edu/pdf/trs40_pm.pdf [18 October, 2004].

Cody, R. P.; Weisel, C. P.; Birnbaum, G.; Lioy, P. J. (1992) The effect of ozone associated with summertime photochemical smog on the frequency of asthma visits to hospital emergency departments. Environ. Res. 58: 184-194.

Cross, D.; Nelson, H. S. (1991) The role of the peak flow meter in the diagnosis and management of asthma. J. Allergy Clin. Immunol. 87: 120-128.

Cuijpers, C. E. J.; Swaen, G. M. H.; Wesseling, G.; Wouters, E. F. M. (1994) Acute respiratory effects of summer smog in primary school children. Toxicol. Lett. 72: 227-235.

Dab, W.; Medina, S.; Quénel, P.; Le Moullec, Y.; Le Tertre, A.; Thelot, B.; Monteil, C.; Lameloise, P.; Pirard, P.; Momas, I.; Ferry, R.; Festy, B. (1996) Short term respiratory health effects of ambient air pollution: results of the APHEA project in Paris. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Commun. Health 50(suppl. 1): S42-S46.

Dejmek, J.; Selevan, S. G.; Beneš, I.; Solanský, I.; Šräm, R. J. (1999) Fetal growth and maternal exposure to particulate matter during pregnancy. Environ. Health Perspect. 107: 475-480.

Dejmek, J.; Solanský, I.; Beneš, I.; Leníček, J.; Šräm, R. J. (2000) The impact of polycyclic aromatic hydrocarbons and fine particles on pregnancy outcome. Environ. Health Perspect. 108: 1159-1164.

De Leon, S. F.; Thurston, G. D.; Ito, K. (2003) Contribution of respiratory disease to nonrespiratory mortality associations with air pollution. Am. J. Respir. Crit. Care Med. 167: 1117-1123.

Delfino, R. J.; Coate, B. D.; Zeiger, R. S.; Seltzer, J. M.; Street, D. H.; Koutrakis, P. (1996) Daily asthma severity in relation to personal ozone exposure and outdoor fungal spores. Am. J. Respir. Crit. Care Med. 154: 633-641.

Delfino, R. J.; Zeiger, R. S.; Seltzer, J. M.; Street, D. H.; Matteucci, R. M.; Anderson, P. R.; Koutrakis, P. (1997a) The effect of outdoor fungal spore concentrations on daily asthma severity. Environ. Health Perspect. 105: 622-635.

Delfino, R. J.; Murphy-Moulton, A. M.; Burnett, R. T.; Brook, J. R.; Becklake, M. R. (1997b) Effects of air pollution on emergency room visits for respiratory illnesses in Montreal, Quebec. Am. J. Respir. Crit. Care Med. 155: 568-576.

Delfino, R. J.; Zeiger, R. S.; Seltzer, J. M.; Street, D. H. (1998a) Symptoms in pediatric asthmatics and air pollution: differences in effects by symptom severity, anti-inflammatory medication use and particulate averaging time. Environ. Health Perspect. 106: 751-761.

Delfino, R. J.; Murphy-Moulton, A. M.; Becklake, M. R. (1998b) Emergency room visits for respiratory illnesses among the elderly in Montreal: association with low level ozone exposure. Environ. Res. 76: 67-77.

Delfino, R. J.; Gone, H.; Linn, W. S.; Pellizzari, E. D.; Hu, Y. (2003) Asthma symptoms in Hispanic children and daily ambient exposures to toxic and criteria air pollutants. Environ. Health Perspect. 111: 647-656.

Delfino, R. J., Quintana, P. J. E.; Floro, J.; Gastañaga, V. M.; Samimi, B. S.; Kleinman, M. T.; Liu, L.-J. S.; Bufalino, C.; Wu, C.-F.; McLaren, C. E. (2004) Association of FEV_1 in asthmatic children with personal and microenvironmental exposure to airborne particulate matter. Environ. Health Perspect. 112: 932-941.

Desqueyroux, H.; Pujet, J.-C.; Prosper, M.; Squinazi, F.; Momas, I. (2002a) Short-term effects of low-level air pollution on respiratory health of adults suffering from moderate to severe asthma. Environ. Res. A 89: 29-37.

Desqueyroux, H.; Pujet, J.-C.; Prosper, M.; Le Moullec, Y.; Momas, I. (2002b) Effects of air pollution on adults with chronic obstructive pulmonary disease. Arch. Environ. Health 57: 554-560.

Detels, R.; Tashkin, D. P.; Sayre, J. W.; Rokaw, S. N.; Coulson, A. H.; Massey, F. J., Jr.; Wegman, D. H. (1987) The UCLA population studies of chronic obstructive respiratory disease: 9. lung function changes associated with chronic exposure to photochemical oxidants; a cohort study among never-smokers. Chest 92: 594-603.

Devlin, R. B.; McDonnell, W. F.; Mann, R.; Becker, S.; House, D. E.; Schreinemachers, D.; Koren, H. S. (1991) Exposure of humans to ambient levels of ozone for 6.6 hours causes cellular and biochemical changes in the lung. Am. J. Respir. Cell Mol. Biol. 4: 72-81.

Díaz, J.; García, R.; Ribera, P.; Alberdi, J. C.; Hernández, E.; Pajares, M. S.; Otero, A. (1999) Modeling of air pollution and its relationship with mortality and morbidity in Madrid, Spain. Int. Arch. Occup. Environ. Health 72: 366-376.

Dietert, R. R.; Etzel, R. A.; Chen, D.; Halonen, M.; Holladay, S. D.; Jarabek, A. M.; Landreth, K.; Peden, D. B.; Pinkerton, K.; Smialowicz, R. J.; Zoetis, T. (2000) Workshop to identify critical window of exposure for children's health: immune and respiratory systems work group summary. Environ. Health Perspect. Suppl. 108(3): 483-490.

Dockery, D. W.; Schwartz, J.; Spengler, J. D. (1992) Air pollution and daily mortality: associations with particulates and acid aerosols. Environ. Res. 59: 362-373.

Dockery, D. W.; Luttmann-Gibson, H.; Rich, D. Q.; Link, M. S.; Mittleman, M. A.; Gold, D. R.; Koutrakis, P.; Schwartz, J. D.; Verrier, R. L. (2005) Association of air pollution with increased incidence of ventricular tachyarrhythmias recorded by implanted cardioverter defibrillators. Environ. Health Perspect. 113: 670-674.

Dominici, F.; McDermott, A.; Zeger, S. L.; Samet, J. M. (2002) On the use of generalized additive models in time-series studies of air pollution and health. Am. J. Epidemiol. 156: 193-203.

Dominici, F.; McDermott, A.; Daniels, M.; Zeger, S. L.; Samet, J. M. (2003) Mortality among residents of 90 cities. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 9-24. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [12 May, 2004].

Drechsler-Parks, D. M. (1995) The dose-response relationship in older men exposed to ozone. Exp. Gerontol. 30: 65-75.

Fairley, D. (1999) Daily mortality and air pollution in Santa Clara County, California: 1989-1996. Environ. Health Perspect. 107: 637-641.

Fairley, D. (2003) Mortality and air pollution for Santa Clara County, California, 1989-1996. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 97-106. Available: http://www.healtheffects.org/news.htm [16 May, 2003].

Flachaire, E.; Nuñez, O. (2002) Estimation of income distribution and detection of subpopulations: an explanatory model. Paris, France: Document de travail de la MSE, EUREQua 2002.86, Université Paris1 Panthéon-Sorbonne. Available: http://eurequa.univ-paris1.fr/membres/flachaire/research/Flachaire_Nunez_02.pdf [14 June, 2005].

Fortoul, T. I.; Valverde, M.; López M. D. C.; Bizarro, P.; López, I.; Sanchez, I.; Colín-Barenque, L.; Avila-Costa, M. R.; Rojas, E.; Ostrosky-Shejet, P. (2003) Single-cell gel electrophoresis assay of nasal epithelium and leukocytes from asthmatic and nonasthmatic subjects in Mexico City. Arch. Environ. Health 58: 348-352.

Friedman, M. S.; Powell, K. E.; Hutwagner, L.; Graham, L. M.; Teague, W. G. (2001) Impact of changes in transportation and commuting behaviors during the 1996 summer olympic games in Atlanta on air quality and childhood asthma. JAMA J. Am. Med. Assoc. 285: 897-905.

Frischer, T. M.; Kühr, J.; Pullwitt, A.; Meinert, R.; Forster, J.; Studnicka, M.; Koren, H. (1993) Ambient ozone causes upper airways inflammation in children. Am. Rev. Respir. Dis. 148: 961-964.

Frischer, T.; Pullwitt, A.; Kuehr, K.; Meinert, R.; Haschke, N.; Studnicka, M.; Lubec, G. (1997) Aromatic hydroxylation in nasal lavage fluid following ambient ozone exposure. Free Radical Biol. Med. 22: 201-207.

Frischer, T.; Studnicka, M.; Gartner, C.; Tauber, E.; Horak, F.; Veiter, A.; Spengler, J.; Kühr, J.; Urbanek, R. (1999) Lung function growth and ambient ozone: a three-year population study in school children. Am. J. Respir. Crit. Care Med. 160: 390-396.

Frischer, T.; Studnicka, M.; Halmerbauer, G.; Horak, F.; Gartner, C.; Tauber, E.; Koller, D. Y. (2001) Ambient ozone exposure is associated with eosinophil activation in healthy children. Clin. Exp. Allergy 31: 1213-1219.

Fuhlbrigge, A.; Kitch, B.; Paltiel, A. D.; Kuntz, K. M.; Neumann, P. J.; Dockery, D. W.; Weiss, S. T. (2001) FEV_1 is associated with risk of asthma attacks in a pediatric population. J. Allergy Clin. Immunol. 107: 61-67.

Galbraith, R. F. (1994) Some applications of radial plots. J. Am. Stat. Assoc. 89: 1232-1242.

Galizia, A.; Kinney, P. L. (1999) Long-term residence in areas of high ozone: associations with respiratory health in a nationwide sample of nonsmoking young adults. Environ. Health Perspect. 107: 675-679.

Gamble, J. L. (1998) Effects of ambient air pollution on daily mortality: a time series analysis of Dallas, Texas, 1990-1994. Presented at: 91st annual meeting and exhibition of the Air & Waste Management Association; June; San Diego, CA. Pittsburgh, PA: Air & Waste Management Association; paper no. 98-MP26.03.

Garcia-Aymerich, J.; Tobias, A.; Antó, J. M.; Sunyer, J. (2000) Air pollution and mortality in a cohort of patients with chronic obstructive pulmonary disease: a time series analysis. J. Epidemiol. Community Health 54: 73-74.

Garty, B. Z.; Kosman, E.; Ganor, E.; Berger, V.; Garty, L.; Wietzen, T.; Waisman, Y.; Mimouni, M.; Waisel, Y. (1998) Emergency room visits of asthmatic children, relation to air pollution, weather, and airborne allergens. Ann. Allergy Asthma Immunol. 81: 563-570.

Gauderman, W. J.; McConnell, R.; Gilliland, F.; London, S.; Thomas, D.; Avol, E.; Vora, H.; Berhane, K.; Rappaport, E. B.; Lurmann, F.; Margolis, H. G.; Peters, J. (2000) Association between air pollution and lung function growth in southern California children. Am. J. Respir. Crit. Care Med. 162: 1383-1390.

Gauderman, W. J.; Gilliland, G. F.; Vora, H.; Avol, E.; Stram, D.; McConnell, R.; Thomas, D.; Lurmann, F.; Margolis, H. G.; Rappaport, E. B.; Berhane, K.; Peters, J. M. (2002) Association between air pollution and lung function growth in southern California children: results from a second cohort. Am. J. Respir. Crit. Care Med. 166: 76-84.

Gauderman, W. J.; Avol, E.; Gilliland, F.; Vora, H.; Thomas, D.; Berhane, K.; McConnell, R.; Kuenzli, N.; Lurmann, F.; Rappaport, E.; Margolis, H.; Bates, D.; Peters, J. (2004a) The effect of air pollution on lung development from 10 to 18 years of age. N. Engl. J. Med. 351: 1057-1067.

Gauderman, W. J.; Avol, E.; Gilliland, F. (2004b) Air pollution and lung function [reply letter]. N. Engl. J. Med. 351: 2653.

Gent, J. F.; Triche, E. W.; Holford, T. R.; Belanger, K.; Bracken, M. B.; Beckett, W. S.; Leaderer, B. P. (2003) Association of low-level ozone and fine particles with respiratory symptoms in children with asthma. JAMA J. Am. Med. Assoc. 290: 1859-1867.

George, E. I. (1999) Comment on "Hoeting, J. A.; Madigan, D.; Raftery, A. E.; Volinsky, C. T. 1999. Bayesian model averaging: a tutorial. Stat. Sci. 14: 409-412."

Gielen, M. H.; Van Der Zee, S. C.; Van Wijnen, J. H.; Van Steen, C. J.; Brunekreef, B. (1997) Acute effects of summer air pollution on respiratory health of asthmatic children. Am. J. Respir. Crit. Care Med. 155: 2105-2108.

Gilliland, F. D.; Berhane, K.; Rappaport, E. B.; Thomas, D. C.; Avol, E.; Gauderman, W. J.; London, S. J.; Margolis, H. G.; McConnell, R.; Islam, K. T.; Peters, J. M. (2001) The effects of ambient air pollution on school absenteeism due to respiratory illnesses. Epidemiology 12: 43-54.

Gold, D. R.; Damokosh, A. I.; Pope, C. A., III; Dockery, D. W.; McDonnell, W. F.; Serrano, P.; Retama, A.; Castillejos, M. (1999) Particulate and ozone pollutant effects on the respiratory function of children in southwest Mexico City. Epidemiology 10: 8-16.

Gold, D. R.; Litonjua, A.; Schwartz, J.; Lovett, E.; Larson, A.; Nearing, B.; Allen, G.; Verrier, M.; Cherry, R.; Verrier, R. (2000) Ambient pollution and heart rate variability. Circulation 101: 1267-1273.

Gold, D. R.; Schwartz, J.; Litonjua, A.; Verrier, R.; Zanobetti, A. (2003) Ambient pollution and reduced heart rate variability. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 107-112. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [18 October, 2004].

Goldberg, M. S.; Burnett, R. T. (2003) Revised analysis of the Montreal time-series study. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 113-132. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [13 August 2003).

Goldberg, M. S.; Burnett, R. T.; Brook, J.; Bailar, J. C., III; Valois, M.-F.; Vincent, R. (2001) Associations between daily cause-specific mortality and concentrations of ground-level ozone in Montreal, Quebec. Am. J. Epidemiol. 154: 817-826.

Goldberg, M. S.; Burnett, R. T.; Valois, M.-F.; Flegel, K.; Bailar, J. C., III; Brook, J.; Vincent, R.; Radon, K. (2003) Associations between ambient air pollution and daily mortality among persons with congestive heart failure. Environ. Res. 91: 8-20.

Gong, H., Jr.; Wong, R.; Sarma, R. J.; Linn, W. S.; Sullivan, E. D.; Shamoo, D. A.; Anderson, K. R.; Prasad, S. B. (1998a) Cardiovascular effects of ozone exposure in human volunteers. Am. J. Respir. Crit. Care Med. 158: 538-546.

Gong, H., Jr.; Simmons, M. S.; Linn, W. S.; McDonnell, W. F.; Westerdahl, D. (1998b) Relationship between acute ozone responsiveness and chronic loss of lung function in residents of a high-ozone community. Arch. Environ. Health 53: 313-319.

Gonzales, M.; Ngo, L.; Hammond, S. K.; Tager, I. (2003) Validation of a questionnaire and microenvironmental model for estimating past exposures to ozone. Int. J. Environ. Health Res. 13: 249-260.

Goodman, S. N. (2005) The methodologic ozone effect. Epidemiology 16: 430-435.

Goss, C. H.; Newsom, S. A.; Schildcrout, J. S.; Sheppard, L.; Kaufman, J. D. (2004) Effect of ambient air pollution on pulmonary exacerbations and lung function in cystic fibrosis. Am. J. Respir. Crit. Care Med. 169: 816-821.

Gouveia, N.; Fletcher, T. (2000a) Respiratory diseases in children and outdoor air pollution in Sao Paulo, Brazil: a time series analysis. Occup. Environ. Med. 57: 477-483.

Gouveia, N.; Fletcher, T. (2000b) Time series analysis of air pollution and mortality: effects by cause, age and socioeconomic status. J. Epidemiol. Community Health 54: 750-755.

Gouveia, N.; Bremner, S. A.; Novaes, H. M. D. (2004) Association between ambient air pollution and birth weight in São Paulo, Brazil. J. Epidemiol. Community Health 58: 11-17.

Greer, J. R.; Abbey, D. E.; Burchette, R. J. (1993) Asthma related to occupational and ambient air pollutants in nonsmokers. J. Occup. Med. 35: 909-915.

Gryparis, A.; Forsberg, B.; Katsouyanni, K.; Analitis, A.; Touloumi, G.; Schwartz, J.; Samoli, E.; Medina, S.; Anderson, H. R.; Niciu, E. M.; Wichmann, H.-E.; Kriz, B.; Kosnik, M.; Skorkovsky, J.; Vonk, J. M.; Dörtbudak, Z. (2004) Acute effects of ozone on mortality from the "air pollution and health: a European approach" project. Am. J. Respir. Crit. Care Med. 170: 1080-1087.

Gwynn, R. C.; Thurston, G. D. (2001) The burden of air pollution: impacts among racial minorities. Environ. Health Perspect. Suppl. 109(4): 501-506.

Gwynn, R. C.; Burnett, R. T.; Thurston, G. D. (2000) A time-series analysis of acidic particulate matter and daily mortality and morbidity in the Buffalo, New York, region. Environ. Health Perspect. 108: 125-133.

Ha, E.-H.; Hong, Y.-C.; Lee, B.-E.; Woo, B.-H.; Schwartz, J.; Christiani, D. C. (2001) Is air pollution a risk factor for low birth weight in Seoul? Epidemiology 12: 643-648.

Hagen, J. A.; Nafstad, P.; Skrondal, A.; Bjørkly, S.; Magnus, P. (2000) Associations between outdoor air pollutants and hospitalization for respiratory diseases. Epidemiology 11: 136-140.

Hajat, S.; Haines, A.; Goubet, S. A.; Atkinson, R. W.; Anderson, H. R. (1999) Association of air pollution with daily GP consultations for asthma and other lower respiratory conditions in London. Thorax 54: 597-605.

Hajat, S.; Anderson, H. R.; Atkinson, R. W.; Haines, A. (2002) Effects of air pollution on general practitioner consultations for upper respiratory diseases in London. Occup. Environ. Med. 59: 294-299.

Hankinson, J. L.; Odencrantz, J. R.; Fedan, K. B. (1999) Spirometric reference values from a sample of the general U.S. population. Am. J. Respir. Crit. Care Med. 159: 179-187.

Health Effects Institute. (2003) Revised analyses of time-series studies of air pollution and health. Boston, MA: Health Effects Institute; special report. Available: Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [27 June 2003].

Hedley, A. J.; Wong, C.-M.; Thach, T. Q.; Ma, S.; Lam, T.-H.; Anderson, H. R. (2002) Cardiorespiratory and all-cause mortality after restrictions on sulphur content of fuel in Hong Kong: an intervention study. Lancet 360: 1646-1652.

Hernández-Garduño, E.; Pérez-Neria, J.; Paccagnella, A. M.; Piña-García, M.; Munguía-Castro, M.; Catalán-Vázquez, M.; Rojas-Ramos, M. (1997) Air pollution and respiratory health in Mexico City. J. Occup. Environ. Med. 39: 299-307.

Higgins, I. T. T.; D'Arcy, J. B.; Gibbons, D. I.; Avol, E. L.; Gross, K. B. (1990) Effect of exposures to ambient ozone on ventilatory lung function in children. Am. Rev. Respir. Dis. 141: 1136-1146.

Hill, A. B. (1965) The environment and disease: association or causation? Proc. R. Soc. Med. 58: 295-300.

Hiltermann, T. J. N.; Stolk, J.; Van der Zee, S. C.; Brunekreef, B.; De Bruijne, C. R.; Fischer, P. H.; Ameling, C. B.; Sterk, P. J.; Hiemstra, P. S.; Van Bree, L. (1998) Asthma severity and susceptibility to air pollution. Eur. Respir. J. 11: 686-693.

Hoek, G. (2003) Daily mortality and air pollution in The Netherlands. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 133-142. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [12 May, 2004].

Hoek, G.; Brunekreef, B. (1995) Effect of photochemical air pollution on acute respiratory symptoms in children. Am. J. Respir. Crit. Care Med. 151: 27-32.

Hoek, G.; Brunekreef, B.; Verhoeff, A.; Van Wijnen, J.; Fischer, P. (2000) Daily mortality and air pollution in the Netherlands. J. Air Waste Manage. Assoc. 50: 1380-1389.

Hoek, G.; Brunekreef, B.; Fischer, P.; Van Wijnen, J. (2001) The association between air pollution and heart failure, arrhythmia, embolism, thrombosis, and other cardiovascular causes of death in a time series study. Epidemiology 12: 355-357.

Hoeting, J. A.; Madigan, D.; Raftery, A. E.; Volinsky, C. T. (1999) Bayesian model averaging: a tutorial. Stat. Sci. 14: 382-417.

Holguín, F.; Téllez-Rojo, M. M.; Hernández, M.; Cortez, M.; Chow, J. C.; Watson, J. G.; Mannino, D.; Romieu, I. (2003) Air pollution and heart rate variability among the elderly in Mexico City. Epidemiology 14: 521-527.

Holmén, A.; Blomqvist, J.; Frindberg, H.; Johnelius, Y.; Eriksson, N. E.; Henricson, K. Å.; Herrström, P.; Högstedt, B. (1997) Frequency of patients with acute asthma in relation to ozone, nitrogen dioxide, other pollutants of ambient air and meteorological observations. Int. Arch. Occup. Environ. Health 69: 317-322.

Hong, Y.-C.; Lee, J.-T.; Kim, H.; Ha, E.-H.; Schwartz, J.; Christiani, D. C. (2002) Effects of air pollutants on acute stroke mortality. Environ. Health Perspect. 110: 187-191.

Höppe, P.; Praml, G.; Rabe, G.; Lindner, J.; Fruhmann, G.; Kessel, R. (1995a) Environmental ozone field study on pulmonary and subjective responses of assumed risk groups. Environ. Res. 71: 109-121.

Höppe, P.; Lindner, J.; Praml, G.; Brönner, N. (1995b) Effects of environmental ozone on the lung function of senior citizens. Int. J. Biometeorol. 38: 122-125.

Höppe, P.; Peters, A.; Rabe, G.; Praml, G.; Lindner, J.; Jakobi, G.; Fruhmann, G.; Nowak, D. (2003) Environmental ozone effects in different population subgroups. Int. J. Hyg. Environ. Health 206: 505-516.

Horak, F., Jr.; Studnicka, M.; Gartner, C.; Spengler, J. D.; Tauber, E.; Urbanek, R.; Veiter, A.; Frischer, T. (2002a) Particulate matter and lung function growth in children: a 3-yr follow-up study in Austrian schoolchildren. Eur. Respir. J. 19: 838-845.

Horak, F., Jr.; Studnicka, M.; Gartner, C.; Spengler, J. D.; Tauber, E.; Urbanek, R.; Veiter, A.; Frischer, T. (2002b) Particulate matter and lung function growth in children: a 3-yr follow-up study in Austrian schoolchildren [author response]. Eur. Respir. J. 20: 1355.

Huang, Y.; Dominici, F.; Bell, M. L. (2005) Bayesian hierarchical distributed lag models for summer ozone exposure and cardio-respiratory mortality. Environmetrics 16: 547-562.

Hubbell, B. J.; Hallberg, A.; McCubbin, D. R.; Post, E. (2005) Health-related benefits of attaining the 8-hr ozone standard. Environ. Health Perspect. 113: 73-82.

Hwang, J.-S.; Chan, C.-C. (2002) Effects of air pollution on daily clinic visits for lower respiratory tract illness. Am. J. Epidemiol. 155: 1-10.

Ihorst, G.; Frischer, T.; Horak, F.; Schumacher, M.; Kopp, M.; Forster, J.; Mattes, J.; Kuehr, J. (2004) Long- and medium-term ozone effects on lung growth including a broad spectrum of exposure. Eur. Respir. J. 23: 292-299.

Ilabaca, M.; Olaeta, I.; Campos, E.; Villaire, J.; Tellez-Rojo, M. M.; Romieu, I. (1999) Association between levels of fine particulate and emergency visits for pneumonia and other respiratory illnesses among children in Santiago, Chile. J. Air Waste Manage. Assoc. 49: 154-163.

Ito, K. (2003) Associations of particulate matter components with daily mortality and morbidity in Detroit, Michigan. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 143-156. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [12 May, 2004].

Ito, K. (2004) Revised ozone risk estimates for daily mortality in Detroit, Michigan [personal communication with attachments to Jee Young Kim]. New York, NY: New York University School of Medicine, Nelson Institute of Environmental Medicine; October 31.

Ito, K.; Thurston, G. D. (1996) Daily PM_{10}/mortality associations: an investigation of at-risk subpopulations. J. Exposure Anal. Environ. Epidemiol. 6: 79-95.

Ito, K.; De Leon, S. F.; Lippmann, M. (2005) Associations between ozone and daily mortality, analysis and meta-analysis. Epidemiology 16: 446-457.

Jaffe, D. H.; Singer, M. E.; Rimm, A. A. (2003) Air pollution and emergency department visits for asthma among Ohio Medicaid recipients, 1991-1996. Environ. Res. 91: 21-28.

Jalaludin, B. B.; Chey, T.; O'Toole, B. I.; Smith, W. T.; Capon, A. G.; Leeder, S. R. (2000) Acute effects of low levels of ambient ozone on peak expiratory flow rate in a cohort of Australian children. Int. J. Epidemiol. 29: 549-557.

Jalaludin, B. B.; O'Toole, B. I.; Leeder, S. R. (2004) Acute effects of urban ambient air pollution on respiratory symptoms, asthma medication use, and doctor visits for asthma in a cohort of Australian children. Environ Res. 95: 32-42.

Jedrychowski, W.; Maugeri, U.; Bianchi, I.; Flak, E. (2001) Transient or persistent asthma-like symptoms and lung growth over 2-year follow-up in pre-adolescent children. J. Epidemiol. Biostat. 6: 229-233.

Jenkins, P. L.; Phillips, T. J.; Mulberg, E. J.; Hui, S. P. (1992) Activity patterns of Californians: use of and proximity to indoor pollutant sources. Atmos. Environ. Part A 26: 2141-2148.

Jones, M. C. (1983) The projection pursuit algorithm for exploratory data analysis [Ph.D. thesis]. Bath, England: University of Bath.

Jones, G. N.; Sletten, C.; Mandry, C.; Brantley, P. J. (1995) Ozone level effect on respiratory illness: an investigation of emergency department visits. South. Med. J. 88: 1049-1056.

Just, J.; Ségala, C.; Sahraoui, F.; Priol, G.; Grimfeld, A.; Neukirch, F. (2002) Short-term health effects of particulate and photochemical air pollution in asthmatic children. Eur. Respir. J. 20: 899-906.

Kaiser, R.; Romieu, I.; Medina, S.; Schwartz, J.; Krzyzanowski, M.; Künzli, N. (2004) Air pollution attributable postneonatal infant mortality in U.S. metropolitan areas: a risk assessment study. Environmental Health: a global access science source. Available: http;//www.ehjournal.net/content/3/1/4 [5 July, 2005].

Kim, S.-Y.; Lee, J.-T.; Hong, Y.-C.; Ahn, K.-J.; Kim, H. (2004) Determining the threshold effect of ozone on daily mortality: an analysis of ozone and mortality in Seoul, Korea, 1995-1999. Environ. Res. 94: 113-119.

Kinney, P. L.; Lippmann, M. (2000) Respiratory effects of seasonal exposures to ozone and particles. Arch. Environ. Health 55: 210-216.

Kinney, P. L.; Özkaynak, H. (1991) Associations of daily mortality and air pollution in Los Angeles County. Environ. Res. 54: 99-120.

Kinney, P. L.; Ito, K.; Thurston, G. D. (1995) A sensitivity analysis of mortality/PM_{10} associations in Los Angeles. In: Phalen, R. F.; Bates, D. V., eds. Proceedings of the colloquium on particulate air pollution and human mortality and morbidity; January 1994; Irvine, CA. Inhalation Toxicol. 7: 59-69.

Kinney, P. L.; Thurston, G. D.; Raizenne, M. (1996a) The effects of ambient ozone on lung function in children: a reanalysis of six summer camp studies. Environ. Health Perspect. 104: 170-174.

Kinney, P. L.; Nilsen, D. M.; Lippmann, M.; Brescia, M.; Gordon, T.; McGovern, T.; El Fawal, H.; Devlin, R. B.; Rom, W. N. (1996b) Biomarkers of lung inflammation in recreational joggers exposed to ozone. Am. J. Respir. Crit. Care Med. 154: 1430-1435.

Kinney, P. L.; Aggarwal, M.; Nikiforov, S. V.; Nadas, A. (1998) Methods development for epidemiologic investigations of the health effects of prolonged ozone exposure. Part III: an approach to retrospective estimation of lifetime ozone exposure using a questionnaire and ambient monitoring data (U.S. sites). Cambridge, MA: Health Effects Institute; research report no. 81; pp. 79-108.

Klemm, R. J.; Mason, R. M., Jr. (2000) Aerosol Research and Inhalation Epidemiological Study (ARIES): air quality and daily mortality statistical modeling—interim results. J. Air. Waste Manage. Assoc. 50: 1433-1439.

Klemm, R. J.; Lipfert, F. W.; Wyzga, R. E.; Gust, C. (2004) Daily mortality and air pollution in Atlanta: two years of data from ARIES. Inhalation Toxicol. 16(suppl. 1): 131-141.

Kochi, I.; Hubbell, B.; Kramer, R. (2005) An empirical Bayes approach to combining and comparing estimates of the value of a statistical life for environmental policy analysis. Available: http://www.nicholas.duke.edu/people/faculty/kramer/VSLrevised%204-6-05.pdf [11 August, 2005].

Koken, P. J.; Piver, W. T.; Ye, F.; Elixhauser, A.; Olsen, L. M.; Portier, C. J. (2003) Temperature, air pollution, and hospitalization for cardiovascular diseases among elderly people in Denver. Environ. Health Perspect. 111: 1312-1317.

Koop, G.; Tole, L. (2004) Measuring the health effects of air pollution: to what extent can we really say that people are dying from bad air? J. Environ. Econ. Manage. 47: 30-54.

Kopp, M. V.; Ulmer, C.; Ihorst, G.; Seydewitz, H. H.; Frischer, T.; Forster, J.; Kuehr, J. (1999) Upper airway inflammation in children exposed to ambient ozone and potential signs of adaptation. Eur. Respir. J. 14: 854-861.

Kopp, M. V.; Bohnet, W.; Frischer, T.; Ulmer, C.; Studnicka, M.; Ihorst, G.; Gardner, C.; Forster, J.; Urbanek, R.; Kuehr, J. (2000) Effects of ambient ozone on lung function in children over a two-summer period. Eur. Respir. J. 16: 893-900.

Koren, H. S.; Hatch, G. E.; Graham, D. E. (1990) Nasal lavage as a tool in assessing acute inflammation in response to inhaled pollutants. Toxicology 60: 15-25.

Korrick, S. A.; Neas, L. M.; Dockery, D. W.; Gold, D. R.; Allen, G. A.; Hill, L. B.; Kimball, K. D.; Rosner, B. A.; Speizer, F. E. (1998) Effects of ozone and other pollutants on the pulmonary function of adult hikers. Environ. Health Perspect. 106: 93-99.

Krzyzanowski, M.; Quackenboss, J. J.; Lebowitz, M. D. (1992) Relation of peak expiratory flow rates and symptoms to ambient ozone. Arch. Environ. Health 47: 107-115.

Künzli, N.; Lurmann, F.; Segal, M.; Ngo, L.; Balmes, J.; Tager, I. B. (1997) Association between lifetime ambient ozone exposure and pulmonary function in college freshmen—results of a pilot study. Environ. Res. 72: 8-23.

Kuo, H. W.; Lai, J. S.; Lee, M. C.; Tai, R. C.; Lee, M. C. (2002) Respiratory effects of air pollutants among asthmatics in central Taiwan. Arch. Environ. Health 57: 194-200.

Kwon, H.-J.; Cho, S.-H.; Nyberg, F.; Pershagen, G. (2001) Effects of ambient air pollution on daily mortality in a cohort of patients with congestive heart failure. Epidemiology 12: 413-419.

Lagerkvist, B. J.; Bernard, A.; Blomberg, A.; Bergstrom, E.; Forsberg, B.; Holmstrom, K.; Karp, K.; Lundstrom, N.-G.; Segerstedt, B.; Svensson, M.; Nordberg, G. (2004) Pulmonary epithelial integrity in children: relationship to ambient ozone exposure and swimming pool attendance. Environ. Health Perspect. 112: 1768-1771.

Langstaff, J. (2003) Percentiles of 1996-2000 ozone concentrations [memorandum to Joe Pinto]. Research Triangle Park, NC: U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards; September 17.

Lebowitz, M. D.; Camilli, A. E.; Bronnimann, D.; Quackenboss, J. (1987) The significance and meaningfulness of intraindividual changes in objective test results as responses to air contaminants. Presented at: 80th annual meeting of the Air Pollution Control Association; June; New York, NY. Pittsburgh, PA: Air Pollution Control Association; paper no. 87-32.1.

Lebowitz, M. D.; Quackenboss, J. J.; Krzyzanowski, M. (1991) Acute respiratory effects of prolonged ambient ozone. In: Berglund, R. L.; Lawson, D. R.; McKee, D. J., eds. Tropospheric ozone and the environment: papers from an international conference; March 1990; Los Angeles, CA. Pittsburgh, PA: Air & Waste Management Association; pp. 111-119. (A&WMA transactions series no. TR-19).

Lee, J.-T.; Schwartz, J. (1999) Reanalysis of the effects of air pollution on daily mortality in Seoul, Korea: a case-crossover design. Environ. Health Perspect. 107: 633-636.

Lee, J.-T.; Shin, D.; Chung, Y. (1999) Air pollution and daily mortality in Seoul and Ulsan, Korea. Environ. Health Perspect. 107: 149-154.

Lee, J.-T.; Kim, H.; Song, H.; Hong, Y.-C.; Cho, Y.-S.; Shin, S.-Y.; Hyun, Y.-J.; Kim, Y.-S. (2002) Air pollution and asthma among children in Seoul, Korea. Epidemiology 13: 481-484.

Lee, K.; Parkhurst, W. J.; Xue, J.; Özkaynak, H.; Neuberg, D.; Spengler, J. D. (2004) Outdoor/indoor/presonal ozone exposures of children in Nashville, Tennessee. J. Air Waste Manage. Assoc. 54: 352-359.

Le Tertre, A.; Quenel, P.; Eilstein, D.; Medina, S.; Prouvost, H.; Pascal, L.; Boumghar, A.; Saviuc, P.; Zeghnoun, A.; Filleul, L.; Declercq, C.; Cassadou, S.; Le Goaster, C. (2002a) Short-term effects of air pollution on mortality in nine French cities: a quantitative summary. Arch. Environ. Health 57: 311-319.

Le Tertre, A.; Medina, S.; Samoli, E.; Forsberg, B.; Michelozzi, P.; Boumghar, A.; Vonk, J. M.; Bellini, A.; Atkinson, R.; Ayres, J. G.; Sunyer, J.; Schwartz, J.; Katsouyanni, K. (2002b) Short term effects of particulate air pollution on cardiovascular diseases in eight European cities. J. Epidemiol. Community Health 56: 773-779.

Levy, J. I.; Carrothers, T. J.; Tuomisto, J. T.; Hammitt, J. K.; Evans, J. S. (2001) Assessing the public health benefits of reduced ozone concentrations. Environ. Health Perspect. 109: 1215-1226.

Levy, J. I.; Chemerynski, S. M.; Sarnat, J. A. (2005) Ozone exposure and mortality, an empiric Bayes metaregression analysis. Epidemiology 16: 458-468.

Liao, D.; Duan, Y.; Whitsel, E. A.; Zheng, Z.-J.; Heiss, G.; Chinchilli, V. M.; Lin, H.-M. (2004) Association of higher levels of ambient criteria pollutants with impaired cardiac autonomic control: a population-based study. Am. J. Epidemiol. 159: 768-777.

Lierl, M. B.; Hornung, R. W. (2003) Relationship of outdoor air quality to pediatric asthma exacerbations. Ann. Allergy Asthma Immunol. 90: 28-33.

Lin, C. A.; Martins, M. A.; Farhat, S. C. L.; Pope, C. A., III; Conceição, G. M. S.; Anastácio, V. M.; Hatanaka, M.; Andrade, W. C.; Hamaue, W. R.; Böhm, G. M.; Saldiva, P. H. N. (1999) Air pollution and respiratory illness of children in São Paulo, Brazil. Paediatr. Perinat. Epidemiol. 13: 475-488.

Lin, M.-C.; Yu, H.-S.; Tsai, S.-S.; Cheng, B.-H.; Hsu, T.-Y.; Wu, T.-N.; Yang, C.-Y. (2001) Adverse pregnancy outcome in a petrochemical polluted area in Taiwan. J. Toxicol. Environ. Health Part A 63: 565-574.

Lin, M.; Chen, Y.; Burnett, R. T.; Villeneuve, P. J.; Krewski, D. (2003) Effect of short-term exposure to gaseous pollution on asthma hospitalisation in children: a bi-directional case-crossover analysis. J. Epidemiol. Community Health 57: 50-55.

Lin, M.; Chen, Y.; Villeneuve, P. J.; Burnett, R. T.; Lemyre, L.; Hertzman, C.; McGrail, K. M.; Krewski, D. (2004) Gaseous air pollutants and asthma hospitalization of children with low household income in Vancouver, British Columbia, Canada. Am. J. Epidemiol. 159: 294-303.

Linn, W. S.; Shamoo, D. A.; Anderson, K. R.; Peng, R.-C.; Avol, E. L.; Hackney, J. D.; Gong, H., Jr. (1996) Short-term air pollution exposures and responses in Los Angeles area schoolchildren. J. Exposure Anal. Environ. Epidemiol. 6: 449-472.

Linn, W. S.; Szlachcic, Y.; Gong, H., Jr.; Kinney, P. L.; Berhane, K. T. (2000) Air pollution and daily hospital admissions in metropolitan Los Angeles. Environ. Health Perspect. 108: 427-434.

Lipfert, F. W.; Hammerstrom, T. (1992) Temporal patterns in air pollution and hospital admissions. Environ. Res. 59: 374-399.

Lipfert, F. W.; Morris, S. C.; Wyzga, R. E. (2000a) Daily mortality in the Philadelphia metropolitan area and size-classified particulate matter. J. Air Waste Manage. Assoc. 50: 1501-1513.

Lipfert, F. W.; Perry, H. M., Jr.; Miller, J. P.; Baty, J. D.; Wyzga, R. E.; Carmody, S. E. (2000b) The Washington University-EPRI veterans' cohort mortality study: preliminary results. In: Grant, L. D., ed. PM2000: particulate matter and health. Inhalation Toxicol. 12(suppl. 4): 41-73.

Lipfert, F. W.; Perry, H. M., Jr.; Miller, J. P.; Baty, J. D.; Wyzga, R. E.; Carmody, S. E. (2003) Air pollution, blood pressure, and their long-term associations with mortality. Inhalation Toxicol. 15: 493-512.

Lippmann, M. (1988) Health significance of pulmonary function responses to airborne irritants. JAPCA 38: 881-887.

Lippmann, M.; Spektor, D. M. (1998) Peak flow rate changes in O3 exposed children: spirometry vs miniWright flow meters. J. Exposure Anal. Environ. Epidemiol. 8: 101-107.

Lippmann, M.; Ito, K.; Nádas, A.; Burnett, R. T. (2000) Association of particulate matter components with daily mortality and morbidity in urban populations. Cambridge, MA: Health Effects Institute; research report no. 95.

Lipsett, M.; Hurley, S.; Ostro, B. (1997) Air pollution and emergency room visits for asthma in Santa Clara County, California. Environ. Health Perspect. 105: 216-222.

Liu, L.-J. S.; Koutrakis, P.; Leech, J.; Broder, I. (1995) Assessment of ozone exposures in the greater metropolitan Toronto area. J. Air Waste Manage. Assoc. 45: 223-234.

Loomis, D. P.; Borja-Aburto, V. H.; Bangdiwala, S. I.; Shy, C. M. (1996) Ozone exposure and daily mortality in Mexico City: a time-series analysis. Cambridge, MA: Health Effects Institute; research report no. 75.

Loomis, D.; Castillejos, M.; Gold, D. R.; McDonnell, W.; Borja-Aburto, V. H. (1999) Air pollution and infant mortality in Mexico City. Epidemiology 10: 118-123.

Luginaah, I. N.; Fung, K. Y.; Gorey, K. M.; Webster, G.; Wills, C. (2005) Association of Ambient Air Pollution with Respiratory Hospitalization in a Government Designated "Area of Concern": The Case of Windsor, Ontario. Environ. Health Perspect. 113: 290-296.

Lumley, T.; Sheppard, L. (2000) Assessing seasonal confounding and model selection bias in air pollution epidemiology using positive and negative control analyses. Environmetrics 11: 705-717.

Maisonet, M.; Bush, T. J.; Correa, A.; Jaakkola, J. J. K. (2001) Relation between ambient air pollution and low birth weight in the northeastern United States. Environ. Health Perspect. Suppl. 109(3): 351-356.

Mann, J. K.; Tager, I. B.; Lurmann, F.; Segal, M.; Quesenberry, C. P., Jr.; Lugg, M. M.; Shan, J.; Van den Eeden, S. K. (2002) Air pollution and hospital admissions for ischemic heart disease in persons with congestive heart failure or arrhythmia. Environ. Health Perspect. 110: 1247-1252.

Martins, L. C.; Latorre, M. R. D. O.; Saldiva, P. H. N.; Braga, A. L. F. (2002) Air pollution and emergency room visits due to chronic lower respiratory diseases in the elderly: an ecological time-series study in São Paulo, Brazil. J. Occup. Environ. Med. 44: 622-627.

McConnell, R.; Berhane, K.; Gilliland, F.; London, S. J.; Vora, H.; Avol, E.; Gauderman, W. J.; Margolis, H. G.; Lurmann, F.; Thomas, D. C.; Peters, J. M. (1999) Air pollution and bronchitic symptoms in southern California children with asthma. Environ. Health Perspect. 107: 757-760.

McConnell, R.; Berhane, K.; Gilliland, F.; London, S. J.; Islam, T.; Gauderman, W. J.; Avol, E.; Margolis, H. G.; Peters, J. M. (2002) Asthma in exercising children exposed to ozone: a cohort study. Lancet 359: 386-391.

McConnell, R.; Berhane, K.; Gilliland, F.; Molitor, J.; Thomas, D.; Lurmann, F.; Avol, E.; Gauderman, W. J.; Peters, J. M. (2003) Prospective study of air pollution and bronchitic symptoms in children with asthma. Am. J. Respir. Crit. Care Med. 168:790-797.

McDonnell, W. F.; Abbey, D. E.; Nishino, N.; Lebowitz, M. D. (1999) Long-term ambient ozone concentration and the incidence of asthma in nonsmoking adults: the ahsmog study. Environ. Res. 80: 110-121.

Metzger, K. B.; Tolbert, P. E.; Klein, M.; Peel, J. L.; Flanders, W. D.; Todd, K. H.; Mulholland, J. A.; Ryan, P. B.; Frumkin , H. (2004) Ambient air pollution and cardiovascular emergency department visits. Epidemiology 15: 46-56.

Mitchell, H.; Senturia, Y.; Gergen, P.; Baker, D.; Joseph, C.; McNiff-Mortimer, K.; Wedner, H. J.; Crain, E.; Eggleston, P.; Evans, R., III; Kattan, M.; Kercsmar, C.; Leickly, F.; Malveaux, F.; Smartt, E.; Weiss, K. (1997) Design and methods of the National Cooperative Inner-City Asthma Study. Pediatr. Pulmonol. 24: 237-252.

Moolgavkar, S. H. (2003) Air pollution and daily mortality in two U.S. counties: season-specific analyses and exposure-response relationships. Inhalation Toxicol. 15: 877-907.

Moolgavkar, S. H.; Luebeck, E. G. (1996) A critical review of the evidence on particulate air pollution and mortality. Epidemiology 7: 420-428.

Moolgavkar, S. H.; Luebeck, E. G.; Hall, T. A.; Anderson, E. L. (1995) Air pollution and daily mortality in Philadelphia. Epidemiology 6: 476-484.

Moolgavkar, S. H.; Luebeck, E. G.; Anderson, E. L. (1997) Air pollution and hospital admissions for respiratory causes in Minneapolis-St. Paul and Birmingham. Epidemiology 8: 364-370.

Morgan, G.; Corbett, S.; Wlodarczyk, J. (1998a) Air pollution and hospital admissions in Sydney, Australia, 1990 to 1994. Am. J. Public Health 88: 1761-1766.
Morgan, G.; Corbett, S.; Wlodarczyk, J.; Lewis, P. (1998b) Air pollution and daily mortality in Sydney, Australia, 1989 through 1993. Am. J. Public Health 88: 759-764.
Mortimer, K. M.; Tager, I. B.; Dockery, D. W.; Neas, L. M.; Redline, S. (2000) The effect of ozone on inner-city children with asthma: identification of susceptible subgroups. Am. J. Respir. Crit. Care Med. 162: 1838-1845.
Mortimer, K. M.; Neas, L. M.; Dockery, D. W.; Redline, S.; Tager, I. B. (2002) The effect of air pollution on inner-city children with asthma. Eur. Respir. J. 19: 699-705.
Naeher, L. P.; Holford, T. R.; Beckett, W. S.; Belanger, K.; Triche, E. W.; Bracken, M. B.; Leaderer, B. P. (1999) Healthy women's PEF variations with ambient summer concentrations of PM_{10}, $PN_{2.5}$, SO_4^{2-}, H^+, and O_3. Am. J. Respir. Crit. Care Med. 160: 117-125.
National Institutes of Health. (1997) Guidelines for the diagnosis and management of asthma: expert panel report 2. Bethesda, MD: U.S. Department of Health and Human Services, National Heart, Lung, and Blood Institute; publication no. 97-4051. Available: http://www.nhlbi.nih.gov/guidelines/asthma/asthgdln.pdf (11 April 2003).
Nauenberg, E.; Basu, K. (1999) Effect of insurance coverage on the relationship between asthma hospitalizations and exposure to air pollution. Public Health Rep. 114: 135-148.
Navidi, W.; Thomas, D.; Langholz, B.; Stram, D. (1999) Statistical methods for epidemiologic studies of the health effects of air pollution. Cambridge, MA: Health Effects Institute; research report no. 86.
Neas, L. M.; Dockery, D. W.; Koutrakis, P.; Tollerud, D. J.; Speizer, F. E. (1995) The association of ambient air pollution with twice daily peak expiratory flow rate measurements in children. Am. J. Epidemiol. 141: 111-122.
Neas, L. M.; Dockery, D. W.; Koutrakis, P.; Speizer, F. E. (1999) Fine particles and peak flow in children: acidity *versus* mass. Epidemiology 10: 550-553.
Neidell, M. J. (2004) Air pollution, health, and socio-economic status: the effect of outdoor air quality on childhood asthma. J. Health Econ. 23: 1209-1236.
Newhouse, C. P.; Levetin, B. S.; Levetin, E. (2004) Correlation of environmental factors with asthma and rhinitis symptoms in Tulsa, OK. Ann. Allergy Asthma Immunol. 92: 356-366.
Nutman, A.; Solomon, Y.; Mendel, S.; Nutman, J.; Hines, E.; Topilsky, M.; Kivity, S. (1998) The use of a neural network for studying the relationship between air pollution and asthma-related emergency room visits. Respir. Med. 92: 1199-1202.
Oftedal, B.; Nafstad, P.; Magnus, P.; Bjørkly, S.; Skrondal, A. (2003) Traffic related air pollution and acute hospital admission for respiratory diseases in Drammen, Norway 1995-2000. Eur. J. Epidemiol. 18: 671-675.
O'Neill, M. S.; Loomis, D.; Borja-Aburto, V. H. (2004) Ozone, area social conditions, and mortality in Mexico City. Environ. Res. 94: 234-242.
Ostro, B. (1995) Fine particulate air pollution and mortality in two Southern California counties. Environ. Res. 70: 98-104.
Ostro, B.; Sanchez, J. M.; Aranda, C.; Eskeland, G. S. (1996) Air pollution and mortality: results from a study of Santiago, Chile. In: Lippmann, M., ed. Papers from the ISEA-ISEE annual meeting; September 1994; Research Triangle Park, NC. J. Exposure Anal. Environ. Epidemiol. 6: 97-114.
Ostro, B.; Lipsett, M.; Mann, J.; Braxton-Owens, H.; White, M. (2001) Air pollution and exacerbation of asthma in African-American children in Los Angeles. Epidemiology 12: 200-208.
Palli, D.; Saieva, C.; Grechi, D.; Masala, G.; Zanna, I.; Barbaro, A.; Decarli, A.; Munnia, A.; Peluso, M. (2004) DNA bulky adducts in a Mediterranean population correlate with environmental ozone concentration, an indicator of photochemical smog. Int. J. Cancer 109: 17-23.
Park, H.; Lee, B.; Ha, E.-H.; Lee, J.-T.; Kim, H.; Hong, Y.-C. (2002) Association of air pollution with school absenteeism due to illness. Arch. Pediatr. Adolesc. Med. 156: 1235-1239.
Park, S. K.; O'Neill, M. S.; Vokonas, P. S.; Sparrow, D.; Schwartz, J. (2005) Effects of air pollution on heart rate variability: the VA normative aging study. Environ. Health Perspect. 113: 304-309.
Pearce, N.; Beasley, R.; Burgess, C.; Crane, J. (1998) Asthma epidemiology: principles and methods. New York, NY: Oxford University Press.
Peel, J. L.; Tolbert, P. E.; Klein, M.; Metzger, K. B.; Flanders, W. D.; Knox, T.; Mulholland, J. A.; Ryan, P. B.; Frumkin, H. (2005) Ambient air pollution and respiratory emergency department visits. Epidemiology 16: 164-174.
Pereira, L. A. A.; Loomis, D.; Conceição, G. M. S.; Braga, A. L. F.; Arcas, R. M.; Kishi, H. S.; Singer, J. M.; Böhm, G. M.; Saldiva, P. H. N. (1998) Association between air pollution and intrauterine mortality in São Paulo, Brazil. Environ. Health Perspect. 106: 325-329.

Pereira, F. A. C.; De Assunção, J. V.; Saldiva, P. H. N.; Pereira, L. A. A.; Mirra, A. P.; Braga, A. L. F. (2005) Influence of air pollution on the incidence of respiratory tract neoplasm. J. Air Waste Manage. Assoc. 55: 83-87.

Peters, J. M.; Avol, E.; Navidi, W.; London, S. J.; Gauderman, W. J.; Lurmann, F.; Linn, W. S.; Margolis, H.; Rappaport, E.; Gong, H., Jr.; Thomas, D. C. (1999a) A study of twelve southern California communities with differing levels and types of air pollution. I. Prevalence of respiratory morbidity. Am. J. Respir. Crit. Care Med. 159: 760-767.

Peters, J. M.; Avol, E.; Gauderman, W. J.; Linn, W. S.; Navidi, W.; London, S. J.; Margolis, H.; Rappaport, E.; Vora, H.; Gong, H., Jr.; Thomas, D. C. (1999b) A study of twelve southern California communities with differing levels and types of air pollution. II. Effects on pulmonary function. Am. J. Respir. Crit. Care Med. 159: 768-775.

Peters, A.; Liu, E.; Verrier, R. L.; Schwartz, J.; Gold, D. R.; Mittleman, M.; Baliff, J.; Oh, J. A.; Allen, G.; Monahan, K.; Dockery, D. W. (2000a) Air pollution and incidence of cardiac arrhythmia. Epidemiology 11: 11-17.

Peters, A.; Skorkovsky, J.; Kotesovec, F.; Brynda, J.; Spix, C.; Wichmann, H. E.; Heinrich, J. (2000b) Associations between mortality and air pollution in central Europe. Environ. Health Perspect. 108: 283-287.

Peters, A.; Dockery, D. W.; Muller, J. E.; Mittleman, M. A. (2001) Increased particulate air pollution and the triggering of myocardial infarction. Circulation 103: 2810-2815.

Petroeschevsky, A.; Simpson, R. W.; Thalib, L.; Rutherford, S. (2001) Associations between outdoor air pollution and hospital admissions in Brisbane, Australia. Arch. Environ. Health 56: 37-52.

Plunkett, L. M.; Turnbull, D.; Rodricks, J. V. (1992) Differences between adults and children affecting exposure assessment. In: Guzelian, P. S.; Henry, D. J.; Olin, S. S., eds. Similarities and differences between children and adults: implications for risk assessment. Washington, DC: ILSI Press, pp. 79-96.

Poloniecki, J. D.; Atkinson, R. W.; Ponce de Leon, A.; Anderson, H. R. (1997) Daily time series for cardiovascular hospital admissions and previous day's air pollution in London, UK. Occup. Environ. Med. 54: 535-540.

Ponce de Leon, A.; Anderson, H. R.; Bland, J. M.; Strachan, D. P.; Bower, J. (1996) Effects of air pollution on daily hospital admissions for respiratory disease in London between 1987-88 and 1991-92. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Commun. Health 50(suppl. 1): S63-S70.

Pönkä, A.; Virtanen, M. (1996) Asthma and ambient air pollution in Helsinki. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Community Health 50(suppl. 1): S59-S62.

Pönkä, A.; Savela, M.; Virtanen, M. (1998) Mortality and air pollution in Helsinki. Arch. Environ. Health 53: 281-286.

Pope, C. A., III; Burnett, R. T.; Thun, M. J.; Calle, E. E.; Krewski, D.; Ito, K.; Thurston, G. D. (2002) Lung cancer, cardiopulmonary mortality, and long-term exposure to fine particulate air pollution. JAMA J. Am. Med. Assoc. 287: 1132-1141.

Prescott, G. J.; Cohen, G. R.; Elton, R. A.; Fowkes, F. G. R.; Agius, R. M. (1998) Urban air pollution and cardiopulmonary ill health: a 14.5 year time series study. Occup. Environ. Med. 55: 697-704.

Raizenne, M.; Stern, B.; Burnett, R.; Spengler, J. (1987) Acute respiratory function and transported air pollutants: observational studies. Presented at: 80th annual meeting of the Air Pollution Control Association; June; New York, NY. Pittsburgh, PA: Air Pollution Control Association; paper no. 87-32.6.

Raizenne, M. E.; Burnett, R. T.; Stern, B.; Franklin, C. A.; Spengler, J. D. (1989) Acute lung function responses to ambient acid aerosol exposures in children. Environ. Health Perspect. 79: 179-185.

Ramadour, M.; Burel, C.; Lanteaume, A.; Vervloet, D.; Charpin, D.; Brisse, F.; Dutau, H.; Charpin, D. (2000) Prevalence of asthma and rhinitis in relation to long-term exposure to gaseous air pollutants. Allergy (Copenhagen) 55: 1163-1169.

Ramsay, T. O.; Burnett, R. T.; Krewski, D. (2003) The effect of concurvity in generalized additive models linking mortality to ambient particulate matter. Epidemiology 14: 18-23.

Rich, K. E.; Petkau, J.; Vedal, S.; Brauer, M. (2004) A case-crossover analysis of particulate air pollution and cardiac arrhythmia in patients with implantable cardioverter defibrillators. Inhalation Toxicol. 16: 363-372.

Rich, D. Q.; Schwartz, J.; Mittleman, M. A.; Link, M.; Luttmann-Gibson, H.; Catalano, P. J.; Speizer, F. E.; Dockery, D. W. (2005) Association of short-term ambient air pollution concentrations and ventricular arrhythmias. Am. J. Epidemiol. 161: 1123-1132.

Ritz, B.; Yu, F. (1999) The effect of ambient carbon monoxide on low birth weight among children born in southern California between 1989 and 1993. Environ. Health Perspect. 107: 17-25.

Ritz, B.; Yu, F.; Chapa, G.; Fruin, S. (2000) Effect of air pollution on preterm birth among children born in Southern California between 1989 and 1993. Epidemiology 11: 502-511.

Ritz, B.; Yu, F.; Fruin, S.; Chapa, G.; Shaw, G. M.; Harris, J. A. (2002) Ambient air pollution and risk of birth defects in Southern California. Am. J. Epidemiol. 155: 17-25.

Roemer, W. H.; Van Wijnen, J. H . (2001) Daily mortality and air pollution along busy streets in Amsterdam, 1987-1998. Epidemiology 12: 649-653.

Romieu, I.; Meneses, F.; Sienra-Monge, J. J. L.; Huerta, J.; Velasco, S. R.; White, M. C.; Etzel, R. A.; Hernandez-Avila, M. (1995) Effects of urban air pollutants on emergency visits for childhood asthma in Mexico City. Am. J. Epidemiol. 141: 546-553.

Romieu, I.; Meneses, F.; Ruiz, S.; Sienra, J. J.; Huerta, J.; White, M. C.; Etzel, R. A. (1996) Effects of air pollution on the respiratory health of asthmatic children living in Mexico City. Am. J. Respir. Crit. Care Med. 154: 300-307.

Romieu, I.; Meneses, F.; Ruiz, S.; Huerta, J.; Sienra, J. J.; White, M.; Etzel, R.; Hernandez, M. (1997) Effects of intermittent ozone exposure on peak expiratory flow and respiratory symptoms among asthmatic children in Mexico City. Arch. Environ. Health 52: 368-376.

Romieu, I.; Meneses, F.; Ramirez, M.; Ruiz, S.; Padilla, R. P.; Sienra, J. J.; Gerber, M.; Grievink, L.; Dekker, R.; Walda, I.; Brunekreef, B. (1998) Antioxidant supplementation and respiratory functions among workers exposed to high levels of ozone. Am. J. Respir. Crit. Care Med. 158: 226-232.

Romieu, I.; Sienra-Monge, J. J.; Ramírez-Aguilar, M.; Téllez-Rojo, M. M.; Moreno-Macías, H.; Reyes-Ruiz, N. I.; Del Río-Navarro, B. E.; Ruiz-Navarro, M. X.; Hatch, G.; Slade, R.; Hernández-Avila, M. (2002) Antioxidant supplementation and lung functions among children with asthma exposed to high levels of air pollutants. Am. J. Respir. Crit. Care Med. 166: 703-709.

Romieu, I.; Sienra-Monge, J. J.; Ramírez-Aguilar, M.; Moreno-Macias, H.; Reyes-Ruiz, N. I.; Estela del Rio-Navarro, B.; Hernández-Avila, M.; London, S. J. (2004) Genetic polymorphism of *GSTM1* and antioxidant supplementation influence lung function in relation to ozone exposure in asthmatic children in Mexico City. Thorax 59: 8-10.

Rona, R. J.; Gulliford, M. C.; Chinn, S. (1993) Effects of prematurity and intrauterine growth on respiratory health and lung function in childhood. Br. Med. J. 306: 817-820.

Ross, M. A.; Persky, V. W.; Scheff, P. A.; Chung, J.; Curtis, L.; Ramakrishnan, V.; Wadden, R. A.; Hryhorczuk, D. O. (2002) Effect of ozone and aeroallergens on the respiratory health of asthmatics. Arch. Environ. Health 57: 568-578.

Ruidavets, J.-B.; Cournot, M.; Cassadou, S.; Giroux, M.; Meybeck, M.; Ferrières, J. (2005) Ozone air pollution is associated with acute myocardial infarction. Circulation 111: 563-569.

Saez, M.; Tobias, A.; Muñoz, P.; Campbell, M. J. (1999) A GEE moving average analysis of the relationship between air pollution and mortality for asthma in Barcelona, Spain. Stat. Med. 18: 2077-2086.

Saez, M.; Ballester, F.; Barceló, M. A.; Pérez-Hoyos, S.; Bellido, J.; Tenías, J. M.; Ocaña, R.; Figueiras, A.; Arribas, F.; Aragonés, N.; Tobías, A.; Cirera, L.; Cañada, A.; on behalf of the EMECAM Group. (2002) A combined analysis of the short-term effects of photochemical air pollutants on mortality within the EMECAM project. Environ. Health Perspect. 110: 221-228.

Saldiva, P. H. N.; Lichtenfels, A. J. F. C.; Paiva, P. S. O.; Barone, I. A.; Martins, M. A.; Massad, E.; Pereira, J. C. R.; Xavier, V. P.; Singer, J. M.; Böhm, G. M. (1994) Association between air pollution and mortality due to respiratory diseases in children in São Paulo, Brazil: a preliminary report. Environ. Res. 65: 218-225.

Saldiva, P. H. N.; Pope, C. A., III; Schwartz, J.; Dockery, D. W.; Lichtenfels, A. J.; Salge, J. M.; Barone, I.; Böhm, G. M. (1995) Air pollution and mortality in elderly people: a time-series study in São Paulo, Brazil. Arch. Environ. Health 50: 159-163.

Samet, J. M.; Zeger, S. L.; Dominici, F.; Curriero, F.; Coursac, I.; Dockery, D. W.; Schwartz, J.; Zanobetti, A. (2000) The national morbidity, mortality, and air pollution study. Part II: morbidity, mortality, and air pollution in the United States. Cambridge, MA: Health Effects Institute; research report no. 94, part II.

Sarnat, J. A.; Schwartz, J.; Catalano, P. J.; Suh, H. H. (2001) Gaseous pollutants in particulate matter epidemiology: confounders or surrogates? Environ. Health Perspect. 109: 1053-1061.

Sartor, F.; Snacken, R.; Demuth, C.; Walckiers, D. (1995) Temperature, ambient ozone levels, and mortality during summer, 1994, in Belgium. Environ. Res. 70: 105-113.

Scarlett, J. F.; Abbott, K. J.; Peacock, J. L.; Strachan, D. P.; Anderson, H. R. (1996) Acute effects of summer air pollution on respiratory function in primary school children in southern England. Thorax 51: 1109-1114.

Schildcrout, J. S.; Heagerty, P. J. (2005) Regressions analysis of longitudinal binary data with time-dependent environmental covariates: bias and efficiency. Biostatistics: doi: 10.1093/biostatistics/kxi033.

Schindler, C.; Künzli, N.; Bongard, J.-P.; Leuenberger, P.; Karrer, W.; Rapp, R.; Monn, C.; Ackermann-Liebrich, U.; Swiss Study on Air Pollution and Lung Diseases in Adults Investigators. (2001) Short-term variation in air pollution and in average lung function among never-smokers. Am. J. Respir. Crit. Care Med. 163: 356-361.

Schouten, J. P.; Vonk, J. M.; de Graaf, A. (1996) Short term effects of air pollution on emergency hospital admissions for respiratory disease: results of the APHEA project in two major cities in The Netherlands, 1977-89. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Community Health 50(suppl. 1): S22-S29.

Schwartz, J. (1991) Particulate air pollution and daily mortality in Detroit. Environ. Res. 56: 204-213.

Schwartz, J. (1996) Air pollution and hospital admissions for respiratory disease. Epidemiology 7: 20-28.

Schwartz, J. (2005) How sensitive is the association between ozone and daily deaths to control for temperature? Am. J. Respir. Crit. Care Med. 171: 627-631.

Schwartz, J.; Spix, C.; Touloumi, G.; Bachárová, L.; Barumamdzadeh, T.; le Tertre, A.; Piekarksi, T.; Ponce de Leon, A.; Pönkä, A.; Rossi, G.; Saez, M.; Schouten, J. P. (1996) Methodological issues in studies of air pollution and daily counts of deaths or hospital admissions. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Commun. Health 50(suppl. 1): S3-S11.

Schwartz, J.; Litonjua, A.; Suh, H.; Verrier, M.; Zanobetti, A.; Syring, M.; Nearing, B.; Verrier, R.; Stone, P.; MacCallum, G.; Speizer, F. E.; Gold, D. R. (2005) Traffic related pollution and heart rate variability in a panel of elderly subjects. Thorax 60: 455-461.

Selwyn, B. J.; Stock, T. H.; Hardy, R. J.; Chan, F. A.; Jenkins, D. E.; Kotchmar, D. J.; Chapman, R. S. (1985) Health effects of ambient ozone exposure in vigorously exercising adults. In: Lee, S. D., ed. Evaluation of the scientific basis for ozone/oxidants standards: proceedings of an APCA international specialty conference; November 1984; Houston, TX. Pittsburgh, PA: Air Pollution Control Association; pp. 281-296. (APCA international specialty conference transactions: TR-4).

Sheppard, L. (2003) Ambient air pollution and nonelderly asthma hospital admissions in Seattle, Washington, 1987-1994. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 227-230. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [18 October, 2004].

Sheppard, L. (2005) Acute air pollution effects: consequences of exposure distribution and measurements. J. Toxicol. Environ. Health Part A 68: 1127-1135.

Sheppard, L.; Levy, D.; Norris, G.; Larson, T. V.; Koenig, J. Q. (1999) Effects of ambient air pollution on nonelderly asthma hospital admissions in Seattle, Washington, 1987-1994. Epidemiology 10: 23-30.

Sheppard, L.; Slaughter, J. C.; Schildcrout, J.; Liu, L.-J. S.; Lumley, T. (2005) Exposure and measurement contributions to estimates of acute air pollution effects. J. Exposure Anal. Environ. Epidemiol. 15: 366-376.

Sherwin, R. P.; Richters, V.; Kraft, P.; Richters, A. (2000) Centriacinar region inflammatory disease in young individuals: a comparative study of Miami and Los Angeles residents. Virchows Arch. 437: 422-428.

Shumway, R. H.; Azari, A. S.; Pawitan, Y. (1988) Modeling mortality fluctuations in Los Angeles as functions of pollution and weather effects. Environ. Res. 45: 224-241.

Silverman, B. W. (1986) Density estimation. London, England: Chapman & Hall.

Simpson, R. W.; Williams, G.; Petroeschevsky, A.; Morgan, G.; Rutherford, S. (1997) Associations between outdoor air pollution and daily mortality in Brisbane, Australia. Arch. Environ. Health 52: 442-454.

Spektor, D. M.; Lippmann, M.; Lioy, P. J.; Thurston, G. D.; Citak, K.; James, D. J.; Bock, N.; Speizer, F. E.; Hayes, C. (1988a) Effects of ambient ozone on respiratory function in active, normal children. Am. Rev. Respir. Dis. 137: 313-320.

Spektor, D. M.; Lippmann, M.; Thurston, G. D.; Lioy, P. J.; Stecko, J.; O'Connor, G.; Garshick, E.; Speizer, F. E.; Hayes, C. (1988b) Effects of ambient ozone on respiratory function in healthy adults exercising outdoors. Am. Rev. Respir. Dis. 138: 821-828.

Spektor, D. M.; Lippmann, M. (1991) Health effects of ambient ozone on healthy children at a summer camp. In: Berglund, R. L.; Lawson, D. R.; McKee, D. J., eds. Tropospheric ozone and the environment: papers from an international conference; March 1990; Los Angeles, CA. Pittsburgh, PA: Air & Waste Management Association; pp. 83-89. (A&WMA transactions series no. TR-19).

Stieb, D. M.; Burnett, R. T.; Beveridge, R. C.; Brook, J. R. (1996) Association between ozone and asthma emergency department visits in Saint John, New Brunswick, Canada. Environ. Health Perspect. 104: 1354-1360.

Stieb, D. M.; Judek, S.; Burnett, R. T. (2002) Meta-analysis of time-series studies of air pollution and mortality: effects of gases and particles and the influence of cause of death, age, and season. J. Air Waste Manage. Assoc. 52: 470-484.

Stieb, D. M.; Judek, S.; Burnett, R. T. (2003) Meta-analysis of time-series studies of air pollution and mortality: update in relation to the use of generalized additive models. J. Air Waste Manage. 53: 258-261.

Sunyer, J.; Basagaña, X. (2001) Particles, and not gases, are associated with the risk of death in patients with chronic obstructive pulmonary disease. Int. J. Epidemiol. 30: 1138-1140.

Sunyer, J.; Castellsagué, J.; Sáez, M.; Tobias, A.; Antó, J. M. (1996) Air pollution and mortality in Barcelona. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Community Health 50(suppl. 1): S76-S80.

Sunyer, J.; Spix, C.; Quénel, P.; Ponce-de-León, A.; Pönka, A.; Barumandzadeh, T.; Touloumi, G.; Bacharova, L.; Wojtyniak, B.; Vonk, J.; Bisanti, L.; Schwartz, J.; Katsouyanni, K. (1997) Urban air pollution and emergency admissions for asthma in four European cities: the APHEA project. Thorax 52: 760-765.

Sunyer, J.; Basagaña, X.; Belmonte, J.; Antó, J. M. (2002) Effect of nitrogen dioxide and ozone on the risk of dying in patients with severe asthma. Thorax 57: 687-693.

Tager, I. B. (1993) Introduction to working group on tropospheric ozone, Health Effects Institute Environmental Epidemiology Planning Project. Environ. Health Perspect. 101(suppl. 4): 205-207.

Tager, I. B. (1999) Air pollution and lung function growth. Is it ozone? [editorial]. Am. J. Respir. Crit. Care Med. 160: 387-389.

Tager, I. B.; Künzli, N.; Lurmann, F.; Ngo, L.; Segal, M.; Balmes, J. (1998) Methods development for epidemiologic investigations of the health effects of prolonged ozone exposure. Part II: an approach to retrospective estimation of lifetime ozone exposure using a questionnaire and ambient monitoring data (California sites). Cambridge, MA: Health Effects Institute; research report no. 81; pp. 27-78.

Taggart, S. C. O.; Custovic, A.; Francis, H. C.; Faragher, E. B.; Yates, C. J.; Higgins, B. G.; Woodcock, A. (1996) Asthmatic bronchial hyperresponsiveness varies with ambient levels of summertime air pollution. Eur. Respir. J. 9: 1146-1154.

Téllez-Rojo, M. M.; Romieu, I.; Ruiz-Velasco, S.; Lezana, M.-A.; Hernández-Avila, M. M. (2000) Daily respiratory mortality and PM_{10} pollution in Mexico City: importance of considering place of death. Eur. Respir. J. 16: 391-396.

Tenías, J. M.; Ballester, F.; Rivera, M. L. (1998) Association between hospital emergency visits for asthma and air pollution in Valencia, Spain. Occup. Environ. Med. 55: 541-547.

Tenías, J. M.; Ballester, F.; Pérez-Hoyos, S.; Rivera, M. L. (2002) Air pollution and hospital emergency room admissions for chronic obstructive pulmonary disease in Valencia, Spain. Arch. Environ. Health 57: 41-47.

Thompson, A. J.; Shields, M. D.; Patterson, C. C. (2001) Acute asthma exacerbations and air pollutants in children living in Belfast, Northern Ireland. Arch. Environ. Health 56: 234-241.

Thurston, G. D.; Ito, K. (2001) Epidemiological studies of acute ozone exposures and mortality. J. Exposure Anal. Environ. Epidemiol. 11: 286-294.

Thurston, G. D.; Ito, K.; Kinney, P. L.; Lippmann, M. (1992) A multi-year study of air pollution and respiratory hospital admissions in three New York State metropolitan areas: results for 1988 and 1989 summers. J. Exposure Anal. Environ. Epidemiol. 2: 429-450.

Thurston, G. D.; Ito, K.; Hayes, C. G.; Bates, D. V.; Lippmann, M. (1994) Respiratory hospital admissions and summertime haze air pollution in Toronto, Ontario: consideration of the role of acid aerosols. Environ. Res. 65: 271-290.

Thurston, G. D.; Lippmann, M.; Scott, M. B.; Fine, J. M. (1997) Summertime haze air pollution and children with asthma. Am. J. Respir. Crit. Care Med. 155: 654-660.

Tobías, A.; Campbell, M. J.; Sáez, M. (1999) Modelling asthma epidemics on the relationship between air pollution and asthma emergency visits in Barcelona, Spain. Eur. J. Epidemiol. 15: 799-803.

Tolbert, P. E.; Mulholland, J. A.; Maclntosh, D. L.; Xu, F.; Daniels, D.; Devine, O. J.; Carlin, B. P.; Klein, M.; Dorley, J.; Butler, A. J.; Nordenberg, D. F.; Frumkin, H.; Ryan, P. B.; White, M. C. (2000) Air quality and pediatric emergency room visits for asthma in Atlanta, Georgia. Am. J. Epidemiol. 151: 798-810.

Touloumi, G.; Katsouyanni, K.; Zmirou, D.; Schwartz, J.; Spix, C.; Ponce de Leon, A.; Tobias, A.; Quennel, P.; Rabczenko, D.; Bacharova, L.; Bisanti, L.; Vonk, J. M.; Ponka, A. (1997) Short-term effects of ambient oxidant exposure on mortality: a combined analysis within the APHEA project. Am. J. Epidemiol. 146: 177-185.

Tsai, S.-S.; Goggins, W. B.; Chiu, H.-F.; Yang, C.-Y. (2003a) Evidence for an association between air pollution and daily stroke admissions in Kaohsiung, Taiwan. Stroke 34: 2612-2616.

Tsai, S.-S.; Huang, C.-H.; Goggins, W. B.; Wu, T.-N.; Yang, C.-Y. (2003b) Relationship between air pollution and daily mortality in a tropical city: Kaohsiung, Taiwan. J. Toxicol. Environ. Health Part A 66: 1341-1349.

Tyler, W. S.; Tyler, N. K.; Last, J. A.; Gillespie, M. J.; Barstow, T. J. (1988) Comparison of daily and seasonal exposures of young monkeys to ozone. Toxicology 50: 131-144.

U.S. Environmental Protection Agency. (1996a) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: www.epa.gov/ncea/ozone.htm.

U.S. Environmental Protection Agency. (1996b) Air quality criteria for particulate matter. Research Triangle Park, NC: National Center for Environmental Assessment-RTP Office; report nos. EPA/600/P-95/001aF-cF. 3v.

U.S. Environmental Protection Agency. (2004) Air quality criteria for particulate matter. Research Triangle Park, NC: National Center for Environmental Assessment; report no. EPA/600/P-99/002aF-bF. 2v. Available: http://cfpub.epa.gov/ncea/ [9 November, 2004].

Ulmer, C.; Kopp, M.; Ihorst, G.; Frischer, T.; Forster, J.; Kuehr, J. (1997) Effects of ambient ozone exposures during the spring and summer of 1994 on pulmonary function of schoolchildren. Pediatr. Pulmonol. 23: 344-353.

Vaughan, T. R.; Weber, R. W.; Tipton, W. R.; Nelson, H. S. (1989) Comparison of PEFR and FEV_1 in patients with varying degrees of airway obstruction: effect of modest altitude. Chest 95: 558-562.

Vedal, S.; Brauer, M.; White, R.; Petkau, J. (2003) Air pollution and daily mortality in a city with low levels of pollution. Environ. Health Perspect. 111: 45-51.

Vedal, S.; Rich, K.; Brauer, M.; White, R.; Petkau, J. (2004) Air pollution and cardiac arrhythmias in patients with implantable cardiovascular defibrillators. Inhalation Toxicol. 16: 353-362.

Verhoeff, A. P.; Hoek, G.; Schwartz, J.; Van Wijnen, J. H. (1996) Air pollution and daily mortality in Amsterdam. Epidemiology 7: 225-230.

Virnig, B. A.; McBean, M. (2001) Administrative data for public health surveillance and planning. Annu. Rev. Public Health 22: 213-230.

Wang, X.; Ding, H.; Ryan, L.; Xu, X. (1997) Association between air pollution and low birth weight: a community-based study. Environ. Health Perspect. 105: 514-520.

Ward, D. J.; Roberts, K. T.; Jones, N.; Harrison, R. M.; Ayres, J. G.; Hussain, S.; Walters, S. (2002) Effects of daily variation in outdoor particulates and ambient acid species in normal and asthmatic children. Thorax 57: 489-502.

Weisel, C. P.; Cody, R. P.; Lioy, P. J. (1995) Relationship between summertime ambient ozone levels and emergency department visits for asthma in central New Jersey. Environ. Health Perspect. 103(suppl. 2): 97-102.

Weisel, C. P.; Cody, R. P.; Georgopoulos, P. G.; Purushothaman, V.; Weiss, S. H.; Bielory, L.; Gregory, P.; Stern, A. H. (2002) Concepts in developing health-based indicators for ozone. Int. Arch. Occup. Environ. Health 75: 415-422.

White, M. C.; Etzel, R. A.; Wilcox, W. D.; Lloyd, C. (1994) Exacerbations of childhood asthma and ozone pollution in Atlanta. Environ. Res. 65: 56-68.

Wiley, J. A.; Robinson, J. P.; Piazza, T.; Garrett, K.; Cirksena, K.; Cheng, Y.-T.; Martin, G. (1991a) Activity patterns of California residents. Final report. Sacramento, CA: California Air Resources Board; report no. ARB/R93/487. Available from: NTIS, Springfield, VA.; PB94-108719.

Wiley, J. A.; Robinson, J. P.; Cheng, Y.-T.; Piazza, T.; Stork, L.; Pladsen, K. (1991b) Study of children's activity patterns: final report. Sacramento, CA: California Air Resources Board; report no. ARB-R-93/489.

Wilson, A. M.; Wake, C. P.; Kelly, T.; Salloway, J. C. (2005) Air pollution, weather, and respiratory emergency room visits in two northern New England cities: an ecological time-series study. Environ. Res. 97: 312-321.

Wong, T. W.; Lau, T. S.; Yu, T. S.; Neller, A.; Wong, S. L.; Tam, W.; Pang, S. W. (1999a) Air pollution and hospital admissions for respiratory and cardiovascular diseases in Hong Kong. Occup. Environ. Med. 56: 679-683.

Wong, C.-M.; Ma, S.; Hedley, A. J. Lam, T.-H. (1999b) Does ozone have any effect on daily hospital admissions for circulatory diseases? J. Epidemiol. Community Health 53: 580-581.

Wong, C.-M.; Atkinson, R. W.; Anderson, H. R.; Hedley, A. J.; Ma, S.; Chau, P. Y.-K.; Lam, T.-H. (2002) A tale of two cities: effects of air pollution on hospital admissions in Hong Kong and London compared. Environ. Health Perspect. 110: 67-77.

World Health Organization. (2004) Meta-analysis of time-series studies and panel studies of particulate matter (PM) and ozone (O_3): report of a WHO task group. Copenhagen, Denmark: WHO Regional Office for Europe; document no. EUR/04/5042688. Available: http://www.euro.who.int/document/E82792.pdf [18 November, 2004].

Woodruff, T. J.; Grillo, J.; Schoendorf, K. C. (1997) The relationship between selected causes of postneonatal infant mortality and particulate air pollution in the United States. Environ. Health Perspect. 105: 608-612.

Xu, X.; Ding, H.; Wang, X. (1995) Acute effects of total suspended particles and sulfur dioxides on preterm delivery: a community-based cohort study. Arch. Environ. Health 50: 407-415.

Yang, Q.; Chen, Y.; Shi, Y.; Burnett, R. T.; McGrail, K. M.; Krewski, D. (2003) Association between ozone and respiratory admissions among children and the elderly in Vancouver, Canada. Inhalation Toxicol. 15: 1297-1308.

Yang, C.-Y.; Chen, Y.-S.; Yang, C.-H.; Ho, S.-C. (2004a) Relationship between ambient air pollution and hospital admissions for cardiovascular diseases in Kaohsiung, Taiwan. J. Toxicol. Environ. Health Part A 67: 483-493.

Yang, C.-Y.; Chang, C.-C.; Chuang, H.-Y.; Tsai, S.-S.; Wu, T.-N.; Ho, C.-K. (2004b) Relationship between air pollution and daily mortality in a subtropical city: Taipei, Taiwan. Environ. Int. 30: 519-523.

Zeger, S. L.; Thomas, D.; Dominici, F.; Samet, J. M.; Schwartz, J.; Dockery, D.; Cohen, A. (2000) Exposure measurement error in time-series studies of air pollution: concepts and consequences. Environ. Health Perspect. 108: 419-426.

Zhu, L.; Carlin, B. P.; Gelfand, A. E. (2003) Hierarchical regression with misaligned spatial data: relating ambient ozone and pediatric asthma ER visits in Atlanta. Environmetrics 14: 537-557.

Zidek, J. V. (1997) Interpolating air pollution for health impact assessment. In: Barnett, E. V.; Turkman, K. F., eds. Pollution Assessment and Control. New York, NY: John Wiley & Sons. (Statistics for the Environment, no. 3).

Zidek, J. V.; Wong, H.; Le, N. D.; Burnett, R. (1996) Causality, measurement error and multicollinearity in epidemiology. Environmetrics 7: 441-451.

Zidek, J. V.; White, R.; Le, N. D.; Sun, W.; Burnett, R. T. (1998) Imputing unmeasured explanatory variables in environmental epidemiology with application to health impact analysis of air pollution. Environ. Ecol. Stat. 5: 99-115.

Zmirou, D.; Barumandzadeh, T.; Balducci, F.; Ritter, P.; Laham, G.; Ghilardi, J.-P. (1996) Short term effects of air pollution on mortality in the city of Lyon, France, 1985-90. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Community Health 50(suppl. 1): S30-S35.

Zmirou, D.; Schwartz, J.; Saez, M.; Zanobetti, A.; Wojtyniak, B.; Touloumi, G.; Spix, C.; Ponce de León, A.; Le Moullec, Y.; Bacharova, L.; Schouten, J.; Pönkä, A.; Katsouyanni, K. (1998) Time-series analysis of air pollution and cause-specific mortality. Epidemiology 9: 495-503.

8. INTEGRATIVE SYNTHESIS: OZONE EXPOSURE AND HEALTH EFFECTS

8.1 INTRODUCTION

This integrative synthesis is structured to provide a coherent framework for the assessment of health risks associated with human exposures to ambient surface-level (tropospheric) ozone (O_3) in the United States. The main goal of the chapter is to integrate newly available scientific information with key findings and conclusions from the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996a), so as to address issues central to the EPA's assessment of evidence needed to support the current review of the primary O_3 NAAQS. The integrated assessment of key findings and conclusions provided here and elsewhere in this document with regard to O_3 exposure and health effects will be drawn upon and their policy implications considered in an Ozone Staff Paper prepared by EPA's Office of Air Quality Planning and Standards (OAQPS). The analyses provided in that Staff Paper aim to "bridge the gap" between scientific assessments in this criteria document and judgments required of the EPA administrator in evaluating whether to retain or, possibly, to revise the current primary O_3 NAAQS. Other types of scientific information concerning ambient O_3 welfare effects (i.e., tropospheric O_3 effects on vegetation and ecosystems, relationships to surface-level solar UV flux/climate changes, and effects on man-made materials) are assessed in ensuing Chapters 9, 10, and 11. That information will also be considered in the OAQPS staff paper in posing options regarding the secondary O_3 NAAQS.

As discussed in Chapter 2 of this document, O_3 found in the earth's troposphere generally originates from photochemical reactions that are predominantly catalyzed by the interaction of sunlight with precursor pollutants, especially nitrogen oxides (NO_x) and hydrocarbons such as volatile organic compounds (VOCs), emitted by surface-level mobile and stationary sources. Other photochemical oxidants, such as peroxyacetyl nitrate (PAN) and hydrogen peroxide (H_2O_2), are also generated along with O_3 by such atmospheric interactions. In addition to the tropospheric O_3 generated by these interactions, some O_3 is found near the earth's surface as the result of its downward transport from the stratosphere. However, in contrast to stratospheric O_3, which plays an important role in maintaining the habitability of the planet by shielding the

surface from harmful solar ultraviolet (UV) radiation, tropospheric O_3 at the surface can exert adverse effects on humans, nonhuman animal species, and vegetation. As was the case for previous O_3-related NAAQS criteria revisions, the present criteria document focuses mainly on the assessment of health and welfare effects resulting from exposures to surface-level concentrations of tropospheric O_3, whereas less attention is accorded to the distinctly much more limited available information on other photochemical oxidants, e.g., PAN or H_2O_2.

Based on the criteria review completed in 1978, the original primary and secondary NAAQS set in 1971 for total photochemical oxidants were revised in 1979 to focus on O_3 as the indicator for new primary and secondary standards that were attained when the expected number of days per calender year with maximum 1-h average O_3 concentrations >0.12 ppm did not exceed one. The NAAQS for ambient O_3 were revised in 1997 by replacing the 1-h standards with an 8-h primary standard that is met when the 3-year average of the annual fourth highest daily maximum 8-h average concentration is <0.08 ppm. The new 1997 primary standard was based on various scientific supportive data from experimental human exposure, animal toxicological and epidemiological studies, as assessed in the 1996 O_3 AQCD and in the 1996 O_3 Staff Paper (U.S. Environmental Protection Agency, 1996b).

8.1.1 Chapter Organization

In addition to providing the above brief background information regarding prior O_3 NAAQS reviews (including the 1997 EPA revision of the O_3 NAAQS), this first section (8.1 Introduction) of the integrative synthesis chapter aims to orient the reader to the organization and content of the chapter. The next section (Section 8.2) focuses on air quality trends and current ambient O_3levels to help provide context for the ensuing discussions of O_3 exposures and associated health effects. The subsequent sections (8.3, 8.4, and 8.5) then integrate newly available key scientific information assessed in Chapters 4 through 7 of this document, including integration of information on O_3 dosimetry, toxicological information derived from controlled human exposure and laboratory animal studies, and epidemiologic evidence.

These sections collectively address the following topics: (1) ambient O_3 exposures, personal exposures, and dosimetric considerations; (2) experimental studies on toxicological responses to acute O_3 exposures in humans (clinical studies) and both acute and chronic effects

in animals; (3) epidemiological evidence for associations between O_3 exposure of human populations and health effects and the strength and robustness of these associations; (4) integration of the experimental and epidemiological evidence; (5) biological mechanisms and other evidence useful in judging the plausibility of adverse health effects being associated with human exposures to ambient O_3 levels encountered in the United States; and (6) identification of susceptible and vulnerable populations likely at increased risk for O_3-related health effects and numbers of people potentially falling in such categories in the United States.

The present chapter mainly focuses on discussion of new scientific information that has become available since the 1996 O_3 criteria review that supported EPA's revision of the O_3 NAAQS in 1997. This includes assessment of information published or accepted for publication in peer-reviewed open literature mainly through December 2004, with a few particularly pertinent and important studies published beyond that point also being considered.

Important data gaps and uncertainties that still exist with regard to various important issues and research needs are also briefly noted for some key areas. Detailed discussion of such research needs is beyond the scope of this document; however, such discussion is typically undertaken later as part of EPA efforts focused on identification of O_3 research needs and development of associated research planning documents.

8.2 AMBIENT OZONE AIR QUALITY IN UNITED STATES

8.2.1 Current Ozone Concentrations and Spatial Patterns

Ambient air O_3 is monitored in the United States during 'ozone seasons', which vary in length depending on location. The ozone season extends all year in the Southwest. In most other areas of the country, O_3 is monitored typically from April to October. However, O_3 is monitored throughout the year in many urban areas, in as much as O_3 is present the year round not only in polluted areas but in clean areas as well. The median of the daily maximum 8-h average O_3 concentration in the United States, averaged over May to September from 2000 to 2004 for all U.S. counties, was 0.049 ppm. In 95% of all counties, the median of the daily maximum 8-h average O_3 concentration was less than 0.057 ppm. However, it should be noted that most monitors are located in the East. The daily maximum 1-hour concentrations were

typically much higher in large urban areas or in areas downwind of them. For example, in Houston, TX they approached 0.20 ppm during this period. Daily 1-hour maximum ozone concentrations were lower in the rest of the country, but were still above 0.12 ppm in many locations. Eight hour daily maximum concentrations were not as high, but tend to be highly correlated with 1-hour daily maximums.

Within individual MSAs, O_3 concentrations tend to be well correlated across monitoring sites, although spatial variations in concentrations can be substantial. In many city centers, O_3 concentrations tend to be lower than in either upwind or downwind areas, largely due to reaction of O_3 with NO emitted by motor vehicles. For example, much lower O_3 concentrations overall are found in downtown Los Angeles (e.g., in Lynwood) than at sites located further downwind (e.g., in San Bernadino). The much higher downwind levels are formed from photochemical reactions involving the urban emissions, including products formed as the result of reactions titrating O_3 in the urban core. Thus, O_3 concentrations tend to be higher downwind of urban centers, and they decrease again in going to areas that are more remote from precursor sources. Likewise, surface-level O_3 can be depleted in rural areas close to NO sources, such as highways and powerplants.

8.2.2 Diurnal and Seasonal Variations

Ozone concentrations typically tend to peak in early to mid-afternoon in areas where there is strong photochemical activity and to peak later in the afternoon or during early evening in areas where transport is more important in determining the O_3 abundance. Summertime maxima in O_3 concentrations occur in those U.S. areas where substantial photochemical activity acts on O_3 precursors emitted as the result of human activities. Monthly maxima can occur anytime from June through August. However, springtime maxima are observed in some National Parks, mainly in the western United States, and at a number of other relatively unpolluted monitoring sites throughout the Northern Hemisphere. For example, the highest O_3 concentrations at Yellowstone National Park tend to occur during April and May. Typically, monthly minima tend to occur from November through February at polluted sites and during the fall at relatively remote sites.

8.2.3 Long-Term Trends

National attention started to be focused in the 1940s on O_3 and associated photochemical smog in the Los Angeles area. Prior to the adoption of stringent emissions controls, peak levels of O_3 were consistently higher in the Los Angeles area than are currently observed. For example, in 1958, peak O_3 concentrations measured in Los Angeles were about 0.6 ppm but have declined since then, although not at a steady rate. Peak O_3 levels of 0.2 to 0.5 ppm were still found at some locations in the Los Angeles basin during the 1970s. For example, on two days (October 13 and 14) during a 1978 episode, Tuazon et al. (1981) observed peak 1-h averaged values of O_3 of nearly 0.4 ppm and nearly 0.5 ppm. Currently, peak 1-h and 8-h average O_3 concentrations are about 0.17 and 0.15 ppm in the Los Angeles basin (cf. Figures 3-10 and 3-11). High O_3 levels were also earlier found throughout the rest of the United Sates as well, but peak O_3 levels have also gradually declined across the country during the 1980s. However, during one particularly hot summer (of 1988) in the East, peak 1-h O_3 concentrations of about 0.2 ppm were observed in many eastern U.S. cities (U.S. Environmental Protection Agency, 1990).

Historically high O_3 concentrations, as noted above, have not only been observed in the United States. For example, during an episode in Great Britain in 1976, peak O_3 levels exceeded 0.25 ppm and daily maximum 8-h O_3 concentrations were above 0.1 ppm for 18 consecutive days at one rural site (Wayne, 1991). Also, concentrations of O_3 in the range found in Los Angeles during the 1970s are still found in Mexico City.

Nationwide, 2nd highest 1-h ozone concentrations in the United States have decreased dramatically during the past several decades, i.e., by approximately 29 percent from 1980 to 2003 and 16 percent from 1990 to 2003. Also, 4th highest 8-h O_3 concentrations decreased by approximately 21 percent since 1980 and 9 percent since 1990 (U.S. Environmental Protection Agency, 2003). Trends in metrics for evaluating compliance with the O_3 NAAQS (i.e., changes in the 4th highest O_3 concentration) can be found in EPA's "National Air Quality and Emissions Trends Reports". These reports indicate that the 4th highest O_3 concentrations are still decreasing nationwide, but the rate of decrease has slowed since 1990. However, such trends have not been uniform across the United States. In general, reductions in the O_3 metrics given above have been largest in New England and in states along the West Coast and smallest in midwestern states. Downward trends in California O_3 concentrations have been driven mainly by notable decreases in Southern California, with reductions in other areas not being as large.

Trends in peak O_3 metrics do not necessarily reflect changes in O_3 values across the middle of the distribution of O_3 concentrations. Of note, O_3 concentrations towards the center of its nationwide distribution have not shown much change, and there are some indications that O_3 concentrations at the lower end of the distribution may even be increasing.

8.2.4 Interrelationships Between Ozone and Other Ambient Pollutants

Data on ambient concentrations of other oxidants (e.g., H_2O_2, PAN) and oxidation products (e.g., HNO_3, H_2SO_4) in the atmosphere are not nearly as abundant as they are for O_3. Because data for such species are usually obtained only as part of specialized field studies, it is difficult to relate observed ambient O_3 concentrations to ambient levels of other oxidant species or oxidation products. In general, such secondary species are expected to be at least moderately positively correlated with O_3. On the other hand, primary species are expected to be more highly correlated with each other than with secondary species, provided that the primary species originate from common sources in given areas. Measurements of gas phase oxidants conducted as part of the Southern Oxidants Study (SOS) indicated combined hydroperoxide (H_2O_2, CH_3OOH, and $HOCH_2OOH$) concentrations typically in the range of several ppb. Concentrations of PAN, PPN and MPAN also observed during the SOS likewise indicated combined concentrations in the range of several ppb. Oxidants are also present in airborne cloud droplets, rain drops, and particulate matter (PM). A few measurements of reactive oxygen species (expressed as equivalent H_2O_2) in ambient fine PM indicated levels of less than 1% of those for ambient O_3 on a molar basis. However, it should be noted that these measurements are potentially subject to both positive and negative artifacts.

Because PM is not a single distinct chemical species, but rather a mix of primary and secondary species, relationships between ambient O_3 and PM concentrations can be quite complex. As an example of this complexity, $PM_{2.5}$ concentrations positively correlated with O_3 during the summer, but negatively correlated with O_3 during the winter at Ft. Meade, MD. Also, Ito et al. (2005) examined relationships between PM_{10} and O_3 on a seasonal basis in several urban areas (cf. Figure 7-24). Seasonal relationships with ambient O_3 similar to those at Ft. Meade were found, reflecting the dominant contribution of $PM_{2.5}$ to PM_{10} in the urban areas studied (although PM_{10} generally contains a higher fraction than does $PM_{2.5}$ of primary [mainly crustal] material). Possibly contributing to the higher correlations observed between fine PM

and O_3 in the summer is the fact that O_3 can contribute to formation of submicron particles via interactions with various other atmospheric constituents present, such as terpenes, and other biogenically derived hydrocarbons from trees, other vegetation, and wood products. Formation of ultrafine particles by this mechanism is most likely to occur during afternoons of summer days when temperatures and O_3 concentrations are sufficiently elevated to facilitate O_3 reactions with increased amounts of terpenes emitted from vegetation. Bursts of ultrafine particle formation have been observed repeatedly in both urban and rural air. Woo et al. (2001), for example, reported rapid formation of ultrafine particles in the ambient air of Atlanta typically around noon in both summer and winter. The mechanisms underlying such ultrafine particle formation events may also involve other atmospheric reactions that are related to O_3 formation, such as the nucleation of H_2SO_4 (produced by oxidation of SO_2) and, probably, NH_3.

8.2.5 Policy Relevant Background (PRB) Ozone Concentrations

Background O_3 concentrations used for NAAQS-setting purposes are referred to as Policy Relevant Background (PRB) O_3 concentrations. Policy Relevant Background concentrations are those that would occur in the United States in the absence of anthropogenic emissions in continental North America (defined here as the United States, Canada, and Mexico). Such PRB O_3 concentrations include contributions from natural sources everywhere in the world and from anthropogenic sources outside these three countries. For the purpose of informing O_3 NAAQS decisions, EPA focuses on assessing risks to human health and environmental effects from O_3 levels in excess of PRB concentrations. Issues concerning the methodology for estimating PRB O_3 concentrations are discussed in detail in Section AX3.9 of Annex AX3.

Contributions to PRB O_3 include photochemical reactions involving natural emissions of VOCs, NO_x, and CO, as well as the long-range transport of O_3 and its precursors from outside North America and the stratospheric-tropospheric exchange (STE) of O_3. Processes involved in STE are described in detail in Section AX2.3 of Annex AX2. Natural sources of O_3 precursors include biogenic emissions, wildfires, and lightning. Biogenic emissions from agricultural activities are not considered in the formation of PRB O_3.

Currently, estimates of PRB O_3 concentrations are based on predictions generated by the global scale, three dimensional, chemical transport model GEOS-CHEM (Fiore et al., 2003). Estimates of PRB O_3 concentrations cannot be derived solely from measurements of O_3 at

relatively unpolluted sites because of long-range transport from anthropogenic source regions within North America. It is impossible to determine sources of O_3 at a particular location without ancillary data that could be used as tracers of sources or to calculate photochemical production and loss rates for O_3. Policy relevant background O_3 concentrations vary as a function of season, altitude, and total surface O_3 concentration, with PRB O_3 concentrations at the surface generally falling in the range of 0.015 to 0.035 ppm from 1300 to 1700 local time and tending to decline under conditions conducive to O_3 episodes. The PRB concentrations are highest during spring and decline into summer; and higher values tend also to occur at higher elevations during the spring due to contributions from hemispheric pollution and stratospheric intrusions. The contribution to surface O_3 by stratospheric intrusions is typically well below 0.020 ppm. Stohl (2001) and Sprenger et al. (2003) found that the maximum probability of stratospheric intrusions reaching the 800 hPa level (~1800 m) was less than 1% and that higher probabilities (1 to 2%, and 10%) applied for stratospheric intrusions penetrating to the 600 hPa level (~4100 m) and 500 hPa level (~5400 m), respectively. Thus, stratospheric intrusions only rarely contribute to elevated surface-level O_3 concentrations at low altitude sites but have a higher (albeit still low) probability of elevating them at high-altitude sites.

8.3 FACTORS AFFECTING HUMAN EXPOSURE TO AMBIENT OZONE

Exposure to O_3 and related photochemical oxidants varies over time due to changes in their ambient concentrations and because people move between locations having notably different concentrations. The amount of O_3 delivered to the lung is not only influenced by the ambient concentration but also by the individual's breathing route and rate. Thus, activity level is an important consideration in determining the potential O_3 exposure and dose received.

The use of data from ambient air monitoring stations is still the most common surrogate for assigning exposure estimates in epidemiologic studies. Since the primary source of O_3 exposure is the ambient air, O_3 concentration data from outdoor community monitoring sites should provide a relative assignment of exposure with time, if: concentrations are relatively uniform across the region; time-activities pattern are roughly the same across the study population; and housing characteristics (such as ventilation rates and O_3 sinks contributing to indoor O_3 decay rates) are relatively constant for the study area. However, because these types of factors often do

vary across populations and locations, some error tends to be associated not only with estimates of the magnitude of O_3 exposure but, also, potentially with relative exposure assignments based solely on ambient monitoring data. Nevertheless, ambient O_3 monitoring data appear to provide the most useful index of human O_3 exposure currently available to help characterize health outcomes associated with O_3 exposures of large population groups.

8.3.1 Personal Exposure

Personal O_3 concentrations have been measured for children, outdoor workers, and individuals with COPD, all being populations potentially susceptible to O_3 or other respiratory irritants. Outdoor workers can be expected to have somewhat higher O_3 exposures than other individuals, because they typically spend more time outdoors and often engage in prolonged moderate and heavy exertion activities. Children also tend to be more active outside and, therefore, often manifest a higher breathing rate than most adults. However, available exposure measurement studies are not sufficient to allow for highly confident broad quantitative generalization about the "typical" magnitudes of observed differences in exposure between the general population and such potentially susceptible subpopulations.

8.3.2 Indoor Concentrations

Apart from only a few specific indoor sources such as photo-copying machines, O_3 indoors is derived from the infiltration of ambient air from outdoors. Generally, O_3 enters indoor environments through infiltration from outdoors and through building components, such as windows, doors, and ventilation systems. Ozone concentrations in indoor environments depend primarily on the outdoor O_3 concentration, outdoor/indoor infiltration and the air exchange rate (AER). Once indoors, O_3 reacts on various surfaces and with airborne components of either indoor or outdoor origin.

Indoor O_3 concentrations tend to reflect outdoor concentrations and, hence, are higher when outdoor O_3 is higher. However, because O_3 reacts indoors with surfaces and other contaminants, O_3 concentrations are typically lower indoors than outdoors. Gas phase reactions occurring outdoors also produce other oxidants analogous to the production of photochemical smog. The extent and rate of production of these other species indoors is a function of indoor O_3

concentrations and the presence of other necessary precursors (i.e., VOCs), along with an optimal AER.

Several studies have measured O_3 concentrations in residences, schools, office buildings and museums; and typical concentrations varied across all such locations. However, indoor concentrations generally varied in relationship to the AER in the indoor environment (increasing with higher AER) and generally tended to be notably lower than outdoor ambient O_3 levels. For example, one study examining the relationship between O_3 concentrations indoors and outside of a school in New England reported average O_3 concentrations of 20 ppb (0.020 ppm) indoors and 40 ppb (0.040 ppm) outdoors. With regard to mobile source microenvironments, as is the case for other enclosed environments, O_3 exposures depend on the extent of mixing of outdoor air into the vehicle cabin. Thus, if windows are kept open, O_3 concentrations inside the vehicle may be expected to approach outdoor values; but, if windows are kept closed and there is air conditioning, then interior values can be much lower than those outside, especially if recirculated air is used. For example, in one N.C. study involving police cars with air conditioning and recirculated air, O_3 concentrations in the vehicle cabin (11.7 ppb average) were less than half those outside (28.3 ppb average at outdoor monitoring sites in the area).

Although concentrations of O_3 may be reduced to lower levels once ambient O_3 enters indoor environments, it should be kept in mind that the indoor O_3 may interact with other airborne substances of indoor or outdoor origin that may be present indoors. For example, Wainman et al. (2000) showed that O_3 reacts with d-limonene, a common component of air fresheners to produce submicron particles. These particles are found mainly in the size range from 0.1 to 0.3 μm. Wainman et al. noted that terpenes such as limonene are emitted by wood products; that they are used as solvents, as odorants in cleaning products, and as air fresheners; and, because of their widespread uses, their concentrations are often higher indoors than they are outdoors. In addition to particle formation, Weschler (2004) points out that gas phase products, such as aldehydes and hydroperoxides, produced by reactions of O_3 with terpenes and other unsaturated carbon compounds may also be of concern. During the formation of these products, OH radicals are also produced which can react with compounds that do not react with O_3. To the extent that building ventilation rates etc. remain constant between days characterized by high and low O_3, the concentrations of these other secondary pollutants formed indoors will tend to be correlated with ambient O_3. Thus, ambient O_3 concentrations measured outdoors at community

monitoring sites and/or personal O_3 exposure monitor measurements may serve not only as indices of direct human exposure to O_3 per se, but also as surrogate indices of exposures to broader O_3-containing mixtures of ambient or indoor air contaminants.

8.4 SYNTHESIS OF AVAILABLE INFORMATION ON OZONE-RELATED HEALTH EFFECTS

The integrated synthesis of the latest available information on O_3-related health effects poses large challenges, especially in view of the emergence of certain important new information since the 1996 O_3 AQCD, which adds greatly to the complexity of an integrative assessment. Such information includes new findings from:

- Dosimetry studies that clarify further factors potentially affecting regional distribution of O_3 in the respiratory tract of humans and laboratory animals and providing improved bases by which to attempt animal-to-human extrapolations of experimentally-observed O_3-induced health effects.
- Experimental toxicological studies using controlled human exposures and laboratory animals aimed at delineating exposure-response relationships and understanding potential biochemical mechanisms underlying toxic effects, pathology, and susceptibility;
- Epidemiological studies, reflecting progress in addressing many research needs identified during the last review, as well as raising new issues and reevaluating previously addressed issues that remain important in interpreting the body of epidemiological evidence and characterization of its strengths and limitations.

Previous criteria assessments, including the 1996 O_3 AQCD, found that experimental studies of controlled human and laboratory animal exposures to O_3 provided the most clear cut and compelling evidence with regard to characterizing O_3-related health effects. This section first summarizes key dosimetry and health related findings derived from the 1996 O_3 AQCD and then integrates those findings with new information obtained since 1996 from human and animal experimental studies. Ozone-induced physiological, pathological, cellular and biochemical alterations are evaluated in order to assess human health effects due to ambient O_3 exposures. Also, the influence of O_3-induced changes at cellular and molecular levels are integrated to elucidate scientific bases for the observed physiological and pathological alterations. These research results are evaluated in order both to help assess the biological plausibility of health

outcome associations observed in epidemiologic studies and to assess the coherence of the overall body of evidence relevant to O_3-related health outcome conclusions.

8.4.1 Key Health-Related Findings and Conclusions from the 1996 Ozone Air Quality Criteria Document

Based on extensive dosimetric and experimental data as well as growing epidemiologic evidence available at the time, the 1996 O_3 AQCD arrived at a set of findings and conclusions stated in relation to answering five key questions regarding potential health effects of ambient O_3 exposure. In general, the existing evidence was such to warrant a high degree of confidence in those conclusions derived from experimental (controlled exposure) studies. Considerable confidence could also be placed in the emerging field/panel studies providing observational study results substantiating and extending the controlled exposure study findings. Other epidemiologic studies provided highly suggestive, although less conclusive, indications of increased morbidity (e.g., as indexed by emergency department visits, hospital admissions, etc.) and, possibly, mortality being associated with exposure of human populations to ambient O_3. The main findings and conclusions derived from the 1996 ozone criteria review are recapitulated (largely verbatim) below in relation to the five key questions addressed in the summary and conclusions of the Integrative Synthesis in the 1996 O_3 AQCD.

1. What are the effects of short-term (<8-h) exposures to ozone?

Short-term O_3 exposure of laboratory animals and humans causes changes in pulmonary function, including tachypnea (rapid, shallow breathing), decreased lung volumes and flows, and increased airway responsiveness to nonspecific stimuli. Increased airway resistance occurs in both humans and laboratory animals, but typically at higher exposure levels than other functional endpoints. In addition, adult human subjects experience O_3 induced symptoms of airway irritation such as cough or pain on deep inspiration. The changes in pulmonary function and respiratory symptoms occur as a function of exposure concentration, duration, and level of exercise. Adult human subjects with mild asthma have qualitatively similar responses in lung volume and airway responsiveness to bronchoconstrictor drugs as nonasthmatics. Respiratory symptoms are also similar, but wheezing is a prevalent symptom in O_3-exposed asthmatics in addition to the other demonstrated symptoms of airway irritation. Airway resistance, however,

increases relatively more in asthmatics from an already higher baseline. Recovery from the effects of O_3 on pulmonary function and symptoms is usually complete within 24 h of the end of exposure, although other responses may persist somewhat longer.

- *Increased O_3 levels are associated with increased hospital admissions and emergency department visits for respiratory causes. Analyses from data in the Northeastern United States suggest that O_3 air pollution is associated with a substantial portion (on the order of 10 to 20%) of all summertime respiratory hospital visits and admissions.*
- *Pulmonary function in children at summer camps in southern Ontario, Canada, in the northeastern United States, and in Southern California is associated with O_3 concentration. Meta-analysis indicates that a 0.50-mL decrease in FEV_1 is associated with a 1 ppb increase in O_3 concentration. For preadolescent children exposed to 120 ppb (0.12 ppm) ambient O_3, this amounts to an average decrement of 2.4 to 3.0% in FEV_1. Similar responses are reported for children and adolescents exposed to O_3 in ambient air or O_3 in purified air for 1 to 2 h while exercising.*
- *Pulmonary function decrements are generally observed in healthy subjects (8 to 45 years of age) after 1 to 3 h of exposure as a function of the level of exercise performed and the O_3 concentration inhaled during the exposure. Group mean data from numerous controlled human exposure and field studies indicate that, in general, statistically significant pulmonary function decrements beyond the range of normal measurement variability (e.g., 3 to 5% for FEV_1) occur*
  - *at >0.50 ppm O_3 when at rest,*
  - *at >0.37 ppm O_3 with light exercise (slow walking),*
  - *at >0.30 ppm O_3 with moderate exercise (brisk walking),*
  - *at >0.18 ppm O_3 with heavy exercise (easy jogging), and*
  - *at >0.16 ppm O_3 with very heavy exercise (running).*
- *Smaller group mean changes (e.g., <5%) in FEV_1 have been observed at lower O_3 concentrations than those listed above. For example, FEV_1 decrements have been shown to occur with very heavy exercise in healthy adults at 0.15 to 0.16 ppm O_3, and such effects may occur in healthy young adults at levels as low as 0.12 ppm. Also, pulmonary function decrements have been observed in children and adolescents at concentrations of 0.12 and 0.14 ppm O_3 with heavy exercise. Some individuals within a study may experience FEV_1 decrements in excess of 15% under these exposure conditions, even when the group mean decrement is less than 5%.*
- *For exposures of healthy subjects performing moderate exercise during longer duration exposures (6 to 8 h), 5% group mean decrements in FEV_1 were observed at*
  - *0.08 ppm O_3 after 5.6 h,*
  - *0.10 ppm O_3 after 4.6 h, and*

1 – *0.12 ppm O_3 after 3 h.*

2 • *For these same subjects, 10% group mean FEV_1 decrements were observed at 0.12 ppm O_3 after 5.6 and 6.6 h. As in the shorter duration studies, some individuals experience changes larger than those represented by the group mean changes.*

3 • *An increase in the incidence of cough has been reported at O_3 concentrations as low as 0.12 ppm in healthy adults during 1 to 3 h of exposure with very heavy exercise. Other respiratory symptoms, such as pain on deep inspiration, shortness of breath, and lower respiratory scores (a combination of several symptoms), have been observed at 0.16 to 0.18 ppm O_3 with heavy and very heavy exercise. Respiratory symptoms also have been observed following exposure to 0.08, 0.10, and 0.12 ppm O_3 for 6.6 h with moderate levels of exercise.*

4 • *Increases in nonspecific airway responsiveness in healthy adults have been observed after 1 to 3 h of exposure to 0.40 ppm, but not 0.20 ppm, O_3 at rest and have been observed at concentrations as low as 0.18 ppm, but not to 0.12 ppm, O_3 during exposure with very heavy exercise. Increases in nonspecific airway responsiveness during 6.6-h exposures with moderate levels of exercise have been observed at 0.08, 0.10, and 0.12 ppm O_3.*

5 *Short-term O_3 exposure of laboratory animals and humans disrupts the barrier function of*
6 *the lung epithelium, permitting materials in the airspaces to enter lung tissue, allowing cells and*
7 *serum proteins to enter the airspaces (inflammation), and setting off a cascade of responses.*

8 • *Increased levels of PMNs and protein in lung lavage fluid have been observed following exposure of healthy adults to 0.20, 0.30, and 0.40 ppm with very heavy exercise and have not been studied at lower concentrations for 1- to 3-h exposures. Increases in lung lavage protein and PMNs also have been observed at 0.08 and 0.10 ppm O_3 during 6.6-h exposures with moderate exercise; lower concentrations have not been tested.*

9 *Short-term O_3 exposure of laboratory animals and humans impairs alveolar macrophage*
10 *clearance of viable and nonviable particles from the lungs and decreases the effectiveness of*
11 *host defenses against bacterial lung infections in animals and perhaps humans. The ability of*
12 *alveolar macrophages to engulf microorganisms is decreased in humans exposed to 0.08 and*
13 *0.10 ppm O_3 for 6.6 h with moderate exercise.*

14 *Recent epidemiology studies addressing the effects of short-term ambient exposure to O_3 in*
15 *the population have yielded significant associations with a wide range of health outcomes,*
16 *including lung function decrements, aggravation of preexisting respiratory disease, increases in*
17 *daily hospital admissions and emergency department visits for respiratory causes, and increased*
18 *mortality. Results from lung function epidemiology studies are generally consistent with the*

experimental studies in laboratory animals and humans. An association between daily mortality and O_3 concentration for areas with high O_3 levels (e.g., Los Angeles) has been suggested, although the magnitude of such an effect is unclear.

2. What are the effects of repeated, short-term exposures to ozone?

During repeated short-term exposures, some of the O_3-induced responses are partially or completely attenuated. Over a 5-day exposure, pulmonary function changes are typically greatest on the second day, but return to control levels by the fifth day of exposure. Most of the inflammatory markers (e.g., PMN influx) also attenuate by the fifth day of exposure, but markers of cell damage (e.g., lactate dehydrogenase enzyme activity) do not attenuate and continue to increase. Attenuation of lung function decrements is reversed following 7 to 10 days without O_3. Some inflammatory markers are also reversed during this time period, but others still show attenuation even after 20 days without O_3. The mechanisms and impacts involved in attenuation are not known, although animal studies show that the underlying cell damage continues throughout the attenuation process. In addition, attenuation may alter the normal distribution of O_3 within the lung, allowing more O_3 to reach sensitive regions, possibly affecting normal lung defenses (e.g., PMN influx in response to inhaled microorganisms).

3. What are the effects of long-term exposures to ozone?

Available data indicate that exposure to O_3 for months and years causes structural changes in several regions of the respiratory tract, but effects may be of the greatest importance in the centriacinar regions (where the alveoli and conducting airways meet); this region typically is affected in most chronic airway diseases of the human lung. This information on O_3 effects in the distal lung is extrapolated from animal toxicological studies because, to date, comparable data are not available from humans. The apparent lack of reversal of effects during periods of clean air exposure raises concern that seasonal exposures may have a cumulative impact over many years. The role of adaptive processes in this response is unknown but may be critically dependent on the temporal frequency or profile of exposure. Furthermore, the interspecies diversity in apparent sensitivity to the chronic effects of O_3 is notable, with the rat representing the lower limit of response, and the monkey the upper limit. Epidemiological studies attempting

to associate chronic health effects in humans with long-term O_3 exposure provide only suggestive evidence that such a linkage exists.

Long-term exposure in the females of one strain of mice to high O_3 levels (1 ppm) caused a small, but statistically significant increase in lung tumors. There was no concentration-response relationship, and rats were not affected. Genotoxicity data are either negative or weak. Given the nature of the database, potential carcinogenicity in animals is uncertain. Ozone did not show tumor-promoting activity in a chronic rat study (at 0.5 ppm O_3).

4. What are the effects of binary pollutant mixtures containing ozone?

Combined data from laboratory animal and controlled human exposure studies of O_3 support the hypothesis that coexposure to pollutants, each at low-effect levels, may result in effects of significance. The data from human studies of O_3 in combination with NO_2, SO_2, H_2SO_4, HNO_3, or CO show no more than an additive response for lung spirometry or respiratory symptoms. The larger number of laboratory animal studies with O_3 in mixture with NO_2 and H_2SO_4 show that effects can be additive, synergistic, or even antagonistic, depending on the exposure regimen and the endpoint studied. This issue of exposure to copollutants remains poorly understood, especially with regard to potential chronic effects.

5. What population groups are at risk as a result of exposure to ozone?

Identification of population groups that may show increased sensitivity to O_3 is based on their (1) biological responses to O_3, (2) preexisting lung disease (e.g., asthma), (3) activity patterns, (4) personal exposure history, and (5) personal factors (e.g., age, nutritional status).

The predominant information on the health effects of O_3 noted above comes from clinical and field studies on healthy, nonsmoking, exercising subjects, 8 to 45 years of age. These studies demonstrate that, among this group, there is a large variation in sensitivity and responsiveness to O_3, with at least a 10-fold difference between the most and least responsive individuals. Individual sensitivity to O_3 also may vary throughout the year, related to seasonal variations in ambient O_3 exposure. The specific factors that contribute to this large intersubject variability, however, remain undefined. Although differences may be due to the dosimetry of O_3 in the respiratory tract, available data show little difference on O_3 deposition in the lungs for inhalation through the nose or mouth.

Daily life studies reporting an exacerbation of asthma and decrease in peak expiratory flow rates, particularly in asthmatic children, appear to support the controlled studies; however, those studies may be confounded by temperature, particle or aeroallergen exposure, and asthma severity of the subjects or their medication use. In addition, field studies of summertime daily hospital admissions for respiratory causes show a consistent relationship between asthma and ambient levels of O_3 in various locations in the Northeastern United States, even after controlling for independent contributing factors. Controlled studies on mild asthmatics suggest that they have similar lung volume responses but greater airway resistance changes to O_3 than nonasthmatics. Furthermore, limited data from studies of moderate asthmatics suggest that this group may have greater lung volume responses than nonasthmatics.

Other population groups with preexisting limitations in pulmonary function and exercise capacity (e.g., chronic obstructive pulmonary disease, chronic bronchitis, ischemic heart disease) would be of primary concern in evaluating the health effects of O_3. Unfortunately, not enough is known about the responses of these individuals to make definitive conclusions regarding their relative responsiveness to O_3. Indeed, functional effects in these individuals with reduced lung function may have greater clinical significance than comparable changes in healthy individuals.

Currently available data on personal factors or personal exposure history known or suspected of influencing responses to O_3 follow.

- *Human studies have identified a decrease in pulmonary function responsiveness to O_3 with increasing age, although symptom rates remain similar. Toxicological studies are not easily interpreted but suggest that young animals are not more responsive than adults.*
- *Available toxicological and human data have not conclusively demonstrated that males and females respond differently to O_3. If gender differences exist for lung function responsiveness to O_3, they are not based on differences in baseline pulmonary function.*
- *Data are not adequate to determine whether any ethnic or racial group has a different distribution of responsiveness to O_3. In particular, the responses of nonwhite asthmatics have not been investigated.*
- *Information derived from O_3 exposure of smokers is limited. The general trend is that smokers are less responsive than nonsmokers. This reduced responsiveness may wane after smoking cessation.*

- *Although nutritional status (e.g., vitamin E deficiency) makes laboratory rats more susceptible to O_3-induced effects, it is not clear if vitamin E supplementation has an effect in human populations. Such supplementation has no or minimal effects in animals. The role of such antioxidant vitamins in O_3 responsiveness, especially their deficiency, has not been well studied.*

Based on information presented in this document, the population groups that have demonstrated increased responsiveness to ambient concentrations of O_3 consist of exercising, healthy and asthmatic individuals, including children, adolescents, and adults.

Since the 1996 O_3 AQCD evaluations, a distinctly more extensive database of air pollution epidemiologic studies has become available. A subset of these studies which examined O_3 health effects have reported a variety of O_3-related health effects associations. Based on the physiological, biochemical and molecular changes observed in controlled human exposure studies and animal toxicological studies, new evidence is now available by which to evaluate the biological plausibility and extent of coherence for various health outcomes (such as respiratory and cardiovascular effects, fetal and infant development effects, and mortality) reported in the epidemiologic studies as discussed in ensuing sections. Biological observations pointing towards putative mechanisms of action in developing hypotheses to interpret associated pathological symptoms reported in epidemiologic studies are also critically evaluated in subsequent sections, as are in vitro and in vivo experimental studies using novel molecular technologies to address potential mechanisms of action.

8.4.2 Assessment and Integration of New Experimental Evidence

8.4.2.1 Background on Cross-Cutting Issues

Discussion of several cross-cutting issues that will facilitate a clearer understanding of the ensuing assessment is provided here to enhance an integrated and comprehensive understanding of the experimental and epidemiologic studies on O_3 health effects. An important issue to be considered is the extrapolation of observed effects in animals to humans, from the perspective of dosimetry and the strength and weaknesses of such extrapolation models. The most challenging issue is the integration of (a) epidemiologic (observational) findings that suggest a potential causative role of ambient O_3 (with adjustments for other copollutants) in producing

health effects with (b) physiological, biochemical and toxicological findings from experimental studies.

8.4.2.2 Approaches to Experimental Evaluation of Ozone Health Effects

Three chapters in the current document provide detailed discussion of various experimental approaches utilized to evaluate O_3-related health effects. Chapter 4 discusses dosimetry issues pertinent to both animal and human exposure scenarios. Chapter 5 discusses the experimental studies of physiological, biochemical (cellular and molecular changes) and pathological observations in laboratory animals (including nonhuman primates, dogs, and rodent species) and in vitro studies using cell culture systems (in certain cases, on humans cells recovered from BALF postexposure to O_3). Chapter 6 evaluates studies on human volunteers exposed to O_3 which have investigated a variety of physiological and biochemical endpoints.

In interpreting the results from the experimental approaches, one must consider the following three issues: (1) exposure/dose considerations; (2) role of confounders; and (3) interpretation of results from high dose exposures and animal to human extrapolations. Earlier animal toxicology studies were carried out using relatively high O_3 exposure concentrations/doses that do not necessarily reflect "real-world" exposure scenarios. Those experiments were primarily aimed at understanding the pathophysiology associated with O_3 exposure in healthy animals, to help understand potential mechanisms(s) of action, and to help validate health outcomes reported in epidemiologic studies. Since the 1996 O_3 AQCD, the majority of human and animal studies have used ambient and/or near ambient doses. Earlier controlled chamber exposure studies on human volunteers mainly limited exposures to O_3 alone in comparison to sham (clean air) exposures, thus providing evidence concerning direct effects of O_3 per se versus more closely mimicking real-world atmospheric exposures to multipollutant mixes. Some newer air pollution clinical studies are utilizing various co-exposure regimens to simulate more closely ambient exposure to air pollution mixtures; and the results from these studies will be highly useful in developing better models to interpret the toxicological effects associated with O_3-containing ambient air pollutants mixes.

Interpretations of experimental studies of air pollution, as in the case of environmental comparative toxicology studies, are affected by limitations associated with animal extrapolation models. The differences between humans and rodents with regard to O_3 inhalability, absorption

and distribution profiles based on breathing pattern, exposure dose and differences in lung structure and anatomy (see Chapter 4 and 5 for details) have to be taken into consideration. Also, in spite of a high degree of homology and the existence of a high percentage of orthologous genes across human and rodents, particularly mice, extrapolation of molecular alterations at the gene level suffers from the regulatory control of various signaling units as simple as cis and trans activating transcription factor units. Given these molecular differences, extrapolation of physiological parameters (which are under the control of various biochemical, endocrine and neuronal controls) observed between human and rodents represents a difficult task.

8.4.2.3 Interspecies Comparison of Experimental Results: Dosimetric Considerations

In this section, a brief overview of the experimental results obtained from studies on human and laboratory animals are comparatively analyzed and presented to provide background for assessing biological plausibility and coherence discussed in detail in the following section. Each subsection starts off with an introduction to what was known at the time of the publication of the previous O_3 AQCD (U.S. Environmental Protection Agency, 1996), followed by discussion of new information.

Dosimetry Considerations

Dosimetric studies demonstrate fundamental relationships between ambient exposures and doses to target tissues. While experimental and theoretical dosimetry (modeling) studies of O_3 have proved to be valuable in the assessment of toxicity, they are most useful when conducted as part of an integrated approach to determining the distribution of inhaled O_3 along the upper and lower respiratory tract. Derivation of credible dosimetry estimates greatly facilitates the development of useful extrapolation models by which to compare doses and effects across species and subpopulations.

The state-of-the-art of O_3 dosimetry, as described in 1996 O_3 AQCD, indicated consistency across data and models derived from in vivo human and animal studies, thus increasing the level of confidence in the development of dosimetric extrapolation models. Earlier dosimetry models predicted that the tissue dose of inhaled O_3 was greatest at the bronchoalveolar junction, the region experimentally shown to be most impacted by O_3. Ozone bolus inhalation studies in

humans have indicated that inspired O_3 reaches the distal airways and alveoli of resting humans; and, with increased inspiratory flow rates due to exercise, O_3 penetrates deeper and in greater quantity to the distal regions of the lung. These findings have been corroborated by observations of $^{18}O_3$ (oxygen-18-labeled ozone) in the BALF of humans and rats (Hatch et al.,1994).

Some acute responses to O_3 have compared well across species when controlled for dose, indicating that animals and humans (a) respond to O_3 in a dose-dependent manner, i.e., they exhibit increasing breathing frequency with an accompanying decrease in tidal volume (tachypnea), and (b) show similar changes in alveolar permeability as measured by protein in the bronchoalveolar lavage fluid (BALF). These parallel changes in humans and animals were sufficiently homologous to suggest a common mode of action. It has also been recognized that O_3-induced spirometric changes, the hallmark of response in humans, also occur in exposed rats when hyperventilated with CO_2 stimulation. However, the effect of anesthesia in the rodent model in contrast to the awake human remains uncertain; but, as will be discussed, activity level differences between species appear to strongly influence dose. Nevertheless, most lung function decrements subside with repeated exposures in both humans and animals, with analogous attenuation of certain (but not all) parameters measured in the BALF. The mechanisms associated with attenuation are unclear but may involve endogenous antioxidants. The significance of non-attenuated markers in BALF has been interpreted to relate to potential chronicity of O_3 effects. Studies on long-term exposure in monkeys and rats do show long term changes in the distal lung that appear to be represented by a near-linear dose-response pattern. More thorough analysis of this dose response is needed, however.

As discussed in the 1996 O_3 AQCD, Hatch et al. (1994) compared responses of exercising humans (15-min intervals of rest and exercise at 60 L/min for 2 h) to those of resting rats also exposed to 0.4 ppm $^{18}O_3$ (oxygen-18-labeled ozone) for 2 h. They observed 4 or 5 times the $^{18}O_3$ dose (as adduct) in BALF constituents of humans as compared to those of F344 male rats. This 4- to 5-fold difference appeared to be due to the exercise-stimulated hyperventilation of the humans when compared to the rats. *Only when the resting rats were exposed to 2 ppm O_3 for 2 h at rest did the $^{18}O_3$ labeling of BALF constituents and indices of effect (i.e., BAL cells and protein at 24 h) compare favorably with those of the exercising humans exposed to 0.4 ppm $^{18}O_3$ for 2 h with intermittent exercise.* Thus, the rat and human appear to have similar sensitivities to O_3 when exercise is taken into account as a dose modifier. It was further concluded in the

1996 O_3 AQCD that attempts to compare resting animal data to exercising human data obtained at similar O_3 concentrations would likely underestimate the dose to the lung and, presumably, the resultant risk of effect.

In the past decade, no further reports have been published on O_3 uptake studies in animals, although several controlled human bolus and/or general O_3 uptake studies have provided refined data. The bolus uptake studies suggest that prior exposure to O_3 diminishes bolus uptake. In the earlier document, the effect of mode of breathing (oral or nasal) on O_3 uptake was thought to be minimal, with approximately equal uptake via the nose or mouth. Newer bolus dose studies have demonstrated that the uptake and regional respiratory tract distribution of O_3 is sensitive to mode of breathing (nasal uptake greater than oral) and to air flow rate (uptake decreases with increasing flow). Similarly, the change in breathing with exercise vs rest causes a shift in regional O_3 distribution, allowing deeper respiratory tract penetration, with resultant greater dose and damage to respiratory bronchiolar and alveolar tissues (as predicted by the models described in the 1996 O_3 AQCD).

The efficiency of O_3 uptake is chemically rate dependent. The resultant reaction products (hydrogen peroxide, aldehydes, and hydroxyhydroperoxides) created by ozonolysis of lipids in airway and epithelial lining fluid are thought to mediate O_3 toxicity. The dependence of O_3 absorption on chemical-reaction rates is consistent with the observation of Bush et al. (2001), that the rate of O_3 uptake is lower than for Cl_2 despite the similar gas-phase diffusion coefficients of these two gases. The slower uptake rate of O_3 relative to Cl_2 appears due to the limiting reaction rate of O_3 in the epithelial lining fluid. The work by Rigas et al. (1997) using the O_3 bolus technique in humans, showing uptake to be increased by continuous exposure to NO_2 and SO_2 and decreased by continuous O_3 exposure, suggests an important role for copollutant exposures. Thus, an inflammatory response may magnify the production of O_3-reactive substrates in the epithelial lining fluid when other oxidants are present.

New uptake studies (Ultman et al., 2004) carried out in controlled human clinical studies have observed gender-specific differences in the uptake of O_3, but these differences do not correlate well with spirometric responses. Rather, they appear to be related to breathing pattern and lung size, with females having smaller lungs than males. Other uptake studies carried out in humans using environmentally relevant O_3 concentrations have demonstrated the significance of incorporating inter-subject variability in dose-response relationship prediction and extrapolation.

Thus, a number of variables seem to have a degree of impact on O_3 uptake, notably including age, route of breathing, breathing pattern, gender and certain pre-exposure conditions. These differences are important in order to interrelate biological effect and risk assessment estimates.

The general consistency observed in O_3 uptake in animal and human experimental exposure studies provides increased confidence in the use of theoretical dosimetry modeling and the use of animal toxicological data (see Chapter 4 for detailed discussion). Models have taken into consideration various factors such as age, as well as anatomical, physiological, and biochemical alterations. Incorporation of novel information, such as (a) the identification of primary site of acute cell injury, (b) the site of O_3 reaction/diffusion in the epithelial lining fluid, (c) the roles of intermediate reactive oxygen species (ROS) and lipid ozonation products in oxidative injury, and (d) the roles of metabolic enzyme profiles in developing lung tissue, can be expected to lead to refined novel models and better extrapolation.

8.4.2.4 Critical Analysis of Toxicological Effects of O_3 Exposure

In the following subsections, research results generated from experimental studies on humans and animals during the past decade are assessed (keeping in view the interspecies differences discussed in the preceding section) in evaluating experimental evidence for biological plausibility and coherence for O_3 health effects discussed in the later sections.

8.4.2.4.1 Pulmonary Function

A number of controlled human exposure, animal, and epidemiological studies assessed in the 1996 O_3 AQCD demonstrated alterations in various measurements of pulmonary function. Inhalation of O_3 for several hours while physically active elicits both acute pathophysiologic changes and subjective respiratory tract symptoms. The pulmonary responses observed in healthy human subjects exposed to ambient O_3 concentrations include decreased inspiratory capacity; mild bronchoconstriction; rapid, shallow breathing pattern during exercise; and subjective symptoms of tracheobronchial airway irritation, including cough and pain during inspiration. Acute O_3 exposures also cause decreases in forced vital capacity (FVC), forced expiratory volume in 1 s (FEV_1), and increased airways resistance (SR_{aw}). The severity of symptoms and the magnitude of response depends on inhaled dose, individual O_3 sensitivity, and the extent of tolerance resulting from previous exposures.

A progressive decrease in tidal volume and a "compensatory" increase in frequency of breathing to maintain steady minute ventilation during exposure suggests a direct modulation of ventilatory control. These changes in humans parallel responses of many animal species exposed to O_3 and other lower airway irritants (Tepper et al., 1990). Pulmonary function evaluations carried out in several animal species on acute exposure to O_3 generally show responses similar to those observed in humans, such as increased breathing frequency, decreased tidal volume, increased resistance and decreased FVC. These effects are observed at relatively low O_3 concentrations (0.25 to 0.4 ppm) following several hours of exposure in many species. The alterations in breathing pattern return to normal within hours after exposure and the pattern of attenuation in responses following repeated exposures is similar to that observed in humans. When rats were exposed to concentrations $\geq$1 ppm even breathing mechanics were found to be affected.

The time course of spirometry responses to O_3 exposure depends on the specific exposure conditions. Early controlled human studies reviewed in the 1986 and 1996 O_3 AQCD typically reported statistically significant pulmonary responses in exercising (intermittent or continuous) subjects exposed for 2 h to a concentration in the range of 0.12 to 0.4 ppm O_3 (mimicking midday ambient O_3 peaks reported in Los Angeles, CA). Significant effects were not observed following 2 h exposures in sedentary subjects below 0.5 ppm O_3. Some later human studies reviewed in the 1996 O_3 AQCD utilized 6-8 h exposures with exercise in order to better mimick longer exposures to ambient O_3 (recognizing the more prolonged elevated ambient O_3 levels often observed in some urban areas in the northeastern states) and provided some of the strongest and most quantifiable concentration-response data on the acute health effects of O_3 based on pulmonary function tests.

All evaluations have indicated that there exists considerable interindividual differences in the magnitude of responses to O_3. However, an individual's lung function and to a lesser extent, respiratory symptom responses to O_3 are reproducible over a period of time, indicating that some individuals are consistently more responsive than others to O_3. Figure 8-1 illustrates the variability in FEV_1 responses in young healthy adults following a prolonged (6.6 h) exposure to O_3, as summarized in the 1996 O_3 AQCD. Referring to this figure, the average FEV_1 response following exposure to 0.08 ppm O_3 is small (between a 5 and 10% decrement). However, ~18% of the exposed subjects had moderate FEV_1 decrements of 10 to 20% and ~8% experienced

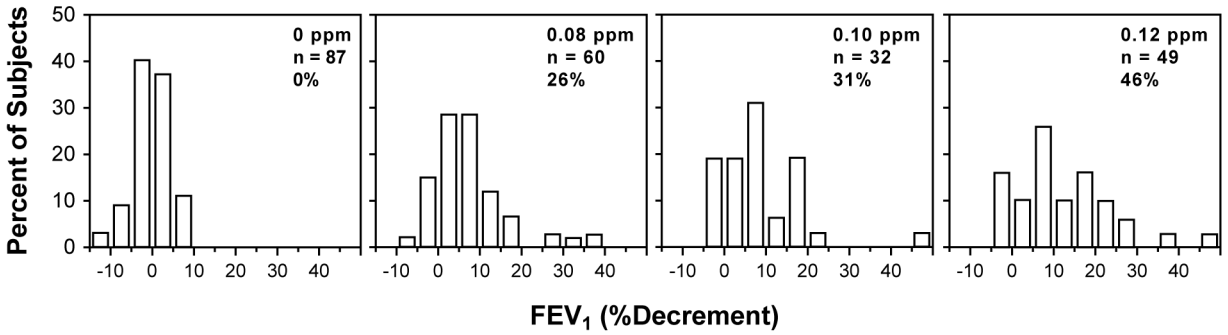


Figure 8-1. Frequency distributions of FEV_1 decrements following 6.6-h exposures to O_3 or filtered air. During each hour of the exposures, subjects were engaged in moderate exercise for 50 minutes. With increasing O_3 concentration, the distribution of responses becomes asymmetric, with a few individuals exhibiting large FEV_1 decrements. The percentage in each panel indicates the portion of subjects having a FEV_1 decrement in excess of 10%.

Source: McDonnell (1996).

large FEV_1 decrements of greater than 20%. This serves to emphasize that while average responses may be small and seem physiologically insignificant, some individuals typically experience distinctly more severe effects. As a further example of intersubject variability, Figure 8-2 illustrates the portion of young healthy adult males (24 yr old) predicted to have FEV_1 decrements of greater than 5, 10, and 15% when exposed to O_3 during moderate exercise, as also presented in the 1996 O_3 AQCD.

New studies (assessed in Chapter 6 and Annex 6 of this document) which evaluated responses in hundreds of subjects clearly indicate that FEV_1 decrements and symptom responses decrease with age beyond young adulthood (18 to 20 years). Hazucha et al. (2003), for example, examined gender and age differences in O_3 responsiveness and found that young females lose O_3 sensitivity faster than young males, but the rate is about the same for both genders by middle age (see Figure 8-3).

The development of effects is time-dependent during both exposure and recovery periods, with considerable overlap of evolving and receding effects. In healthy human subjects exposed to typical ambient concentrations (i.e., <0.2 ppm O_3), spirometric responses largely resolve within a few hours (4 to 6 h) postexposure; but cellular effects persist for longer periods (~24 h). Persisting small residual lung function effects are almost completely resolved within 24 hours.

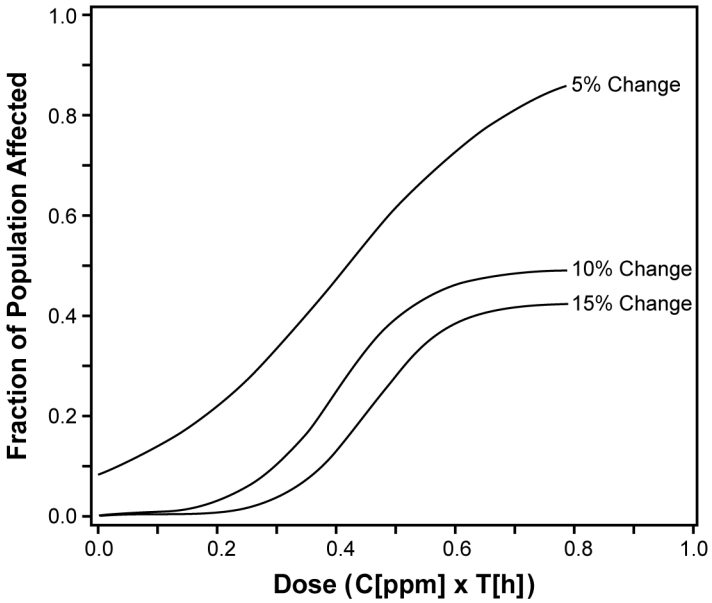


Figure 8-2. Proportion of moderately exercising healthy adults (24 yrs old) predicted to have 5, 10, or 15% decrements in FEV_1 as a function of concentration (0 to 0.12 ppm O_3) times exposure duration (1 to 6.6 h).

Source: McDonnell et al. (1995).

In hyperresponsive individuals, the recovery takes longer (as much as 48 h) to return to baseline values. The majority of these responses are attenuated after repeated exposure, but such tolerance to O_3 is lost within a week postexposure. The biochemical indicators of lung injury and associated morphological changes were not found to be attenuated in the majority of laboratory animals. Unfortunately, no data are available on pulmonary function changes in animals upon chronic exposure to O_3. However, earlier work of repeated exposure of rats to an episodic profile of O_3 demonstrated small but significant decrements in lung function that were consistent with early indicators of focal fibrinogenesis in the proximal alveolar region.

In the 1996 O_3 AQCD, O_3-induced decrease in inspiratory capacity was hypothesized to be the result of neurogenic inhibition of maximal inspiration due to stimulation of C-fiber afferents either directly or from O_3-induced inflammatory mediators. Earlier human studies (Coleridge et al., 1993; Hazucha and Sant'Ambrogio, 1993) reported a role for bronchial C-fibers and rapidly adapting receptors as primary vagal afferents responsible for O_3- induced changes in ventilatory rate and depth. As discussed in Chapter 6, the newer results of Passannante et al.

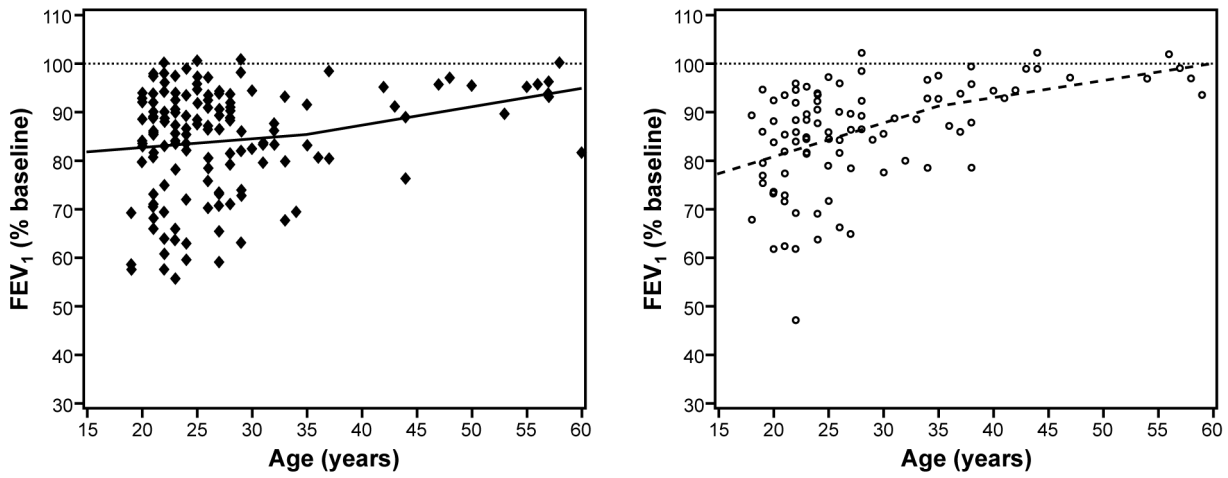


Figure 8-3. Effect of age on FEV_1 responses to O_3 exposure (0.42 ppm for 1.5 h with intermittent exercise). Left and right panels are data for males (n = 146; 19 to 60 yrs old) and females (n = 94; 18 to 59 yrs old), respectively. On average, FEV_1 responses to O_3 exposure decrease with increasing age. However, there is a large amount of intersubject variability in responses, e.g., responses of 20 to 25 year olds range from a small increase to greater than a 50% decrement in FEV_1 following O_3 exposure.

Source: Adapted from Hazucha et al. (2003).

(1998) also support C-fiber stimulation as a primary mechanism of the O_3-induced reduction in inspiratory capacity and suggest a role for nociceptive mechanisms. This neurogenic mechanism also likely has an effect on airway responsiveness and lung inflammation.

Lung function changes due to O_3 exposure have been evaluated in patients with preexisting respiratory diseases under experimental controlled exposure regimens, with or without physical exertion in the form of intermittent exercise. These new studies found minimal O_3-induced effects in COPD patients. For example, Gong et al. (1997a) exposed nine COPD patients (0.24 ppm O_3 for 4 h with intermittent exercise) and observed a nonsignificant FEV_1 decrement of −8% in COPD patients that was not statistically different from the −3% decrement seen in healthy subjects. Augmenting observations discussed in the 1996 O_3 AQCD, newer studies of asthmatics (see Chapter 6) continue to indicate that pulmonary function deficiencies detected by spirometric analyses are somewhat increased relative to healthy controls. A tendency for increased O_3-induced pulmonary function responses were reported in asthmatics relative to

healthy subjects exposed to O_3 concentrations of ≤0.2 ppm for 4- 8 h duration (Scannell et al., 1996). Similarly, Alexis et al. (2000) observed statistically significant O_3-induced decreases in FEV_1 in mild atopic asthmatics that tended to be greater than experienced by healthy control subjects. In a longer exposure duration (7.6 h) study, Horstman et al. (1995) reported that mild-to-moderate asthmatics exposed to 0.16 ppm O_3 had FEV_1 decrements that were significantly greater than in healthy subjects (19% versus 10% respectively). Moreover, Horstman et al. (1995) found that responses of asthmatics were more severe in patients with lower baseline lung function. Though most controlled human exposure studies may not provide the required statistical power (due to the limited number of subjects compared to panel or field studies), they do suggest that asthmatics are at least as sensitive, if not more, than healthy subjects.

In addition to effects of O_3 exposure on the large airways as indicated by spirometric responses, O_3 exposure also affects the function of the small airways and parenchymal lung. Studies reported by Foster et al. (1993, 1997) that examined the effect of O_3 on ventilation distribution in healthy adult males suggest a prolonged O_3 effect on the small airways and ventilation distribution in some individuals. Animal toxicology studies have shown the centriacinar region (CAR) of the lung (the segment between the last conducting airway and the gas exchange region) to be a region highly susceptible to O_3-induced damage (epithelial cell necrosis and remodeling of respiratory bronchioles) and seem to be reasonably predictive of similar morphological changes as being likely to occur in humans. Unfortunately, common pulmonary function tests do not measure acute changes in the small airways of the CAR. Identification of acute effects of O_3 in small airways, if any, would lend additional support for concerns about long-term effects of repeated O_3 exposures.

8.4.2.4.2 Airway Responsiveness

Increased airway responsiveness, also referred to as airway hyperresponsiveness (AHR) or bronchial hyperreactivity, is an indicator of enhanced reactivity of airways to bronchoconstriction induced by a variety of stimuli (exposure to cold air, allergens or exercise). AHR is assessed by airway function (either spirometry or plethysmography) after inhalation exposure to specific (antigen, allergen) or nonspecific (methacholine, histamine) bronchoconstrictor stimuli. It was recognized in the 1996 O_3 AQCD that exposure to O_3 to induce AHR in humans usually resolves in 18-24 h after exposure in a majority of subjects, but

may persist in some individuals for longer periods. Gong et al. (1997b) found that subjects with asthma developed tolerance to repeated O_3 exposures in a manner similar to normal subjects; however, there were more persistent effects of O_3 on airway responsiveness, which only partially attenuated when compared to filtered air controls. Such an occurrence and duration of increased nonspecific airway responsiveness following O_3 exposure could have clinical implications in asthmatics, possibly putting them at potential increased risk for more prolonged bouts of bronchoconstriction in response to various triggering stimuli (e.g., allergens, cold air, etc.).

Studies examining the effects of O_3 on exacerbations of antigen-induced asthma suggested that allergen-specific increased airway responsiveness indeed occurs in mild asthmatics upon exposure to O_3. Jörres et al. (1996) confirmed that higher O_3 concentrations cause increased airway reactivity to specific antigens in subjects with mild allergic asthma and, to a lesser extent, in subjects with allergic rhinitis, after exposure to 0.25 ppm O_3 for 3 h. This enhancement of allergen responsiveness after O_3 exposure appears to be time dependent, suggesting that the timing of allergen challenge in O_3-exposed subjects with allergic asthma is important. Significant, clinically relevant decreases in pulmonary function have been observed in the early phase allergen response in subjects with rhinitis after consecutive (4-day) exposure to 0.125 ppm O_3 (Holz et al. 2002). Similar increased airway responsiveness to house dust mite antigen 16-18 h postexposure to a single dose of O_3 (0.16 ppm for 7.6 h) was also observed in asthmatics. These observations suggest that O_3 exposure may be a clinically important factor that can exacerbate the response to ambient bronchoconstrictor substances in individuals with preexisting allergic asthma and that its influence may be both immediate and persist for relatively long periods of time.

An extensive laboratory animal study database (using rats, mice, guinea pigs, and rabbits), exploring the effects of acute, long-term, and repeated exposures to O_3, indicates that induction of AHR occurs at relatively high O_3 concentrations. These studies provide clues to the roles of physiological and biochemical components involved in this process, but one has to exercise caution in the interpretation of these results, as different mechanisms may be involved in mediating high-dose and low-dose responses. Some of these studies indicated differences in O_3-induced AHR between immature and adult rats and also between obese and lean mice strains. Some of the ex-vivo studies carried out in New Zealand white rabbits using environmentally relevant O_3 concentrations indicated O_3-induced alterations in tracheal epithelial

functions and potential O_3-induced direct vascular constriction. As observed in humans, the acute changes in AHR do not persist upon long-term exposure in animals exposed to near-ambient concentrations of O_3; and attenuation has been observed. Both human and animal studies indicate that airway responses are not associated with inflammation, but they do suggest a likely role for neuronal involvement.

8.4.2.4.3 Morphological and Biochemical Abnormalities

Most of the research results alluded to the ensuing discussion come from toxicology studies using various laboratory animal species that were usually exposed to relatively high, non-ambient concentrations of O_3. However, these exploratory and mechanistic studies may provide important and useful hypotheses to consider in integrating various health outcomes observed or predicted by epidemiologic studies. A limited number of controlled human exposure studies evaluated cellular and biochemical parameters in the BALF. These studies have yielded limited evidence supporting the observations made in animal toxicology studies. Keeping in view the species- specific differences, the morphological and biochemical alterations in humans and animals are integrated in the following paragraphs to develop working hypotheses to interpret human health outcomes.

Lung Injury and Morphological Changes

The 1996 O_3 AQCD stated that short-term O_3 exposure causes similar types of alterations in lung morphology in all laboratory animal species studied, including primates. The cells in the CAR have been recognized as a primary target, possibly because it receives the greatest dose of O_3 delivered to the lower respiratory tract. The ciliated cells in the nasal cavity and airways and Type I epithelial cells in the gas-exchange region are also identified as targets. Differences in the distribution of antioxidants in the CAR of the lung were responsible for the differences in injury and morphological changes observed between nonhuman primates and rodents. Though acute O_3 exposure induces structural changes such as fibrosis in the CAR, these structural alterations appear to be partially transient, with recovery shortly postexposure; but the time for recovery is dependent on species and the dose of O_3.

New studies reviewed in the 1996 O_3 AQCD of lung morphological changes or damage due to long-term or prolonged exposure to O_3 found chronic lesions similar to early lesions of

respiratory bronchiolitis, which have the potential to progress to fibrotic lung disease. Some of the morphological changes associated with long-term exposures, such as increases in hyperplastic epithelial cells, appear to reverse following cessation of O_3 exposure. However, in the underlying interstitium of the CAR, proliferation of fibroblasts creates excess noncellular matrices. These processes are only partially reversible and may progress following cessation of exposure. This suggests initiation of focal interstitial fibrosis, which can progress to chronic degenerative lung disease. Another important observation reported in the 1996 O_3 AQCD was that of greater injury observed in the monkey's lung upon intermittent exposure (simulated ambient) compared to continuous exposure, suggesting a role for loss of tolerance in this process.

Reports of morphological changes following chronic O_3 exposures in animal studies (rodents and primates) published since the 1996 AQCD allude to the earlier findings assessed in that document. In rats, the effects of chronic ~0.5 ppm O_3 exposure included mucous cell metaplasia, hyperplasia of the nasal epithelium, increased mucosubstances, and increased Bcl-2 protein levels. In mice, lifetime exposures of 0.5 ppm O_3 were linked to similar outcomes. Taken together, the rodent studies suggest that O_3 exposure may have the potential to induce similar long-lasting alterations in human airways. A series of new studies that utilized infant rhesus monkeys and simulated seasonal ambient exposure (0.5 ppm 8 h/day for 5 days, every 14 days for 11 episodes) reported remodeling in the distal airways; abnormalities in tracheal basement membrane; eosinophil accumulation in conducting airways; and decrements in airway innervation, again confirming the potential greater injury due to seasonal exposure compared to continuous exposure alluded to in the 1996 O_3 AQCD.

One epidemiologic report by Sherwin et al. (2000) compared results for autopsy of the lungs of Los Angeles and Miami residents and observed a significantly greater extent and severity of centriacinar region alterations in the lungs of Los Angeles residents independent of a smoking effect. These results suggest that the severity of CAR alterations may be related to the higher O_3 levels in Los Angeles. Similar observations of CAR thickening and deposition of collagen seen with chronic O_3 exposure in rat also suggest progressive structural lung injury that can evolve into a more chronic form, such as fibrosis. Again, however, one must be cautious in extrapolating these laboratory animal observations to humans, given the exposure regimens and doses used.

Lung Inflammation and Permeability

The 1996 O_3 AQCD recognized respiratory tract inflammation and increased cellular permeability as two important biological markers of ozone exposure in both animals and humans. These distinct, independent biological events have been observed in all species studied in response to acute exposure to O_3. Increased epithelial permeability and inflammation in the lower respiratory tract are measured by increases in bronchoalveolar lavage fluid (BALF) protein and/or albumin and neutrophils (PMNs), respectively. Nasal lavage (NL) fluid and cells from O_3-exposed humans were used to assess the inflammatory and permeability changes in the upper respiratory tract. Structural changes in nasal mucosa have been demonstrated after O_3 exposure in animals and humans. The presence of PMNs in the lung has long been accepted as a hallmark of inflammation and as an important indicator that O_3 causes inflammation in the lungs. Importantly, respiratory tract inflammation may lead to significant health effects, including impaired host defenses and irreversible structural alterations (as discussed earlier). Ozone-induced mucous membrane cell metaplasia observed in rodents appears to be mediated by inflammation.

Laboratory animals exhibit varying degrees of sensitivity to O_3 exposure (see Chapter 5 for detailed discussion); and this is evident even for the induction of pulmonary inflammation and permeability. Newer animal toxicology studies on O_3-induced inflammation reviewed in Chapter 5 indicate that the lowest ozone concentration that had an effect on mouse lung inflammation was also 0.11 ppm for 24 hours. Shorter durations (8 h) required greater concentrations of ozone (0.26 ppm) for effects on epithelial permeability but had no effect on inflammation. The lowest concentration of ozone that had an effect on epithelial permeability or inflammation in the rat was 0.5 ppm for 3 hours. Subchronic exposures in animals suggest that permeability changes are transient (and species-dependent) and return to control levels even with continuing exposure. Chronic animal O_3 exposure studies suggest a role for persistent inflammation in O_3-induced alterations in lung structure and function. Significant remodeling of epithelium and underlying connective tissues in distal airways have been reported in rat exposed to 0.25 ppm O_3 (12h/day for 6 wk) and in monkeys exposed to 0.2 ppm O_3 (8h/day for 90d). Various factors such as viral infection, chemotactants and oxidized matrix fragments are also implicated in the establishment and persistence of O_3-induced inflammation.

A number of controlled human exposure studies reviewed in the 1996 O_3 AQCD clearly indicated that a single acute exposure (1-4 h) of humans to moderate O_3 concentrations (0.2-0.6 ppm) while exercising at moderate to heavy levels results in a number of cellular and biochemical changes suggesting pulmonary inflammation and increased lung permeability. Both the inflammatory response and increased lung permeability have been observed as early as 1 h and persisted for at least 18 h. Devlin et al. (1991) reported these changes (increased neutrophils, inflammatory mediators such as PGE_2 and IL-6) to occur in humans exposed to 0.08 to 0.12 ppm O_3 with moderate exercise for 6.6 h. The newer studies reviewed in this document (see Chapter 6 for details) have provided additional information on three different aspects of O_3-induced inflammatory responses, such as (1) intersubject variability; (2) differential attenuation profile for various inflammatory markers; and (3) effects of repeated exposures.

Mean changes in inflammatory markers seen with exposure to ambient levels of O_3 (Devlin et al., 1991) exhibited interindividual differences; and, in some individuals, the changes were comparable to those observed in subjects exposed to 0.4 ppm (as reported by Koren et al., 1989), suggesting that some individuals in the population may be quite sensitive at ambient levels of O_3. Mudway and Kelly (2004) examined O_3-induced inflammatory responses (PMN influx) and altered epithelial permeability (protein leakage) via a meta-analysis of 21 controlled human exposure studies. Their analysis of PMN responses is illustrated in Figure 8-4. Tentatively, Mudway and Kelly (2004) suggested that the O_3 dose predicted to produce an average PMN influx exceeding the 95% confidence interval for PMN levels following filtered air (FA) exposures may, in essence, represent a threshold dose. For a 1 h exposure to 0.12 ppm O_3, the threshold dose for early phase PMN responses would not be exceeded unless an individual was engaged in very heavy exercise (V_E = 90 L/min). However, a longer 8 h exposure to 0.08 ppm O_3 could reach the early phase PMN dose threshold during relatively light sustained activity (V_E = 17 L/min). For these same 1- and 8-h exposure scenarios, BALF protein levels would be predicted to increase by about 1.1-fold. Regarding late phase PMN responses, their threshold dose was 26% greater than the early phase responses. Mudway and Kelly (2004) did note that their "threshold" doses were for average PMN responses in healthy adults and that some individuals would respond at lower doses. Indeed, Krishna et al. (1998) and Stenfors et al. (2002) observed significant early and late phase PMN responses, respectively, at doses below the levels tentatively referred to as threshold doses by Mudway and Kelly (2004). Additionally,

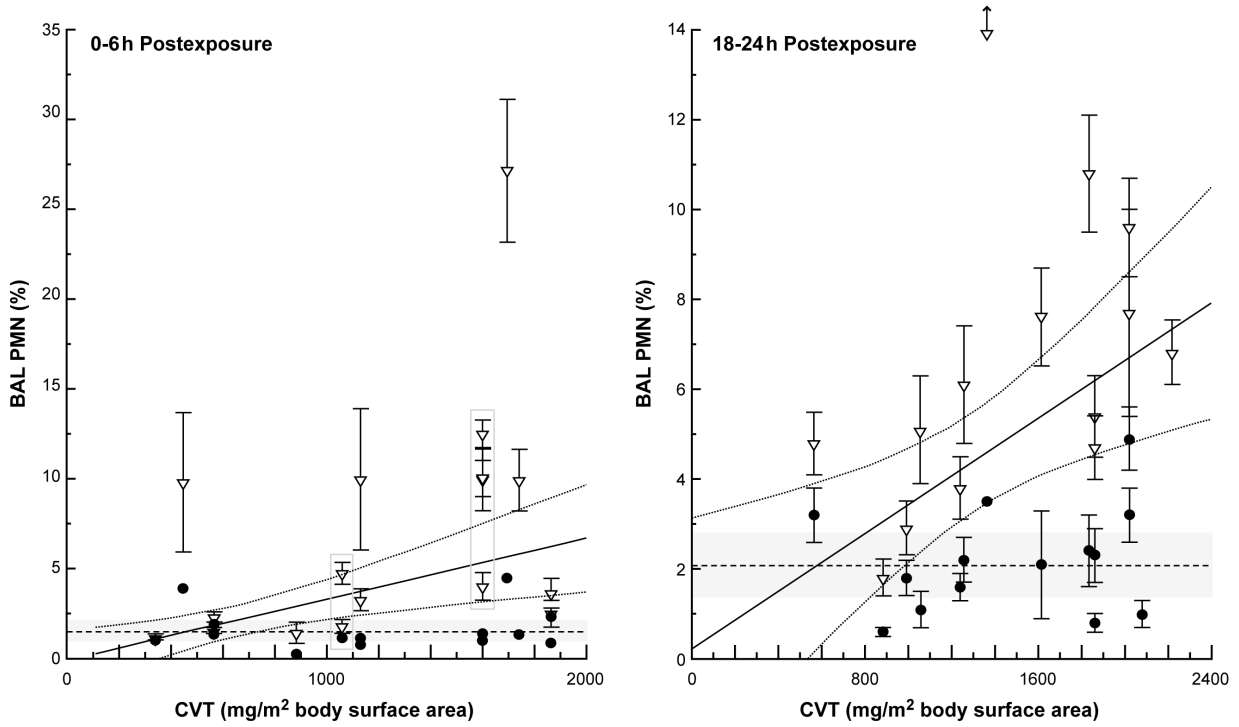


Figure 8-4. Neutrophilia response in the distal airways postexposure (PE) to O_3 or filtered air. Early (0-6 h PE) and delayed (18-24 h PE) responses illustrated in the left and right panel, respectively. Inhaled dose (CVT) is the product of O_3 concentration, minute ventilation per body surface area, and exposure duration. Data are mean (bars are standard errors) of neutrophils (% in BAL) after ozone (▽) or air (●) from 21 studies where subjects (18-40 yrs of age) were exposed to between 0.08 and 0.6 ppm O_3 for 1 to 6.6 h. The dashed lines (- - -) are average percent neutrophils following air exposures and the shaded area is the 95% confidence interval (CI). Solid lines (—) illustrate the linear relationship between neutrophil response and O_3 dose with a 95% CI illustrated by dotted lines (····).

Source: Adapted from Mudway and Kelly (2004).

significant inflammatory responses to O_3 exposures that did not elicit significant spirometric responses have been reported (Holz et al., 2005; McBride et al., 1994).

Soluble mediators of inflammation (e.g., the cytokines IL-6 and IL-8) as well as arachidonic acid metabolites (e.g., PGE_2, $PGF_{2\alpha}$, thromboxane, and leukotrienes [LTs] such as LTB_4) have been measured in the BAL fluid of humans exposed to O_3. In addition to their role in inflammation, many of these compounds have bronchoconstrictive properties and may be

involved in increased airway responsiveness following O_3 exposure. The time course for the inflammatory responses (including recruitment of neutrophils and other soluble mediators) is not clearly established, but a differential attenuation profile for many of these parameters is evident from the meta-analysis of 21 controlled human exposure studies reviewed by Mudway and Kelly (2004).

Repeated exposures in humans also indicate ongoing cellular damage irrespective of attenuation of the inflammatory responses and lung function (Devlin et al. 1997; Jörres et al. 2000). Devlin et al. (1997) examined the inflammatory responses of humans repeatedly exposed to 0.4 ppm O_3 for 5 consecutive days. Several indicators of inflammation (e.g., PMN influx, IL-6, PGE_2, BAL protein, fibronectin) were attenuated after 5 days of exposure (i.e., values were not different from FA). Several other markers (LDH, IL-8, total protein, epithelial cells) did not show attenuation, indicating that tissue damage probably continues to occur during repeated exposure. The recovery of the inflammatory response occurred for some markers after 10 days, but some responses did not return to normal even after 20 days. When re-exposed 2 weeks later, changes in BALF indicated that epithelial cells appeared to be fully repaired (Devlin et al., 1997). Kopp et al. (1999) observed inflammatory responses only after the first O_3 peak in summer; and its absence late in summer (even after exposure to higher levels of O_3) may be due to attenuation of the inflammatory response in the subjects.

Numerous studies reported acute O_3-induced changes in lung epithelial permeability assessed by indirect assay (increased levels of albumin and protein in BALF). Few other studies demonstrated O_3-induced epithelial cell permeability through direct assessment of clearance of ^{99m}Tc-DTPA (technetium-99m labeled diethylene triamine pentaacetic acid). For example, Kehrl et al. (1987) showed increased ^{99m}Tc-DTPA clearance in healthy young adults at 75 minutes postexposure to 0.4 ppm O_3 for 2 h. More recently, Foster and Stetkiewicz (1996) have shown that increased ^{99m}Tc-DTPA clearance persists for at least 18-20 h post-O_3 exposure (130 min to average O_3 concentration of 0.24 ppm), and the effect is greater at the lung apices than at the base.

Interaction of O_3 with the constituents of the extracellular lining fluid and the induction of oxidative stress is implicated in injury and inflammation. Animal toxicology and human in vitro studies that evaluated biochemical mediators implicated in injury and inflammation found alterations in the expression of cytokines, chemokines, and adhesion molecules, indicative of an

ongoing active stress response as well as injury repair and regeneration processes. Both animal and human studies indicate cellular and biochemical changes associated with inflammation and increased permeability, but the relationship between these changes and their role in lung function and airway responses is not known.

Host Defense

Evidence for O_3-induced dysfunction of host defense components and for subsequent enhanced susceptibility to bacterial lung infection stems from studies carried out in laboratory animals. Acute exposures of 0.08 ppm (3 h) O_3 have been implicated in the mortality of mice due to Streptococcal bacterial infection. Changes in antibacterial defenses are dependent on exposure regimens, species and strain of test animal, species of bacteria, and age of animal (with young mice being more susceptible to the effects of O_3, for example). Animal toxicology studies indicated that acute O_3-induced suppression of alveolar phagocytosis and immune functions observed in animals appeared to be transient and were attenuated with continuous or repeated exposures. A single study reviewed in the 1996 O_3 AQCD reported decrements in the ability of alveolar macrophages (AMs) to phagocytose microorganisms upon exposure to 0.08-0.1 ppm O_3 for 6.6 h (Devin et al., 1991). It has also been reported that O_3 exposures can interfere with AM-mediated clearance in the respiratory region of the lung and with mucociliary clearance of the tracheobronchial airways. Ozone-induced perturbations in the clearance process have been found to be dose-dependent, with low dose exposures accelerating clearance and high doses slowing the clearance process. Some respiratory tract regional- and species-specific differences have also been observed.

In vitro cultures of epithelial cells obtained from nonatopic and mild atopic asthmatics, when exposed to O_3 (0.01-0.1 ppm), exhibited significantly increased permeability compared to cells from normal persons, thus indicating a potential inherent susceptibility of cells from asthmatics for O_3-induced permeability. New animal toxicology studies reported O_3-induced modulation of cell-mediated immune responses affecting the onset and persistence of infection in rats (Cohen et al. 2001, 2002). However, there is no compelling evidence from animal toxicological, human clinical or epidemiologic studies that O_3 enhances the incidence of respiratory viral infection in humans.

The available data at this time indicate that acute O_3 exposure has a potential to impair the host defense capability, primarily by interfering with the functions of alveolar macrophages. Any impairment in macrophage function may lead to decreased clearance of microorganisms or nonviable particles. Compromised alveolar macrophage functions in asthmatics may increase their susceptibility to other O_3 effects or to the effects of particles.

Biochemical Alterations

An extensive experimental database, including research assessed in 1996 O_3 AQCD, suggests that potential biochemical alterations in various metabolic pathways (including xenobiotic metabolism) are involved in lung injury, inflammation, and functional alterations. Interaction of O_3 with the lipid constituents of pulmonary surfactant has been proposed as one of the key mechanisms by which O_3 exerts its toxic effects. Experimental evidence clearly indicates a role for the initial interaction of O_3 with lipid constituents of the ELF and generation of lipid ozonation products and secondary redox mediators in the initiation of site-specific cell-injury response cascades. One such lipid ozonation product, 4-hydroxynonenal, has been found to bind to proteins and increased protein adducts in human alveolar macrophages, suggesting a role for 4-hydroxynonenal in acute cell toxicity. Cholesterol, the most abundant neutral lipid in pulmonary surfactant is susceptible to attack by O_3 resulting in multiple oxidized cholesterol products, including the formation of cholesterol epoxide. A 20-fold increase in cholesterol epoxide in the BALF from mice exposed to 0.5 ppm O_3 for 3 h suggests a potential role for this oxidation product in O_3 toxicity (Pulfer et al., 2005). Species- and region-specific increases in lung xenobiotic metabolism have been observed in response to both short- and long-term O_3 exposure. It has been well recognized that antioxidants in the ELF confer protection against O_3 toxicity. But the observation of O_3 reactivity even with environmentally relevant exposures, questions their ability to quench O_3 reactivity. Species-specific and age-dependent changes in the antioxidant metabolism add another dimension to their role in this process. Carefully controlled studies of dietary antioxidant supplementation (Samet et al., 2001; Trenga et al., 2001) reported some protective effects of α-tocopherol and ascorbate for O_3-induced spirometric lung function decrements but not for the intensity of subjective symptoms and inflammatory responses (including cell recruitment, activation, and a release of mediators). Dietary antioxidants have also afforded partial protection to asthmatics by attenuating postexposure

bronchial hyperresponsiveness (Trenga et al., 2001). In addition, two epidemiologic studies of street workers and asthmatic children in Mexico City found that subjects taking antioxidant supplements containing vitamins E and C were protected from O_3-induced changes in lung function (Romieu et al., 1998, 2002).

Based on the above discussion, it is evident that a very extensive experimental database accumulated from animal toxicology studies (including nonhuman primate studies) and limited controlled human exposure studies, has provided important insights into various biochemical, cellular, and molecular alterations in lung tissue exposed to O_3. The majority of these studies, although using acute exposure regimens and relatively high concentrations at times, do provide credible hypotheses regarding potential molecular mechanisms implicated in O_3 toxicity. Utilizing this information in relevant rodent-to-human extrapolation models with appropriate species-specific adjustments may well provide useful information on initial biochemical alterations that may aid in the development of suitable biomarkers for O_3 exposures/effects.

Cardiovascular Effects

Ozone-induced lung injury and permeability changes, as well as O_3-induced alterations in the hemodynamics may lead to O_3 effects on the cardiovascular system. Also, the interaction of O_3 with ELF, lipids, and surfactants, and the lipid ozonation products and ROS generated in this process have the potential to penetrate the epithelial barrier and to initiate toxic effects on the cardiovascular system. An increasing body of animal toxicology evidence suggests that hematological and thermoregulatory alterations (in heart rate variability and/or core body temperature) may mediate acute cardiovascular effects. Studies carried out using isolated perfused rat lung model (Delaunois et al., 1998) indicate inhibition of pulmonary mechanical reactivity to bronchoconstrictors and persistent vasoreactivity of the vascular bed upon exposure to O_3 (0.4 ppm for 4 h). Earlier studies in rats indicate a potential role for platelet activating factor (PAF) in O_3-inflammatory response. Recent observations of O_3-induced generation of oxysterols and β-epoxides from cholesterol in surfactant suggest that these lipid ozonation products like lysophospholipids may initiate PAF-like activity and initiate clotting and thrombolytic effects in the cardiovascular system.

A few human experimental studies have examined the potential effects of O_3 exposure on cardiovascular functions. For example, Gong et al (1998) evaluated various cardiac function and

hemodynamic variables in healthy and hypertensive adult males and observed impairment of alveolar-arterial oxygen transfer, leading them to suggest that such an impairment could lead to decreased oxygen supply to the myocardium. Also, Foster et al (1993; 1997) have reported O_3-induced ventilation-perfusion mismatch. Such an altered ventilation distribution profile observed even in relatively young healthy adults could contribute to the alveolar-arterial oxygen transfer impairment reported by Gong et al. (1998). Taken together, these observations support the regional differences in ventilation and perfusion in severe COPD patients reported by King and Briscoe (1968) and Kronenberg et al. (1973). Such preexisting compromised gas exchange abnormalities would likely make lungs of individuals with COPD more vulnerable to O_3-induced gas exchange inhibition and reduced oxygen saturation.

8.4.3 Assessment of Epidemiological Evidence

Based on the O_3 epidemiologic evidence available at the time, the 1996 O_3 AQCD arrived at the following conclusions:

> An association between daily mortality and O_3 concentration for areas with high O_3 levels (e.g., Los Angeles) has been suggested, although the magnitude of such an effect is unclear. Increased O_3 levels are associated with increased hospital admissions and emergency department visits for respiratory causes. Analyses from data in the northeastern United States suggest that O_3 air pollution is associated with a substantial portion (on the order of 10 to 20%) of all summertime respiratory hospital visits and admissions. Pulmonary function in children at summer camps in southern Ontario, Canada, in the northeastern United States, and in Southern California is associated with O_3 concentration." (U.S. EPA, 1996, p1-29).

The 1996 O_3 AQCD further stated that only suggestive epidemiologic evidence existed for health effects of chronic ambient O_3 exposure in the population, and this was partly due to an inability to isolate potential effects related to O_3 from those of other pollutants, especially PM (U.S. Environmental Protection Agency, 1996).

The discussion in this section of scientific strength and limitations of the growing body of epidemiologic evidence for associations between ambient exposure to O_3 and various health effects discussed is based primarily on Chapter 7 evaluations. The following criteria were considered in assessing the relative scientific quality of the epidemiologic studies: (1) the *strength* of reported associations, in terms of magnitude, statistical significance and statistical power of effects estimates; (2) *robustness* of reported associations (based on defined health endpoint criteria), potential confounding by copollutants; (3) *consistency and coherency* of the

effect associations *(4) temporality,* in terms of lag periods between exposure and observed effects; and (5) *biological plausibility* of the observed O_3-related health effects assessed in terms of their coherence in relation to findings derived from controlled human exposure studies which, overall, provide insights into the plausibility of reported O_3 human health effects reflecting causal relationships.

Many newly available epidemiologic studies have provided additional evidence for O_3-related health effects beyond that previously known. Significant statistical associations have been reported by various investigators between acute O_3 exposure and several respiratory and cardiovascular health endpoints, including: mortality; hospital admissions; emergency department visits; respiratory illness and symptoms; and changes in pulmonary function. Similarly, associations have been reported between long-term exposure to O_3 and increased morbidity; development of respiratory disease; and declines in lung function and lung function growth. The numerous new epidemiological studies that have been conducted in areas across the United States and Canada, as well as in Europe, Latin America, Australia and Asia, are summarized in the Annex to Chapter 7. Based on evidence extracted from the full body of epidemiologic studies carried out and reviewed since the 1996 O_3 AQCD, it has been well demonstrated that deleterious human health outcomes are positively associated with acute ambient O_3 concentrations currently encountered in the United States.

8.4.3.1 Strength and Consistency of Epidemiological Associations

As quoted above, assessments in the 1996 O_3 AQCD supported a consistent relationship between O_3 concentration and respiratory illness, hospital visits and reduced lung function. However, due to insufficient evidence examining O_3-mortality associations and uncertainties regarding weather model specification, the 1996 O_3 AQCD was limited to only a very qualitative assessment of O_3-mortality associations. Since then, Generalized Additive Models (GAMs) have become widely utilized for epidemiologic time-series analysis of health effects attributable to air pollution, increasing our confidence in quantitative estimation of O_3-mortality risks. Some concerns have been raised regarding the use of default convergence criteria in applications of commercially available software employed for GAM analyses to estimate air pollution-related health effects, as discussed in the 2004 PM AQCD (U.S. Environmental Protection Agency, 2004a). However, reanalyses of a number of studies, comparing results using default GAM

convergence criteria to results from analyses using stringent GAM convergence criteria and/or from GLM analyses, found little difference among the O_3 effect estimates obtained (as discussed in Chapter 7 of this document). Overall, the magnitude of the effect-size estimates observed for O_3-mortality relationships tend to be relatively consistent across the newly available studies. The effect estimates for O_3-morbidity endpoints have greater variability, but consistent positive associations between ambient O_3 and various health outcomes have also been observed.

8.4.3.1.1 Acute Exposure Studies

Numerous epidemiological studies carried out over the past decade have added evidence to the knowledge base assessed in the 1996 O_3 AQCD, which included both (a) individual-level camp and exercise studies that established a relationship between ambient O_3 exposure and human lung function decline and (b) aggregate time-series studies that suggested positive relationships for O_3-related respiratory morbidity. The new studies reviewed in Chapter 7 in this document include numerous field/panel studies and time-series studies from various regions. In field/panel studies on the effects of air pollution exposure, the most common health outcomes measured were lung function and respiratory symptoms. The time-series studies examined daily emergency department visits, hospital admissions, and mortality data.

Field/Panel Studies of Acute Exposure Effects

Pulmonary Function And Respiratory Symptoms

Healthy Individuals

Many of the new field/panel studies reviewed in Chapter 7 and the controlled human exposure studies reviewed in Chapter 6 of this document provide additional data supporting two major findings reported in the 1996 O_3 AQCD. First, acute O_3 exposure is associated with a significant decline in lung function parameters. Ozone-related lung function decrements were most notable in children and asthmatics. In addition, adults who work or exercise outdoors also were found to be vulnerable to O_3-associated declines in lung function due to their increased exposure to O_3. Second, acute exposure to O_3 is associated with increased respiratory symptoms, particularly cough, and increased as-needed medication use in asthmatic children. Immediate effects of O_3 were observed on both lung function and respiratory symptoms, with the strongest

associations often observed at a lag of 0- or 1-day. The two health outcomes are further discussed below.

Pulmonary function was determined by either spirometry (forced expiratory volume in 1 s [FEV_1] and forced vital capacity [FVC]) or by peak expiratory flow (PEF) meters. The spirometric parameter, FEV_1 is a strong and consistent measure of lung function. PEF is a closely related but different metric of lung function, and PEF is more feasibly performed in field studies, using inexpensive peak flow meters that produce similar results to PEF measured spirometrically.

In a number of newly available field/panel studies, FEV_1 was measured in panels of exercising children, outdoor workers, and adult hikers exposed to ambient O_3 while experiencing elevated exertion levels. Collectively, the results of the new studies (discussed in Section 7.2.3.1) confirm and extend those from analogous field/panel studies assessed in the 1996 O_3 AQCD and findings from experimental controlled human exposure studies indicating that acute O_3 exposures prolonged over several hours and combined with elevated levels of exertion or exercise magnify O_3 effects on lung function, as evaluated in terms of FEV_1.

For example, six field studies by three different research groups of 7- to 17-year-old, healthy (nonasthmatic) children exposed for several hours to ambient O_3 during increased physical exertion in summer camp activities were assessed in the 1996 O_3 AQCD. When analyzed together by consistent statistical methods, the data from those studies showed an average relationship between afternoon FEV_1 and concurrent 1-h O_3 concentrations of −0.50 mL/ppb (95% CI: −0.63, −0.36), with individual slopes ranging from −0.19 to −1.29 mL/ppb (Kinney et al., 1996). Four new field/panel studies (assessed in Section 7.2.3.1 of this document) that evaluated pulmonary function in healthy school-aged children exposed to mean 1-h max O_3 concentrations ranging from 20 to 112 ppb found exposure-response functions of approximately −0.18 to −1.42 mL/ppb. Also, two other studies assessed in the 1996 O_3 AQCD that measured lung function before and after well-defined exercise events (1/2-h long) in adults during exposures to ambient O_3 across 4 to 135 ppb found exposure-response slopes of −0.4 mL/ppb (95% CI: −0.7, −0.1) (Selwyn et al., 1985) and −1.35 mL/ppb (95% CI: −2.04, −0.66) (Spektor et al., 1988). In comparison, new studies of healthy adult workers (street workers, berry pickers) and hikers engaged in prolonged (≥6 h) strenuous physical exertion at mean exposure levels ranging from 40 to 123 ppb 1-h max O_3 reported exposure-response slopes

of −1.40 to −3.8 mL/ppb (as assessed in Section 7.2.3.1 in Chapter 7 of this document). The most representative data are those of Korrick et al. (1998) from a U.S. study of adult hikers that provided outcome measures stratified by gender, age, smoking-status, and presence of asthma within a population capable of above-normal exertion.

Pulmonary function changes or declines measured in susceptible populations suggest that juvenile asthmatics are at greater risk with the largest O_3-related decline of FEV_1 of −2.08% (95% CI: −6.24, 2.08) per 40 ppb increase in 1/2- h max O_3 at a 2-day lag (Höppe et al., 2003). An extended analysis by Höppe et al. (2003) examined individual susceptibility to O_3 effects. Ozone responders were regarded as those with a greater than 10% change in FEV_1. Compared to athletes and the elderly, a greater percentage of children and asthmatics (20% versus 5%) were found to be sensitive to O_3 effects on lung function. The small sample sizes in these studies limit extrapolation to larger populations; however, the studies indicate a trend that should be evaluated in larger epidemiologic studies.

Asthma Panels

Several studies assessed in the 1996 O_3 AQCD that evaluated elevated respiratory symptoms and/or pulmonary function decrements in asthmatic children showed greater responses in asthmatic than in nonasthmatic subjects, suggesting that asthmatic individuals might constitute a sensitive population group in O_3 epidemiologic studies.

Additional panel studies carried out over the past decade to understand the effect of acute exposure to O_3 in asthmatics evaluated lung function by PEF and/or respiratory symptoms (i.e., cough, wheeze, shortness of breath and medication use) ascertained by questionnaire. Several additional studies (see Figures 7-1 and 7-2), both in the U.S. and in other countries, reported decrements in PEF to be associated with O_3 exposures among asthmatics. One large U.S. multicity study (Mortimer et al., 2002) associated O_3 concentrations with the incidence of ≥10% declines in morning PEF (odds ratio of 1.30 [95% CI: 1.04, 1.61] per 30 ppb increase in 8-h avg O_3 for a 5-day cumulative lag). In a group of adult hikers in Mount Washington, NH (Korrick et al., 1998), asthmatic subjects experienced a four-fold greater decline in FEV_1 compared to healthy individuals with the same exposure to O_3. Asthmatic hikers experienced a mean change of −4.47% (95% CI: −7.65, −1.29) per 30 ppb increase in 8-h avg O_3 while other hikers had a mean change of −1.08% (95% CI: −2.49, 0.33).

Several single-city studies did not observe statistically significant declines in lung function parameters (e.g., Delfino et al., 1997; Hilterman et al., 1998), which might be partially attributable to small sample sizes and/or low levels of O_3. Although results were not always statistically significant in these single-city studies, effect estimates were consistently negative, providing suggestive evidence that O_3 was associated with lung function declines among asthmatics. Collectively, results from the large multicities study by Mortimer et al. (2002), as well as those from the smaller single-city studies suggest that exposure to O_3 may be associated with declines in lung function in this potentially susceptible population.

The majority of studies that evaluated respiratory symptoms (i.e., cough, shortness of breath, and wheeze) and the increased use of asthma medication related to O_3 exposure are also focused on asthmatic children. Two large U.S. studies (Mortimer et al., 2002; Gent et al., 2003) and some international studies (Hilterman et al., 1998; Desqueyroux et al., 2002a,b) suggest positive associations between O_3 ambient concentrations and increased symptoms or asthma medication use. In the multicities study by Mortimer et al. (2002), the odds ratio for the incidence of symptoms (including cough, chest tightness, and wheeze) was 1.35 (95% CI: 1.04, 1.69) per 30 ppb increase in 8-h avg O_3 for a 4-day cumulative lag. As in the case of studies examining the O_3 effect on lung function, not all studies on respiratory symptoms observed consistent positive associations with O_3. For example, Avol et al. (1998) studied symptoms in asthmatic, wheezy, and healthy children aged 10 to 12 years in southern California. Some symptom associations were noted but they were inconsistent, possibly due to relatively low O_3 concentrations during the study period. The authors also noted that the study children did not spend substantial time outdoors engaged in physical activities. Once again, the strong evidence from the large multicities study by Mortimer et al. (2002), along with less consistent but generally supportive evidence from several single-city studies suggest that O_3 exposure may be associated with increased respiratory symptoms and medication use in asthmatic children.

School Absenteeism

Two large U.S. studies (Chen et al., 2000; Gilliland, 2001) and one study from Seoul, Korea (Park et al., 2002) investigated the relationship between ambient O_3 concentrations and school absenteeism. Results from all these studies suggested a positive association between O_3 and absences from school, with each one arriving at these associations using different lag

periods. Chen et al. (2000) reported a 10.4% (95% CI: 2.7, 18.1) excess rate of total daily school absences for 40 ppb increase in daily 1-h max O_3 with a distributed lag of 1 to 14 days. The 12 southern California communities study by Gilliland et al. (2001) also reported larger O_3-related school absences due to respiratory causes, 147% (95% CI: 6, 478) per 30 ppb increase in 8-h -avg O_3 with a 30-day lag, compared to nonrespiratory causes, 61% (95% CI: 9, 137). The studies reported by Park et al. (2002) were analyzed using GAM with default convergence criteria and indicated a positive association with same day O_3 (16% [95% CI: 12, 22] per 30 ppb increase in 8-h -avg O_3). Results from the three studies listed above suggest that ambient O_3 concentrations may be associated with school absenteeism, particularly illness-related absences. However, the associations observed during the long lag period of two to four weeks may reflect confounding by other time-varying factors or be a chance finding from an exploratory analysis. Additional studies and analysis using similar lag periods are needed to more clearly delineate quantitative relationships between ambient O_3 and school absences.

Field Studies on Cardiovascular Effects

A limited number of studies evaluated potential short term effects of air pollution on cardiovascular functions. Several of these studies evaluated the effects of PM, O_3 and other gaseous pollutants. Two major U.S. population-based studies (Liao et al., 2004; Park et al., 2005) suggested an association between short-term O_3 exposure and decreased heart rate variability (HRV). Park et al., (2005) reported stronger associations of HRV with $PM_{2.5}$ and O_3 in people with ischemic heart disease and hypertension. These results are consistent with a Mexico City study that observed an O_3-induced HRV effect in individuals with hypertension (Holguin et al., 2003). Several other studies, on the other hand, did not find any such relationship, but these studies might have had limited power (e.g., low O_3 concentration ranges, small sample sizes) to examine the subtle effects. Studies that evaluated the relationship between air pollutants and the onset of myocardial infarction (Ruidavets et al., 2005; Peters et al., 2001) suggested a positive association with O_3. However, due to lack of information on potential confounding by PM and the limited number of studies available, additional research is needed to confirm these observations.

Only a limited number of epidemiologic studies examined cardiovascular outcomes in relation to O_3 exposures. Among them, the larger population-based studies (Liao et al., 2004;

Park et al., 2005; Ruidavets et al., 2005) observed suggestive evidence of an association of O_3 exposure with decreased HRV and increased incidence of myocardial infarctions.

Time-Series Analyses of Acute Exposure Effects

Emergency Department Visits and Hospital Admissions

Many time-series studies reviewed in the 1996 O_3 AQCD indicated positive associations between O_3 air pollution and increased hospital admissions. Strong evidence establishing a correlation between O_3 exposure and increased exacerbations of preexisting respiratory disease in the general public were reported at 1 h-maximum O_3 concentrations <0.12 ppm.

Several studies published during the past decade examined temporal associations between O_3 exposures and emergency department visits or hospital admissions for respiratory diseases (see Table AX7-2 in Chapter 7 Annex). One of these studies by Peel et al. (2005) reported stronger and more positive associations between O_3 and emergency department visits due to respiratory-related diseases in the warm season. A 3.1% (95% CI: 0.2, 6.2) excess risk in asthma visits was associated with a standardized increment of 30 ppb in 8-h max O_3 (see Section 7.1.3.2). This risk was significantly associated with respiratory infections, when adjusted for PM_{10}, NO_2, and CO in multipollutant models. Several other U.S. and Canadian studies reported positive associations between O_3 concentrations and emergency department visits due to respiratory causes (Figure 8-5). However, several of the European studies observed no association between O_3 concentrations and emergency department visits for respiratory diseases. These inconsistent results might be partially attributable to differences in model specifications and statistical methods used to evaluate seasonal patterns and potential confounding by copollutants. Overall, then, the current body of evidence remains inconclusive regarding ambient O_3 effects on risk of emergency departments visits. Additional studies are needed to establish stronger and more convincing associations between increased concentrations of ambient O_3 and increased risk of emergency department visits.

Studies of acute O_3 exposure effects on respiratory disease-related hospital admissions (summarized in Section 7.3.3) have considered various factors in their analyses. The hospital admission data were assessed based on the type of respiratory disease (such as asthma, COPD), seasonal effects (summer vs. winter, O_3 concentration and temperature), age of the study population, studies carried out in single or multiple cities, effect of confounders and lag days

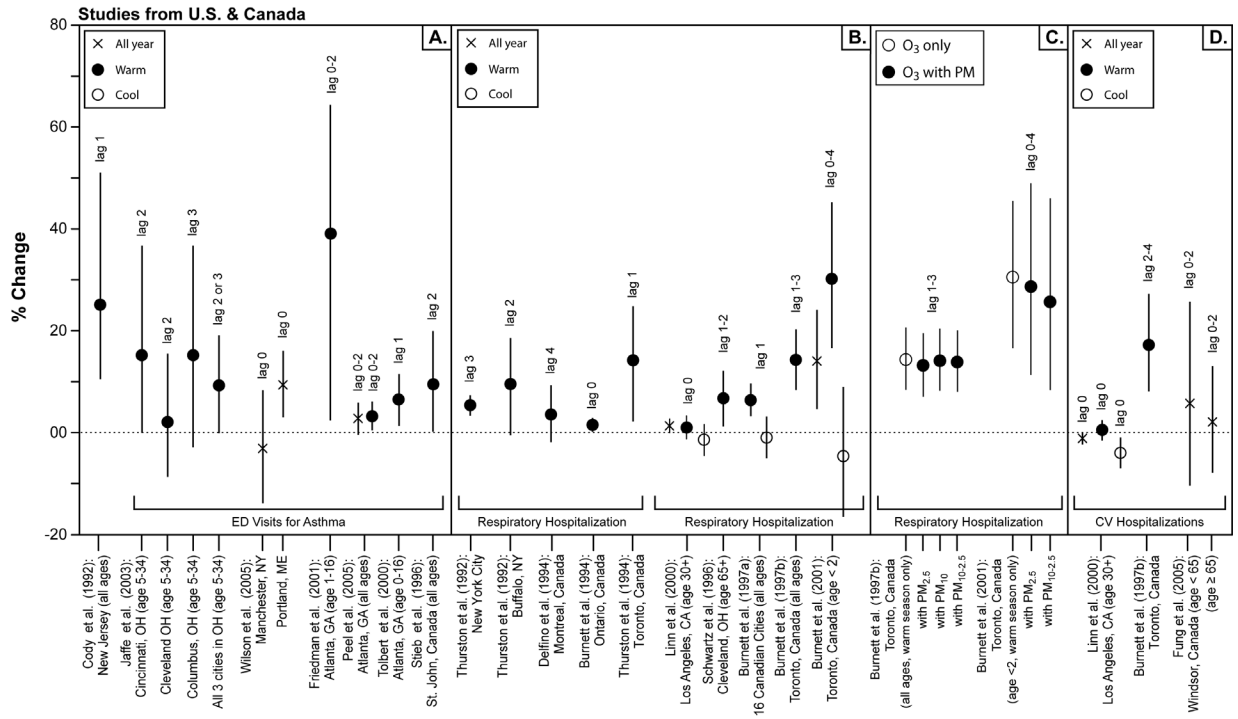


Figure 8-5. Ozone-associated percent change (95% CI) in emergency department visits for asthma (A), total respiratory hospitalization by season (B), respiratory hospitalization with adjustment for PM indices (C) and (D) total cardiovascular hospitalization per standardized increment (see Section 7.1.3.2). Only results for U.S. and Canadian studies are presented.

between exposure and hospital admissions (Figure 8-5). Two large studies (Burnett et al., 1997a; Anderson et al. 1997) that used consistent analytical methodologies found significant associations between O_3 concentrations and increased risk for unscheduled hospital admissions for respiratory-related diseases, despite differences in geographic locations of the study populations. In both studies, larger effects were observed in the warm season. The 16 cities Canadian study by Burnett et al. (1997a) analyzed data for a population of 100,000 over a period of 10 years. During the summer, an excess risk of 6.7% (95% CI: 3.5, 10.0) per 40 ppb standard increment in 1-h max O_3 was observed. The five cities APHEA (Air Pollution on Health: European Approach) study by Anderson et al. (1997) also reported significant associations between O_3 and hospitalizations for COPD. An excess risk of 4.7% (95% CI: 1.6, 7.9) per 40 ppb increase in 1-h max O_3 was observed in the warm season. Several other studies (single city

or two city studies for five or more years) also suggested a positive association between increase in O_3 and increased risk of hospital admissions.

Many other studies reported less consistent or no associations between increases in O_3 concentrations and hospital admissions. A few other studies raise questions and concern about other factors in this relationship. Despite these inconsistencies noted across the studies, the collective evidence supports the findings of significant and robust effects of O_3 on respiratory hospitalization outcomes. Large multicity studies as well as several individual city studies have reported positive O_3 associations with total respiratory, asthma, and COPD hospitalizations, especially in those that analyzed the O_3 effect during the summer or warm season.

A subset of hospital admissions studies examined the effect of O_3 on cardiovascular outcomes (see Figure 8-5). The evidence is inconclusive on the association between O_3 exposure and cardiovascular hospitalizations with regard to year-round data. However, in studies that adjusted for seasonal or meteorological factors, there was suggestive evidence that O_3 was associated with increased risk of cardiovascular hospital admissions in the warm season.

Mortality-Related to Short-term O_3 Exposures

Due to the limited number of studies and uncertainties regarding weather model specifications, no meaningful quantitative assessment of O_3-mortality associations was possible in the 1996 O_3 AQCD. However, newly available large multicity studies designed specifically to examine the effect of O_3 on mortality have provided much more robust and credible information.

Two large multicity studies from the U.S. (Bell et al., 2004; Schwartz et al., 2005) and one from Europe (Gryparis et al., 2004) specifically evaluated O_3 effects on mortality and indicated positive associations between increased O_3 levels and mortality. Among the positive studies, risk estimates for (U.S. and Canadian) single-city studies carried out using a single-pollutant model are in the range of 0.8 to 3% excess deaths per 40 ppb increase in 1-h max O_3 (Figure 8-6), while multicity studies and meta-analyses reported a risk estimate in the range of 0.5 to 2% excess risk with identified heterogeneity (due to model specifications) across cities and studies (Figure 8-6). Models examining different lag times, observed that there was an immediate effect of O_3 on mortality which persisted over several days, resulting in risk effect estimates at a cumulative lag of 0 to 7 days.

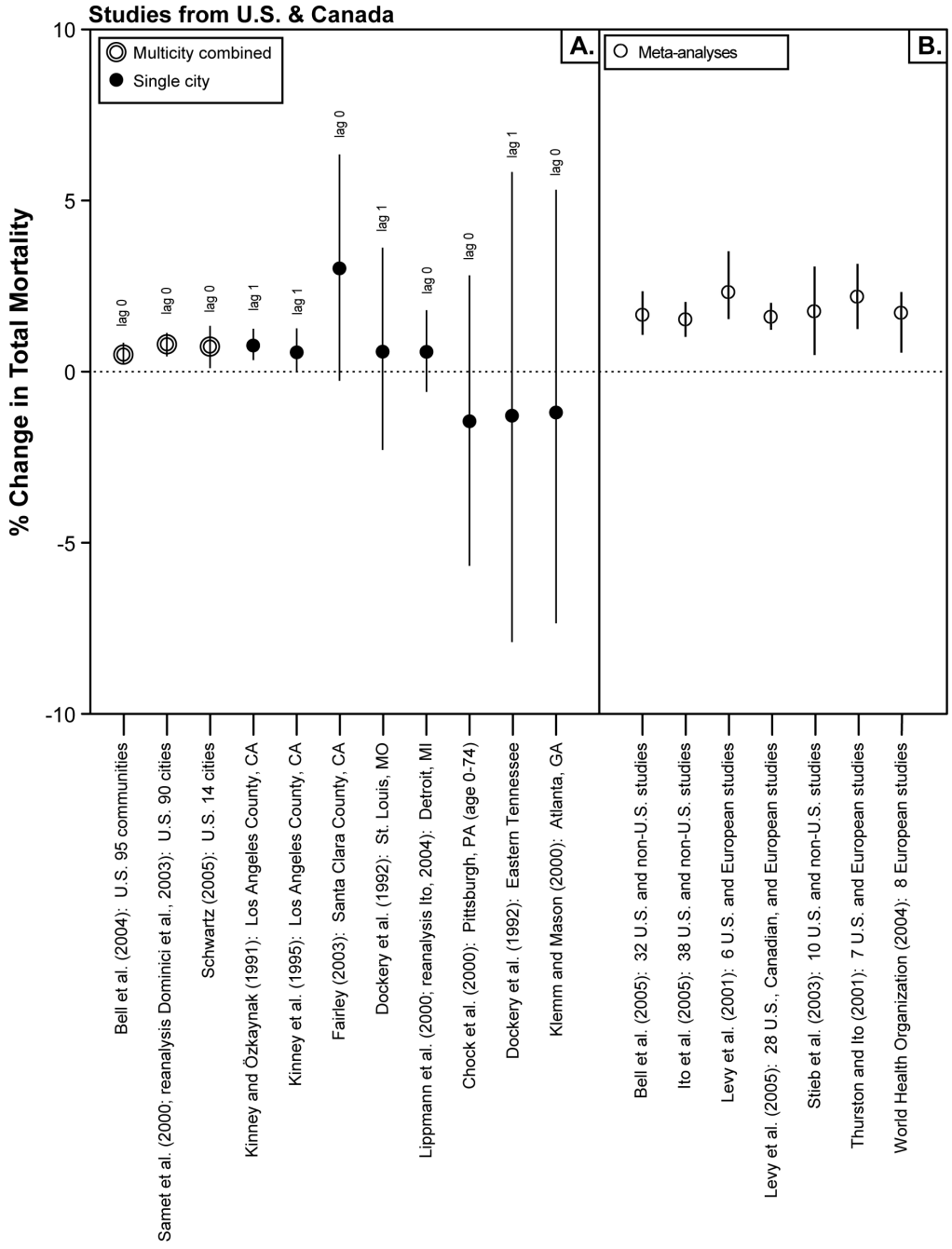


Figure 8-6. **A. All cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) <u>for all year analyses</u> per standardized increment (see Section 7.1.3.2). Analyses include all ages unless otherwise noted. Only results from <u>single-day lag models</u> are presented.**

B. Combined all cause (nonaccidental) O_3 excess mortality risk estimates (95% CI) from recent meta-analyses per standard increment of 40 ppb in 1-h max O_3 or equivalent. Note that all meta-analyses, except Stieb et al. (2003), included studies which used Poisson GAM with default convergence criteria.

Some of the epidemiological studies and, particularly, meta-analyses examined the influence of season on O_3 mortality associations and indicated larger O_3-mortality risk estimates in the warm season compared to the colder season. These estimates appear to be consistent with a causal association when O_3 levels are high in the warm season. This seasonal dependence of O_3-mortality effects complicates the evaluation and interpretation of risk estimates from year-round data without adjustment for temporal trends (Figure 8-7). The confounding effects of copollutants were examined by three recent meta-analyses (Bell et al., 2005; Ito et al., 2005; Levy et al., 2005). These, as well as other multicity and single-city studies, indicated that copollutants do not appear to substantially confound O_3-mortality associations. Meta-analyses on the association between cause-specific mortality and O_3 levels observed larger positive associations with cardiovascular mortality compared to total mortality (Figure 8-8). Additional analyses carried out to examine specific population groups potentially susceptible to O_3-mortality effects did not clearly identify any specific group, but they did suggest that severe asthmatics and elderly populations might be relatively more susceptible to O_3.

8.4.3.1.2 Chronic Ozone Exposure Studies

There were a limited number of studies reported in the 1996 O_3 AQCD that addressed potential health effects of long-term ambient O_3 exposures. Several longitudinal epidemiological studies carried out in the past decade evaluated the potential effects of chronic (several weeks to many years) O_3 exposure on lung function, respiratory symptoms, lung inflammation, asthma prevalence, cancer incidence, and mortality**.** Based on the available data at this time, no clear conclusions can be drawn now regarding the relationship between chronic O_3 exposure and such health outcomes. A limited number of studies also examined the potential effects of ambient O_3 exposure on birth defects, and these also suggest the need for additional studies and a larger database before drawing any conclusions regarding possible associations.

Very few studies have investigated the effects of long-term O_3 exposure on incidence of cancer and mortality. Uncertainties regarding the exposure period of relevance and inconsistencies across mortality outcomes and gender raise concerns regarding plausibility. The largest and most representative U.S. study, by Pope et al. (2002), observed positive but nonsignificant associations between O_3 exposure and all cause, cardiopulmonary, and lung

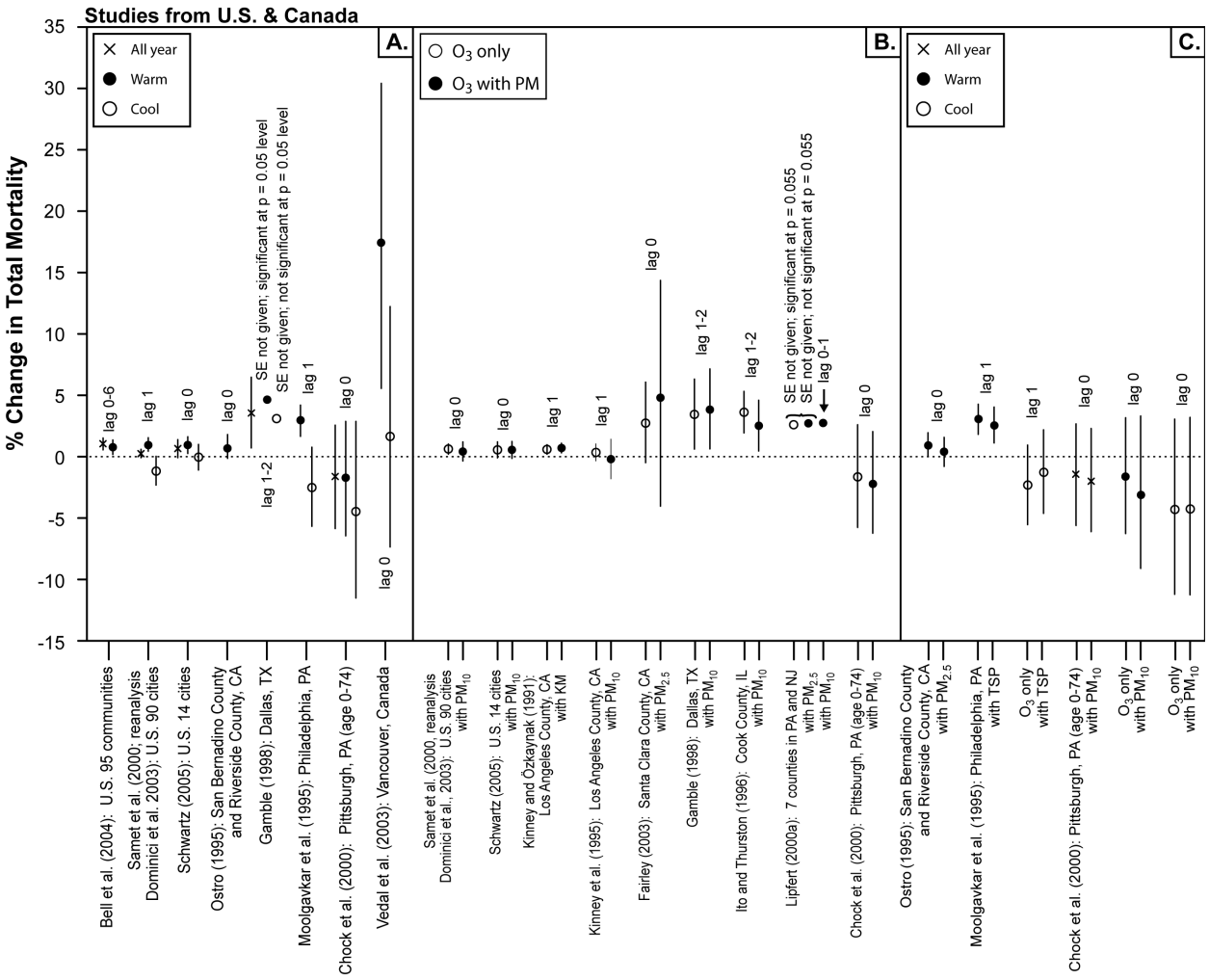


Figure 8-7. All causes (nonaccidental) O_3 excess mortality risk estimates (95% CI) per standardized increment (see Section 7.1.3.2). (A) by season; (B) with adjustment for PM indices in all year analysis and (C) with adjustment for PM indices by season. Note: Only U.S. and Canadian studies presented.

cancer mortality. Thus, the current evidence is inconclusive regarding a potential relationship between chronic O_3 exposure and increased mortality risk.

8.4.3.2 Robustness of Epidemiological Associations

In evaluating the strength of the epidemiological evidence, the magnitude of observed O_3 effect estimates and their statistical significance is important; however, consideration must be given to the precision of the effect estimates and the robustness of the effects associations.

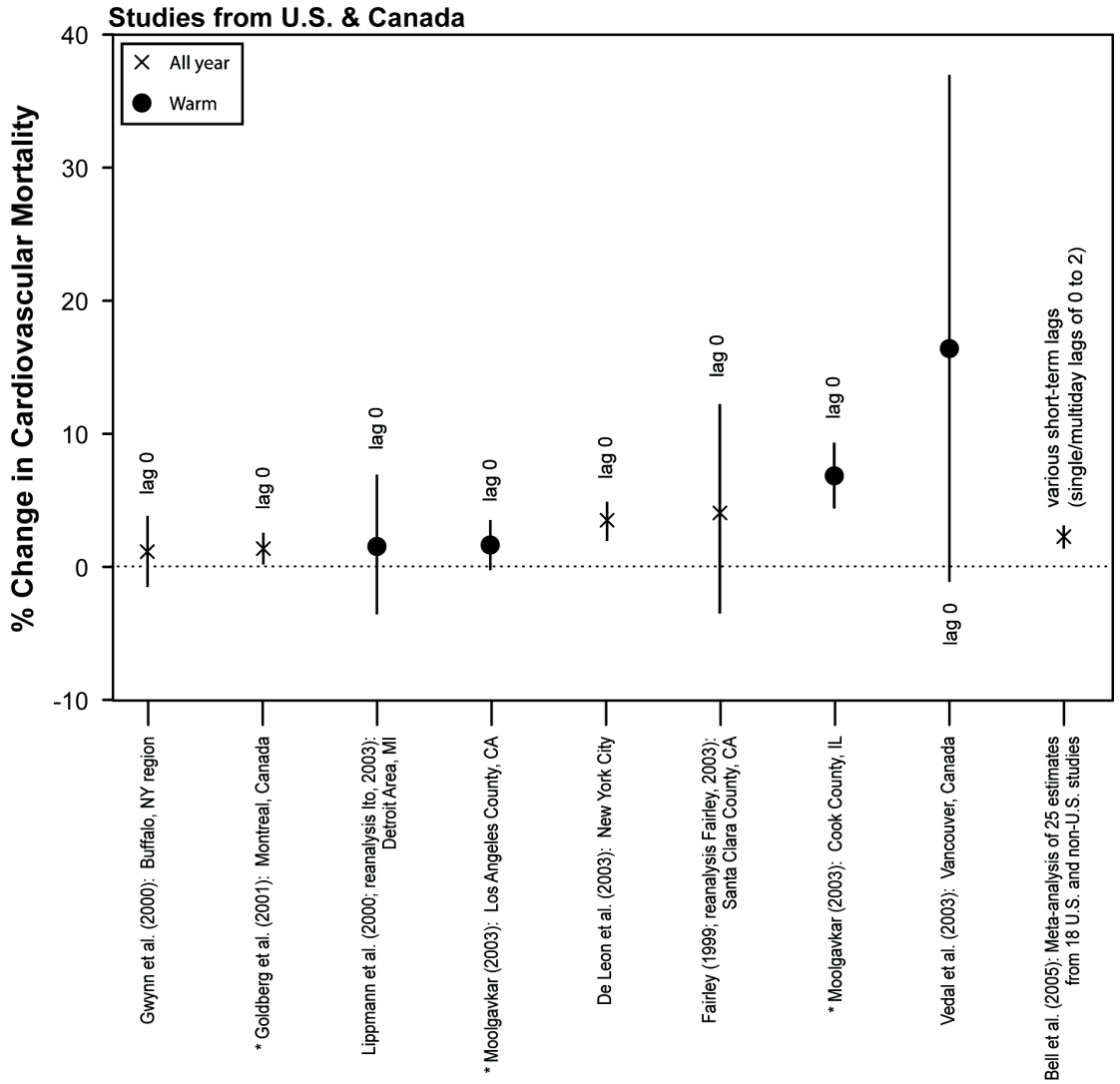


Figure 8-8. Ozone-associated percent change (95% CI) in cardiovascular risk estimates per standardized increment (see Section 7.1.3.2). Analyses are for all ages. Only studies for U.S. and Canada presented. *Analyses using GAM default convergency criteria.

Examining the robustness of the associations includes evaluating the impact of alternative models, model specifications for temporal trends and meteorological factors, and potential confounding by copollutants. Also of interest are issues related to exposure assessment and measurement error. A detailed discussion on each of these topics can be found in Chapter 7 (Sections 7-1 and 7-6). The following sections focus on the extent to which the current epidemiological findings can be considered robust.

8.4.3.2.1 Exposure Issues: Ambient versus Personal

In many air pollution epidemiologic studies, especially time-series studies utilizing administrative data on mortality and hospitalization outcomes, data from central ambient monitoring sites are generally used as the estimate of exposure. Personal exposures of individual study participants generally are not directly observed in epidemiologic studies. The relationship between ambient O_3 concentrations and personal O_3 exposure levels varies, depending on factors such as time spent outdoors, ventilation conditions, personal factors, and air quality indices. There is suggestive evidence that ambient O_3 concentrations from central monitors may serve as valid surrogate measures for aggregate personal O_3 exposures in time-series studies. However, ambient concentrations generally overestimate true personal O_3 exposures. Thus, use of ambient concentrations in risk calculations will likely result in effect estimates that are biased towards the null, resulting in biased descriptions of underlying concentration-response relationships. These effect estimates, though conservative from a testing perspective, must be evaluated and used with caution, as they may lead to an underestimation of the overall impact of air pollution on health effects.

8.4.3.2.2 Confounding by Temporal Trends and Meteorologic Effects

The effect of seasonal differences in the health outcomes and O_3 exposure levels was recognized in the 1996 O_3 AQCD. This issue is discussed in detail in Section 7.6.5 of this document. Two important factors, i.e., temporal trends and meteorological factors must be considered in evaluating O_3 health effects estimates. In the U.S. 95 communities study (Bell et al., 2004), sensitivity analyses indicated that the O_3 risk estimates were robust to tripling the degrees of freedom for smoothing terms used to control for temporal trends. In a case-crossover study by Schwartz (2004), the O_3-mortality risk estimates from an analysis using nonlinear regression splines to control for temperature were similar to those from an analysis that matched on temperature, indicating that the effect estimates were not sensitive to methods used to control confounding by temperature.

Analysis of O_3 health effects is further complicated in view of the fact that the relationship of O_3 with temperature and with other pollutants appears to change across seasons. As shown in Figures 8-5, 8-7 and 8-8, the O_3 effect estimates from warm season data were consistently larger compared to those calculated using all-year data and cool-season data. In a study of daily

hospital admissions (Burnett et al., 2001), season-stratified analyses appeared to effectively control confounding by season.

In summary, adjusting for temporal trends and meteorological factors is critical to obtaining meaningful O_3-effect estimates. Analyses have found that confounding by seasonal variability is controlled effectively by stratifying the data by season. Mortality and morbidity effect estimates computed using year-round data need to be interpreted with caution.

8.4.3.2.3 Assessment of Confounding by Copollutants

The presence and influence of PM and other gaseous copollutants have to be considered in assessing O_3-health effects associations found by observational studies. The potential for copollutant confounding in the epidemiological time-series studies was assessed in some detail in Section 7.6.6. Multipollutant modeling is the most common method used to test for potential confounding in epidemiological studies; however, interpretation of the results is often complicated by the high degree of correlation among air pollutants. Across the various health outcomes, O_3 effects were generally not confounded by PM and other gaseous copollutants. The O_3 effects on lung function, respiratory symptoms, respiratory hospitalizations, and mortality were robust to PM-adjustment in all year and warm season only analyses.

The O_3 mortality risk estimates from two-pollutant models adjusted for PM are presented in Figure 8-7 (U.S. and Canadian studies only). In the two multicity studies analyzed here, the addition of PM_{10} did not substantially change the risk estimates (Samet et al., 2000; Dominci et al., 2003; Schwartz, 2004). The O_3-mortality effects in single-city studies also were robust after adjusting for PM_{10} indices, both in all-year and season-stratified analyses data.

In summary, assessing the health effects attributable to O_3 is very challenging, given the high covariation among the copollutants and the limitations in the statistical methodology to assess independent effects of such correlated variables. Definitive partitioning out of the individual pollutant-specific health outcomes from among an ambient mixture of multiple components is very difficult due to the dynamic nature of their interactions over time. However, the new time-series studies that made an exhaustive survey using populations from multiple U.S. cities do provide substantial epidemiologic evidence indicating that associations for O_3 with mortality and morbidity are robust to confounding by copollutants.

8.4.3.3 Lag Period between Ozone Exposure and Health Response

The lag times between cause and effect depend on underlying biological mechanisms involved in the processes. Different lag periods are appropriate for assessing different health outcomes. As discussed in Section 7.6.4, examining longer lag periods may be needed to understand more fully the O_3-related health outcomes. The most significant associations between O_3 concentrations and mortality and respiratory hospitalization were observed with 0-day and 1-day lags. In the 95 U.S. communities study (Bell et al., 2004) and the related U.S. study of the 19 largest cities (Huang et al., 2005), the risk estimated over multiple days (cumulative lag of 0 to 6 days) using distributed lag models indicated a strong effect of O_3 on mortality. It should be noted that when there is a pattern of effects across lag periods, selecting a single-day lag effect estimate may underestimate the overall effect size and not fully capture the risk distributed over adjacent days. Longer averaging periods may aid in characterizing cumulative O_3-related effects over several days; however, interpreting these results may not be straightforward.

8.4.3.4 Concentration-Response Functions and Threshold

Ozone concentration-response relationships have been explored in several studies with various health outcomes, including mortality, hospitalizations, emergency department visits, lung function, and respiratory symptoms. While some studies found no threshold for O_3 health effects, others have found that a very low-level threshold may be present. A study by Kim et al. (2004) specifically examined the presence of a threshold in O_3-mortality effects in Seoul, Korea by analyzing data using a log linear GAM (linear model), a cubic natural spline model (nonlinear model), and a B-mode splined model (threshold model). An estimated threshold value of 47 ppb was observed for 1-h daily max O_3. This study further observed that if a threshold truly exists, the use of log-linear models may underestimate the O_3 effect on mortality at levels above the threshold.

It should be noted that exposure measurement error may reduce the ability to detect a threshold in O_3 population studies that used ambient O_3 concentrations as an indicator of personal ambient exposure. In addition, due to the variability in individual sensitivities, a threshold may not be seen at the population level. The limited evidence suggests that if there is a

threshold level in O_3 health effects, it is likely near the lower limit of ambient O_3 concentrations in the United States.

8.4.3.5 Consistency of Findings Across Epidemiologic Studies

Most of the multicity and meta-analyses studies consistently found positive associations between O_3 and mortality. Generally consistent O_3 effects on hospitalizations and various respiratory health outcomes also were found. Ozone concentrations tend to be spatially variable in urban areas. The geographic variability in O_3 concentrations and personal exposures may contribute to the heterogeneity in observed O_3 health effects. The degree of influence of the geographic variability on heterogeneity in effects tend to vary by study, as study design affects different aspects of exposure (e.g., time period and duration of exposure). In addition, some of the observed heterogeneity of O_3 effects may be partially attributable to the use of centrally-located ambient monitors to assess exposure. There may be differences in relative personal exposures to O_3 by region due to varying factors, such as use of air conditioning and activity patterns, that affect the relationship between personal exposure and ambient concentrations.

Among the field studies, various respiratory health outcomes were examined, including PEF, other spirometric parameters, respiratory symptoms, and medication use. One field study investigated the O_3 effect in asthmatic children living in eight urban cities in the U.S. (Mortimer et al., 2002). In the analysis pooling data from all eight cities, O_3 was associated with a decrement in morning PEF for a 5-day cumulative lag period. The percent changes in PEF were quite homogenous, with values ranging from −1.08% for Washington, DC to −1.71% for St. Louis. Ozone also was associated with an increased incidence of morning symptoms in the pooled analysis (Mortimer et al., 2002).

More than 80% of the O_3-mortality estimates from the various studies conducted in North America, South America, Europe, and Australia fell between 0.5 and 5% excess risk per 40 ppb increase in 1-h max O_3 using year-round data. In general, the O_3-mortality estimates were greater when using summer only data compared to year-round data. Though not all statistically significant, most of the O_3-mortality estimates were greater than zero, indicating a positive relationship between O_3 exposure and mortality. Three recent mortality meta-analyses that included both U.S. and non-U.S. studies found consistent all-year combined point estimates of 1.6 to 1.8% excess risk per 40 ppb increase in 1-h max O_3. The O_3 risk estimates from the

numerous hospitalization and emergency department visit studies were generally larger in magnitude and more variable from study to study compared to the mortality studies.

Because differences in study design, population, and data analysis may affect risk estimates, one study investigated the geographic heterogeneity of O_3 effects in multiple cities using standardized methods. In the pooled analysis of 95 U.S. communities using all available data, intercity heterogeneity was observed among the 95 communities, which the authors noted as plausible given the city-specific differences in pollution characteristics, the use of air conditioning, time-activity patterns, and socioeconomic factors (Bell et al., 2004).

Overall, the epidemiological studies indicate that there are associations between acute O_3 exposures and morbidity and mortality outcomes in numerous locations across the United States. In general, fairly consistent O_3 effect estimates were observed for the various health outcomes, including pulmonary function, symptoms, hospitalization, and mortality.

8.4.3.6 Summary and Conclusions for Epidemiology Findings

Discussions presented in the previous sections evaluated the merits of the epidemiologic studies to derive judgments about potential causal relationships between O_3 exposures and health outcomes. These evaluations were carried out in the context of the criteria listed in Section 8.2.2. Information with regard to one of the criteria, i.e., coherence and biological plausibility, is discussed in the next section, which undertakes to provide an integrated analysis of the biological evidence from human and animal toxicology studies with the epidemiologic evidence.

The results from the new field/panel studies evaluated in this document provide additional evidence for likely causal relationships being reflected by significant associations between acute O_3 exposure and decrements in lung function. Several new studies also associated acute O_3 exposure with increased respiratory symptoms and use of asthma medication in children and, in some cases, adults. New population based time-series studies also indicated a positive association between acute O_3 exposure and respiratory morbidity indexed by hospital admissions and emergency visits, especially in season-stratified data. The results from large multicity studies and several meta-analyses consistently suggest an elevated risk of mortality for acute exposure to O_3. Additional analyses evaluating the potential susceptibility of individuals with preexisting cardiovascular disease is rather limited. Analysis of the data from chronic mortality and morbidity studies indicate possible associations between O_3 and seasonal changes in lung

function; but, overall, the strength of the evidence does not allow establishment of a likely causal relationship between chronic O_3 exposure and these health outcomes.

Issues regarding strengths of models used in air pollution epidemiology were carefully considered. There have been improvements in the modeling to adjust for potential confounding variables, including temporal trends, meteorological factors, and copollutants. However, more sensitivity analyses would still be useful to examine the extent of adequate adjustment for confounding by these factors. Results from multipollutant models indicate that copollutants, e.g., PM, generally do not confound the association between O_3 and acute health outcomes, suggesting an independent effect of O_3. The limited evidence suggests that if there is a threshold level in O_3 health effects, it is likely near the lower limit of U.S. ambient O_3 concentrations.

In conclusion, the epidemiological evidence continues to support likely causal associations between acute ambient O_3 exposures and increased risk of acute respiratory morbidity and mortality, based on the assessment of strength, robustness, and consistency of results reported from numerous studies reviewed in Chapter 7. There is a lack, however, of sufficient evidence by which to convincingly establish a positive association between chronic O_3 exposure and increased respiratory morbidity and mortality. Additional investigations are needed to further understand the health effects resulting from long-term O_3 exposure.

8.5 BIOLOGICAL PLAUSIBILITY AND COHERENCE OF EVIDENCE FOR OZONE-RELATED HEALTH EFFECTS

This section is organized to integrate epidemiologic studies with toxicologic and mechanistic information obtained from controlled human exposure studies and animal toxicology studies for the two major health endpoints, morbidity and mortality reported to be associated with either short- or long- term exposure to ambient O_3. Morbidity associations have been subdivided into (a) school absenteeism, (b) emergency department visits for asthma, and (c) hospitalizations due to respiratory and cardiovascular illnesses. Mortality associations were also critically evaluated to understand risk estimates for total nonaccidental mortality and mortality in specific susceptible populations. The discussion in each subsection concisely summarizes pertinent key information and then presents the plausibility of effects being reasonably attributed to the conclusions derived for the endpoint assessed. To facilitate an easy discussion and to recapitulate various biological endpoints that have been investigated in human

and animal toxicology studies (see Chapters 4, 5, and 6), the first subsection addresses the plausible interpretative assessments from the salient observations from experimental studies.

Several criteria listed in Section 8.4.2 are used in evaluating the available scientific support for conclusions regarding potential causal relationships between O_3 exposure and specific types of health outcomes. In addition to those criteria addressed in the preceding discussion of epidemiological evidence, certain other critical evaluation measures must be considered to ensure that these observations are biologically relevant and consistent with experimentally demonstrated biological mechanisms of action. For this assessment, the ensuing discussion on biological plausibility and coherence considers (a) the extent to which available epidemiological evidence logically ties to a range of relevant health endpoints (from cardiopulmonary physiological changes to morbidity to mortality) and (b) whether available toxicological and biochemical evidence supports plausible causal relationships for the observed epidemiological associations.

The ensuing discussion on biological plausibility and coherence also considers the following criteria for integrated assessment: (a) adequateness of the statistical power of the epidemiological studies to establish evidence for associations, (b) the location (urban vs. rural), (c) seasonal pattern vs. all year O_3 levels, (d) socioeconomic status of the population, and (e) the lag days (used in the epidemiologic analysis), considered in relation to the time course of likely biological mechanisms potentially implicated in the process.

Animal-to-Human Extrapolation Issues

The physiological and biochemical observations reported in Table 8-1 represent the knowledge base available from toxicological studies in humans and animals that underlie the biological alterations that govern acute O_3-induced health effects. This table is generated from the experimental database (see Annexes for Chapters 5 and 6 for experimental details) that utilized exposure regimens of varied concentration and duration that are environmentally relevant. As noted in the earlier section, most of the observed acute O_3 effects are transient and attenuate over time. However, the time-line for resolution of many of these physiological and biological parameters in normal and human subjects with underlying cardiopulmonary diseases follow different profiles as presented in Figure 8-9. Alterations in the cellular and molecular profiles observed in human airway epithelium upon acute exposure to O_3 evolve over time

Table 8-1. Acute O_3-induced Physiological and Biochemical Changes in Human and Animals

Physiological/Biochemical Alterations	Human Exposure Studies [1,2]	Animal Toxicology Studies [3,4]
Pulmonary Function:	↓ FEV_1 ↑ Frequency of breathing (rapid, shallow) ↓ inspiratory capacity (cough, breathing discomfort, throat irritation, wheezing) Mild bronchoconstriction	↓ FEV_1 ↑ Frequency of breathing (rapid, shallow) ↓ inspiratory capacity
Airway Responsiveness:	↑ (neuronal involvement) Change in lung resistance	↑ (vagal mediation) Change in lung resistance
Inflammation:	Yes ↑ inflammatory mediators	Yes ↑ inflammatory mediators
ROS	↑	↑
Host Defense:	↑ particle clearance ↑ permeability ↓ AM phagocytosis	↑ particle clearance ↑ permeability ↓ clearance of bacteria ↑ severity of infection ↑ mortality & morbidity
Lung injury: Morphology	Yes	Yes
Susceptibility:	Age, Inter individual variability Disease status Polymorphism in certain genes being recognized	Species specific differences Genetic basis for susceptibility indicated
Cardiovascular Changes:	Impairment in arterial O_2 transfer Ventilation-perfusion mismatch (suggesting potential arterial vasoconstriction)	Heart rate variability (HRV) ↓ core body temperature ↑ ANF Role for PAF indicated increased pulmonary vascular resistance

[1] Controlled chamber exposure studies in human volunteers were carried out for a duration of 1-6.6 h with O_3 concentration in the range of 0.08-0.4 ppm with intermittent exercise.
[2] Data on some of the biochemical parameters were obtained from in vitro studies using cells recovered from BALF.
[3] Responses were observed in animal toxicology studies with exposure for a duration of 2-72 h with O_3 concentration in the range of 0.1-2.0 ppm.
[4] Various species (mice, rat, guinea pigs and rabbit) and strains.

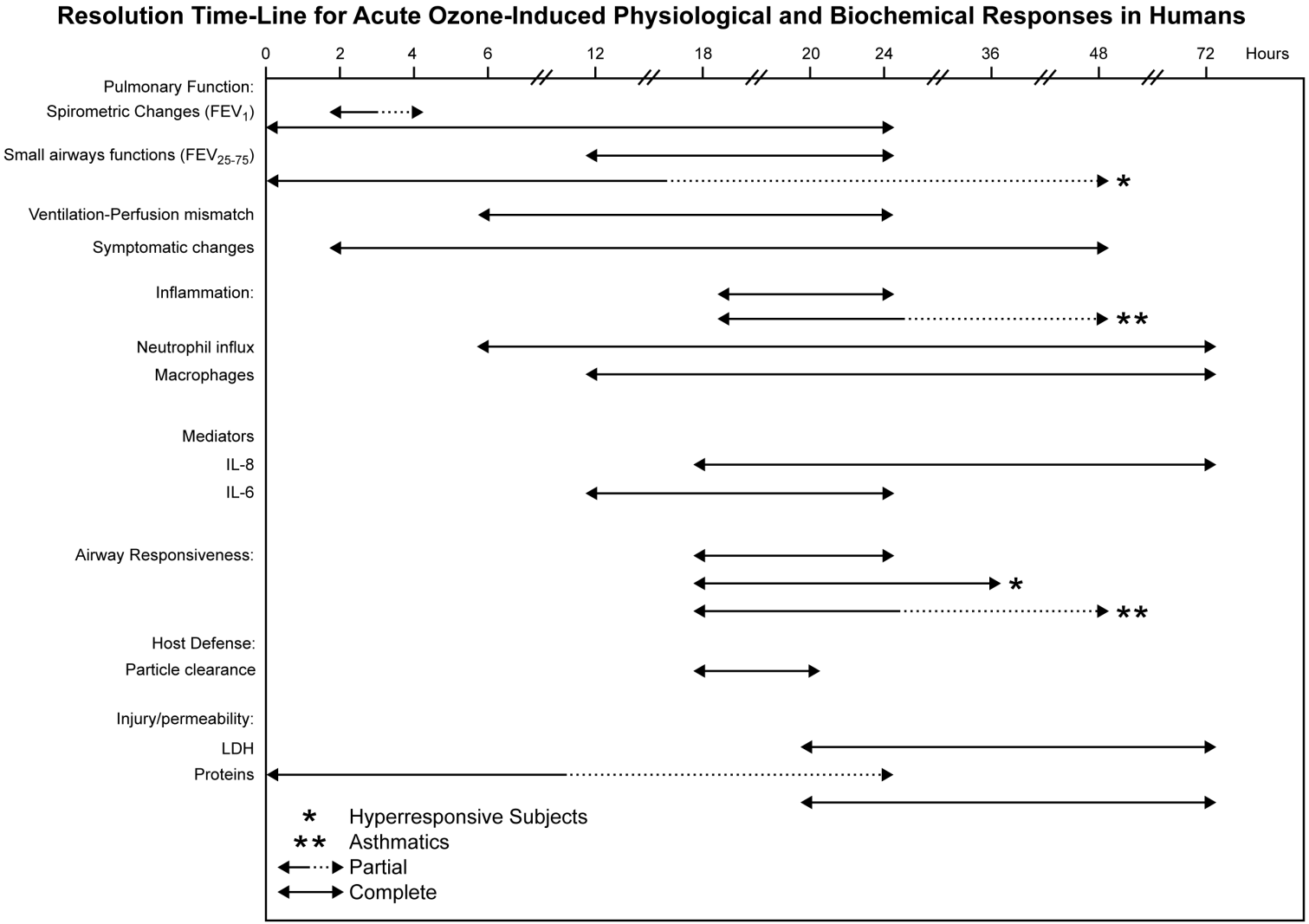


Figure 8-9. Resolution time-line for the physiological and biochemical parameters are derived from studies reported in Chapter 6 and Chapter 6 Annex.

(Figure 8-10), and the knowledge of this profile is valuable in assessing biological plausibility to integrate across evidence for various health endpoints.

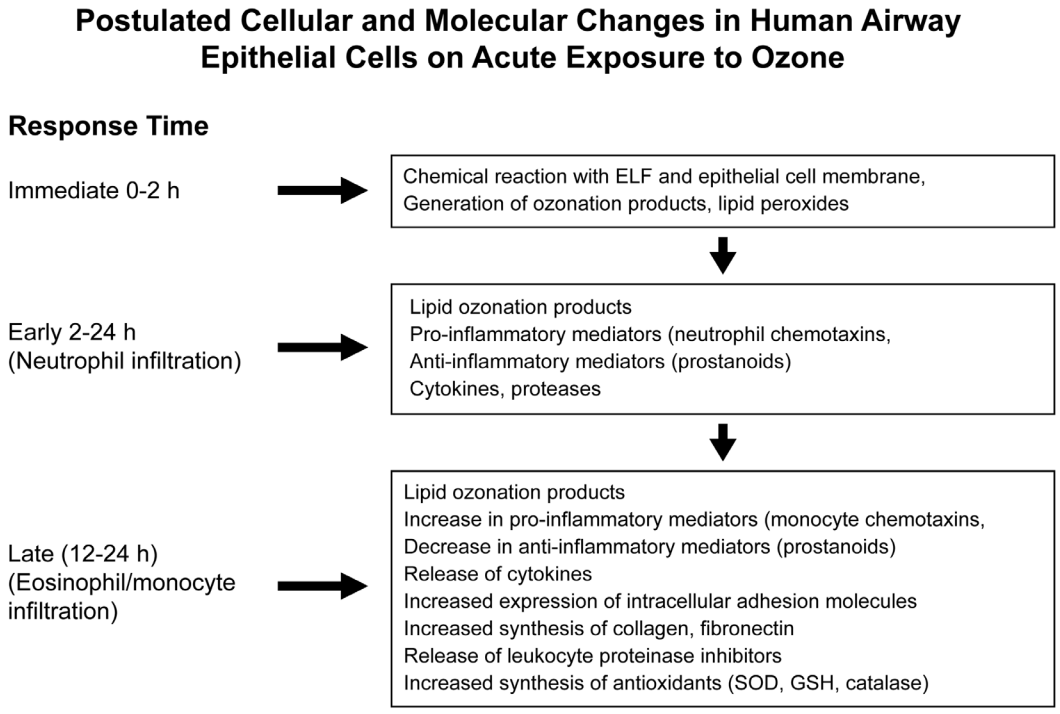


Figure 8-10. O_3-induced cellular and molecular changes and their evolution depicted here is derived from the data reported in Leikauf et al. (1995) and Mudway and Kelly (2000).

Basic similarities in physiological, biochemical, and pathological processes that exist between human and animal species are derived from the high degree of genome sequence homology that exists across species. This homology reinforces the significance of knowledge gained on the initiation, progression and treatment regimes for various disease processes across animal species. This homology is also apparent in acute O_3-induced effects, especially on the respiratory tract of human and animal species as presented in Table 8-1 and Figures 8-9 and 8-10. The commonality of phenomenon observed in humans and rats with regard to respiratory system effects (in terms of spirometry, ventilatory response, host defense and inflammation) and their attenuation adds strength to animal-human extrapolations. Such similarities observed at

higher levels of cellular organization (neutrophilic inflammation, macrophage phagocytosis processes) have increased the value and importance of animal studies in generating important data that is impossible to collect in human studies but which may corroborate both clinical and epidemiologic studies.

Quantitative extrapolation involves a combination of dosimetry, end point homology and species sensitivity, particularly in the case of exposure and health outcome analyses. However, extrapolation models have not been completely validated and, therefore, uncertainties do exist. Based on inflammatory markers in BALF, a 2 ppm O_3 exposure in nonexercising rats approximates to a 0.4 ppm exposure in exercising humans despite species-specific differences (Hatch et al., 1994). This observation lends support to the use of some of the animal toxicology data derived from relatively high O_3 concentration exposure regimens in understanding putative molecular changes associated with acute O_3 exposure in humans. Similarly, the presence of apparent O_3-induced lesions in animals from chronic O_3 exposure studies (12 to 24 months) indicate functional defects that may potentially provide a means to facilitate more direct assessments of long-term health outcomes in humans.

8.5.1 Acute Ozone Exposure-Induced Health Effects

As noted in Section 8.4.2, several new epidemiologic (field/panel) studies show positive associations between short-term exposure to ambient O_3 and human health effects. These health effects as evaluated include school absenteeism, decline in lung function, increased use of asthma medication, and increased hospitalization, especially among individuals with asthma or certain known cardiopulmonary or cardiovascular diseases (see Chapter 7). The patterns of physiological and biochemical alterations reviewed earlier (see Figures 8-9, 8-10 and Table 8-2) tend to support certain hypotheses regarding underlying pathological mechanisms in the development of respiratory effects reported in the epidemiologic studies. Some of these mechanisms (see Table 8-2) include (a) decrements in lung function (capacities and volume), (b) bronchoconstriction, (c) increased airway responsiveness, (d) airway inflammation, (e) epithelial injury, (f) immune system activation, (g) host defense and (h) sensitivity of an individual such as age, genetic susceptibility and the extent of tolerance resulting from previous exposures. The time sequence, magnitude, and overlap of these complex events, both in terms of development

and recovery (see Figure 8-9 and 8-10), indicate the difficulties associated with the interpretation of biological plausibility associated with the cardiopulmonary health effects.

Respiratory Health Effects

Controlled human exposure studies have clearly demonstrated the following three types of respiratory responses to acute O_3 exposures: (1) irritative cough and substernal chest pain upon inspiration; (2) decrements in FVC and FEV_1 due to decreased inspiratory capacity rather than airways obstruction and (3) neutrophilic inflammation of the respiratory tract. Susceptibility or sensitivity to these effects were observed even among a carefully selected homogeneous study population. The sources of this heterogeneity are uncertain. As discussed in the earlier section, changes in baseline levels of various responses, the lag in the recovery phase and the role of residual defects in these mechanisms in hyperresponsive individuals suggest potential for increased health effects in cardiopulmonary compromised individuals such as people with asthma, COPD and cardiovascular diseases. Recent research has emphasized further characterization of the mechanisms and consequences of O_3-induced pulmonary function and inflammatory responses. In addition, animal studies indicate morphological changes associated with acute O_3 exposures.

Ozone-induced altered breathing patterns (rapid shallow breathing) observed in controlled human exposure studies and animals occur without significantly affecting minute ventilation, suggesting compensatory changes in breathing pattern. Such a shift in breathing pattern diminishes deep lung penetration of O_3. Breathing pattern is modulated by changes in peripheral mechanisms, such as direct or indirect stimulation of lung receptors and bronchial C-fibers. The activity of these afferents is integrated with input from sensory pathways and thus determines the depth and frequency of breathing. Stimulation of bronchial C-fibers along with inhibition of inspiration through local axon reflexes can induce neurogenic inflammation via tachykinins and other proinflammatory neuropetides. Ozone-induced increases in the levels of neuropeptide substance P observed in the BALF of human subjects suggests potential neurogenic involvement in vascular permeability, plasma protein extravasation, bronchoconstriction and mucus secretion (Solway and Leff, 1991). Similar neurogenic involvement due to vagally mediated stimulation of C-fibers seen in animal toxicology studies support O_3-induced bronchial hyperresponsiveness observed in humans.

An extensive database of animal, human, and in vitro studies supports the conclusion that O_3 interacts with airway epithelial cell membranes and lining fluid to form lipid ozonation products and ROS. These reactive products initiate a cascade of events leading to oxidative stress, injury, inflammation, airway epithelial damage and increased alveolar permeability to vascular fluids. Inflammation is the outcome of host response to injury and usually resolves completely. Continued irritant challenge may evolve into a chronic inflammatory state with simultaneous alterations in lung structure and function, leading to diseases such as fibrosis and emphysema. Continued inflammation can also alter the lung's ability to respond to infectious agents, allergens, and toxins. Acute inflammatory responses to O_3 exposure are well documented in humans and animals. As presented in Figure 8-10, the early inflammatory response to O_3-induced lung injury is apparent in human subjects within 3 h postexposure. This initial neutrophilic inflammatory response phase is characterized by increases in PMNs in the BALF along with increased levels of inflammatory mediators such as interleukins, prostaglandins and complement component C3a. In vitro studies using human and animal lung cell culture systems have further examined the involvement of various inflammatory mediators and in some instances their downstream signaling pathways. The late inflammatory phase in the lung is characterized by increased levels of monocytes and eosinophils and respective mediators such as cytokines, leukotrines, proteinases, and ROS.

Disruption of the lung's blood barrier by O_3 resulting in vascular permeability changes and plasma protein extravasation. BALF analysis on plasma influx markers such as albumin, proteins, immunoglobulins, and epithelial cell damage markers such as LDH indicate O_3-induced lung epithelial injury. Ozone-induced lung injury and subsequent disruption of the airway epithelial barrier has been implicated in increased mucociliary clearance of particles observed in controlled human studies. Analogously, animal toxicology studies (see Chapter 5) have reported increased mortality to bacterial and viral infections subsequent to O_3 exposure and also increased clearance of particles.

Controlled O_3 exposure studies of healthy humans have indicated a large degree of intersubject variability. The spirometric and symptomatic responses are highly reproducible within the subject; but, within a group, pulmonary function measurements varied from −4% to 56%. These are likely also to be genetic function, but as yet this factor remains of uncertain importance. Analysis of personal characteristics such as age, height, smoking history and

allergies indicated age as an important contributor. Based on FEV_1 measurements as criteria, young adults (18-25 yrs) were found to be more sensitive to O_3 than children or adults. Sensitivity to O_3 had been found to decline with older age (>60 yrs), but it should be noted that the baseline values for various measurable pulmonary functions are different in this group of population.

Cardiovascular Health Effects

There exist few experimental studies in animals and humans that have investigated potential cardiovascular effects with acute O_3 exposures. Ozone induces lung injury, inflammation, and impaired mucociliary clearance with a host of associated biochemical changes all leading to increased lung epithelial permeability. As discussed in the Section 5.4.2 the generation of lipid ozonation products and ROS in lung tissue can influence the pulmonary hemodynamics and ultimately cardiovascular system. Recent reports of interaction of O_3 with cholesterol in the lung surfactant and the generation of highly reactive products such as oxysterols and β-epoxide have indicated a role for cardiovascular effects and atherosclerosis (Pulfer and Murphy, 2004). Ozone-induced changes in heart rate variability, edema of heart tissue, and increased tissue and serum levels of ANF observed in animal toxicology studies lend support to potential cardiovascular effects of acute O_3 exposures. Such effects resulting from stimulation of airway irritant receptors, c-fiber activation, may result from either local or central nervous system involvement. The observation of O_3 -induced changes in the alveolar-arterial oxygen transfer in controlled human exposure studies on subjects with hypertension indicates potential complex ANF effects that need to be investigated further.

Coherence Between Epidemiologic and Experimental Evidence for Acute Respiratory and Cardiovascular Effects

Epidemiologic studies, indicate a positive association between exposure to ambient O_3 and declines in lung function in children and those with cardiopulmonary diseases such as asthma. Meta-analyses of children in summer camp studies (Kinney et al., 1996) and a multicity study by Mortimer et al (2002) supports the earlier observations that children and asthmatics are particularly susceptible to ambient O_3. This association based on decrements in lung function and exacerbating pulmonary disease symptoms suggests that O_3 exposures may result in

increased use of medication in children and asthmatics. Studies that evaluated relationships between exposure to ambient O_3 and school absences in children provide corroborative positive associations. An increased incidence of disease exacerbations in people with cardiovascular diseases could not clearly be established.

Increased incidence of emergency department visits due to specific respiratory illness (e.g., asthma) and hospitalization due to specific causes (e.g., respiratory or cardiovascular disease exacerbation) reported in various studies discussed in Chapter 7 (depicted in Figure 8-1 for studies from the United States and Canada) suggest a causal association supported by animal toxicology data. This association becomes more apparent when the data are analyzed for the influence of seasonal differences in ambient O_3 levels. Several controlled clinical studies reviewed in the 1996 O_3 AQCD on atopic and asthmatic subjects have not shown enhanced responsiveness to acute O_3 exposure compared to healthy subjects. The majority of the newer studies reviewed in Chapter 6 continue to suggest that asthmatics are as sensitive as, if not more sensitive than, normal subjects in manifesting O_3-induced pulmonary function decrements.

Ozone-induced increases in neutrophils, protein, and IL-8 were found to be significantly higher in the BALF from asthmatics compared to healthy subjects. Similarly, subjects with allergic asthma exhibited increased airway responsiveness to inhaled allergens upon acute O_3 exposure. Consistent with these changes it is suggested that asthmatics will be more sensitive to small airway effects of ambient O_3. Asthmatics present a differential response profile for the cellular, molecular, and biochemical parameters (Figure 8-10) in response to acute O_3 exposure. Increases in O_3-induced nonspecific airway responsiveness incidence and duration could have important clinical implications for asthmatics.

Bronchial constriction following provocation with allergens presents a two-phase response. The early response is mediated by release of histamine and leukotrienes that leads to contraction of smooth muscle cells in the bronchi, narrowing the lumen and decreasing the airflow. In asthmatics, these mediators also attract accumulation eosinophils, followed by production of mucus and a late-phase bronchial constriction and reduced airflow. Holz et al (2002) reported an early phase response in subjects with rhinitis after a consecutive 4-day exposure to 0.125 ppm O_3 that resulted in a clinically relevant (>20%) decrease in FEV_1. Allergen challenge in mild asthmatics 24 h postexposure to 0.27 ppm O_3 for 2 h had been found to significantly increase

eosinophil counts in BALF compared to healthy subjects (Vagaggini et al., 2002). Epithelial cells from the mucosal biopsies of allergic asthmatics indicated significant increase in the expression of IL-5, IL-8 and GM-CSF suggesting increased neutrophilic inflammation compared to healthy subjects (Bosson et al., 2003). In vitro exposure studies (0.1 ppm O_3) of nasal epithelial cells from atopic asthma patients were found to release significantly greater amounts of neuropeptides, neurokinin A and Substance P, suggesting activation of neurogenic inflammation (Schierhorn et al., 1999). Collectively, these observations suggest that O_3 exposure may exacerbate pre-existing allergic asthma. People with allergic asthma may represent a segment of the population reported to have increased symptoms of respiratory illness exacerbations, emergency department visits, and hospital admissions in epidemiologic studies.

Recent population time-series studies (Figure 8-5) have also indicated a potential association between acute O_3 exposure and cardiovascular hospitalization. Additional well-designed time-series studies are needed to evaluate cardiovascular morbidity more specifically associated with acute O_3 exposure. The intimate hemodynamic and neurohumoral relationships, and potential cardiac consequences of pulmonary insults are well recognized. Two important observations in human clinical studies: (1) O_3-induced impairment in alveolar-arterial oxygen transfer (Gong et al., 1998) and (2) O_3-induced ventilation-perfusion mismatch (Foster et al., 1993, 1997) are consistent with potential cardiovascular impacts of O_3. If such relationships are validated, they will aid in our understanding the role of O_3-induced reductions in gas exchange and oxygen saturation in COPD patients with already compromised gas exchange process. Cardiovascular disease conditions and COPD are most common among old age groups. Thus, age-associated pulmonary function deficiencies in older people would add an additional burden with respect to O_3-induced effects. The recent observations of air pollution-induced vasoconstriction in controlled human exposure studies by Brook et al. (2002) suggest a possible role for O_3.

Animal toxicology studies indicate acute O_3-induced microvascular leakage and subsequent edema of airways in guinea pigs (Inoue et al., 1997) and increased baseline values for total vascular resistance in rabbit pulmonary vessels in ex vivo studies (Delaunois et al., 1998). These observations support the possibility of potential cardiovascular effects.

8.5.2 Chronic O_3 Exposure-Induced Health Effects

The effects of chronic O_3 exposure in humans have been addressed primarily with cross-sectional epidemiologic studies. Due to lack of precise information on exposure, the possibility of selection bias and the difficulty of controlling for confounders, these findings are inconclusive. However, several new longitudinal epidemiological studies have evaluated the potential associations between chronic exposure to O_3 and morbidity and mortality (see Section 7.5). These studies suggest that long-term exposure may be related to changes in lung function, increased incidence of asthma, mortality, and, possibly, lung cancer. However, based on available evidence, no definitive relationship could be established between chronic O_3 exposure and these health outcomes. There are no data available from controlled human chamber studies that evaluated chronic exposure regimens.

The lack of adequate data from epidemiologic and clinical studies in human has directed attention to the results from chronic exposure studies in animals. Earlier chronic animal studies employed traditional exposure designs using chronic stable exposures. Later studies have attempted to incorporate design features that mimic diurnal and seasonal pattern of O_3 exposure and realistic exposure concentrations. Studies on monkeys that compared these two designs reported increased airway pathology with the latter design. Persistent and irreversible effects observed in chronic animal toxicology studies indicate the need for complementary human data from epidemiologic studies.

Animal toxicology data provide a clearer picture indicating that long-term O_3 exposure at levels found in the ambient air may have lasting effects. Chronic exposure studies in animals have reported biochemical and morphological changes suggestive of irreversible long-term O_3 impacts on the lung. Some of the studies in rats (0.5-1.0 ppm O_3 for 6 h/day) for 20 months and monkeys (0.61 ppm) for one year noted increased deposition of collagen and thickening of the CAR of the deep lung. Differences in this degree of lung damage have been observed with continuous exposure and seasonal pattern. A long term study of infant rhesus monkeys exposed to simulated seasonal O_3 (0.5 ppm 8 h/day for 5 days every 14 days for 11 episodes) resulted in remodeling in the distal airways, abnormalities in tracheal basement membrane, accumulation of eosinophils in conducting airways and decrements in airway innervation. Earlier studies in rats following seasonal episodic profiles also showed small, but significant, decrements in lung function that were consistent with focal fibrinogenesis in the proximal alveolar region. On the

other hand, chronic O_3 exposures in a range of 0.5 to 1.0 ppm induce epithelial hyperplasia that disappears in a few days, and the weight of evidence from new experimental animal studies (using non-lifetime exposures) does not support ambient O_3 as being a pulmonary carcinogen.

Collectively, the evidence from animal studies strongly suggest that O_3 is capable of damaging the distal airways and proximal alveoli, resulting in lung tissue remodeling leading to apparent irreversible changes. Compromised pulmonary function and structural changes due to persistent inflammation may exacerbate the progression and development of chronic lung disease.

8.5.3 Mortality-Related Health Endpoints

An extensive analysis of population time-series studies that evaluated the air pollution related mortality risk estimates presented in Section 7-4 utilized data from single and multicity studies from around the world. Mortality risk estimates derived from studies in U.S. and Canada coupled with meta-analyses (Figures 8-6 and 8-7) all indicate an elevated risk for mortality on acute O_3 exposure after adjustment for the influence of season and PM (Figure 8-7) . Meta-analyses of large U.S. multicity studies also suggest a positive association. Mortality risk estimates derived from the studies that analyzed PM as a potential confounder suggest that the reported estimates are not attributable to confounding by PM. Several single-city studies that specifically evaluated the relationship between cardiovascular mortality and O_3 exposure also indicated a positive association.

The epidemiology results outlined above for mortality suggest a pattern of effects that may be biologically germane to interpretation of its causality, but our knowledge about potential underlying mechanisms remains very limited and suggests a need for further experimental support. The majority of the physiological and biochemical parameters evaluated both in human clinical and animal toxicology studies (Table 8-1; Figure 8-9) suggest a relatively transient nature for O_3-induced biochemical perturbations. Most effects attenuate over time, depending on the preexisting pathophysiology. One can hypothesize a generic pathway of O_3-induced lung damage, potentially involving oxidative lung damage with subsequent inflammation and/or decline in lung function leading to respiratory distress.

Recent analysis of third National Health and Nutrition Examination Followup study data indicated that about 20% of the adult population have reduced FEV_1 values indicative of

impaired lung function. The majority of these individuals have COPD, asthma or fibrotic lung disease (Mannino et al., 2003). These cardiopulmonary disease conditions are associated with persistent low-grade systemic inflammation. It has also been reported that patients with COPD are at increased risk for cardiovascular disease. Lung disease with underlying inflammation may also link to low-grade systemic inflammation associated with atherosclerosis. These effects in disease are independent of cigarette smoking (Sin et al., 2005). Lung function decrements in cardiopulmonary disease has also been associated with inflammatory markers such as C-reactive protein (CRP) in blood. In fact, at the population level, individuals with the lowest FEV_1 have the highest levels of CRP, while those with highest FEV_1 have the lowest values for CRP (Mannino et al., 2003; Sin and Man, 2003). The complex, physiological and biochemical perturbations that exist simultaneously (Figure 8-9 and 8-10) subsequent to acute exposure to O_3 may tilt the biological homeostasis mechanisms leading to adverse health effects in people with compromised cardiopulmonary systems. However, no experimental data are available at this time to support such a hypothesis as being operational in O_3-induced cardiovascular mortality observed in the epidemiological studies. It is possible that reevaluation of some of the epidemiological panel studies that reported changes in CRP in the context of air pollution mortality evaluations focused on PM with correct adjustments for O_3 may shed some light on this potential relationship.

8.6 SUSCEPTIBILITY FACTORS

Many factors such as age, gender, disease, nutritional status, smoking, and genetic variability may contribute to the differential effects of environmental pollutants, including O_3. Genetic factors, such as single nucleotide polymorphisms (SNPs) and developmental defects, can contribute to innate susceptibility, while acquired susceptibility may develop due to personal habits (smoking, diet, exercise) and other risk factors such as age, gender, pregnancy, and copollutants. However, the available information from animal toxicology and epidemiologic studies did not provide sufficiently clear scientific evidence by which to confidentially identify and/or associate any specific factor as contributing to adverse health effects of O_3 (U.S. Environmental Protection Agency, 1996a). However, advances in available research results

since then have improved our ability to delineate likely susceptible or vulnerable populations at increased risk for O_3-induced health effects.

New animal toxicology studies using various strains of mice and rats have identified O_3-sensitive and resistant strains and illustrated the importance of genetic background in determining O_3 susceptibility. Using subacute low exposure regimen (0.3 ppm O_3, 48h) studies on inbred strains that have been designated as inflammation prone or resistant, Kleeberger et al., (1997) identified the pro-inflammatory cytokine gene, *Tnf*-α, as a susceptibility gene. Further characterization of this model indicated a role for TNF receptors (TNFR1, TNFR2) in O_3-induced pulmonary epithelial injury and inflammation (Cho et al., 2001). Studies on five inbred strains of mouse with differing response to O_3 exposure (acute high dose or low dose continuous exposure for 3 days), reported a protective role for clara cell secretory protein (CCSP) against O_3-induced oxidative damage (Broeckaert et al., 2003; Wattiez et al., 2003). The role for these genes and/or their orthologs in human susceptibility to O_3 exposure is yet to be examined.

Apart from age at the time of exposure, controlled human exposure studies have also indicated a high degree of interindividual variability in some of the pulmonary physiological parameters. Recent studies by David et al. (2003) and Romieu et al. (2004) reported a role for genetic polymorphism in antioxidant enzymes and genes involved in inflammation to modulate pulmonary function and inflammatory responses to O_3 exposure. Similar to mouse studies referred above, polymorphism in *Tnf*-α has been implicated in O_3-induced lung function changes in healthy, mild asthmatics and individuals with rhinitis. These observations suggest a potential role for these markers in the innate susceptibility to O_3, however, the validity of these markers and their relevance in the context of prediction to population studies need additional experimentation.

Biochemical and molecular parameters extensively evaluated in these experiments were used to identify specific loci on the chromosomes and, in some cases, to relate the differential expression of specific genes to biochemical and physiological differences observed among these species. Utilizing O_3-sensitive and O_3-resistant species, it has been possible to identify the involvement of AHR and inflammation processes in O_3 susceptibility. However, most of these studies were carried out using relatively high doses of O_3, making the relevance of these studies questionable in human health effects assessment. No doubt, the molecular parameters identified in these studies may serve as useful biomarkers with the availability of suitable technologies and,

ultimately, can likely be integrated with epidemiological studies. Interindividual differences in O_3 responsiveness have been observed across a spectrum of symptoms and lung function responses do not yet allow identification of important underlying factors, except a significant role for age.

8.6.1 Preexisting Disease as a Potential Risk Factor

People with preexisting pulmonary disease may be at increased risk from O_3 exposure. Altered physiological, morphological and biochemical states typical of respiratory diseases like asthma, COPD and chronic bronchitis may render people sensitive to additional oxidative burden induced by O_3 exposure. Based on studies assessed in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996a), asthmatics appear to be at least as, or more, sensitive to the acute effects of O_3 as healthy nonasthmatic subjects. The new results reviewed in Chapters 6 and 7 of this document from controlled exposure and epidemiologic studies also suggest that asthmatics are a potentially sensitive subpopulation for O_3 health effects.

A number of time-series epidemiologic studies have reported increased risk in study subsets of individuals with preexisting lung diseases, among which tend to implicate asthmatics as potentially susceptible individuals. The epidemiologic studies of acute exposure to O_3 discussed in Section 8.4.2 indicate increased risk for exacerbation of disease symptoms during the warm season.

Newly available human exposure studies by Stenfors and coworkers have shown differences regarding PMN influx in BALF between asthmatics and healthy human subjects. In vitro studies (Stenfors et al., 2002) using nasal mucosal biopsies from atopic and nonatopic subjects exposed to 0.1 ppm O_3 found significant differences in the release of IL-4, IL-6, IL-8, and *TNF*-α. A subsequent study by the same group (Schierhorn et al., 2002) found a significant difference in the O_3-induced release of the neuropeptides neurokinin A and substance P from allergic patients, compared to nonallergic controls, suggesting increased activation of sensory nerves by O_3 in the allergic tissues. Another report from Bayram et al. (2002) using in vitro culture of bronchial epithelial cells recovered from atopic and nonatopic asthmatics indicated the existence of a significant difference in permeability (by measuring the paracellular flux of ^{14}C-BSA). Additional controlled O_3 exposure studies in human subjects with intermittent asthma (Hiltermann et al., 1999), and asthmatics (Basha et al., 1994; Scannell et al., 1996)

reported increased secretion of IL-8 suggesting increased neutrophilic inflammation in those subjects. Two studies (Jörres et al. 1996; Holz et al. 2002) observed increased airway responsiveness to repeated daily O_3 exposure to bronchial allergen challenge in subjects with preexisting allergic airway disease.

Newly available reports from controlled human exposure studies (see Chapter 6) utilized subjects with preexisting cardiopulmonary diseases such as COPD, asthma, allergic rhinitis, and hypertension. The data generated from these studies that evaluated pulmonary function changes in spirometry did not clearly find differences between filtered air and O_3 exposure in COPD and asthmatic subjects. However, the data on airway responsiveness, inflammation and various molecular markers of inflammation and bronchoconstriction indicate that people with atopic asthma and allergic rhinitis are potentially susceptible groups for O_3-induced adverse health effects. There was only one study with a limited number of subjects that evaluated the effects of O_3 exposure in hypertensive patients, and it did not find significant O_3-induced changes in clinical parameters, such as heart rate, blood pressure and ECG.

The observation of increased pathology in long-term animal exposure studies in the absence of observable physiological changes also suggests that chronic exposure may increase susceptibility to adverse health effects, but this needs to be validated via long-term epidemiologic studies.

8.6.2 Potential Public Health Impacts

Exposure to ambient O_3 is associated with various health outcomes, including increased incidence of cough, reduction in lung function, increased inflammation, and increased hospital admissions and mortality. In protecting public health, a distinction must be made between health effects that are considered "adverse" and those that are not. What constitutes an adverse health effect varies for different population groups, with some changes in healthy individuals not being viewed as adverse but those of similar type and magnitude in other susceptible individuals being seen as adverse. Hence, the definition of adversity of health effects will be an important issue in ultimately considering and describing the rationale for decisions concerning review of the O_3 NAAQS.

8.6.2.1 General Concepts Related to Defining of Adverse Health Effects

The official statement of the American Thoracic Society (ATS) published on "What Constitutes an Adverse Health Effect of Air Pollution?" (ATS, 2000) updated guidance for defining adverse respiratory health effects published fifteen years earlier (ATS, 1985) to address new investigative approaches used to identify the effects of air pollution and to reflect the concern for the impacts of air pollution on specific susceptible groups.

In the 2000 update, there is an increased focus on quality of life measures as indicators of adversity and also a more specific consideration of population risk. Exposure to air pollution that increases the risk of an adverse effect to the entire population is adverse, even though it may not increase the risk of any identifiable individual to an unacceptable level. For example, a population of asthmatics could have a distribution of lung function such that no identifiable single individual has a level associated with significant impairment. Exposure to air pollution could shift the distribution to lower levels that still do not bring any identifiable individual to a level that is associated with clinically relevant effects. However, this would be considered to be adverse because individuals within the population would have diminished reserve function and, therefore, would be at increased risk if affected by another agent.

Reflecting new investigative approaches, the ATS statement also describes the potential usefulness of research into the genetic basis for disease, including responses to environmental agents, that will provide insights into the mechanistic basis for susceptibility, and provide markers of risk status. Likewise biomarkers, that are indicators of exposure, effect or susceptibility, may someday be useful in defining the point at which a response should be equated with an adverse effect.

The 1996 O_3 AQCD provided information useful in helping to define adverse health effects associated with ambient O_3 exposure by describing gradation of severity and adversity of respiratory-related effects, and those definitions are reproduced and presented here as Tables 8-2 and 8-3. The severity of effects described in those tables and the approaches taken to define the adversity still appear to be valid and reasonable even in the context of the new ATS statement (ATS, 2000).

Table 8-2. Gradation of Individual Responses to Short-Term Ozone Exposure in Healthy Persons [a]*

Functional Response	None	Small	Moderate	Large
FEV_1	Within normal range (±3%)	Decrements of 3 to ≤10%	Decrements of >10 but <20%	Decrements of ≥20%
Nonspecific bronchial responsiveness [b]	Within normal range	Increases of <100%	Increases of ≤300%	Increases of >300%
Duration of response	None	<4 hours	>4 hours but ≤24 hours	>24 hours
Symptomatic Response	**Normal**	**Mild**	**Moderate**	**Severe**
Cough	Infrequent cough	Cough with deep breath	Frequent spontaneous cough	Persistent uncontrollable cough
Chest pain	None	Discomfort just noticeable on exercise or deep breath	Marked discomfort on exercise or deep breath	Severe discomfort on exercise or deep breath
Duration of response	None	<4 hours	>4 hours but ≤24 hours	>24 hours
Impact of Responses	**Normal**	**Normal**	**Mild**	**Moderate**
Interference with normal activity	None	None	A few sensitive individuals choose to limit activity	Many sensitive individuals choose to limit activity

[a] See text for discussion; see Appendix A for abbreviations and acronyms.
[b] An increase in nonspecific bronchial responsiveness of 100% is equivalent to a 50% decrease in PD_{20} or PD_{100}.
*This table is reproduced from the 1996 O_3 AQCD (Table 9-1, page 9-24) (U.S. Environmental Protection Agency, 1996a).

8.6.2.2 Estimation of Potential Numbers of Persons in At-Risk Susceptible Population Groups in the United States

Although O_3-related increases in individual health risks may appear to be small, they are likely significant from an overall public health perspective due to the large number of individuals in potential risk groups. Numerous subpopulations may be identified as having increased susceptibility or vulnerability to adverse health effects from O_3, including older adults, children, individuals with preexisting cardiopulmonary disease, those of lower socioeconomic status, and those with higher exposure levels. Clearly, the impact of O_3 on public health can be very extensive.

Table 8-3. Gradation of Individual Responses to Short-Term Ozone Exposure in Persons with Impaired Respiratory Systems [a]*

Functional	None	Small	Moderate	Large
FEV_1 change	Decrements of <3%	Decrements of 3 to ≤10%	Decrements of >10 but <20%	Decrements of ≥20%
Nonspecific bronchial responsiveness [b]	Within normal range	Increases of <100%	Increases of ≤300%	Increases of >300%
Airway resistance (SR_{aw})	Within normal range (±20%)	SR_{aw} increased <100%	SR_{aw} increased up to 200% or up to 15 cm H_2O/s	SR_{aw} increased >200% or more than 15 cm H_2O/s
Duration of response	None	<4 hours	>4 hours but ≤24 hours	>24 hours
Symptomatic	**Normal**	**Mild**	**Moderate**	**Severe**
Wheeze	None	With otherwise normal breathing	With shortness of breath	Persistent with shortness of breath
Cough	Infrequent cough	Cough with deep breath	Frequent spontaneous cough	Persistent uncontrollable cough
Chest pain	None	Discomfort just noticeable on exercise or deep breath	Marked discomfort on exercise or deep breath	Severe discomfort on exercise or deep breath
Duration of response	None	< 4 hours	>4 hours, but ≤24 hours	>24 hours
Impact of Responses	**Normal**	**Mild**	**Moderate**	**Severe**
Interference with normal activity	None	Few individuals choose to limit activity	Many individuals choose to limit activity	Most individuals choose to limit activity
Medical treatment	No change	Normal medication as needed	Increased frequency of medication use or additional medication	Physician or emergency room visit

[a] See text for discussion; see Appendix A for abbreviations and acronyms.
[b] An increase in nonspecific bronchial responsiveness of 100% is equivalent to a 50% decrease in PD_{20} or PD_{100}.
*This table is reproduced from the 1996 O_3 AQCD (Table 9-2, page 9-25) (U.S. Environmental Protection Agency, 1996a).

One consideration in the assessment of potential public health impacts is the size of various population groups that may be at increased risk for health effects associated with O_3-related air pollution exposure. Table 8-4 summarizes information on the prevalence of chronic respiratory and circulatory conditions in the U.S. population in 2002 and 2003 (Dey and Bloom, 2005; Lethbridge-Çejku et al., 2004). Individuals with preexisting cardiopulmonary disease constitute a fairly large proportion of the population, with tens of millions of people included in each disease category. For respiratory conditions, approximately 11% of U.S. adults and 13% of children have been diagnosed with asthma, and 6% of adults have COPD (chronic bronchitis and emphysema). Table 8-5 provides further information on the number of various specific respiratory conditions per 100 persons by age among the U.S. population during the mid-1990s. Approximately 23 million people, or 11% of the U.S. adult population, have some type of heart disease, with 6% reporting diagnoses of coronary heart disease. Approximately 21% of the U.S. adult population has hypertension. Cardiovascular conditions are more common among older age groups, while asthma prevalence is higher in children.

In addition, subpopulations based on age group or socioeconomic status also comprise substantial segments of the population that may be potentially at risk for O_3-related health impacts. Based on U.S. census data from 2003, about 26% of the U.S. population are under 18 years of age and 12% are 65 years of age or older. Approximately 12% of the U.S. population (including 18% of children) are below the poverty level and 16% do not have health insurance coverage. Hence, large proportions of the U.S. population are included in groups that are considered likely to have increased susceptibility and vulnerability for health effects from ambient O_3 exposure.

The health statistics data illustrate what is known as the "pyramid" of effects. At the top of the pyramid, there are approximately 2.5 millions deaths from all causes per year in the U.S. population, with about 900,000 deaths due to circulatory diseases and 100,000 deaths from chronic lower respiratory diseases (Kochanek et al., 2004). For circulatory disease morbidity, there are approximately 6 million hospital discharges per year (DeFrances et al., 2005), 4.5 million emergency department visits (McCaig and Burt, 2005), and 80 million ambulatory care visits (Woodwell and Cherry, 2004). For respiratory health diseases, there are nearly 4 million hospital discharges per year (DeFrances et al., 2005), 14 million emergency department visits (McCaig and Burt, 2005), 112 million ambulatory care visits (Woodwell and

Table 8-4. Prevalence of Selected Cardiorespiratory Disorders by Age Group and by Geographic Region in the United States (2002 [U.S. Adults] and 2003 [U.S. Children] National Health Interview Survey)

			Age (Years)				Region			
	Adults (18+ Years)		18-44	45-64	65-74	75+	Northeast	Midwest	South	West
Chronic Condition/Disease	Cases ($\times 10^6$)	%	%	%	%	%	%	%	%	%
Respiratory Conditions										
Asthma	21.9	10.6	11.5	10.6	8.4	7.6	11	10.9	9.8	11.8
COPD										
Chronic Bronchitis	9.1	4.4	3.5	5.5	5.5	5.3	3.8	4	5.4	3.8
Emphysema	3.1	1.5	0.3	2	4.9	4.7	1.5	1.8	1.7	1.1
Circulatory Conditions										
All Heart Disease	22.7	11.2	4	12.7	26.3	36.6	10.5	11.7	11.6	10.7
Coronary Heart Disease	12.5	6.2	0.9	7.1	18.7	24.5	5.7	6.2	6.8	5.8
Hypertension	43.3	21.2	7.4	29	49.6	51.8	19.7	21.1	23.3	18.9
Stroke	4.8	2.4	0.4	2.5	6.4	11.1	2.4	2.3	2.4	2.7

			Age (Years)			Region			
	Children (<18 years)		0-4	5-11	12-17	Northeast	Midwest	South	West
Chronic Condition/Disease	Cases ($\times 10^6$)	%	%	%	%	%	%	%	%
Respiratory Conditions									
Asthma	9.1	12.5	7.5	14	14.7	14	13.5	11.8	11.2

Source: Lethbridge-Çejku et al. (2004) for data on adults (18+ years); Dey and Bloom (2005) for data on children (<18 years).

Table 8-5. Acute Respiratory Conditions per 100 Persons/Year by Age Group in the United States (1996 National Health Interview Survey)

Type of Acute Condition	All Ages	Under 5 Years	5-17 Years	18-24 Years	25-44 Years	45+ Years		
						Total	45-64 Years	65+ Years
Respiratory Conditions	78.9	129.4	101.5	86	76.9	53.3	55.9	49
Common Cold	23.6	48.6	33.8	23.8	18.7	16.1	16.4	15.7
Other Acute Upper Respiratory Infections	11.3	13.1	15	16.1	11.6	7	7.5	6.1
Influenza	36	53.7	44.3	40.5	38.1	23.3	26.1	18.6
Acute Bronchitis	4.6	7.2[a]	4.3	3.9[a]	5.1	3.8	3.5	4.4[a]
Pneumonia	1.8	3.9[a]	1.7[a]	1.4[a]	1.3[a]	2.0[a]	0.9[a]	3.8[a]
Other Respiratory Conditions	1.7	2.9[a]	2.4[a]	0.4[a]	2.0[a]	1.1[a]	1.5[a]	0.5[a]

[a]Figure does not meet standard of reliability or precision.

Source: Adams et al. (1999).

Cherry, 2004), and an estimated 700 million restricted activity days per year due to respiratory conditions (Adams et al., 1999). Combining small risk estimates with relatively large baseline levels of health outcomes can result in quite large public health impacts. Thus, even a small percentage reduction in O_3 health impacts on cardiopulmonary diseases would reflect a large number of avoided cases.

Another key input for public health impact assessment is the range of concentration-response functions for various health outcomes. Epidemiologic studies have reported associations between short-term exposure to O_3 with mortality, hospitalizations for cardiopulmonary diseases, reduced lung function, incidence of respiratory symptoms, and changes in heartbeat rhythm and rate. Effect estimates for morbidity responses to short-term changes in O_3 tend to be larger in magnitude than those for mortality.

A limited number of studies assessed the impact of reductions in air pollution levels on health outcomes. A study by Neidell (2004) examined the relationship between air pollutants and asthma hospitalizations in California. The most recent EPA O_3 report (U.S. Environmental Protection Agency, 2004b) indicated that the fourth highest daily 8-h max O_3 levels in the pacific

southwest region had decreased by 16% from 1990 to 2003. This downward trend in O_3 levels was mostly influenced by the improvements in Los Angeles and other southern California metropolitan areas. Results from this study noted declines in levels of air pollutants since 1992 and decreased asthma admissions in 1998 for children aged 1 to 18 years ranging from 5 to 14%, depending on the age group. The greatest decline (>10%) in air pollution-related asthma admissions was observed among 3- to 12-year old children. Although this benefit analysis was not specific to O_3, it provides evidence of decreased morbidity resulting from reduced air pollutant concentrations, including O_3.

An intervention study in Atlanta, GA, during the 1996 Summer Olympic Games examined the impact of a citywide decrease in automobile traffic on air quality and childhood asthma (Friedman et al., 2001). Citywide acute care visits and hospitalizations for asthma during the 17 days of the Olympic Games were compared to a baseline period consisting of the four weeks before and after the Olympic Games. During the Olympic Games, levels for all pollutants generally declined, but the most dramatic change was observed for O_3. The 1-h max O_3 concentration in Atlanta decreased 27.9% from a mean of 81.3 ppb during the baseline period to 58.6 ppb during the Olympic Games. The number of asthma acute care events also decreased by 41.6% in the Georgia Medicaid claims file, compared to a 3.1% decline in nonasthma acute care events. Combining data from the baseline and intervention periods, a 31% (95% CI: 0.8, 69.9) excess risk of asthma events was observed per 40 ppb increase in 1-h max O_3 at a cumulative lag of 0- to 2-days. Although a 16.1% decrease in PM_{10} concentrations also occurred during the Olympic Games, there was no association between PM_{10} and asthma acute care events.

Many studies have examined O_3-related health effects, yet only a few have addressed the question as to the extent to which reductions in ambient O_3 actually lead to reductions in adverse health outcomes attributable to O_3. While the findings from these studies suggest that decreases in ambient O_3 levels will likely lead to a reduction in asthma-related hospital admissions and emergency visits, more studies are needed to diminish uncertainty regarding this issue of accountability.

In addition to attribution of risks for the various health outcomes to O_3 and other copollutants, important considerations in assessing the impact of O_3 on public health include the size of the population at risk as well as the concentration-response relationship and the potential

identification of threshold levels. Taken together, it can be concluded that exposure to ambient O_3 likely has a significant impact on public health in the United States.

8.7 SUMMARY AND CONCLUSIONS FOR OZONE HEALTH EFFECTS

This section summarizes the main conclusions derived from this integrated synthesis of information regarding health effects associated with ambient O_3 exposures. The conclusions derived are based on an integrated analysis of animal, human clinical toxicological and epidemiological studies that have evaluated health effects associated with short-term, repeated, and long-term exposures to O_3 alone or in combination with other ambient pollutants. Experimental evidence from human and animal toxicological studies presented in Chapters 4, 5, and 6 was utilized to provide biological plausibility for the health effects observed in epidemiologic studies. These empirical efforts are also aimed at identifying susceptible populations that are at potentially greater risk for effects of O_3 exposure.

1. Health effects of acute (short-term) exposures to Ozone

Numerous field panel and time-series epidemiologic studies (using better weather models and adjustments to confounding copollutants than compared to those assessed in the 1996 O_3 AQCD) have evaluated the effects of short-term exposure to O_3 on a wide range of health endpoints, from lung function decrements to mortality. Results from the majority of studies continue to support the conclusions reported in the 1996 O_3 AQCD.

- Panel studies typically have evaluated the effects of short-term O_3 exposure on both healthy individuals and people with cardiopulmonary diseases. These evaluations included measurement of lung function changes, respiratory symptoms and use of asthma medication.

- Clinical controlled exposure studies in humans indicate changes in lung function and respiratory symptoms that vary as a function of exposure concentration, duration and level of exercise.

- Newer meta-analyses confirmed the interindividual differences in lung function decrements reported in the 1996 O_3 AQCD. Age-specific differences in the lung function responses were also observed. Spirometric responses (due to decrements in lung function) in healthy adults exposed to near ambient O_3 levels typically resolve to near baseline values within 4-6 h.

- Meta-analyses of four controlled human exposure studies (two new and two reported in the 1996 O_3 AQCD) reporting the effects of prolonged (6.6 h) exposures to 0.08 ppm O_3 during moderate exercise on pulmonary function in young healthy adults (M = 90, F = 30; mean age, 23 yrs) indicate an absolute FEV_1 decrease of 6%, whereas FEV_1 increased by 1% following free air (FA) exposures.

- Inflammatory responses (PMN, inflammation mediators such as cytokines and chemokines) and permeability changes (proteins, albumin), typically measured in BALF, also exhibit intersubject variability. Recent meta-analyses on numerous clinical studies indicate interindividual differences in response to short-term O_3 exposures.

- Inflammatory and permeability responses also resolve (in some instances complete recovery) and exhibit differential attenuation profiles between normal healthy subjects and people with preexisting respiratory diseases. Some lung inflammation markers may not resolve readily and mild persistent inflammation has been reported.

- Field/panel studies of healthy individuals and asthmatics have revealed a positive association between short-term exposure to O_3 and decrements in lung function.

- An association between on short-term O_3 exposures and school absenteeism (due to respiratory illness) has also been suggested.

- With regard to cardiac impacts, a limited number of field studies that examined the relationship between short-term O_3 exposures and cardiovascular effects (heart rate variability, myocardial infarction) suggest an association.

- A large multicity and several single-city studies have indicated a positive association between increased O_3 levels (especially during the warm season) and increased risk for hospital admissions. On the other hand epidemiologic data on emergency department visits do not suggest such an association with increase in ambient O_3 levels.

- Data from two large multicity studies from the U.S. and several single-city studies suggest a positive association between increase in O_3 levels and all cause (non-accidental) daily mortality. Meta-analyses on the influence of season suggest a causal association. Additional meta-analyses on cause-specific mortality are suggestive of a likely positive association between increases in ambient O_3 levels and cardiovascular mortality.

- Short-term O_3-induced lung function decrements, respiratory symptoms, inflammation and permeability changes observed in animal toxicology studies are consistent with human studies.

2. Health effects of repeated short-term exposures to Ozone

The results of new controlled human exposure studies of repeated short-term O_3 exposures continue to support the health effects findings/conclusions reported in the 1996 O_3 AQCD.

- Repeated exposure studies at higher concentrations typically show that FEV_1 response to O_3 is enhanced on the second of several days of exposure. Such an enhanced response was not observed at lower O_3 concentrations. With repeated O_3 exposures over several days, spirometric and symptom responses become attenuated, but this tolerance is lost after about a week without exposure.

- In humans repeatedly exposed to 0.4 ppm O_3 for 5 consecutive days, several indicators of inflammation (e.g., PMN influx, IL-6, PGE_2, BAL protein, fibronectin) were attenuated after 5 days of exposure. Lung injury and permeability markers (LDH, IL-8, total protein, epithelial cells) did not show attenuation, indicating that tissue damage probably continues to occur during repeated exposure. The recovery of the inflammatory response occurred for some markers after 10 days, but some responses were not normalized even after 20 days.

The continued presence of cellular injury markers indicates a persistent effect that may not necessarily be recognized due to the attenuation of spirometric and symptom responses.

- Repeated daily exposure to lower concentrations of O_3 (0.125 ppm for 4 days) causes an increased response to bronchial allergen challenge in subjects with preexisting allergic airway disease, with or without asthma. In these subjects, changes in airway responsiveness after O_3 exposure appear to be resolved more slowly than changes in FEV_1 or respiratory symptoms.

3. Health effects of long-term exposures to Ozone

Assessment of human health effects associated with long-term O_3 exposures is hampered by the lack of pertinent data from human clinical and epidemiologic studies. Chronic animal toxicology studies continue to support structural alterations in several regions of the respiratory tract and identify the CAR as the most affected region.

- Animal toxicology studies that utilized exposure regimens to simulate seasonal exposure pattern also report increased lung injury compared to conventional chronic stable exposures. One long term study of infant rhesus monkeys exposed to simulated seasonal O_3 patterns (0.5 ppm 8h/day for 5 days, every 14 days for 11 episodes) demonstrated: (1) remodeling in the distal airways; (2) abnormalities in tracheal basement membrane; (3) eosinophil accumulation in conducting airways; (4) decrements in airway innervation. These findings advance earlier information regarding possible injury-repair processes occurring with seasonal O_3 exposures.

- Effects of O_3 on the upper respiratory tract of F344 rats exposed to O_3 (0.12, 0.5, or 1.0 ppm for 20 months) included marked mucous cell metaplasia in the rats exposed to 0.5 and 1.0 pm O_3, but not at 0.12 ppm O_3. The persistent nature of the O_3-induced mucous cell metaplasia suggests that O_3 exposure may have the potential to induce similar long-lasting alterations in the airways of humans. Hyperplasia in the nasal epithelium of rats exposed to 0.25 and 0.5 ppm, 8h/day, 7 days/week, for 13 weeks has been reported.

- Pathophysiological changes associated with chronic O_3 exposures observed in animal studies suggest possible similar alterations in humans. The pulmonary function changes observed in children in polluted metropolitan areas and lung structural alterations reported in autopsy study in Los Angeles suggest a role for long-term ambient O_3 exposure and need to be critically evaluated with proper study design.

4. Susceptibility factors associated with exposure to ozone

Various factors such as age, gender, nutrition, socioeconomic, activity patterns, and disease status have been shown to influence the response to environmental air pollutants. Controlled human exposure studies clearly established differential biological response to O_3 based on physical activity (exertion) and age. These studies also demonstrated a large variation in sensitivity and responsiveness to O_3. The specific factors that contribute to this intersubject variability are yet to be identified.

- Increased hospital admissions for asthma and COPD in summer (with increased levels of ambient O_3) suggest that people with these respiratory diseases as potential sub-population for O_3-induced health effects.

- Similarly, based on O_3-induced differential responses in lung inflammation and in airway hyperresponsiveness, asthmatics (including children) appear to have potentially increased susceptibility to O_3. However, there is no supportive data from controlled human studies suggesting individuals with COPD are more sensitive to O_3-induced health effects.

- Animal toxicology studies provided supportive evidence to the observations of varied susceptibility. Various strains of mice and rats have demonstrated the importance of genetic background in O_3 susceptibility. Moreover, genetic and molecular characterization studies in laboratory animals identified genetic loci responsible for both sensitivity and resistance.

- Consistent with the 1996 O_3 AQCD, the scarcity of data prevents determination of the role of ethnic or racial background and nutrition status on O_3-induced health effects. However, as presented in this document, exercising (moderate to high physical exertion) healthy,

adolescents, and asthmatics appear to demonstrate increased responsiveness to ambient concentrations of O_3 and may be susceptible for O_3-induced health effects.

5. Health effects of binary pollutant mixtures containing ozone

A limited number of controlled human exposure studies and few animal toxicology studies with the binary mixtures containing O_3 suggest potential interactions depending on the exposure regimens and copollutant constituents.

- Continuous exposure to SO_2 and NO_2 increased inhaled bolus O_3 absorption, while continuous exposure to O_3 decreased O_3 bolus absorption. Asthmatics exhibited enhanced airway reactivity to house dust mite following exposures to O_3, NO_2, and the combination of the two gases. Spirometric response, however, was impaired only by O_3 and O_3+NO_2 at higher concentrations.

- Animal toxicology studies with O_3 in mixture with NO_2, formaldehyde, and PM demonstrated additive, synergistic or antagonistic effects depending on the exposure regimen and the endpoints evaluated.

- One controlled exposure study of children, designed to approximate exposure conditions of an epidemiologic study by matching the population and exposure atmosphere (0.1 ppm O_3, 0.1 ppm SO_2 and 101 $\mu g/^{m2}$ H_2SO_4), failed to support the findings of the epidemiologic study. This study points out the difficulties in attempting to link the outcomes of epidemiologic and controlled studies with binary pollutant mixtures.

REFERENCES

Adams, P. F.; Hendershot, G. E.; Marano, M. A. (1999) Current estimates from the National Health Interview Survey, 1996. Hyattsville, MD: U.S. Department of Health and Human Services, Public Health Service, National Center for Health Statistics; DHHS publication no. (PHS) 99-1528. (Vital and health statistics: v. 10, data from the National Health Survey, no. 200). Available: http://www.cdc.gov/nchs/products/pubs/pubd/series/sr10/pre-200/pre-200.htm [12 March, 2001].

Alexis, N.; Urch, B.; Tarlo, S.; Corey, P.; Pengelly, D.; O'Byrne, P.; Silverman, F. (2000) Cyclooxygenase metabolites play a different role in ozone-induced pulmonary function decline in asthmatics compared to normals. Inhalation Toxicol. 12: 1205-1224.

American Thoracic Society. (1985) Guidelines as to what constitutes an adverse respiratory health effect, with special reference to epidemiologic studies of air pollution. Am. Rev. Respir. Dis. 131: 666-668.

American Thoracic Society. (1991) Lung function testing: selection of reference values and interpretative strategies Am. Rev. Respir. Dis. 144: 1202-1218.

American Thoracic Society. (2000) What constitutes an adverse health effect of air pollution? Am. J. Respir. Crit. Care Med. 161: 665-673.

Anderson, H. R.; Spix, C.; Medina, S.; Schouten, J. P.; Castellsague, J.; Rossi, G.; Zmirou, D.; Touloumi, G.; Wojtyniak, B.; Ponka, A.; Bacharova, L.; Schwartz, J.; Katsouyanni, K. (1997) Air pollution and daily admissions for chronic obstructive pulmonary disease in 6 European cities: results from the APHEA project. Eur. Respir. J. 10: 1064-1071.

Avol, E. L.; Navidi, W. C.; Rappaport, E. B.; Peters, J. M. (1998) Acute effects of ambient ozone on asthmatic, wheezy, and healthy children. Cambridge, MA: Health Effects Institute; research report no. 82.

Basha, M. A.; Gross, K. B.; Gwizdala, C. J.; Haidar, A. H.; Popovich, J., Jr. (1994) Bronchoalveolar lavage neutrophilia in asthmatic and healthy volunteers after controlled exposure to ozone and filtered purified air. Chest 106: 1757-1765.

Bayram, H.; Rusznak, C.; Khair, O. A.; Sapsford, R. J.; Abdelaziz, M. M. (2002) Effect of ozone and nitrogen dioxide on the permeability of bronchial epithelial cell cultures of non-asthmatic and asthmatic subjects. Clin. Exp. Allergy 32: 1285-1292.

Bell, M. L.; McDermott, A.; Zeger, S. L.; Samet, J. M.; Dominici, F. (2004) Ozone and short-term mortality in 95 US urban communities, 1987-2000. JAMA J. Am. Med. Assoc. 292: 2372-2378.

Bell, M. L.; Dominici, F.; Samet, J. M. (2005) A meta-analysis of time-series studies of ozone and mortality with comparison to the national morbidity, mortality, and air pollution study. Epidemiology 16: 436-445.

Bosson, J.; Stenfors, N.; Bucht, A.; Helleday, R.; Pourazar, J.; Holgate, S. T.; Kelly, F. J.; Sandström, T.; Wilson, S.; Frew, A. J.; Blomberg, A. (2003) Ozone-induced bronchial epithelial cytokine expression differs between healthy and asthmatic subjects. Clin. Exp. Allergy 33: 777-782.

Broeckaert, F.; Clippe, A.; Wattiez, R.; Falmagne, P.; Bernard, A. (2003) Lung hyperpermeability, Clara-cell secretory potein (CC16), and susceptibility to ozone of five inbred strains of mice. Inhalation Toxicol. 15: 1209-1230.

Brook, R. D.; Brook, J. R.; Urch, B.; Vincent, R.; Rajagopalan, S.; Silverman, F. (2002) Inhalation of fine particulate air pollution and ozone causes acute arterial vasoconstriction in healthy adults. Circulation 105: 1534-1536.

Brook, R. D.; Franklin, B.; Cascio, W.; Hong, Y.; Howard, G.; Lipsett, M.; Luepker, R.; Mittleman, M.; Samet, J.; Smith, S. C., Jr.; Tager, I. (2004) Air pollution and cardiovascular disease. A statement for healthcare professionals from the Expert Panel on Population and Prevention Science of the American Heart Association. Circulation 109: 2655-2671.

Burnett, R. T.; Dales, R. E.; Raizenne, M. E.; Krewski, D.; Summers, P. W.; Roberts, G. R.; Raad-Young, M.; Dann, T.; Brook, J. (1994) Effects of low ambient levels of ozone and sulfates on the frequency of respiratory admissions to Ontario hospitals. Environ. Res. 65: 172-194.

Burnett, R. T.; Brook, J. R.; Yung, W. T.; Dales, R. E.; Krewski, D. (1997) Association between ozone and hospitalization for respiratory diseases in 16 Canadian cities. Environ. Res. 72: 24-31.

Burnett, R. T.; Cakmak, S.; Brook, J. R.; Krewski, D. (1997) The role of particulate size and chemistry in the association between summertime ambient air pollution and hospitalization for cardiorespiratory diseases. Environ. Health Perspect. 105: 614-620.

Burnett, R. T.; Smith-Doiron, M.; Stieb, D.; Raizenne, M. E.; Brook, J. R.; Dales, R. E.; Leech, J. A.; Cakmak, S.; Krewski, D. (2001) Association between ozone and hospitalization for acute respiratory diseases in children less than 2 years of age. Am. J. Epidemiol. 153: 444-452.

Bush, M. L.; Zhang, W.; Ben-Jebria, A.; Ultman, J. S. (2001) Longitudinal distribution of ozone and chlorine in the human respiratory tract: simulation of nasal and oral breathing with the single-path diffusion model. Toxicol. Appl. Pharmacol. 173: 137-145.

Chen, L.; Jennison, B. L.; Yang, W.; Omaye, S. T. (2000) Elementary school absenteeism and air pollution. Inhalation Toxicol. 12: 997-1016.

Cho, H.-Y.; Zhang, L.-Y.; Kleeberger, S. R. (2001) Ozone-induced lung inflammation and hyperreactivity are mediated via tumor necrosis factor-α receptors. Am. J. Physiol. 280: L537-L546.

Chock, D. P.; Winkler, S. L.; Chen, C. (2000) A study of the association between daily mortality and ambient air pollutant concentrations in Pittsburgh, Pennsylvania. J. Air Waste Manage. Assoc. 50: 1481-1500.

Cody, R. P.; Weisel, C. P.; Birnbaum, G.; Lioy, P. J. (1992) The effect of ozone associated with summertime photochemical smog on the frequency of asthma visits to hospital emergency departments. Environ. Res. 58: 184-194.

Cohen, M. D.; Sisco, M.; Li, Y.; Zelikoff, J. T.; Schlesinger, R. B. (2001) Ozone-induced modulation of cell-mediated immune responses in the lungs. Toxicol. Appl. Pharmacol. 171: 71-84.

Coleridge, J. C. G.; Coleridge, H. M.; Schelegle, E. S.; Green, J. F. (1993) Acute inhalation of ozone stimulates bronchial C-fibers and rapidly adapting receptors in dogs. J. Appl. Physiol. 74: 2345-2352.

David, G. L.; Romieu, I.; Sienra-Monge, J. J.; Collins, W. J.; Ramirez-Aguilar, M.; Del Rio-Navarro, B. E.; Reyes-Ruiz, N. I.; Morris, R. W.; Marzec, J. M.; London, S. J. (2003) Nicotinamide adenine dinucleotide (Phosphate) reduced:quinone oxidoreductase and glutathione s-transferase m1 polymorphism and childhood asthma. Am. J. Respir. Crit. Care Med. 168: 1199-1204.

DeFrances, C. J.; Hall, M. J.; Podgornik, M. N. (2005) 2003 National Hospital Discharge Survey. Hyattsville, MD: National Center for Health Statistics; DHHS publication no. (PHS) 2004-1250. (Advance data from vital and health statistics; no. 359). Available: http://www.cdc.gov/nchs/data/ad/ad359.pdf [3 August, 2005].

Delaunois, A.; Segura, P.; Montaño, L. M.; Vargas, M. H.; Ansay, M.; Gustin, P. (1998) Comparison of ozone-induced effects on lung mechanics and hemodynamics in the rabbit. Toxicol. Appl. Pharmacol. 150: 58-67.

De Leon, S. F.; Thurston, G. D.; Ito, K. (2003) Contribution of respiratory disease to nonrespiratory mortality associations with air pollution. Am. J. Respir. Crit. Care Med. 167: 1117-1123.

Delfino, R. J.; Becklake, M. R.; Hanley, J. A. (1994) The relationship of urgent hospital admissions for respiratory illnesses to photochemical air pollution levels in Montreal. Environ. Res. 67: 1-19.

Delfino, R. J.; Zeiger, R. S.; Seltzer, J. M.; Street, D. H.; Matteucci, R. M.; Anderson, P. R.; Koutrakis, P. (1997) The effect of outdoor fungal spore concentrations on daily asthma severity. Environ. Health Perspect. 105: 622-635.

Desqueyroux, H.; Pujet, J.-C.; Prosper, M.; Squinazi, F.; Momas, I. (2002a) Short-term effects of low-level air pollution on respiratory health of adults suffering from moderate to severe asthma. Environ. Res. A 89: 29-37.

Desqueyroux, H.; Pujet, J.-C.; Prosper, M.; Le Moullec, Y.; Momas, I. (2002b) Effects of air pollution on adults with chronic obstructive pulmonary disease. Arch. Environ. Health 57: 554-560.

Devlin, R. B.; McDonnell, W. F.; Mann, R.; Becker, S.; House, D. E.; Schreinemachers, D.; Koren, H. S. (1991) Exposure of humans to ambient levels of ozone for 6.6 hours causes cellular and biochemical changes in the lung. Am. J. Respir. Cell Mol. Biol. 4: 72-81.

Devlin, R. B.; Folinsbee, L. J.; Biscardi, F.; Hatch, G.; Becker, S.; Madden, M. C.; Robbins, M.; Koren, H. S. (1997) Inflammation and cell damage induced by repeated exposure of humans to ozone. Inhalation Toxicol. 9: 211-235.

Dey, A. N.; Bloom, B. (2005) Summary health statistics for U.S. children: National Health Interview Survey, 2003. Hyattsville, MD: U.S. Department of Health & Human Services, National Center for Health Statistics. (Vital and health statistics, series 10, no. 223). Available: http://www.cdc.gov/nchs/data/series/sr_10/sr10_223.pdf [3 August, 2005].

Dockery, D. W.; Schwartz, J.; Spengler, J. D. (1992) Air pollution and daily mortality: associations with particulates and acid aerosols. Environ. Res. 59: 362-373.

Dominici, F.; McDermott, A.; Daniels, M.; Zeger, S. L.; Samet, J. M. (2003) Mortality among residents of 90 cities. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 9-24. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [12 May, 2004].

Fairley, D. (1999) Daily mortality and air pollution in Santa Clara County, California: 1989-1996. Environ. Health Perspect. 107: 637-641.

Fairley, D. (2003) Mortality and air pollution for Santa Clara County, California, 1989-1996. In: Revised analyses of time-series studies of air pollution and health. Special report. Boston, MA: Health Effects Institute; pp. 97-106. Available: http://www.healtheffects.org/Pubs/TimeSeries.pdf [18 October, 2004].

Fiore, A. M.; Jacob, D. J.; Mathur, R.; Martin, R. V. (2003) Application of empirical orthogonal functions to evaluate ozone simulations with regional and global models. J. Geophys. Res. (Atmos.) 108(D14): 10.1029/2002JD003151.

Foster, W. M.; Stetkiewicz, P. T. (1996) Regional clearance of solute from the respiratory epithelia: 18-20 h postexposure to ozone. J. Appl. Physiol. 81: 1143-1149.

Foster, W. M.; Costa, D. L.; Langenback, E. G. (1987) Ozone exposure alters tracheobronchial mucociliary function in humans. J. Appl. Physiol. 63: 996-1002.

Foster, W. M.; Silver, J. A.; Groth, M. L. (1993) Exposure to ozone alters regional function and particle dosimetry in the human lung. J. Appl. Physiol. 75: 1938-1945.

Foster, W. M.; Weinmann, G. G.; Menkes, E.; Macri, K. (1997) Acute exposure of humans to ozone impairs small airway function. Ann. Occup. Hyg. 41(suppl. 1): 659-666.

Friedman, M. S.; Powell, K. E.; Hutwagner, L.; Graham, L. M.; Teague, W. G. (2001) Impact of changes in transportation and commuting behaviors during the 1996 summer olympic games in Atlanta on air quality and childhood asthma. JAMA J. Am. Med. Assoc. 285: 897-905.

Fung, K. Y.; Luginaah, I.; Gorey, K. M.; Webster, G. (2005) Air pollution and daily hospital admissions for cardiovascular diseases in Windsor, Ontario. Can. J. Public Health 96: 29-33.

Gamble, J. L. (1998) Effects of ambient air pollution on daily mortality: a time series analysis of Dallas, Texas, 1990-1994. Presented at: 91st annual meeting and exhibition of the Air & Waste Management Association; June; San Diego, CA. Pittsburgh, PA: Air & Waste Management Association; paper no. 98-MP26.03.

Gent, J. F.; Triche, E. W.; Holford, T. R.; Belanger, K.; Bracken, M. B.; Beckett, W. S.; Leaderer, B. P. (2003) Association of low-level ozone and fine particles with respiratory symptoms in children with asthma. JAMA J. Am. Med. Assoc. 290: 1859-1867.

Gilliland, F. D.; Berhane, K.; Rappaport, E. B.; Thomas, D. C.; Avol, E.; Gauderman, W. J.; London, S. J.; Margolis, H. G.; McConnell, R.; Islam, K. T.; Peters, J. M. (2001) The effects of ambient air pollution on school absenteeism due to respiratory illnesses. Epidemiology 12: 43-54.

Goldberg, M. S.; Burnett, R. T.; Brook, J.; Bailar, J. C., III; Valois, M.-F.; Vincent, R. (2001) Associations between daily cause-specific mortality and concentrations of ground-level ozone in Montreal, Quebec. Am. J. Epidemiol. 154: 817-826.

Gong, H., Jr.; Shamoo, D. A.; Anderson, K. R.; Linn, W. S. (1997a) Responses of older men with and without chronic obstructive pulmonary disease to prolonged ozone exposure. Arch. Environ. Health 52: 18-25.

Gong, H., Jr.; McManus, M. S.; Linn, W. S. (1997b) Attenuated response to repeated daily ozone exposures in asthmatic subjects. Arch. Environ. Health 52: 34-41.

Gong, H., Jr.; Wong, R.; Sarma, R. J.; Linn, W. S.; Sullivan, E. D.; Shamoo, D. A.; Anderson, K. R.; Prasad, S. B. (1998) Cardiovascular effects of ozone exposure in human volunteers. Am. J. Respir. Crit. Care Med. 158: 538-546.

Gryparis, A.; Forsberg, B.; Katsouyanni, K.; Analitis, A.; Touloumi, G.; Schwartz, J.; Samoli, E.; Medina, S.; Anderson, H. R.; Niciu, E. M.; Wichmann, H.-E.; Kriz, B.; Kosnik, M.; Skorkovsky, J.; Vonk, J. M.; Dörtbudak, Z. (2004) Acute effects of ozone on mortality from the "air pollution and health: a European approach" project. Am. J. Respir. Crit. Care Med. 170: 1080-1087.

Gwynn, R. C.; Burnett, R. T.; Thurston, G. D. (2000) A time-series analysis of acidic particulate matter and daily mortality and morbidity in the Buffalo, New York, region. Environ. Health Perspect. 108: 125-133.

Hatch, G. E.; Slade, R.; Harris, L. P.; McDonnell, W. F.; Devlin, R. B.; Koren, H. S.; Costa, D. L.; McKee, J. (1994) Ozone dose and effect in humans and rats: a comparison using oxygen-18 labeling and bronchoalveolar lavage. Am. J. Respir. Crit. Care Med. 150: 676-683.

Hazucha, M. J.; Sant'Ambrogio, G. (1993) Effects of ozone on the activity of slowly (SAR) and rapidly adapting (RAR) receptors in cats. FASEB J. 7: 407A.

Hazucha, M. J.; Folinsbee, L. J.; Bromberg, P. A. (2003) Distribution and reproducibility of spirometric response to ozone by gender and age. J. Appl. Physiol. 95: 1917-1925.

Hiltermann, T. J. N.; Stolk, J.; Van der Zee, S. C.; Brunekreef, B.; De Bruijne, C. R.; Fischer, P. H.; Ameling, C. B.; Sterk, P. J.; Hiemstra, P. S.; Van Bree, L. (1998) Asthma severity and susceptibility to air pollution. Eur. Respir. J. 11: 686-693.

Hiltermann, J. T. N.; Lapperre, T. S.; Van Bree, L.; Steerenberg, P. A.; Brahim, J. J.; Sont, J. K.; Sterk, P. J.; Hiemstra, P. S.; Stolk, J. (1999) Ozone-induced inflammation assessed in sputum and bronchial lavage fluid from asthmatics: a new noninvasive tool in epidemiologic studies on air pollution and asthma. Free Radical Biol. Med. 27: 1448-1454.

Holz, O.; Mücke, M.; Paasch, K.; Böhme, S.; Timm, P.; Richter, K.; Magnussen, H.; Jörres, R. A. (2002) Repeated ozone exposures enhance bronchial allergen responses in subjects with rhinitis or asthma. Clin. Exp. Allergy. 32: 681-689.

Holz, O.; Tal-Singer, R.; Kanniess, F.; Simpson, K. J.; Gibson, A.; Vessey, R. S. J.; Janicki, S.; Magnussen, H.; Jörres, R. A.; Richter, K. (2005) Validation of the human ozone challenge model as a tool for assessing anti-inflammatory drugs in early development. J. Clin. Pharmacol. 45: 498-503.

Höppe, P.; Peters, A.; Rabe, G.; Praml, G.; Lindner, J.; Jakobi, G.; Fruhmann, G.; Nowak, D. (2003) Environmental ozone effects in different population subgroups. Int. J. Hyg. Environ. Health 206: 505-516.

Horstman, D. H.; Ball, B. A.; Brown, J.; Gerrity, T.; Folinsbee, L. J. (1995) Comparison of pulmonary responses of asthmatic and nonasthmatic subjects performing light exercise while exposed to a low level of ozone. Toxicol. Ind. Health 11: 369-385.

Huang, Y.; Dominici, F.; Bell, M. L. (2005) Bayesian hierarchical distributed lag models for summer ozone exposure and cardio-respiratory mortality. Environmetrics 16: 547-562.

Inoue, H.; Aizawa, H.; Matsumoto, K.; Shigyo, M.; Takata, S.; Hara, M.; Hara, N. (1997) Effect of β-agonists on histamine-induced airway microvascular leakage in ozone-exposed guinea pigs. Am. J. Respir. Crit. Care Med. 156: 723-727.

Ito, K.; Thurston, G. D. (1996) Daily PM_{10}/mortality associations: an investigation of at-risk subpopulations. J. Exposure Anal. Environ. Epidemiol. 6: 79-95.

Ito, K.; De Leon, S. F.; Lippmann, M. (2005) Associations between ozone and daily mortality, analysis and meta-analysis. Epidemiology 16: 446-457.

Jaffe, D. H.; Singer, M. E.; Rimm, A. A. (2003) Air pollution and emergency department visits for asthma among Ohio Medicaid recipients, 1991-1996. Environ. Res. 91: 21-28.

Jörres, R.; Nowak, D.; Magnussen, H.; Speckin, P.; Koschyk, S. (1996) The effect of ozone exposure on allergen responsiveness in subjects with asthma or rhinitis. Am. J. Respir. Crit. Care Med. 153: 56-64.

Jörres, R. A.; Holz, O.; Zachgo, W.; Timm, P.; Koschyk, S.; Müller, B.; Grimminger, F.; Seeger, W.; Kelly, F. J.; Dunster, C.; Frischer, T.; Lubec, G.; Waschewski, M.; Niendorf, A.; Magnussen, H. (2000) The effect of repeated ozone exposures on inflammatory markers in bronchoalveolar lavage fluid and mucosal biopsies. Am. J. Respir. Crit. Care Med. 161: 1855-1861.

Kehrl, H. R.; Vincent, L. M.; Kowalsky, R. J.; Horstman, D. H.; O'Neil, J. J.; McCartney, W. H.; Bromberg, P. A. (1987) Ozone exposure increases respiratory epithelial permeability in humans. Am. Rev. Respir. Dis. 135: 1124-1128.

Kim, S.-Y.; Lee, J.-T.; Hong, Y.-C.; Ahn, K.-J.; Kim, H. (2004) Determining the threshold effect of ozone on daily mortality: an analysis of ozone and mortality in Seoul, Korea, 1995-1999. Environ. Res. 94: 113-119.

King, T. K. C.; Briscoe, W. A. (1968) The distribution of ventilation, perfusion, lung volume and transfer factor (diffusing capacity) in patients with obstructive lung disease. Clin. Sci. 35: 153-170.

Kinney, P. L.; Özkaynak, H. (1991) Associations of daily mortality and air pollution in Los Angeles County. Environ. Res. 54: 99-120.

Kinney, P. L.; Ito, K.; Thurston, G. D. (1995) A sensitivity analysis of mortality/PM_{10} associations in Los Angeles. In: Phalen, R. F.; Bates, D. V., eds. Proceedings of the colloquium on particulate air pollution and human mortality and morbidity; January 1994; Irvine, CA. Inhalation Toxicol. 7: 59-69.

Kinney, P. L.; Thurston, G. D.; Raizenne, M. (1996) The effects of ambient ozone on lung function in children: a reanalysis of six summer camp studies. Environ. Health Perspect. 104: 170-174.

Kleeberger, S. R.; Levitt, R. C.; Zhang, L.-Y.; Longphre, M.; Harkema, J.; Jedlicka, A.; Eleff, S. M.; DiSilvestre, D.; Holroyd, K. J. (1997) Linkage analysis of susceptibility to ozone-induced lung inflammation in inbred mice. Nat. Genet. 17: 475-478.

Klemm, R. J.; Mason, R. M., Jr. (2000) Aerosol Research and Inhalation Epidemiological Study (ARIES): air quality and daily mortality statistical modeling–interim results. J. Air. Waste Manage. Assoc. 50: 1433-1439.

Kochanek, K. D.; Murphy, S. L.; Anderson, R. N.; Scott, C. (2004) Deaths: final data for 2002. Hyattsville, MD: U.S. Department of Health & Human Services, National Center for Health Statistics; DHHS publication no. (PHS) 2005-1120. (National vital statistics reports: v. 53, no. 5). Available: http://www.cdc.gov/nchs/data/nvsr/nvsr53/nvsr53_05.pdf [3 August, 2005].

Kopp, M. V.; Ulmer, C.; Ihorst, G.; Seydewitz, H. H.; Frischer, T.; Forster, J.; Kuehr, J. (1999) Upper airway inflammation in children exposed to ambient ozone and potential signs of adaptation. Eur. Respir. J. 14: 854-861.

Koren, H. S.; Devlin, R. B.; Graham, D. E.; Mann, R.; McGee, M. P.; Horstman, D. H.; Kozumbo, W. J.; Becker, S.; House, D. E.; McDonnell, W. F.; Bromberg, P. A. (1989) Ozone-induced inflammation in the lower airways of human subjects. Am. Rev. Respir. Dis. 139: 407-415.

Korrick, S. A.; Neas, L. M.; Dockery, D. W.; Gold, D. R.; Allen, G. A.; Hill, L. B.; Kimball, K. D.; Rosner, B. A.; Speizer, F. E. (1998) Effects of ozone and other pollutants on the pulmonary function of adult hikers. Environ. Health Perspect. 106: 93-99.

Krishna, M. T.; Madden, J.; Teran, L. M.; Biscione, G. L.; Lau, L. C. K.; Withers, N. J.; Sandstrom, T.; Mudway, I.; Kelly, F. J.; Walls, A.; Frew, A. J.; Holgate, S. T. (1998) Effects of 0.2 ppm ozone on biomarkers of inflammation in bronchoalveolar lavage fluid and bronchial mucosa of healthy subjects. Eur. Respir. J. 11: 1294-1300.

Kronenberg, R. S.; Drage, C. W.; Ponto, R. A.; Williams, L. E. (1973) The effect of age on the distribution of ventilation and perfusion in the lung. Am. Rev. Respir. Dis. 108: 576-586.

Leikauf, G. D.; Simpson, L. G.; Santrock, J.; Zhao, Q.; Abbinante-Nissen, J.; Zhou, S.; Driscoll, K. E. (1995) Airway epithelial cell responses to ozone injury. Environ. Health Perspect. 103(suppl. 2): 91-95.

Lethbridge-Çejku, M.; Schiller, J. S.; Bernadel, L. (2004) Summary health statistics for U.S. adults: National Health Interview Survey, 2002. Hyattsville, MD: Centers for Disease Control and Prevention; DHHS publication no. (PHS) 2004-1550. (Vital and health statistics, series 10, number 222). Available: http://www.cdc.gov/nchs/data/series/sr_10/sr10_222.pdf [3, August, 2005].

Levy, J. I.; Carrothers, T. J.; Tuomisto, J. T.; Hammitt, J. K.; Evans, J. S. (2001) Assessing the public health benefits of reduced ozone concentrations. Environ. Health Perspect. 109: 1215-1226.

Levy, J. I.; Chemerynski, S. M.; Sarnat, J. A. (2005) Ozone exposure and mortality, an empiric Bayes metaregression analysis. Epidemiology 16: 458-468.

Liao, D.; Duan, Y.; Whitsel, E. A.; Zheng, Z.-J.; Heiss, G.; Chinchilli, V. M.; Lin, H.-M. (2004) Association of higher levels of ambient criteria pollutants with impaired cardiac autonomic control: a population-based study. Am. J. Epidemiol. 159: 768-777.

Linn, W. S.; Szlachcic, Y.; Gong, H., Jr.; Kinney, P. L.; Berhane, K. T. (2000) Air pollution and daily hospital admissions in metropolitan Los Angeles. Environ. Health Perspect. 108: 427-434.

Lipfert, F. W.; Morris, S. C.; Wyzga, R. E. (2000) Daily mortality in the Philadelphia metropolitan area and size-classified particulate matter. J. Air Waste Manage. Assoc. 50: 1501-1513.

Lippmann, M.; Ito, K.; Nádas, A.; Burnett, R. T. (2000) Association of particulate matter components with daily mortality and morbidity in urban populations. Cambridge, MA: Health Effects Institute; research report no. 95.

Mannino, D. M.; Ford, E. S.; Redd, S. C. (2003) Obstructive and restrictive lung disease and markers of inflammation: data from the Third National Health and Nutrition Examination. Am. J. Med. 114: 758-762.

McBride, D. E.; Koenig, J. Q.; Luchtel, D. L.; Williams, P. V.; Henderson, W. R., Jr. (1994) Inflammatory effects of ozone in the upper airways of subjects with asthma. Am. J. Respir. Crit. Care Med. 149: 1192-1197.

McCaig, L. F.; Burt, C. W. (2005) National Hospital Ambulatory Medical Care Survey: 2003 Emergency Department Summary. Hyattsville, MD: National Center for Health Statistics; DHHS publication no. (PHS) 2005-1250. (Advance data from vital and health statistics; no. 358). Available: http://www.cdc.gov/nchs/data/ad/ad358.pdf [3 August, 2005].

McDonnell, W. F. (1996) Individual variability in human lung function responses to ozone exposure. Environ. Toxicol. Pharmacol. 2: 171-175.

Moolgavkar, S. H. (2003) Air pollution and daily mortality in two U.S. counties: season-specific analyses and exposure-response relationships. Inhalation Toxicol. 15: 877-907.

Moolgavkar, S. H.; Luebeck, E. G.; Hall, T. A.; Anderson, E. L. (1995) Air pollution and daily mortality in Philadelphia. Epidemiology 6: 476-484.

Mortimer, K. M.; Neas, L. M.; Dockery, D. W.; Redline, S.; Tager, I. B. (2002) The effect of air pollution on inner-city children with asthma. Eur. Respir. J. 19: 699-705.

Mudway, I. S.; Kelly, F. J. (2000) Ozone and the lung: a sensitive issue. Mol. Aspects. Med. 21: 1-48.

Mudway, I. S.; Kelly, F. J. (2004) An investigation of inhaled ozone dose and the magnitude of airway inflammation in healthy adults. Am. J. Respir. Crit. Care Med. 169: 1089-1095.

Neidell, M. J. (2004) Air pollution, health, and socio-economic status: the effect of outdoor air quality on childhood asthma. J. Health Econ. 23: 1209-1236.

Ostro, B. (1995) Fine particulate air pollution and mortality in two Southern California counties. Environ. Res. 70: 98-104.

Park, H.; Lee, B.; Ha, E.-H.; Lee, J.-T.; Kim, H.; Hong, Y.-C. (2002) Association of air pollution with school absenteeism due to illness. Arch. Pediatr. Adolesc. Med. 156: 1235-1239.

Park, S. K.; O'Neill, M. S.; Vokonas, P. S.; Sparrow, D.; Schwartz, J. (2005) Effects of air pollution on heart rate variability: the VA normative aging study. Environ. Health Perspect. 113: 304-309.

Passannante, A. N.; Hazucha, M. J.; Bromberg, P. A.; Seal, E.; Folinsbee, L.; Koch, G. (1998) Nociceptive mechanisms modulate ozone-induced human lung function decrements. J. Appl. Physiol. 85: 1863-1870.

Peel, J. L.; Tolbert, P. E.; Klein, M.; Metzger, K. B.; Flanders, W. D.; Knox, T.; Mulholland, J. A.; Ryan, P. B.; Frumkin, H. (2005) Ambient air pollution and respiratory emergency department visits. Epidemiology 16: 164-174.

Peters, A.; Dockery, D. W.; Muller, J. E.; Mittleman, M. A. (2001) Increased particulate air pollution and the triggering of myocardial infarction. Circulation 103: 2810-2815.

Pope, C. A., III; Burnett, R. T.; Thun, M. J.; Calle, E. E.; Krewski, D.; Ito, K.; Thurston, G. D. (2002) Lung cancer, cardiopulmonary mortality, and long-term exposure to fine particulate air pollution. JAMA J. Am. Med. Assoc. 287: 1132-1141.

Pulfer, M. K.; Murphy, R. C. (2004) Formation of biologically active oxysterols during ozonolysis of cholesterol present in lung surfactant. J. Biol. Chem. 279: 26,331-26,338.

Pulfer, M. K.; Taube, C.; Gelfand, E.; Murphy, R. C. (2005) Ozone exposure in vivo and formation of biologically active oxysterols in the lung. J. Pharmacol. Exp. Ther. 312: 256-264.

Rigas, M. L.; Ben-Jebria, A.; Ultman, J. S. (1997) Longitudinal distribution of ozone absorption in the lung: effects of nitrogen dioxide, sulfur dioxide, and ozone exposures. Arch. Environ. Health 52: 173-178.

Romieu, I.; Meneses, F.; Ramirez, M.; Ruiz, S.; Padilla, R. P.; Sienra, J. J.; Gerber, M.; Grievink, L.; Dekker, R.; Walda, I.; Brunekreef, B. (1998) Antioxidant supplementation and respiratory functions among workers exposed to high levels of ozone. Am. J. Respir. Crit. Care Med. 158: 226-232.

Romieu, I.; Sienra-Monge, J. J.; Ramírez-Aguilar, M.; Téllez-Rojo, M. M.; Moreno-Macías, H.; Reyes-Ruiz, N. I.; Del Río-Navarro, B. E.; Ruiz-Navarro, M. X.; Hatch, G.; Slade, R.; Hernández-Avila, M. (2002) Antioxidant supplementation and lung functions among children with asthma exposed to high levels of air pollutants. Am. J. Respir. Crit. Care Med. 166: 703-709.

Romieu, I.; Sienra-Monge, J. J.; Ramírez-Aguilar, M.; Moreno-Macias, H.; Reyes-Ruiz, N. I.; Estela del Rio-Navarro, B.; Hernández-Avila, M.; London, S. J. (2004) Genetic polymorphism of *GSTM1* and antioxidant supplementation influence lung function in relation to ozone exposure in asthmatic children in Mexico City. Thorax 59: 8-10.

Ruidavets, J.-B.; Cournot, M.; Cassadou, S.; Giroux, M.; Meybeck, M.; Ferrieres, J. (2005) Ozone air pollution is associated with acute myocardial infarction. Circulation 111: 563-569.

Samet, J. M.; Zeger, S. L.; Dominici, F.; Curriero, F.; Coursac, I.; Dockery, D. W.; Schwartz, J.; Zanobetti, A. (2000) The national morbidity, mortality, and air pollution study. Part II: morbidity, mortality, and air pollution in the United States. Cambridge, MA: Health Effects Institute; research report no. 94, part II.

Samet, J. M.; Hatch, G. E.; Horstman, D.; Steck-Scott, S.; Arab, L.; Bromberg, P. A.; Levine, M.; McDonnell, W. F.; Devlin, R. B. (2001) Effect of antioxidant supplementation on ozone-induced lung injury in human subjects. Am. J. Respir. Crit. Care Med. 164: 819-825.

Scannell, C.; Chen, L.; Aris, R. M.; Tager, I.; Christian, D.; Ferrando, R.; Welch, B.; Kelly, T.; Balmes, J. R. (1996) Greater ozone-induced inflammatory responses in subjects with asthma. Am. J. Respir. Crit. Care Med. 154: 24-29.

Schierhorn, K.; Hanf, G.; Fischer, A.; Umland, B.; Olze, H.; Kunkel, G. (2002) Ozone-induced release of neuropeptides from human nasal mucosa cells. Int. Arch. Allergy Immunol. 129: 145-151.

Schierhorn, K.; Zhang, M.; Matthias, C.; Kunkel, G. (1999) Influence of ozone and nitrogen dioxide on histamine and interleukin formation in a human nasal mucosa culture system. Am. J. Respir. Cell Mol. Biol. 20: 1013-1019.

Schwartz, J. (2004) The effects of particulate air pollution on daily deaths: a multi-city case crossover analysis. Occup. Environ. Med. 61: 956-961.

Schwartz, J. (2005) How sensitive is the association between ozone and daily deaths to control for temperature? Am. J. Respir. Crit. Care Med. 171: 627-631.

Schwartz, J.; Spix, C.; Touloumi, G.; Bachárová, L.; Barumamdzadeh, T.; le Tertre, A.; Piekarksi, T.; Ponce de Leon, A.; Pönkä, A.; Rossi, G.; Saez, M.; Schouten, J. P. (1996) Methodological issues in studies of air pollution and daily counts of deaths or hospital admissions. In: St Leger, S., ed. The APHEA project. Short term effects of air pollution on health: a European approach using epidemiological time series data. J. Epidemiol. Commun. Health 50(suppl. 1): S3-S11.

Selwyn, B. J.; Stock, T. H.; Hardy, R. J.; Chan, F. A.; Jenkins, D. E.; Kotchmar, D. J.; Chapman, R. S. (1985) Health effects of ambient ozone exposure in vigorously exercising adults. In: Lee, S. D., ed. Evaluation of the scientific basis for ozone/oxidants standards: proceedings of an APCA international specialty conference; November 1984; Houston, TX. Pittsburgh, PA: Air Pollution Control Association; pp. 281-296. (APCA international specialty conference transactions: TR-4).

Sherwin, R. P.; Richters, V.; Kraft, P.; Richters, A. (2000) Centriacinar region inflammatory disease in young individuals: a comparative study of Miami and Los Angeles residents. Virchows Arch. 437: 422-428.

Sin, D. D.; Man, S. F. P. (2003) Why are patients with chronic obstructive pulmonary disease at increased risk of cardiovascular diseases? Circulation 107: 1514-1519.

Sin, D. D.; Wu, L.-L.; Man, S. F. P. (2005) The relationship between reduced lung function and cardiovascular mortality: a population-based study and a systematic review of the literature. Chest 127: 1952-1959.

Solway, J.; Leff, A. R. (1991) Sensory neuropeptides and airway function. J. Appl. Physiol. 71: 2077-2087.

Spektor, D. M.; Lippmann, M.; Thurston, G. D.; Lioy, P. J.; Stecko, J.; O'Connor, G.; Garshick, E.; Speizer, F. E.; Hayes, C. (1988) Effects of ambient ozone on respiratory function in healthy adults exercising outdoors. Am. Rev. Respir. Dis. 138: 821-828.

Sprenger, M.; Maspoli, M. C.; Wernli, H. (2003) Tropopause folds and cross-tropopause exchange: a global investigation based upon ECMWF analyses for the time period March 2000 to February 2001. J. Geophys. Res. (Atmos.) 108(D12): 10.1029/2002JD002587.

Stenfors, N.; Pourazar, J.; Blomberg, A.; Krishna, M. T.; Mudway, I.; Helleday, R.; Kelly, F. J.; Frew, A. J.; Sandström, T. (2002) Effect of ozone on bronchial mucosal inflammation in asthmatic and healthy subjects. Respir. Med. 96: 352-358.

Stieb, D. M.; Burnett, R. T.; Beveridge, R. C.; Brook, J. R. (1996) Association between ozone and asthma emergency department visits in Saint John, New Brunswick, Canada. Environ. Health Perspect. 104: 1354-1360.

Stieb, D. M.; Judek, S.; Burnett, R. T. (2003) Meta-analysis of time-series studies of air pollution and mortality: update in relation to the use of generalized additive models. J. Air Waste Manage. 53: 258-261.

Stohl, A. (2001) A 1-year Lagrangian "climatology" of airstreams in the Northern Hemisphere troposphere and lowermost stratosphere. J. Geophys. Res. (Atmos.) 106: 7263-7279.

Tepper, J. S.; Wiester, M. J.; Weber, M. F.; Ménache, M. G. (1990) Measurements of cardiopulmonary response in awake rats during acute exposure to near-ambient concentrations of ozone. J. Appl. Toxicol. 10: 7-15.

Thurston, G. D.; Ito, K. (2001) Epidemiological studies of acute ozone exposures and mortality. J. Exposure Anal. Environ. Epidemiol. 11: 286-294.

Thurston, G. D.; Ito, K.; Kinney, P. L.; Lippmann, M. (1992) A multi-year study of air pollution and respiratory hospital admissions in three New York State metropolitan areas: results for 1988 and 1989 summers. J. Exposure Anal. Environ. Epidemiol. 2: 429-450.

Thurston, G. D.; Ito, K.; Hayes, C. G.; Bates, D. V.; Lippmann, M. (1994) Respiratory hospital admissions and summertime haze air pollution in Toronto, Ontario: consideration of the role of acid aerosols. Environ. Res. 65: 271-290.

Tolbert, P. E.; Mulholland, J. A.; Maclntosh, D. L.; Xu, F.; Daniels, D.; Devine, O. J.; Carlin, B. P.; Klein, M.; Dorley, J.; Butler, A. J.; Nordenberg, D. F.; Frumkin, H.; Ryan, P. B.; White, M. C. (2000) Air quality and pediatric emergency room visits for asthma in Atlanta, Georgia. Am. J. Epidemiol. 151: 798-810.

Trenga, C. A.; Koenig, J. Q.; Williams, P. V. (2001) Dietary antioxidants and ozone-induced bronchial hyperresponsiveness in adults with asthma. Arch. Environ. Health 56: 242-249.

Tuazon, E. C.; Winer, A. M.; Pitts, J. N., Jr. (1981) Trace pollutant concentrations in a multiday smog episode in the California South Coast Air Basin by long path length Fourier transform infrared spectroscopy. Environ. Sci. Technol. 15: 1232-1237.

U.S. Environmental Protection Agency. (1990) National air quality and emissions trends report, 1988. Research Triangle Park, NC: Office of Air Quality Planning and Standards; report no. EPA/450/4-90/002.

U.S. Environmental Protection Agency. (1996a) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: http://cfpub2.epa.gov/ncea/.

U.S. Environmental Protection Agency. (1996b) Review of national ambient air quality standards for ozone: assessment of scientific and technical information. OAQPS staff paper. Research Triangle Park, NC: Office of Air Quality Planning and Standards; report no. EPA/452/R-96/007. Available from: NTIS, Springfield, VA; PB96-203435. Available: http://www.epa.gov/ttn/naaqs/standards/ozone/s_o3_pr_sp.html (9 September 2003).

U.S. Environmental Protection Agency. (2003) National air quality and emissions trends report. 2003 special studies edition. Research Triangle Park, NC: Office of Air Quality Standards; Emissions Monitoring and Analysis Division; report no. EPA 454/R-03-005. Available: http://www.epa.gov/air/airtrends/aqtrnd03/toc.html (27 August, 2004).

U.S. Environmental Protection Agency. (2004a) Air quality criteria for particulate matter. Research Triangle Park, NC: National Center for Environmental Assessment; report no. EPA/600/P-99/002aF-bF. 2v. Available: http://cfpub.epa.gov/ncea/ [9 November, 2004].

U.S. Environmental Protection Agency. (2004b) The ozone report: measuring progress through 2003. Research Triangle Park, NC: Office of Air Quality Planning and Standards; report no. EPA-454/K04-001. Available: http://www.epa.gov/air/airtrends/pdfs/2003ozonereport.pdf [12 May, 2005].

Ultman, J. S.; Ben-Jebria, A.; Arnold, S. F. (2004) Uptake distribution of ozone in human lungs: intersubject variability in physiologic response. Boston, MA: Health Effects Institute.

Vagaggini, B.; Taccola, M.; Clanchetti, S.; Carnevali, S.; Bartoli, M. L.; Bacci, E.; Dente, F. L.; Di Franco, A.; Giannini, D.; Paggiaro, P. L. (2002) Ozone exposure increases eosinophilic airway response induced by previous allergen challenge. Am. J. Respir. Crit. Care Med. 166: 1073-1077.

Vedal, S.; Brauer, M.; White, R.; Petkau, J. (2003) Air pollution and daily mortality in a city with low levels of pollution. Environ. Health Perspect. 111: 45-51.

Wainman, T.; Zhang, J.; Weschler, C. J.; Lioy, P. J. (2000) Ozone and limonene in indoor air: a source of submicron particle exposure. Environ. Health Perspect. 108: 1139-1145.

Wattiez, R.; Noél-Georis, I.; Cruyt, C.; Broeckaert, F.; Bernard, A.; Falmagne, P. (2003) Susceptibility to oxidative stress: proteomic analysis of bronchoalveolar lavage from ozone-sensitive and ozone-resistant strains of mice. Proteomics 3: 658-665.

Wayne, R. P. (1991) Chemistry of Atmospheres: an introduction to the chemistry of the atmospheres of Earth, the planets, and their satellites. 2nd ed. New York, NY: Oxford University Press, Inc.

Weschler, C. J. (2004) Chemical reactions among indoor pollutants: what we've learned in the new millennium. Indoor Air 14(suppl. 7): 184-194.

Wilson, A. M.; Wake, C. P.; Kelly, T.; Salloway, J. C. (2005) Air pollution, weather, and respiratory emergency room visits in two northern New England cities: an ecological time-series study. Environ. Res. 97: 312-321.

Woo, K.-S.; Chen, D.-R.; Pui, D. Y. H.; McMurry, P. H. (2001) Measurement of Atlanta aerosol size distributions: observations of ultrafine particle events. Aerosol Sci. Technol. 34: 75-87.

Woodwell, D. A.; Cherry, D. K. (2004) National Ambulatory Medical Care Survey: 2002 summary. Hyattsville, MD: National Center for Health Statistics; DHHS publication no. (PHS) 2004-1250. (Advance data from vital and health statistics; no. 346). Available: http://www.cdc.gov/nchs/data/ad/ad346.pdf [3 August, 2005].

World Health Organization. (2004) Meta-analysis of time-series studies and panel studies of particulate matter (PM) and ozone (O_3): report of a WHO task group. Copenhagen, Denmark: WHO Regional Office for Europe; document no. EUR/04/5042688. Available: http://www.euro.who.int/document/E82792.pdf [18 November, 2004].

9. ENVIRONMENTAL EFFECTS: OZONE EFFECTS ON VEGETATION AND ECOSYSTEMS

9.1 INTRODUCTION

A number of ozone (O_3) effects studies were published between 1996 and 2004, and they are reviewed in this document in the context of the previous O_3 air quality criteria documents (AQCDs) (U.S. Environmental Protection Agency, 1978, 1986, 1992, 1996). Data published since 1996 continue to support the conclusions of previous O_3 AQCDs that there is strong evidence that ambient O_3 concentrations cause foliar injury along with growth and yield damage to numerous common and economically valuable plant and tree species. Research to date has continued to be focused at the species level with very few studies at the ecosystem level. The lack of quantification of biotic and abiotic factors impinging on the individual to population organizational levels results in a limited ability to scale O_3 responses to the ecosystem level. Therefore, a high degree of uncertainty remains in our ability to assess ozone risk to ecological resources and the services they provide.

In general, there has been a shift away from chamber studies in favor of more field-based approaches, although chamber exposures still dominate the effects literature. Field-based approaches include surveys of visible injury, as well as physiological and growth studies using the non-chambered free-air CO_2 exposure (FACE) systems. The FACE systems have substantiated earlier growth and yield results for crop and tree species obtained in open-top chamber (OTC) systems. Increased emphasis has also been placed on quantifying aspects of ozone uptake to better link ambient exposure monitoring with plant/tree response. Much of the progress in quantifying uptake has occurred in Europe in the development of their ozone air quality management tool, the "critical level". The research has developed exposure-response functions for several crops and tree seedlings using OTC studies, as well as in developing and testing models that simulate uptake. Evaluation of this new information has added to our knowledge and provides new research directions, but has not fundamentally altered the conclusions of 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996).

It is well known that O_3 is phytotoxic and that toxicity occurs only if O_3 or its reaction products reach the target tissues in the plant cell. Recent studies have provided an increased

understanding of how ozone interacts with the plant at the cellular level. This increased understanding of cellular-level O_3 effects have translated into better models, more detailed schema of how O_3 alters much of the basic metabolism of plants and how to construct an index that more aptly captures the species, climate, and site factors that alter uptake. These results have and will continue to lead to a better quantification of exposure and effect. However, the translation of these mechanisms into how O_3 is involved in altered cell metabolism and subsequent reductions in whole-plant productivity and ecosystem-level responses remain to be more been fully resolved.

The ensuing sections of this chapter (Section 9.2 to 9.8) are not intended to provide a complete review of the environmental effects of O_3, but rather an assessment of key information published since the 1996 O_3 AQCD. More detailed discussion of the research since 1996 is provided in Chapter 9 Annex Sections AX9.1 to AX9.7 (in Volume 3 of this document). The framework for Chapter 9 follows the environmental effects chapter of the 1996 O_3 AQCD. First, an overview of various methodologies that have been, and continue to be, central to the quantification of O_3 effects on vegetation is provided in Section 9.2 below (see Section AX9.1 for more detailed discussion). The adequacy of each methodology is discussed in the context of developing statistically robust data appropriate for assessing the risk of O_3 to vegetation resources. In Section 9.3, research is then reviewed from the molecular to the biochemical and physiological levels in plants that are impacted, which offers insight into the mode of action of O_3 (see also AX9.2). The manner in which plants respond to O_3, as influenced by the numerous biotic and abiotic factors present in the environment, is next discussed (see also AX9.3). Quantifying these various modifiers is critical to being able to scale the response of individual plants to the community level and across varied landscapes and climates and is needed for regional to national assessments of risk. The development of indices of exposure or O_3 uptake is discussed in the context of their adequacy to realistically describe the ambient concentration-response relationships (see also AX9.4). The exposure-response relationships for a large number of crop species and cultivars, native vegetation, and tree species are reviewed, tabulated, and compared to form the basis for an assessment of the potential risk of current levels of O_3 on vegetation resources (see also AX9.5). Available research by which to assess the impact of O_3 on ecosystems is also reviewed, along with the potential data available for estimating the loss of

various ecosystem services (see also AX9.6). Finally, available research on the economic impact of ozone effects on vegetation resources is briefly discussed (see also AX9.7).

9.2 METHODOLOGIES USED IN VEGETATION RESEARCH

New methodological advancements since 1996 have not fundamentally altered our understanding of O_3 effects on plants or ecosystems. Most of the new information confirms earlier conclusions and provides additional support for OTC use in assessing sensitive species and developing exposure-response relationships. A more in-depth discussion of this topic can be found in Annex Section AX9.1.

The majority of ozone effects studies are fumigation studies conducted in controlled chambers, as noted in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). That document noted that OTCs represented the best technology for determining statistically robust exposure response models of O_3 and crop yield and plant biomass at that time. While OTCs are still the best method for conducting controlled exposures of varying length and frequency for developing exposure-response relationships, several new approaches have been applied to O_3 effects research — most notably free-air exposure or "plume" systems. Free-air exposure systems (FACE) eliminate many of the concerns raised about closed or open-top chamber experiments including small plot size, altered microclimate within OTCs, and the effect of charcoal filtering on overall air quality within OTCs. FACE systems have increased our understanding in some areas; and results from FACE studies have, on the whole, confirmed what was already understood or hypothesized about how plants and plant assemblages respond to O_3. Some shortcomings of using plume systems in O_3 research have also been identified, namely the relatively poor control of exposure levels, the presence of "hotspots" and the inability to decrease O_3 concentrations to below ambient levels when ambient concentrations are phytotoxic. Nonetheless, the application of FACE systems and other open-air systems to ozone exposure research have greatly helped our scaling efforts and are, perhaps, the best approach for studying the response of plant species mixtures to O_3 (Nussbaum and Fuhrer, 2000).

One of the advantages of the application of plume systems to O_3 research is the ability to compare response of plants in open-field systems with results from OTCs. In particular, studies with quaking aspen (*Populus tremuloides* L.) performed in OTCs, FACE, and also sites along an

ambient ozone gradient showed that ozone symptom expression was generally similar across these methodologies, supporting the previously observed level of variation among aspen clones in OTC studies (Isebrands et al., 2000, 2001; Karnosky et al., 1999). While this perhaps represents the first time direct comparisons were made, it supports the use of OTC data in the development of O_3 response functions for individual species. Concerns raised earlier about microclimate differences in chambers and the role of "chamber effects" (Fuhrer, 1994; Manning and Krupa, 1992) still persist; however, most evidence suggests that chamber effects result in altered O_3 uptake without altering the fundamental response of plants to ozone, thus reducing uncertainty in the use of data from OTCs. Extrapolation of the results from chamber studies depends on fully characterizing temperature, light, turbulence, and other chamber characteristics during exposures (Nussbaum and Fuhrer, 2000), but study design is equally important. Conducting studies with a large number of plant species across regions of the country where those species are indigenous is important in capturing regional climatic differences to reduce the uncertainty associated with extrapolating composited response functions across regions and to identify relative risk to vegetation in relation to given O_3 exposure values (U.S. Environmental Protection Agency, 1996).

The lack of rural monitors continues to be a major problem in the characterization of O_3 exposures in remote areas, as well as in linking effects to exposure in natural ecosystems. Since the 1996 O_3 AQCD, the use of passive samplers has expanded monitoring efforts to include remote areas that were previously uncharacterized. This has greatly enhanced our ability to link ozone symptomology with elevated O_3 exposure in such remote areas. However, passive samplers do not capture the temporal dynamics of exposure. Therefore, passive samplers cannot substitute for active monitors when attempting to link exposure dynamics to plant response or when developing exposure/dose-response relationships of much value as inputs for the standard setting process. To overcome this problem, Krupa et al. (2001, 2003) used models and data from a collocated O_3 monitor to estimate the underlying frequency distribution of hourly O_3 concentrations from passive samplers. Future development of passive monitor technology and data synthesis techniques holds promise, particularly since it is unlikely that extensive O_3 monitoring networks will be established in rural areas in the near future.

Exclusion methods that employ protective chemicals such as ethylenediurea (EDU) are the least disruptive of ambient culture conditions in the field, as noted in the 1996 O_3 AQCD.

However, the level of protection afforded by EDU is site- and species-specific and is subject to local meteorologic conditions. In addition, new evidence suggests that EDU does not always have greater effects at higher O_3 exposures and that the degree of protection by EDU largely depends on environmental conditions. Because of the variability observed in the level of protection provided, and the fact that mechanisms of protection afforded by EDU and other exclusion methods are unknown, caution is needed in applying this approach to the study of O_3 effects in the field.

Advancements in biomonitoring have been made since the 1996 O_3 AQCD, primarily in the area of identification and symptom verification of sensitive species (Flagler, 1998; Krupa et al., 1998; Innes et al., 2001; Smith et al., 2003). The U.S. Department of Agriculture (USDA) Forest Service continues its program to monitor ozone effects in forested ecosystems throughout the United States. Currently, 33 states participate in the program, which uses a grid system to identify the location of plants showing foliar injury. Although results cannot be used for developing exposure-response relationships or for quantifying responses to O_3, they can provide an annual assessment and correlative information regarding the extent of O_3 injury occurring across many regions of the United States.

9.3 SPECIES RESPONSE/MODE-OF-ACTION

There are several steps in the process of O_3 uptake and toxicity that are now better understood than in 1996. These advancements are important in refining hypotheses on O_3 uptake and mode of action on plants and in developing a flux-based index for use in quantifying response and, ultimately, for potential use in developing a secondary national ambient air quality standard (SNAAQS). The new information available on the mode of action of O_3 is, in part, a result of improved molecular tools for following rapid changes that occur within the leaf (Pell et al., 1997; Sandermann, 2000; Ward et al., 1991). This new information is discussed in greater detail in Annex Section AX9.2.

Clearly, many changes occur within hours or possibly days following O_3 exposure (Sandermann, 1998). However, other O_3 effects take longer to occur and tend to be most obvious only under exposure to low O_3 concentrations for long periods (Andersen et al., 1997; Hogsett et al., 1989; Langebartels et al., 1997). These low-exposure chronic effects have been

linked to the senescence process or some physiological response very closely linked to senescence (e.g., translocation, reabsorption, allocation of nutrients and carbon).

Langebartels et al. (1997) discussed "memory" or "carry-over effects" within the plant to explain sensitivity to frost in the winter following summertime O_3 exposure. Others have argued that this sensitivity is due to the nutrient status of the tree during the over-wintering phase of its life and to chronic (on-going, less severe levels with fewer peaks at very high levels) exposure to ambient O_3 inducing (1) mineral nutrient deficiency; (2) alterations of normal metabolism, including translocation and allocation of carbohydrates and probably nitrogen; and/or (3) disturbance of normal transpiration and diurnal cycling, leading to water stress (Schmieden and Wild, 1995). While general nutrient concentrations within the foliage may not occur, localized deficiencies might. This is difficult to observe or prove without a great deal of work on all portions of a tree and without a general hypothesis of what is occurring.

It is important to note that the dramatic strides made over the last few years in understanding the genetic make-up of plants, gene control, and signal transduction/control will accelerate in the future and translate into better models of the hypotheses listed above as well as more detailed schemes of how O_3 alters basic plant metabolism. Thus, while our understanding of how O_3 interacts with the plant at the cellular level has dramatically improved (Assmann, 2003; Assmann and Wang, 2001; Rao and Davis, 2001), the translation of those mechanisms into how O_3 is involved with altered cell metabolism and the subsequent reductions in whole plant productivity and other physiological facts has not yet been fully achieved. As the understanding of wounding responses in plants and more information on genome details and varied plant mutants becomes available, the cellular and physiological responses of plants to O_3 exposures are slowly becoming clearer. However, more studies on a larger variety of species are needed before this type of information can be incorporated into indices of response and for consideration in developing SNAAQS.

9.4 MODIFICATION OF FUNCTIONAL AND GROWTH RESPONSES

It has been known for decades that several factors, both biotic and abiotic, alter plant response to ozone. However, only a few studies reported since the 1996 O_3 AQCD have improved our understanding of the role of these interactions in modifying plant O_3 response.

Quantifying how these interactions alter plant O_3 response is a critical first step to reducing the uncertainty in extrapolating individual plant responses to higher levels of biological organization, e.g., ecosystems. None of the recent studies have significantly improved our ability to quantify the degree to which these factors modify plant O_3 response; however, they have reinforced the conclusions of the 1996 O_3 AQCD with regard to factors known to alter plant response to O_3. This new information is discussed in greater detail in Annex Section AX9.3.

In the area of biotic interactions, new evidence with regard to insect pests and diseases (see Docherty et al. (1997) and Flückiger et al. (2002) for recent reviews) has not reduced the uncertainties noted in the 1996 O_3 AQCD. Most interactions thought to affect crops, forest trees and other natural vegetation have yet to be studied. Recent studies have supported the earlier conclusion that O_3 often increases the likelihood and success of insect attacks, but only with respect to chewing insects (e.g., Percy et al., 2002; Kopper and Lindroth, 2003). With the economically important group of sucking insects (such as aphids), no clear trends have been revealed in the latest studies (see reviews by Docherty et al., 1997; Flückiger et al., 2002). Hence, although it seems likely that some insect problems could increase as a result of greater O_3 levels, we are still far from being able to predict the nature of any particular O_3-plant-insect interaction, its likelihood, or its severity.

The situation is somewhat clearer with respect to interactions involving facultative necrotrophic plant pathogens, with O_3 exposure generally contributing to increased disease (Flückiger et al., 2002). With obligate biotrophic fungal, bacterial, and nematode diseases, however, twice as many reports indicate O_3-induced inhibitions than enhancements. This pattern is supported by the concept put forth by Dowding (1988) that pathogens that benefit from damage to cells are enhanced by pollution stress of their hosts, whereas pathogens and pests that require healthy hosts are depressed by pollution stress. The frequent reports that infection by obligate biotrophs reduces the severity of O_3-induced foliar injury (e.g., Schraudner et al., 1996) does not result in true "protection", as the disease *per se* causes negative effects on the host plant. With obligate biotrophs, the nature of any interaction with O_3 is probably dictated by the unique, highly specific biochemical relationships between the pathogen and the host plant. At this time, therefore, although some diseases may become more widespread or severe as a result

of exposure to O_3, it is still not possible to predict exactly which diseases are likely to present the greatest risks to crops and forests.

Recent studies have not greatly added to our understanding of the nature of interactions between O_3 and root symbionts, but have served to support conclusions put forth in the 1996 O_3 AQCD. Several studies have indicated that the functioning of tree root symbioses with mycorrhizae may be adversely affected by O_3 (e.g., Kytöviita et al., 2001), but there is also evidence that the presence of mycorrhizae may overcome O_3-enhanced root diseases (Bonello et al., 1993). There is also evidence that O_3 may encourage the spread of mycorrhizae to the roots of uninfected trees. The role of O_3 in altering root symbionts, its interactions with soil organisms, and the subsequent feedback effects on plant growth represent one of the greatest areas of uncertainty in assessing the influence of O_3 on ecosystems (Andersen, 2003).

The few recent studies of the impact of O_3 on intraspecific plant competition confirmed that grasses frequently show greater resilience than other types of plants. In grass-legume pastures, the leguminous species tend to suffer greater growth inhibition (Johnson et al., 1996; Nussbaum et al., 2000). The suppression of Ponderosa pine (*Pinus ponderosa* Laws.) seedling growth by blue wild-rye grass (*Elymus glaucus* Buckl.) was markedly increased by O_3 (Andersen et al., 2001). However, we are far from being able to predict the outcome of the impact of O_3 on specific competitive situations, such as successional plant communities or crop-weed interactions.

Physical or abiotic factors play a large role in modifying plant response to O_3, and new information is available that supports the conclusions of the 1996 O_3 AQCD. Although some recent field studies have indicated that O_3 impact significantly increases with increased ambient temperature (Ball et al., 2000; Mills et al., 2000), other studies have indicated that temperature has little effect (Balls et al., 1996; Fredericksen et al., 1996). Temperature affects the rates of all physiological processes based on enzyme-catalysis and diffusion; each process and overall growth (the integral of all processes) has a distinct optimal temperature range. It is important to note that a plant's response to changes in temperature will depend on whether it is growing near its optimum temperature for growth or near its maximum temperature (Rowland-Bamford, 2000). But temperature is unquestionably an important variable affecting plant O_3 response in the presence of the elevated CO_2 levels contributing to global climate change. In contrast, evidence continues to accumulate to indicate that O_3 exposure sensitizes plants to low

temperature stress (Colls and Unsworth, 1992) and, also, that O_3 decreases below-ground carbohydrate reserves, which may lead to responses in perennial species ranging from rapid demise to impaired growth in subsequent seasons (i.e., carry-over effects) (Andersen et al., 1997).

Light, a component of the plant's physical environment, is an essential "resource" whose energy content drives photosynthesis and CO_2 assimilation. It has been suggested that increased light intensity may increase O_3 sensitivity of light-tolerant species while decreasing that of shade-tolerant species, but this appears to be an oversimplification with many exceptions. As pointed out by Chappelka and Samuelson (1998) and Topa et al. (2001), the interaction between O_3 sensitivity and light environment is complicated by developmental stage as well as the light environment of individual leaves in the canopy.

Although the relative humidity of the ambient air has generally been found to increase the adverse effects of O_3 by increasing stomatal conductance, and thereby increasing O_3 flux, abundant evidence also indicates that the ready availability of soil moisture results in greater O_3 sensitivity (Mills, 2002). The partial "protection" against the adverse effects of O_3 afforded by drought (as noted in previous O_3 AQCDs) has been observed in field experiments and modeled in computer simulations (Broadmeadow and Jackson, 2000). There is also compelling evidence that O_3 can predispose plants to drought stress (Maier-Maercker, 1998). Hence, the response will depend to some extent upon the sequence in which the stresses occur, but, even though the nature of the response is largely species-specific, successful applications of model simulations have led to larger-scale predictions of the consequences of O_3 × drought interactions. However, regardless of the interaction, the net result on short-term growth is negative; although in tree species, other responses such as increased water use efficiency could benefit long-term survival.

Somewhat analogous to temperature, it appears that any shift away from the nutritional optimum may lead to greater O_3 sensitivity; but the shift would have to be substantial before a significant effect on O_3 response was observed. Mineral nutrients in the soil, other gaseous air pollutants, and agricultural chemicals constitute chemical factors in the environment. The evidence regarding interactions with specific nutrients is still contradictory: some experimental evidence indicates that low general fertility increases sensitivity to O_3 (Whitfield et al., 1998; Landolt et al., 1997), although others have found less sensitivity with decreased fertility (Cardoso-Vilhena and Barnes, 2001). Simulation modeling of trees suggests that nutrient

deficiency and O_3 may act less than additively, but too many examples of contrary trends exist to permit any sweeping conclusions at this time.

Interactions of O_3 with other air pollutants have received relatively little recent attention since 1996 (see Barnes and Wellburn [1998] and Fangmeier et al. [2002] for recent reviews). The situation with SO_2 remains inconsistent, but SO_2 seems unlikely to pose any additional risk to those related to other individual pollutants. With NO and NO_2, the situation is complicated by their nutritional value as a N source. Much more investigation is needed before we will be able to predict the outcomes of different O_3-NO-NO_2 scenarios. The latest research into O_3 × acid rain interactions has confirmed that, at realistic acidities, significant interactions are unlikely (Momen et al., 1997; 1999; Laurence et al., 1997; Sayre and Fahey, 1999). A continuing lack of information precludes our offering any generalizations about interactive effects of O_3 with NH_3, HF, or heavy metals. More evidence has been reported for protective effects against O_3 afforded by the application of fungicides (Wu and Tiedemann, 2002).

Considerable emphasis during the last decade has been placed on research evaluating potential O_3 interactions with the components of global climate change: increased atmospheric CO_2, increased mean global temperatures, and increased surface level UV-B radiation. However, it must be noted that most of these studies have tended to regard increased CO_2 levels and increased mean temperatures as unrelated phenomena. Experiments into the effects of doubled CO_2 levels at today's mean ambient temperatures are of questionable value in trying to assess the impact of *climate change* on responses to O_3. To date, the limited experimental evidence and that obtained by computer simulation suggest that even though an enriched CO_2 atmosphere (~600 ppm) would more than offset the impact of O_3 on responses as varied as wheat (*Triticum aestivum* L.) yield or young Ponderosa pine growth, the concurrent increase in temperature would reduce, but probably not eliminate, the net gain (Batts et al., 1997; Van Oijen and Ewart, 1999; Constable et al., 1996). There is also some recent evidence that O_3 and UV-B interact in their effects on plant injury and photosynthesis (Schnitzler et al., 1999), but additional research is needed to fully understand how O_3 interacts with multiple climate change factors.

9.5 EFFECTS-BASED AIR QUALITY EXPOSURE INDICES

Since the 1996 O_3 AQCD , there has been no direct experimental testing of the adequacy of exposure indices proposed in 1996; therefore, there is no new information to alter the basic conclusions put forth in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996) with regard to exposure indices. A more detailed discussion of effects-based air quality indices can be found in Annex Section AX9.4.

Exposure indices are metrics that relate measured plant damage (i.e., reduced growth) to monitored ambient O_3 concentrations over time to provide a consistent metric for reviewing and comparing exposure-response effects obtained from various studies. The 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996) focused on the research used to develop various exposure indices to quantify growth and yield effects in crops, perennials, and trees (primarily seedlings), and not foliar injury. The proposed indices included various functional and statistical summaries of monitored hourly O_3 concentrations over designated time periods. The indices were developed through regression analyses of earlier exposure studies and was accomplished by ordering the measured responses of growth and/or yield of crops and tree (seedling) species in response to O_3. Their development focused on consideration and inclusion of some, but not all, the factors that affect O_3 uptake and expression of effects (e.g., Lee et al. [1988]).

The few studies that have been published since the 1996 O_3 AQCD continue to support the earlier conclusions, including the importance of peak concentrations, and the duration and occurrence of O_3 exposures in altering plant growth and yield. In addition, a large body of new research, mostly out of Europe, addresses the need for an index related to the actual uptake of O_3 by the plant and the flux of O_3 from the atmosphere to the O_3 affected plant tissues. Despite additional research linking estimates of flux with plant response since 1996, information is still insufficient to identify a flux-based model that incorporates the necessary complexity across space and time to be non-site or non-species specific. Based on the current state of knowledge, exposure indices that cumulate and differentially weight the higher hourly average O_3 concentrations, but include the mid-level values, still represent the best approach for relating vegetation effects to O_3 exposure in the United States.

The new studies have also substantiated earlier conclusions on the role of exposure components including concentration, duration, and exposure patterns in determining plant growth response to O_3 (Oksanen and Holopainen, 2001; Yun and Laurence, 1999). Recent

studies using different exposure patterns have confirmed earlier studies on the role of higher concentrations and exposure duration (Nussbaum et al., 1995). A role for higher concentrations is inferred based on improved air quality in regions in the Western United States (Lefohn and Shadwick, 2000). For example, the O_3 reductions in the San Bernardino Mountain area since the late 1970s are associated with reductions in the higher hourly average O_3 concentrations, the number of hours of concentrations $\geq$0.95 ppm, and the cumulative concentration-weighted exposure index (Lee et al., 2003). The mid-range concentrations appeared to be relatively unchanged or even slightly increasing over the period of 1980 to 2000. General forest improvement has been reported following a decrease of O_3 along a decreasing gradient of exposure (Miller and Rechel, 1999; Arbaugh et al., 2003; Tingey et al., 2004). These studies suggest the focus should be on the higher O_3 concentrations, while including the lower levels, when estimating the effects of O_3 precursor emission reduction strategies on vegetation.

New studies have demonstrated the potential disconnection of peak events and maximal stomatal conductance at xeric to mesic sites in California (Panek et al., 2002; Grulke et al., 2002; Panek, 2004). In addition, a few studies have indicated that O_3 uptake during nighttime hours is greater than previously thought (Grulke et al., 2004; Massman, 2004); and a review of the literature suggests a large number of species exhibit some degree of conductance at night (Musselman and Minnick, 2000). These studies suggest a reconsideration of cumulating exposure 24 h/day and not just during daylight hours in exposure index determinations. This lack of coincidence in temporal patterns of conductance and peak ambient concentrations introduces uncertainty in assessing the impact of O_3. The use of an exposure index that does not consider regionally unique climate and site factors that modify stomatal conductance may, as a result, under- or over- estimate growth effects. The shortcomings of an ambient exposure-based index is especially apparent when assessing the potential impact of O_3 across broad climatic regions of the United States or Europe. Various means to overcome this potential problem were addressed with several new studies; one solution would be to add other components to the present statistical summaries of exposure indices (e.g., meteorological) to develop flux-based indices. However, the increased biological and meteorological information in these indices may make them more regional in their applicability.

A number of studies have taken a flux-based approach to improve upon the concentration-based (i.e., exposure indices) approach as a means to address the issue of

assessing risk of O_3 across different climatic regions. The European acceptance and use of the flux-based critical values is, in part, a recognition of the landscape scaling problems associated with ambient exposure-based indices. A great deal of progress has occurred in developing and testing stomatal models that may be generally applicable across certain vegetation types (Danielsson et al., 2003; Emberson et al., 2000; Grünhage and Jäger, 2003; Matyssek et al., 2004; Pleijel et al., 2000). While a flux-based approach is preferred, a cautionary argument was advanced in a few publications based on the nonlinear relationship between O_3 uptake and foliar injury (growth was not assessed). The concern is that not all O_3 stomatal uptake results in a yield reduction, which depends to some degree on the amount of internal detoxification occurring with each particular species. Those species having high amounts of detoxification potential may, in fact, show little relationship between O_3 stomatal uptake and plant response (Musselman and Massman, 1999).

Given the current state of knowledge and the best available data, exposure indices that cumulate and differentially weight the higher hourly average concentrations, and also include the mid-level values, continue to offer the most defensible approach for use in developing response functions and comparing studies as well as future indices for vegetation protection. A large database exists that has been used for establishing exposure-response relationships; however, at this time, such a database does not exist for relating O_3 flux to growth response.

It is anticipated that as the overlapping relationships of conductance, concentration, and defense mechanisms are better defined, the flux-based indices may be able to predict vegetation injury and/or damage across varied landscapes and climates with more accuracy than the exposure-response models. However, it is unclear that such is the case at this time. The translation of these indices from research and assessment tools to air quality standards has the additional need to be simple, understandable, and adaptive to a manageable monitoring program.

9.6 OZONE EXPOSURE-PLANT RESPONSE RELATIONSHIPS

Data published since 1996 continue to support the conclusions of previous O_3 AQCDs that there is strong evidence that ambient concentrations of O_3 cause foliar injury and growth and yield damage to numerous common and economically valuable plant and tree species. For annual vegetation, the data summarized in Table AX9-16 (see Annex Section AX9.5) show a

range of growth and yield responses both within species and among species. Nearly all of these data were derived from studies in OTCs, with only two studies using open-air systems in the United Kingdom (Ollerenshaw et al., 1999; Ollerenshaw and Lyons, 1999). It continues to be difficult to compare studies that report O_3 exposure using different indices, such as AOT40, SUM06, W126, or 7-h or 12-h mean values.

The AOT40, SUM06, and W126 indices are defined as follows:

AOT60: the seasonal sum of the difference between an hourly concentration above the threshold value of 60 ppb, minus the threshold value of 60 ppb;

SUM06: the seasonal sum of hourly concentrations at or above the threshold value of 60 ppb; and

W126: a sigmoid functional weighting of all hourly concentrations for the season.

When such index comparisons can be made, the results of recent research confirm earlier results summarized in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). A summary of earlier literature concluded that a 7-h, 3-month mean of 49 ppb O_3 corresponding to a SUM06 exposure of 26 ppm·h, would cause 10% loss in 50% of 49 experimental cases (Tingey et al., 1991). Recent data summarized in Table 9-16 support this conclusion and more generally indicate that ambient O_3 exposures can reduce the growth and yield of annual species. Some annual species such as soybean [*Glycine max* (L.) Merr.] are more sensitive, and greater losses in such species may be expected (Table 9-16). Thus, the recent scientific literature supports the conclusions of the 1996 O_3 AQCD that ambient O_3 concentrations are reducing the yield of major crops in the United States.

Much research in Europe has used the cutoff-concentration weighted, cumulative-exposure statistic AOT40; and substantial effort has gone into developing "Level-1" critical levels for vegetation using this index. Based on regression analysis of 15 OTC studies of spring wheat including one U.S. study and 14 studies from locations ranging from southern Sweden to Switzerland, an AOT40 value of 5.7 ppm·h was found to correspond to a 10% yield loss, and a value of 2.8 ppm·h corresponded to a 5% yield loss (Fuhrer et al., 1997). Because a 4 to 5% decrease could be detected with a confidence level of 99%, 3 ppm·h was selected by the European Union as the AOT40 critical level in 1996 (Kärenlampi and Skärby, 1996).

In addition to reductions in crop yield, O_3 may also reduce the quality or nutritive value of annual species. Many studies have found O_3 effects on various measures of plant organs that affect quality, with most of those studies focusing on characteristics important for food or fodder. These studies indicate that ambient O_3 may have economically important effects on the quality of crop and forage species. Previous O_3 AQCDs have concluded that visible symptoms on marketable portions of crops and ornamental plants can occur with seasonal 7-h mean O_3 exposures of 40 to 100 ppb (U.S. Environmental Protection Agency, 1978; 1986; 1992; 1996). The recent scientific literature does not refute this conclusion.

The use of OTCs may reverse the usual vertical gradient in O_3 that occurs within a few meters above the ground surface (see Annex Section AX9.1). This reversal suggests that OTC studies may, to some degree, overestimate the effects of an O_3 concentration as measured several meters above the ground. However, such considerations do not invalidate the conclusion of the 1996 O_3 AQCD that ambient O_3 exposures are sufficient to reduce the yield of major crops in the United States.

As found for single-season agricultural crops, yields of multiple-year forage crops are reduced at ozone exposures that occur over large areas of the United States. This result is similar to that reported in the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996). When species are grown in mixtures, O_3 exposure can lead to the increased growth of O_3-tolerant species while exacerbating the growth decrease of O_3-sensitive species. Because of this competitive interaction, the total growth of the mixed-species community may not be affected by O_3 exposure. However, in some cases, mixtures of grasses and clover species have shown significant decreases in total biomass growth in response to O_3 exposure in studies in the United States and in Sweden. In Europe, a provisional AOT40 critical level of 7 ppm·h over 6 months has been proposed by the European Union as a value to protect sensitive herbaceous perennial plant species from the adverse effects of O_3.

For deciduous tree species, recent evidence from FACE and OTC studies supports results observed in previous OTC studies. For example, a series of O_3-FACE studies was undertaken in Rhinelander, WI (Isebrands et al., 2000, 2001). These studies showed that O_3 symptom expression was generally similar in OTCs, FACE, and also at sites along an ambient O_3 gradient, supporting the previously observed variation among aspen clones obtained using OTCs

(Karnosky et al., 1999). As has been observed in previous O_3 AQCDs, root growth is often found to be the most sensitive biomass response to O_3.

Results since 1996 support the conclusion of the 1996 O_3 AQCD (U.S. Environmental Protection Agency, 1996) that deciduous trees are generally less sensitive to O_3 than are most annual plants, with the exception of a few very sensitive genera such as *Populus* and sensitive species such as black cherry (*Prunus serotina* Ehrh.). However, the data presented in Table AX9-18 (see Annex Section AX9.5) suggest that ambient exposures that occur in the United States can sometimes reduce the growth of seedlings of deciduous species. Results from multi-year studies sometimes show a pattern of increased effects in subsequent years. In some cases, however, growth decreases due to O_3 may become less significant or even disappear over time. While some mature trees show greater O_3 sensitivity than do seedlings in physiological parameters such as net photosynthetic rate, these effects may not translate into measurable reductions in biomass growth. However, because even multi-year experiments do not expose trees to O_3 for more than a small fraction of their life span and because competition may, in some cases, exacerbate the effects of O_3 on individual species, determining O_3 effects on mature trees remains a significant challenge.

In Europe, a Level I critical level has been set for forest trees based on OTC studies of European beech (*Fagus sylvatica* L.) seedlings: defined as an AOT40 value of 10 ppm·h for daylight hours for a 6-month growing season (Kärenlampi and Skärby, 1996). However, other studies show that other species, such as silver birch(*Betula pendula* Roth.), may be more sensitive to O_3 than beech (Pääkkönen et al., 1996).

As found for other tree species, various evergreen tree species and genotypes have widely varying O_3 sensitivities. Based on OTC studies with seedlings, major evergreen species in the United States are generally less sensitive than are most deciduous trees, and slower-growing evergreen species are less sensitive than are faster-growing species. There is evidence that interacting stress, such as competition, may increase the O_3 sensitivity of trees. As in studies of deciduous species, most experiments with evergreen species have only covered a very small portion of the life span of a tree and have been conducted with seedlings, so estimating effects on mature evergreens is difficult.

For all types of perennial vegetation, cumulative effects over more than one growing season may be important; and studies for only a single season may underestimate effects.

Mature trees may be more or less sensitive to O_3 than are seedlings, depending on the species, but information on physiological traits can be used to predict such differences in specific cases. In some cases, mature trees may be more sensitive to O_3 than seedlings due to differences in gas exchange rates, differences in growth rates, greater cumulative exposure, or the interaction of O_3 with other stressors.

9.7 EFFECTS OF OZONE EXPOSURE ON NATURAL ECOSYSTEMS

There is evidence that tropospheric O_3 is an important stressor of ecosystems, with documented impacts on the biotic condition, ecological processes, and chemical/physical nature of natural ecosystems (See Table AX9-22; Annex Section AX9.6). In turn, the effects of O_3 on individual plants and processes are scaled up through the ecosystem affecting processes such as energy and material flow, inter- and intraspecies competition, and net primary productivity (NPP). Thus, effects on individual keystone species and their associated microflora and fauna, which have been shown experimentally, may cascade through the ecosystem to the landscape level, although this has not yet been demonstrated. By affecting water balance, cold hardiness, tolerance to wind and by predisposing plants to insect and disease pests, O_3 may even impact the occurrence and impact of natural disturbance (e.g., fire, erosion). Despite the probable occurrence of such effects, there are essentially no instances where ecosystem level, highly integrated studies have conclusively shown that ozone is indeed altering ecosystem structure and/or function.

Systematic injury surveys demonstrate that foliar injury occurs to O_3 sensitive species in many regions of the United States (Smith et al., 2003; Coulston et al., 2003; Chappelka et al., 1997) (Campbell et al., 2000) and Europe (Braun et al., 1999). However, the frequent lack of correspondence between foliar symptoms and growth effects means that other methods must be used to estimate the regional effects of O_3 on tree growth rates (e.g., Rebbeck, 1996; Kouterick et al., 2000). Investigations of the radial growth of mature trees in combination with data from many controlled studies with seedlings and a few studies with mature trees suggest that ambient O_3 is reducing the growth of mature trees in some locations (Somers et al., 1998). Studies using models based on tree physiology and forest stand dynamics suggest that modest effects of O_3 on growth may accumulate over time and may interact with other stressors (Laurence et al., 2001)

(Laurence et al., 2003). For mixed-species stands, such models predict that overall stand growth rate is generally not likely to be affected. However, competitive interactions among species may change as a result of growth reductions of O_3-sensitive species (Weinstein et al., 2001). These results suggest that O_3 exposure over decades may be altering the species composition of forests in some regions.

Despite increased understanding of possible ecosystem effects of ozone, the data base demonstrating and quantifying the degree to which O_3 is altering natural ecosystems is sparse. Much of the speculation of ozone impact on ecosystems must be inferred from a number of case studies of forest plot field-based data reporting on a number of different species. One means to discuss our current knowledge is by listing the areas in which more information is needed. These include:

Ecosystem processes. Very little is known about the effects of O_3 on water, carbon, and nutrient cycling, particularly at the stand and community levels. Effects on below-ground ecosystem processes in response to O_3 exposure alone, and in combination with other stressors, are critical to projections at the watershed and landscape levels. Little is yet known about the effects of O_3 on structural or functional components of soil food webs or how these impacts could affect plant species diversity (Andersen, 2003).

Biodiversity and genetic diversity. The study of genetic aspects of O_3 impacts on natural ecosystems has been largely based on correlations, and it remains to be shown conclusively whether O_3 affects biodiversity or genetic diversity (Pitelka, 1988; Winner et al., 1991; Davison and Barnes, 1998). Studies of competitive interactions under elevated O_3 levels are needed (Laurence and Andersen, 2003). Reexaminations via new sampling of population studies to bring in a time component to previous studies showing spatial variability in population responses to O_3 are also needed. These studies could be strengthened by modern molecular methodologies to quantify impacts on diversity.

Natural ecosystem interactions with the atmosphere. Little is known about feedbacks between O_3 and climate change on production of volatile organic compounds, which, in turn, could affect O_3 production (Fuentes et al., 2001). At moderate to high O_3 exposure sites, aberrations in stomatal behavior could significantly affect individual tree water balance of O_3-sensitive trees; and if the sensitive tree species is dominant, the hydrologic balance at the watershed and landscape levels could be affected. This has not been addressed in any model,

because O_3-exposure effects, if included at all in the modeling effort, have assumed a linear relationship between assimilation and stomatal conductance. Interaction studies with other components of global change (i.e., warming, increasing atmospheric CO_2, N deposition, etc.) or with various biotic stressors are needed to better predict complex interactions likely in the future (Laurence and Andersen, 2003). Whether O_3 will negate the positive effects of an elevated CO_2 environment on plant carbon and water balances is not yet known, nor is it known if these effects will scale up through the ecosystem. How O_3 affects the progress of pest epidemics and insect outbreaks as concentrations increase is unclear (Ball et al., 1998). Information concerning the impact of O_3 on plant pest and insect reproductive processes and reproductive development under realistic field or forest conditions are needed as well as examination of reproductive effects under interacting pollutants (Black et al., 2000).

Scaling. The vast majority of O_3 studies of trees have been conducted with young, immature trees and in trees that have not yet formed a closed canopy. Questions remain as to the comparability of O_3 effects on juvenile and mature trees and on trees grown in the open versus those in a closed forest canopy in a competitive environment (Chappelka and Samuelson, 1998; Kolb and Matyssek, 2001; Samuelson and Kelly, 2001). Merging the effects of O_3 across spatial scales is also difficult. Scaling responses of a single or a few plants to effects on communities and ecosystems are complicated matters that will require a combination of manipulative experiments with model ecosystems; community and ecosystem studies along natural O_3 gradients; and extensive modeling efforts to project landscape level, regional, national and international impacts of O_3. Linking these various studies via impacts on common research quantification across various scales using measures of such factors as leaf area index or spectral reflective data, which could eventually be remotely sensed (Kraft et al., 1996; Panek et al., 2003), would provide powerful new tools for ecologists.

Identifying endpoints. In general, methodologies to determine the important values of services and benefits derived from natural ecosystems are lacking. Identifying and quantifying factors that could be used in comprehensive risk assessment for O_3 effects on natural ecosystems would increase societal awareness of the importance of protecting ecosystems (Heck et al., 1998).

9.8 ECONOMICS

Substantial progress has been made over the past two decades in our understanding of the effects of ozone and other oxidants on vegetation, particularly for agriculturally important plant species. See Annex Section AX9.7 for a more detailed discussion. The physical and economic effects on agriculture are well documented and provide useful information for the consideration of establishing air quality standards for crops (e.g., Spash, 1997).

Since the completion of the National Crop Loss Assessment Newwork (NCLAN) program in the late 1980s, the number of economic assessments of air pollution studies focusing on terrestrial ecosystems in general, and agriculture in particular, has declined. For example, for the period of 1980 to 1990, 33 economic studies of O_3 and other air pollutant effects on U.S. crops were published in peer-reviewed journal outlets (Spash, 1997). However, in preparing this section of the current O_3 AQCD, only four peer-reviewed economic assessments were found for the decade of 1991 to 2000 that addressed vegetation in the United States. In addition, one peer-reviewed article (Kuik et al., 2000) was found dealing with agriculture in the Netherlands. Recent interest in global climate change and the potential effects of global warming on O_3 and other photochemical oxidants, has renewed interest in the effects of air pollution on both managed and unmanaged terrestrial ecosystems (Adams et al., 1998). In addition, concern is growing for regarding the effects of air pollutants on natural ecosystems and on the services they provide (Daily, 1997). Unfortunately, this interest has not yet translated into additional peer-reviewed publications addressing O_3 or other air pollutants effects on ecosystems.

A study by Murphy et al. (1999) of the economic effects of tropospheric O_3 on U.S. agriculture is of note here, because it confirms the general magnitude of economic effects reported by the two key studies performed a decade earlier (Adams, 1986; 1985). Specifically, Murphy et al. (1999) evaluated benefits to eight major crops associated with several scenarios concerning the reduction or elimination of O_3 precursor emissions from motor vehicles in the United States. Their analysis reported a $2.8 to 5.8 billion (1990 dollars) benefit from complete elimination of O_3 exposures from all sources, i.e., ambient O_3 reduced to a background level assumed to be 0.025 to 0.027 ppm. While the analytical framework is similar to Adams et al. (1986) in the use of NCLAN-based yield response functions and a mathematical programming-based economic optimization model, the study is novel in its focus on the role of motor vehicle emissions of VOCs/NO_x in total anthropogenic O_3 levels. The study is also

notable in its careful attention to federal farm program effects, particularly the deficiency payment component.

There have been a number of recent studies of air pollutant effects on tree species in the literature. Some have reported changes in total biomass and focused on European species (Kurczynska et al., 1997). Other studies have assessed changes in composition of forest species (biodiversity) or forest health due to exposure to air pollutants (Bringmark and Bringmark, 1995; McLaughlin and Percy, 1999; Vacek et al., 1999). As noted previously, changes in forest biomass and composition are more difficult to value than marketable products. However, measures of forest composition or health have implications for an area of increasing policy concern, that being the effect of air pollutants and other environmental stressors on unmanaged (natural) ecosystems and the services they provide (Goulder and Kennedy, 1997; Pimentel et al., 1997). Considerable discussion has occurred among ecologists and economists as to the appropriate means for valuing these services (Anderson, 1990; Carpenter and Dixon, 1985; Common and Perrings, 1992). A number of conceptual articles have been published on this issue in both economic and ecological journals (Bergstrom, 1990; Castle, 1993; Pearce, 1993; Suter, II, 1990).

Effects on forests and natural ecosystems remain problematic, due to limitations in biological response data and economic methods. The problem is even more acute for valuing natural ecosystem service flows. The current limitations surrounding forests and natural ecosystems present a rich research agenda. Areas of greatest potential value in terms of regional policymaking need to be prioritized. Such priority setting can be assisted by sensitivity analyses with existing economic models. By measuring the changes in economic effects arising from changes in key parameters, it is possible to identify those research data gaps most likely to affect economic values.

REFERENCES

Adams, R. M. (1986) Agriculture, forestry, and related benefits of air pollution control: a review and some observations. Am. J. Agric. Econ. 68: 464-472.

Adams, R. M.; Hamilton, S. A.; McCarl, B. A. (1985) An assessment of the economic effects of ozone on U.S. agriculture. J. Air Pollut. Control Assoc. 35: 938-943.

Adams, R. M.; Hamilton, S. A.; McCarl, B. A. (1986) The benefits of pollution control: the case of ozone and U.S. agriculture. Am. J. Agric. Econ. 68: 886-893.

Adams, R. M.; Hurd, B. H.; Lenhart, S.; Leary, N. (1998) Effects of global climate change on agriculture: an interpretative review. Clim. Res. 11: 19-30.

Anderson, E. (1990) The ethical limitations of the market. Econ. Philos. 6: 179-205.

Andersen, C. P. (2003) Source-sink balance and carbon allocation below ground in plants exposed to ozone. New Phytol. 157: 213-228.

Andersen, C. P.; Wilson, R.; Plocher, M.; Hogsett, W. E. (1997) Carry-over effects of ozone on root growth and carbohydrate concentrations of ponderosa pine seedlings. Tree Physiol. 17: 805-811.

Andersen, C. P.; Hogsett, W. E.; Plocher, M.; Rodecap, K.; Lee, E. H. (2001) Blue wild-rye grass competition increases the effect of ozone on ponderosa pine seedlings. Tree Physiol. 21: 319-327.

Arbaugh, M.; Bytnerowicz, A.; Grulke, N.; Fenn, M.; Poth, M.; Temple, P.; Miller, P. (2003) Photochemical smog effects in mixed conifer forests along a natural gradient of ozone and nitrogen deposition in the San Bernardino Mountains. Environ. Int. 29: 401-406.

Assmann, S. M. (2003) Open stomata 1 opens the door to ABA signaling in *Arabidopsis* guard cells. Trends Plant Sci. 8: 151-153.

Assmann, S. M.; Wang, X. Q. (2001) From milliseconds to millions of years: guard cells and environmental responses. Curr. Opin. Plant Biol. 4: 421-428.

Ball, G. R.; Benton, J.; Palmer-Brown, D.; Fuhrer, J.; Skärby, L.; Gimeno, B. S.; Mills, G. (1998) Identifying factors which modify the effects of ambient ozone on white clover (*Trifolium repens*) in Europe. Environ. Pollut. 103: 7-16.

Ball, G. R.; Palmer-Brown, D.; Fuhrer, J.; Skärby, L.; Gimeno, B. S.; Mills, G. (2000) Identification of non-linear influences on the seasonal ozone dose-response of sensitive and resistant clover clones using artificial neural networks. Ecol. Model. 129: 153-168.

Balls, G. R.; Palmer-Brown, D.; Sanders, G. E. (1996) Investigating microclimatic influences on ozone injury in clover (*Trifolium subterraneum*) using artificial neural networks. New Phytol. 132: 271-280.

Barnes, J. D.; Wellburn, A. R. (1998) Air pollutant combinations. In: De Kok, L. J.; Stulen, I., eds. Responses of plant metabolism to air pollution and global change. Leiden, The Netherlands: Backhuys Publishers; pp. 147-164.

Batts, G. R.; Morison, J. I. L.; Ellis, R. H.; Hadley, P.; Wheeler, T. R. (1997) Effects of CO_2 and temperature on growth and yield of crops of winter wheat over four seasons. Eur. J. Agron. 7: 43-52.

Bergstrom, J. C. (1990) Concepts and measures of the economic value of environmental quality: a review. J. Environ. Manage. 31: 215-228.

Black, V. J.; Black, C. R.; Roberts, J. A.; Stewart, C. A. (2000) Impact of ozone on the reproductive development of plants. New Phytol. 147: 421-447.

Bonello, P.; Heller, W.; Sandermann, H., Jr. (1993) Ozone effects on root-disease susceptibility and defence responses in mycorrhizal and non-mycorrhizal seedlings of Scots pine (*Pinus sylvestris* L). New Phytol. 124: 653-663.

Braun, S.; Rihm, B.; Schindler, C.; Flückiger, W. (1999) Growth of mature beech in relation to ozone and nitrogen deposition: an epidemiological approach. Water Air Soil Pollut. 116: 357-364.

Bringmark, E.; Bringmark, L. (1995) Disappearance of spatial variability and structure in forest floors - a distinct effect of air pollution? Water Air Soil Pollut. 85: 761-766.

Broadmeadow, M. S. J.; Jackson, S. B. (2000) Growth responses of *Quercus petraea*, *Fraxinus excelsior* and *Pinus sylvestris* to elevated carbon dioxide, ozone and water supply. New Phytol. 146: 437-451.

Campbell, S.; Temple, P.; Pronos, J.; Rochefort, R.; Andersen, C. (2000) Monitoring for ozone injury in west coast (Oregon, Washington, California) forests in 1998. Portland, OR: U.S. Department of Agriculture, Forest Service, Pacific Northwest Research Station; general technical report no. PNW-GTR-495. Available: http://www.fs.fed.us/pnw/gtrs.htm [11 April, 2003].

Cardoso-Vilhena, J.; Barnes, J. (2001) Does nitrogen supply affect the response of wheat (*Triticum aestivum* cv Hanno) to the combination of elevated CO_2 and O_3? J. Exp. Bot. 52: 1901-1911.

Carpenter, R. A.; Dixon, J. A. (1985) Ecology meets economics: a guide to sustainable development. Environment 27: 6-32.
Castle, E. N. (1993) A pluralistic, pragmatic and evolutionary approach to natural resource management. For. Ecol. Manage. 56: 279-295.
Chappelka, A. H.; Samuelson, L. J. (1998) Ambient ozone effects on forest trees of the eastern United States: a review. New Phytol. 139: 91-108.
Chappelka, A.; Renfro, J.; Somers, G.; Nash, B. (1997) Evaluation of ozone injury on foliage of black cherry (*Prunus serotina*) and tall milkweed (*Asclepias exaltata*) in Great Smoky Mountains National Park. Environ. Pollut. 95: 13-18.
Colls, J. J.; Unsworth, M. H. (1992) Air pollution interactions with natural stressors. In: Barker, J. R.; Tingey, D. T., eds. Air pollution effects on biodiversity. New York, NY: Van Nostrand Reinhold; pp. 93-108.
Common, M.; Perrings, C. (1992) Towards an ecological economics of sustainability. Ecol. Econ. 6: 7-34.
Constable, J. V. H.; Taylor, G. E., Jr.; Laurence, J. A.; Weber, J. A. (1996) Climatic change effects on the physiology and growth of *Pinus ponderosa*: expectations from simulation modeling. Can. J. For. Res. 26: 1315-1325.
Coulston, J. W.; Smith, G. C.; Smith, W. D. (2003) Regional assessment of ozone sensitive tree species using bioindicator plants. Environ. Monit. Assess. 83: 113-127.
Daily, G. C. (1997) Introduction: what are ecosystem services? In: Daily, G. C., ed. Nature's services: societal dependence on natural ecosystems. Washington, DC: Island Press; pp. 1-10.
Danielsson, H.; Karlsson, G. P.; Karlsson, P. E.; Pleijel, J. (2003) Ozone uptake modelling and flux-response relationships—an assessment of ozone-induced yield loss in spring wheat. Atmos. Environ. 37: 475-485.
Davison, A. W.; Barnes, J. D. (1998) Effects of ozone on wild plants. New Phytol. 139: 135-151.
Docherty, M.; Salt, D. T.; Holopainen, J. K. (1997) The impacts of climate change and pollution on forest pests. In: Watt, A. D.; Stork, N. E.; Hunter, M. D., eds. Forests and insects. Chapman and Hall; pp. 229-247.
Dowding, P. (1988) Air pollutant effects on plant pathogens. In: Schulte-Hostede, S.; Darrall, N. M.; Blank, L. W.; Wellburn, A. R., eds. Air pollution and plant metabolism. London, United Kingdom: Elsevier Applied Science Publishers; pp. 329-355.
Emberson, L.; Ashmore, M. R.; Cambridge, H. M.; Simpson, D.; Tuovinen, J. P. (2000) Modelling stomatal ozone flux across Europe. Environ. Pollut. 109: 403-413.
Fangmeier, A.; Bender, J.; Weigel, H. J.; Jäger, H. J. (2002) Effects of pollutant mixtures. In: Bell, J. N. B.; Treshow, M., eds. Air Pollution and Plant Life. 2nd ed. Chichester, United Kingdom: John Wiley & Sons Ltd.; pp. 251-272.
Flagler, R. B. (1998) Recognition of air pollution injury to vegetation: a pictorial atlas. 2nd ed. Pittsburgh, PA: Air & Waste Management Association.
Flückiger, W.; Braun, S.; Hiltbrunner, E. (2002) Effects of air pollutants on biotic stress. In: Bell, J. N. B.; Treshow, M., eds. Air pollution and plant life. 2nd ed. Chichester, United Kingdom: John Wiley & Sons Ltd.; pp. 379-406.
Fredericksen, T. S.; Skelly, J. M.; Snyder, K. R.; Steiner, K. C. (1996) Predicting ozone uptake from meteorological and environmental variables. J. Air Waste Manage. Assoc. 46: 464-469.
Fuentes, J. D.; Hayden, B. P.; Garstang, M.; Lerdau, M.; Fitzjarrald, D.; Baldocchi, D. D.; Monson, R.; Lamb, B.; Geron, C. (2001) New directions: VOCs and biosphere-atmosphere feedbacks. Atmos. Environ. 35: 189-191.
Fuhrer, J. H. (1994) Effects of ozone on managed pasture. 1. Effects of open-top chambers on microclimate, ozone flux and plant growth. Environ. Pollut. 86: 297-305.
Fuhrer, J.; Skarby, L.; Ashmore, M. R. (1997) Critical levels for ozone effects on vegetation in Europe. Environ. Pollution 97: 91-106.
Goulder, L. H.; Kennedy, D. (1997) Valuing ecosystem services: philosophical bases and empirical methods. In: Daily, G. C. Nature's Services: Societal Dependence on Natural Ecosystems. Washington, DC: Island Press; pp. 23-47.
Grulke, N. E.; Preisler, H. K.; Rose, C.; Kirsch, J.; Balduman, L. (2002) O_3 uptake and drought stress effects on carbon acquisition of ponderosa pine in natural stands. New Phytol. 154: 621-631.
Grulke, N. E.; Alonso, R.; Nguyen, T.; Cascio, C.; Dobrowolski, W. (2004) Stomata open at night in pole-sized and mature ponderosa pine: implications for O_3 exposure metrics. Tree Physiol. 24: 1001-1010.
Grünhage, L.; Jäger, H. J. (2003) From critical levels to critical loads for ozone: a discussion of a new experimental and modelling approach for establishing flux-response relationships for agricultural crops and native plant species. Environ. Pollut. 125: 99-110.

Heck, W. W.; Furiness, C. S.; Cowling, E. B.; Sims, C. K. (1998) Effects of ozone on crop, forest, and natural ecosystems: assessment of research needs. EM (October): 11-22.

Hogsett, W. E.; Tingey, D. T.; Hendricks, C.; Rossi, D. (1989) Sensitivity of western conifers to SO_2 and seasonal interaction of acid fog and ozone. In: Olson, R. K.; Lefohn, A. S., eds. Effects of air pollution on western forests [an A&WMA symposium; June; Anaheim, CA]. Air Pollution Control Association; pp. 469-491 (APCA transactions series: no. 16).; pp. Anaheim,CA-Anaheim491.

Innes, J. L.; Skelly, J. M.; Schaub, M. (2001) Ozone and broadleaved species A guide to the identification of ozone-induced foliar injury. Bern, Switzerland: Paul Haupt Publishers.

Isebrands, J. G.; Dickson, R. E.; Rebbeck, J.; Karnosky, D. F. (2000) Interacting effects of multiple stresses on growth and physiological processes in northern forest trees. In: Mickler, R. A.; Birsdey, R. A.; Hom, J., eds. Responses of northern U.S. forests to environmental change. New York, NY: Springer-Verlag; pp. 149-180. (Ecological studies: v. 139).

Isebrands, J. G.; McDonald, E. P.; Kruger, E.; Hendrey, G.; Percy, K.; Pregitzer, K.; Sober, J.; Karnosky, D. F. (2001) Growth responses of *Populus tremuloides* clones to interacting carbon dioxide and tropospheric ozone. Environ. Pollut. 115: 359-371.

Johnson, B. G.; Hale, B. A.; Ormrod, D. P. (1996) Carbon dioxide and ozone effects on growth of a legume-grass mixture. J. Environ. Qual. 25: 908-916.

Kärenlampi, L.; Skärby, L. (1996) Critical levels for ozone in Europe: testing and finalizing the concepts UN-ECE workshop report. In: Proceedings of UN-ECE convention on long-range transboundary air pollution workshop; April; Kuopio, Finland; Kupio, Finland: University of Kuopio, Department of Ecology and Environmental Science.

Karnosky, D. F.; Mankovska, B.; Percy, K.; Dickson, R. E.; Podila, G. K.; Sober, J.; Noormets, A.; Hendrey, G.; Coleman, M. D.; Kubiske, M.; Pregitzer, K. S.; Isebrands, J. G. (1999) Effects of tropospheric ozone on trembling aspen and interaction with CO_2: results from an O_3-gradient and a FACE experiment. Water Air Soil Pollut. 116: 311-322.

Kolb, T. E.; Matyssek, R. (2001) Limitations and perspectives about scaling ozone impacts in trees. Environ. Pollut. 115: 373-393.

Kopper, B. J.; Lindroth, R. L. (2003) Effects of elevated carbon dioxide and ozone on the phytochemistry of aspen and performance of an herbivore. Oecologia 134: 95-103.

Kouterick, K. B.; Skelly, J. M.; Fredericksen, T. S.; Steiner, K. C.; KoIb, T. E.; Ferdinand, J. A. (2000) Foliar injury, leaf gas exchange and biomass responses of black cherry (*Prunus serotina* Ehrh) half-sibling families to ozone exposure. Environ. Pollut. 107: 117-126.

Kraft, M.; Weigel, H. J.; Mejer, G. J.; Brandes, F. (1996) Reflectance measurements of leaves for detecting visible and non-visible ozone damage to crops. J. Plant Physiol. 148: 148-154.

Krupa, S. V.; Tonneijck, A. E. G.; Manning, W. J. (1998) Ozone. In: Flagler, R. B., ed. Recognition of air pollution injury to vegetation - a pictorial atlas. Air & Waste Management Association; pp. 2-11.

Krupa, S. V.; Nosal, M.; Peterson, D. L. (2001) Use of passive ozone O_3 samplers in vegetation effects assessment. Environ. Pollut. 112: 303-309.

Krupa, S. V.; Nosal, M.; Ferdinand, J. A.; Stevenson, R. E.; Skelly, J. M. (2003) A multi-variate statistical model integrating passive sampler and meteorology data to predict the frequency distributions of hourly ambient ozone (O_3) concentrations. Environ. Pollut. 124: 173-178.

Kuik, O. J.; Helming, J. F. M.; Dorland, C.; Spaninks, F. A. (2000) The economic benefits to agriculture of a reduction of low-level ozone pollution in The Netherlands. Eur. Rev. Agric Econ. 27: 75-90.

Kurczyńska, E. U.; Dmuchowski, W.; Wloch, W.; Bytnerowicz. A. (1997) The influence of air pollutants on needles and stems of scots pine (*Pinus Sylvestris* L.) trees. Environ. Pollut. 98: 325-334.

Kytöviita, M. M.; Le Thiec, D.; Dizengremel, P. (2001) Elevated CO_2 and ozone reduce nitrogen acquisition by *Pinus halepensis* from its mycorrhizal symbiont. Physiol. Plant. 111: 305-312.

Landolt, W.; Günthardt-Goerg, M. S.; Pfenninger, I.; Einig, W.; Hampp, R.; Maurer, S.; Matyssek, R. (1997) Effect of fertilization on ozone-induced changes in the metabolism of birch (*Betula pendula*) leaves. New Phytol. 137: 389-397.

Langebartels, C.; Ernst, D.; Heller, W.; Lutz, C.; Payer, H. D.; Sandermann, H., Jr. (1997) Ozone responses of trees: results from controlled chamber exposures at the GSF phytotron. In: Sandermann, H.; Wellburn, A. R.; Heath, R. L., eds. Forest decline and ozone. New York, NY: Springer-Verlag; pp. 163-200 (Ecological studies: v. 127).

Laurence, J. A.; Andersen, C. P. (2003) Ozone and natural systems: understanding exposure, response, and risk. Environ. Int. 29: 155-160.

Laurence, J. A.; Amundson, R. G.; Kohut, R. J.; Weinstein, D. A. (1997) Growth and water use of red spruce (*Picea rubens* Sarg) exposed to ozone and simulated acidic precipitation for four growing seasons. For. Sci. (Bethesda, Md.) 43: 355-361.

Laurence, J. A.; Retzlaff, W. A.; Kern, J. S.; Lee, E. H.; Hogsett, W. E.; Weinstein, D. A. (2001) Predicting the regional impact of ozone and precipitation on the growth of loblolly pine and yellow poplar using linked TREGRO and ZELG models. For. Ecol. Manage. 146: 247-263.

Laurence, J. A.; Retzlaff, W. A.; Kern, J. S.; Lee, E. H.; Hogsett, W. E.; Weinstein, D. A. (2003) Corrigendum to "Predicting the regional impact of ozone and precipitation on the growth of loblolly pine and yellow-poplar using linked TREGRO and ZELIG models" [Forest Ecol Manage 146 (2001) 247-263]. For. Ecol. Manage. 174: 607.

Lee, E. H.; Tingey, D. T.; Hogsett, W. E. (1988) Evaluation of ozone exposure indices in exposure-response modeling. Environ. Pollut. 53: 43-62.

Lee, E. H.; Tingey, D. T.; Hogsett, W. E.; Laurence, J. A. (2003) History of tropospheric ozone for the San Bernardino Mountains of southern California, 1963-1999. Atmos. Environ. 37: 2705-2717.

Lefohn, A. S.; Shadwick, D. S. (2000) Differences in trending estimates in the United States using several ozone metrics. In: Proceedings of the 93rd annual meeting of the Air & Waste Management Association; June; Salt Lake City, UT; Pittsburgh, PA: Air & Waste Management Association; paper no. AS 1d-645.

Maier-Maercker, U. (1998) Predisposition of trees to drought stress by ozone. Tree Physiol. 19: 71-78.

Manning, W. J.; Krupa, S. V. (1992) Experimental methodology for studying the effects of ozone on crops and trees. In: Lefohn, A. S., ed. Surface level ozone exposures and their effects on vegetation. Chelsea, MI: Lewis Publishers, Inc.; pp. 93-156.

Massman, W. J. (2004) Toward an ozone standard to protect vegetation based on effective dose: a review of deposition resistance and a possible metric. Atmos. Environ. 38: 2323-2337.

Matyssek, R.; Wieser, G.; Nunn, A. J.; Kozovits, A. R.; Reiter, I. M.; Heerdt, C.; Winkler, J. B.; Baumgarten, M.; Häberle, K. H.; Grams, T. E. E.; Werner, H.; Fabian, P.; Havranek, W. M. (2004) Comparison between AOT40 and ozone uptake in forest trees of different species, age and site conditions. Atmos. Environ. 38: 2271-2281.

McLaughlin, S.; Percy, K. (1999) Forest health in North America: some perspectives on actual and potential roles of climate and air pollution. Water Air Soil Pollut. 116: 151-197.

Miller, P. R.; Rechel, J. (1999) Temporal changes in crown condition indices, needle litterfall, and collateral needle injuries of Ponderosa and Jeffrey pines. In: Miller, P. R.; McBride, J. R., eds. Oxidant air pollution impacts in the Montane forests of southern California: a case study of the San Bernardino Mountains. New York, NY: Springer; pp. 164-178.

Mills, G. (2002) Modification of plant response by environmental conditions. In: Bell, J. N. B.; Treshow, M., eds. Air pollution and plant life. 2nd ed. Chichester, United Kingdom: John Wiley & Sons Ltd.; pp. 343-358.

Mills, G.; Ball, G.; Hayes, F.; Fuhrer, J.; Skarby, L.; Gimeno, B.; De Temmerman, L.; Heagle, A.; Members of the ICP Vegetation programme. (2000) Development of a multi-factor model for predicting the effects of ambient ozone on the biomass of white clover. Environ. Pollut. 109: 533-542.

Momen, B.; Anderson, P. D.; Helms, J. A.; Houpis, J. L. J. (1997) Acid rain and ozone effects on gas exchange of *Pinus ponderosa*: a comparison between trees and seedlings. Int. J. Plant Sci. 158: 617-621.

Momen, B.; Anderson, P. D.; Helms, J. A. (1999) Temperature dependency of acid-rain effect on photosynthesis of *Pinus ponderosa*. For. Ecol. Manage. 113: 223-230.

Murphy, J. J.; Deluki, M. A.; McCubbin, D. R.; Kim, H. J. (1999) The cost of crop damage caused by ozone air pollution from motor vehicles. J. Environ. Manage. 55: 273-289.

Musselman, R. C.; Massman, W. J. (1999) Ozone flux to vegetation and its relationship to plant response and ambient air quality standards. Atmos. Environ. 33: 65-73.

Musselman, R. C.; Minnick, T. J. (2000) Nocturnal stomatal conductance and ambient air quality standards for ozone. Atmos. Environ. 34: 719-733.

Nussbaum, S.; Fuhrer, J. (2000) Difference in ozone uptake in grassland species between open-top chambers and ambient air. Environ. Pollut. 109: 463-471.

Nussbaum, S.; Geissmann, M.; Fuhrer, J. (1995) Ozone exposure-response relationships for mixtures of perennial ryegrass and white clover depend on ozone exposure patterns. Atmos. Environ. 29: 989-995.
Nussbaum, S.; Bungener, P.; Geissmann, M.; Fuhrer, J. (2000) Plant-plant interactions and soil moisture might be important in determining ozone impacts on grasslands. New Phytol. 147: 327-335.
Oksanen, E.; Holopainen, T. (2001) Responses of two birch (*Betula pendula* Roth) clones to different ozone profiles with similar AOT40 exposure. Atmos. Environ. 35: 5245-5254.
Ollerenshaw, J. H.; Lyons, T. (1999) Impacts of ozone on the growth and yield of field-grown winter wheat. Environ. Pollut. 106: 67-72.
Ollerenshaw, J. H.; Lyons, T.; Barnes, J. D. (1999) Impacts of ozone on the growth and yield of field-grown winter oilseed rape. Environ. Pollut. 104: 53-59.
Pääkkönen, E.; Holopainen, T.; Kärenlampi, L. (1996) Relationships between open-field ozone exposures and growth and senescence of birch (*Betula pendula* and *Betula pubescens*). In: Kärenlampi, L.; Skärby, L. eds. Critical levels for ozone in Europe: testing and finalizing the concepts: UN-ECE workshop report. UN-ECE Convention on Long-Range Transboundary Air Pollution; April; Kuopio, Finland; Kuopio, Finland: University of Kuopio, Department of Ecology and Environmental Science; pp. 298-302.
Panek, J. A. (2004) Ozone uptake, water loss and carbon exchange dynamics in annually drought-stressed *Pinus ponderosa* forests: measured trends and parameters for uptake modeling. Tree Physiol. 24: 277-290.
Panek, J.; Kurpius, M. R.; Goldstein, A. H. (2002) An evaluation of ozone exposure metrics for a seasonally drought-stressed ponderosa pine ecosystem. Environ. Pollut. 117: 93-100.
Panek, J. A.; Baldocchi, D. D.; Goldstein, A. H. (2003) The need for spatially and functionally integrated models of ozone deposition to Sierra Nevada forests. In: Bytnerowicz, A.; Arbaugh, M. J.; Alonso, R., eds. Ozone air pollution in the Sierra Nevada: distribution and effects on forests. New York, NY: Elsevier Science Ltd; pp. 325-357. (Developments in environmental science: v. 2).
Pearce, D. W. (1993) Economic values and the natural world. London, United Kingdom: Earthscan Publications, Ltd.
Pell, E. J.; Schlagnhaufer, C. D.; Arteca, R. N. (1997) Ozone-induced oxidative stress: mechanisms of action and reaction. Physiol. Plant. 100: 264-273.
Percy, K. E.; Awmack, C. S.; Lindroth, R. L.; Kubiske, M. E.; Kopper, B. J.; Isebrands, J. G.; Pregitzer, K. S.; Hendry, G. R.; Dickson, R. E.; Zak, D. R.; Oksanen, E.; Sober, J.; Harrington, R.; Karnosky, D. F. (2002) Altered performance of forest pests under atmospheres enriched with CO_2 and O_3. Nature (London) 420: 403-407.
Pimentel, D.; Wilson, C.; McCullum, C.; Huang, R.; Dwen, P.; Flack, J.; Tran, Q.; Saltman, T.; Cliff, B. (1997) Economic and environmental benefits of biodiversity. BioScience 47: 747-757.
Pitelka, L. F. (1988) Evolutionary responses of plants to anthropogenic pollutants. Trends Ecol. Evol. 3: 233-236.
Pleijel, H.; Danielsson, H.; Karlsson, G. P.; Gelang, J.; Karlsson, P. E.; Selldén, G. (2000) An ozone flux-response relationship for wheat. Environ. Pollut. 109: 453-462.
Rao, M. V.; Davis, K. R. (2001) The physiology of ozone induced cell death. Planta 213: 682-690.
Rebbeck, J. (1996) Chronic ozone effects on three northeastern hardwood species: growth and biomass. Can. J. For. Res. 26: 1788-1798.
Rowland-Bamford, A. J. (2000) Plant responses to changing carbon dioxide and temperature. In: Singh, S. N., ed. Trace gas emissions and plants. Dordrecht, The Netherlands: Kluwer Academic Publishers; pp. 63-74.
Samuelson, L.; Kelly, J. M. (2001) Scaling ozone effects from seedlings to forest trees. Tansley review no 21. New Phytol. 149: 21-41.
Sandermann, H., Jr. (1998) Ozone: an air pollutant acting as a plant-signaling molecule. Naturwissenschaften 85: 369-375.
Sandermann, H., Jr. (2000) Ozone/biotic disease interactions: molecular biomarkers as a new experimental tool. Environ. Pollut. 108: 327-332.
Sayre, R. G.; Fahey, T. J. (1999) Effects of rainfall acidity and ozone on foliar leaching in red spruce *Picea rubens*. Can. J. For. Res. 29: 486-496.
Schmieden, U.; Wild, A. (1995) The contribution of ozone to forest decline. Physiol. Plant. 94: 371-378.
Schnitzler, J. P.; Langebartels, C.; Heller, W.; Liu, J.; Lippert, M.; Dohring, T.; Bahnweg, G.; Sandermann, H. (1999) Ameliorating effect of UV-B radiation on the response of Norway spruce and Scots pine to ambient ozone concentrations. Global Change Biol. 5: 83-94.
Schraudner, M.; Langebartels, C.; Sandermann, H., Jr. (1996) Plant defence systems and ozone. Biochem. Soc. Trans. 24: 456-461.

Smith, G.; Coulston, J.; Jepsen, E.; Prichard, T. (2003) A national ozone biomonitoring program - results from field surveys of ozone sensitive plants in northeastern forests (1994-2000). Environ. Monit. Assess. 87: 271-291.

Somers, G. L.; Chappelka, A. H.; Rosseau, P.; Renfro, J. R. (1998) Empirical evidence of growth decline related to visible ozone injury. For. Ecol. Manage. 104: 129-137.

Spash, C. L. (1997) Assessing the economic benefits to agriculture from air pollution control. J. Econ. Surv. 11: 47-70.

Suter, G. W., II. (1990) Endpoints for regional ecological risk assessments. Environ. Manage. (N. Y.) 14: 9-23.

Tingey, D. T.; Hogsett, W. E.; Lee, E. H.; Herstrom, A. A.; Azevedo, S. H. (1991) An evaluation of various alternative ambient ozone standards based on crop yield loss data. In: Berglund, R. L.; Lawson, D. R.; McKee, D. J. eds. Tropospheric ozone and the environment: papers from an international conference; March 1990; Los Angeles, CA; Pittsburgh, PA: Air & Waste Management Association (A&WMA transactions series no. TR-19); pp. 272-288.

Tingey, D. T.; Hogsett, W. E.; Lee, E. H.; Laurence, J. A. (2004) Stricter ozone ambient air quality standard has beneficial effect on ponderosa pine in California. Environ. Manage. 34: 397-405.

Topa, M. A.; Vanderklein, D. W.; Corbin, A. (2001) Effects of elevated ozone and low light on diurnal and seasonal carbon gain in sugar maple. Plant Cell Environ. 24: 663-677.

U.S. Environmental Protection Agency (1978) Air quality criteria for ozone and other photochemical oxidants. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report no. EPA-600/8-78-004. Available from: NTIS, Springfield, VA; PB80-124753.

U.S. Environmental Protection Agency (1986) Air quality criteria for ozone and other photochemical oxidants. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report nos. EPA-600/8-84-020aF-eF. 5v. Available from: NTIS, Springfield, VA; PB87-142949.

U.S. Environmental Protection Agency (1992) Summary of selected new information on effects of ozone on health and vegetation: supplement to 1986 air quality criteria for ozone and other photochemical oxidants. Research Triangle Park, NC: Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office; report no. EPA/600/8-88/105F. Available from: NTIS, Springfield, VA; PB92-235670.

U.S. Environmental Protection Agency (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: http://cfpub2.epa.gov/ncea/.

Vacek, S.; Bastl, M.; Leps, J. (1999) Vegetation changes in forests of the Krkonose Mountains over a period of air pollution stress (1980-1995). Plant Ecol. 143: 1-11.

Van Oijen, M.; Ewart, F. (1999) The effects of climatic variation in Europe on the yield response of spring wheat cv Minaret to elevated CO_2 and O_3: an analysis of open-top chamber experiments by means of two crop growth simulation models. Eur. J. Agron. 10: 249-264.

Ward, E. R.; Uknes, S. J.; Williams, S. C.; Dincher, S. S.; Wiederhold, D. L.; Alexander, D. C.; Ahi-Goy, P.; Métraux, J. P.; Ryals, J. A. (1991) Coordinate gene activity in response to agents that induce systemic acquired resistance. Plant Cell 3: 1084-1094.

Weinstein, D. A.; Gollands, B.; Retzlaff, W. A. (2001) The effects of ozone on a lower slope forest of the Great Smoky Mountain National Park: simulations linking an individual tree model to a stand model. For. Sci. 47: 29-42.

Whitfield, C. P.; Davison, A. W.; Ashenden, T. W. (1998) The effects of nutrient limitation on the response of *Plantago major* to ozone. New Phytol. 140: 219-230.

Winner, W. E.; Coleman, J. S.; Gillespie, C.; Mooney, H. A.; Pell, E. J. (1991) Consequences of evolving resistance to air pollutants. In: Taylor, G. E.; Pitelka, L. F.; Clegg, M. T., eds. Ecological genetics and air pollution. Springer-Verlag; pp. 177-202.

Wu, Y. X.; Tiedemann, A. V. (2002) Impact of fungicides on active oxygen species and antioxidant enzymes in spring barley (*Hordeum vulgare* L) exposed to ozone. Environ. Pollut. 116: 37-47.

Yun, S. C.; Laurence, J. A. (1999) The response of sensitive and tolerant clones of *Populus tremuloides* to dynamic ozone exposure under controlled environmental conditions. New Phytol. 143: 305-313.

10. TROPOSPHERIC OZONE EFFECTS ON UV-B FLUX, AND ITS ROLE IN CLIMATE CHANGE

10.1 INTRODUCTION

In addition to exerting direct effects on human health and vegetation/ecosystems, as discussed in earlier chapters, tropospheric ozone (O_3) influences the ground-level flux of solar ultraviolet (UV) radiation, as well as other processes that alter the Earth's radiative balance and contribute to climate change. This chapter discusses tropospheric O_3 and (1) its role in in determining surface-level UV flux and, (2) its involvement in global climate change.

10.2 THE ROLE OF TROPOSPHERIC OZONE IN DETERMINING GROUND-LEVEL UV-B FLUX

Atmospheric O_3 plays a crucial role in reducing the exposure of living organisms to solar UV radiation. Approximately 90% of the total atmospheric O_3 burden is located in the stratosphere; therefore, photochemical processes that alter the concentration of stratospheric O_3 are of particular concern within the global community. The importance of stratospheric O_3 depletion due to the release of long-lived anthropogenic chlorinated- and fluorinated hydrocarbons was recognized over a period of several years during the 1970s and early 1980s and led to the international treaty for the protection of stratospheric O_3: the 1987 Montreal Protocol on Substances that Deplete the Ozone Layer.

While roughly representing only 10% of the total atmospheric O_3 burden, tropospheric O_3, like other tropospheric pollutants, can influence the flux of biologically-damaging UV radiation at the Earth's surface. This section summarizes the available information on the factors governing UV flux at the Earth's surface, first, followed by a discussion the status of scientific understanding of the factors governing human UV exposure and, then, of the links between UV exposure and human disease.

10.2.1 Factors Governing Ultraviolet Radiation Flux at the Earth's Surface

The Montreal Protocol requires routine review of the latest scientific information available on the status of the O_3 layer and of UV radiation levels at the Earth's surface. The World Meteorology Organization (WMO) and U.N. Environmental Program (UNEP) are responsible for assessing the state of the science regarding the O_3 layer and for reporting on trends in surface UV radiation levels. The latest WMO/UNEP assessment was published in 2002 (WMO/UNEP, 2002).

An outcome of the on-going atmospheric chemistry research effort devoted to tracking stratospheric O_3 depletion and its effects is a body of literature, though limited, that describes the effects of tropospheric pollutants, particulate matter (PM), and O_3, on ground-level UV radiation flux. Drawing from the WMO/UNEP assessment and more recent literature, this section describes the current level of scientific understanding of the factors influencing ground-level UV radiation flux such as geophysical factors, tropospheric O_3, PM, and cloud cover. Figure 10-1 visually summarizes some of the factors that influence the flux of UV-B at the Earth's surface.

10.2.1.1 UV Radiation:: Wavelengths, Energies and Depth of Atmospheric Penetration

Designations for portions of the electromagnetic spectrum have evolved over time and are usually associated with general functions or effects caused by photons within a given wavelength range. The energy possessed by a photon is inversely proportional to its wavelength. For example, gamma rays, having wavelengths ≤ 0.1 nm, are especially damaging high-energy photons emitted during radioactive decay and by stellar activity. Radiowaves, having wavelengths $\geq 10^8$ nm, are very low in energy and function as carriers for broadcast communications.

The wavelengths ranging between 50 and 400 nm in length are denoted "ultraviolet." Solar radiation of wavelengths <280 nm, including UV-C (200 to 280 nm), is almost entirely blocked by the Earth's upper atmosphere due to photoionization and photodissociation processes. Figure 10-2 compares the solar flux above the atmosphere with ground-level flux. Solar UV-B radiation (280 - 320 nm) is absorbed or scattered in part within the atmosphere, while UV-A radiation (320 - 400 nm) can be scattered but not absorbed to any meaningful degree by atmospheric gases. Both UV-B and UV-A photons contain sufficient energy to break (photolyze) chemical bonds and are associated with human health- and ecosystem-damaging

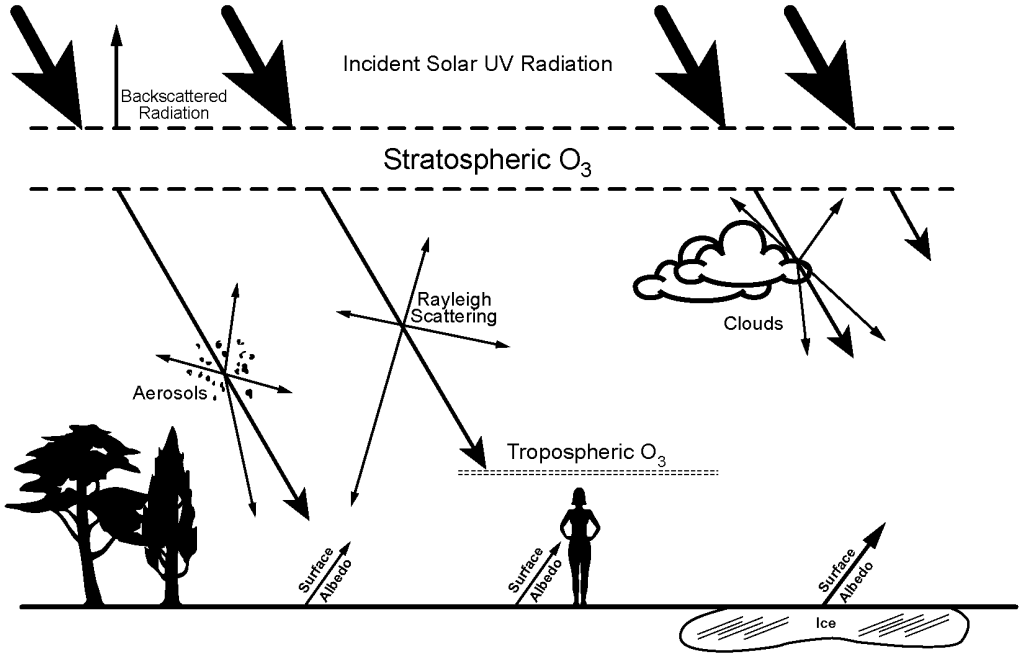


Figure 10-1. Complexity of factors that determine human exposure to UV radiation. In addition to the geophysical/atmospheric factors (e.g., stratospheric and tropospheric O_3, clouds, aerosols, and Rayleigh scattering) that affect the solar flux of UV radiation at surface level, there are human physical, behavioral and demographic factors that influence human exposure to UV radiation.

effects. However, because UV-B is more energetic, it is potentially capable of producing substantially more biological damage than UV-A.

10.2.1.2 Temporal Variations in Solar Flux

The magnitude of the solar radiation flux entering the atmosphere depends upon long-term solar activity, sunspot cycle (11 years), solar rotation (27 days) and the position of the Earth in its orbit around the sun. A variety of changes in solar irradiance can be found in historical data, from 1700 to the present. Solanki and Fligge (2000) concluded that solar irradiation changes on time-scales of days to centuries can be attributed to variations in solar magnetic features. Since the last Maunder minimum in 1700, solar irradiance has increased slightly, at ~3.0% for wavelengths in the UV-C range and at ~1.3% for wavelengths in the UV-B and UV-A ranges.

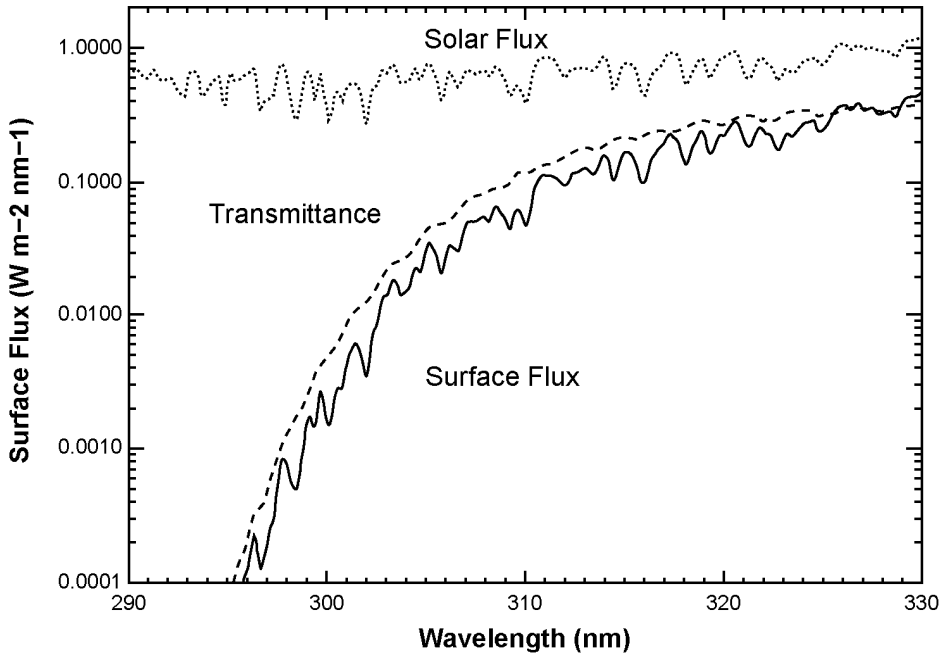


Figure 10-2. Comparison of solar flux above the atmosphere with flux at the Earth's surface. The dotted line represents extraterrestrial solar flux measured by the satellite UARS SOLSTICE instrument (dotted line). The dashed line represents calculated atmospheric transmittance and the solid line is the calculated absolute flux of UV radiation for a solar zenith angle of 50deg, total column O_3 of 275 DU, and a surface reflectivity of 8%. The fine structure on the surface flux trace results from Fraunhofer lines (absorption specific wavelengths within the solar atmosphere).

Source: Krotkov et al. (1998).

Including visible wavelengths, Solanki and Fligge (2000) estimated that the overall increase in solar irradiance was ~0.3%. Rozema et al. (2001) pointed out that any increase in wavelengths <300 nm (UV-C) would initiate additional O_3 formation in the stratosphere. This suggests that any increase in UV-B and/or UV-A solar flux would be offset by a more absorptive stratosphere.

Solar rotation and sunspot activity have the greatest effects on radiation flux originating in the highest levels of the solar atmosphere. The amplitude of the associated cyclical changes in solar shortwave radiation flux follows an inverse relationship between photon wavelength and the solar altitude at which it was emitted. The maximum level of radiation (solar-max) differs

from the minimum (solar-min) by as much as 10% for wavelengths near 160 nm. This peak-to-trough difference declines to around 1% for 300 nm (UV-B range) (Salby, 1996).

The combined effects of the Earth's obliquity (the angle of the Earth's axis of rotation with respect to the plane of its orbit around the sun) and its precession (the rotation of the Earth's axis with respect to a perpendicular line through the plane of its solar orbit) yield variations of up to 30% in total summertime solar flux, depending on latitude (Hartmann, 1994).

Zenith Angle: Latitude, Season, and Time of Day

The sun's relative elevation is measured with respect to the vertical and is known as its "zenith angle." This angle varies hourly, seasonally, and with latitude. Daily and seasonal changes in solar zenith angle result in the largest changes in the magnitude of solar radiation flux, with higher zenith angles corresponding to lower solar flux. The largest natural fluxes occur in the tropical regions, where solar noon occurs at a zenith angle at or near 0°. Seasonal variation in solar flux ranges from small changes at the equator to very large changes at high latitudes. Daily variations in solar flux, from sunrise to sunset, show added wavelength dependence as a function of zenith angle, because transmission of some wavelengths are sensitive to atmospheric pathlength due to scattering and absorption processes. These processes will be discussed further below.

10.2.1.3 Atmospheric Radiative Interactions with Solar Ultraviolet Radiation

Radiative Interactions in the Stratosphere

As noted, earlier, the stratosphere contains 90% or more of the total column density of O_3, the principle gas phase absorber of UV-B. Ozone interacts with UV radiation by scattering the photon, or absorbing and transforming its energy. Upon absorbing a UV photon, O_3 may photodissociate, or become electronically and vibrationally excited.

Photoabsorption by O_3 occurs with very high efficiency. After electronically excited O_3 (O_3*) is formed, it will either lose its excess electronic energy via a collision with another gas molecule (M) or dissociate into ground-state oxygen, O_2, and an electronically excited oxygen radical, $O(^1D)$ (See Reactions 1 and 2). Intermolecular collisions degrade the excess electronic energy of the O_3* molecule by transferring it to other molecules as vibrational, rotational, and/or translational energies, that warm the atmosphere. An $O(^1D)$ radical can react with H_2O to form

two hydroxyl (OH) radicals (Reaction 3). See Chapter 2 for further discussion of odd oxygen and HO_x photochemistry.

$$O_3 + h\nu \rightarrow O_3^* \rightarrow O(^1D) + O_2 \quad (10\text{-}1)$$

$$\rightarrow O_3^* + M \rightarrow O_3 + M^* \quad (10\text{-}2)$$

$$O(^1D) + H_2O \rightarrow 2OH \quad (10\text{-}3)$$

Either of these photochemical processes transforms the energy of the UV photon into heat, a form of energy that, in this context, lacks the potential for human health or ecosystems damage.

The WMO/UNEP (2002) scientific assessment reported that global average total column O_3 had declined by 3% from pre-1980 levels, due to the presence of anthropogenic O_3-depleting substances in the atmosphere. Ozone depletion has a strong latitude and seasonal dependence. The seasonality of total O_3 changes differ between the Northern and Southern Hemispheres. In the northern midlatitudes, total column O_3 declined by ~4% during the winter/spring seasons and by approximately half that amount in the summer/fall of the 1997-2001 time period, relative to pre-1980 total column O_3 levels. In southern midlatitudes, total column O_3 declined ~6% during all seasons.

The concentration of O_3 in a vertical column extending from the Earth's surface is expressed in Dobson Units (DU) corresponding to the column height in hundredths of a millimeter of O_3 at standard temperature and pressure (273 K and 1 atmosphere) (Wayne, 2000). One DU = 2.587×10^{15} molecules of O_3/cm^2. The total O_3 column effectively prevents any UV-C from reaching the surface and reduces the penetration of UV-B to the surface, but it does little to attenuate the intensity of UV-A except at the shorter wavelengths close to the cutoff for UV-B. Cutchis (1974) calculated that with overhead sun, a 10% decrease in the O_3 column would lead to 20, 250, and 500% increases in flux at 305, 290, and 287 nm, respectively, values that have been supported by ground observations in Toronto, ON (49° N; Kerr and McElroy, 1993). Rapid changes of this magnitude appear to happen naturally. As seen in data collected

by the Total Ozone Mapping Satellite (TOMS) (Figure 10-3), the total O_3 column undergoes wide natural variation on short timescales (Cockell, 2001).

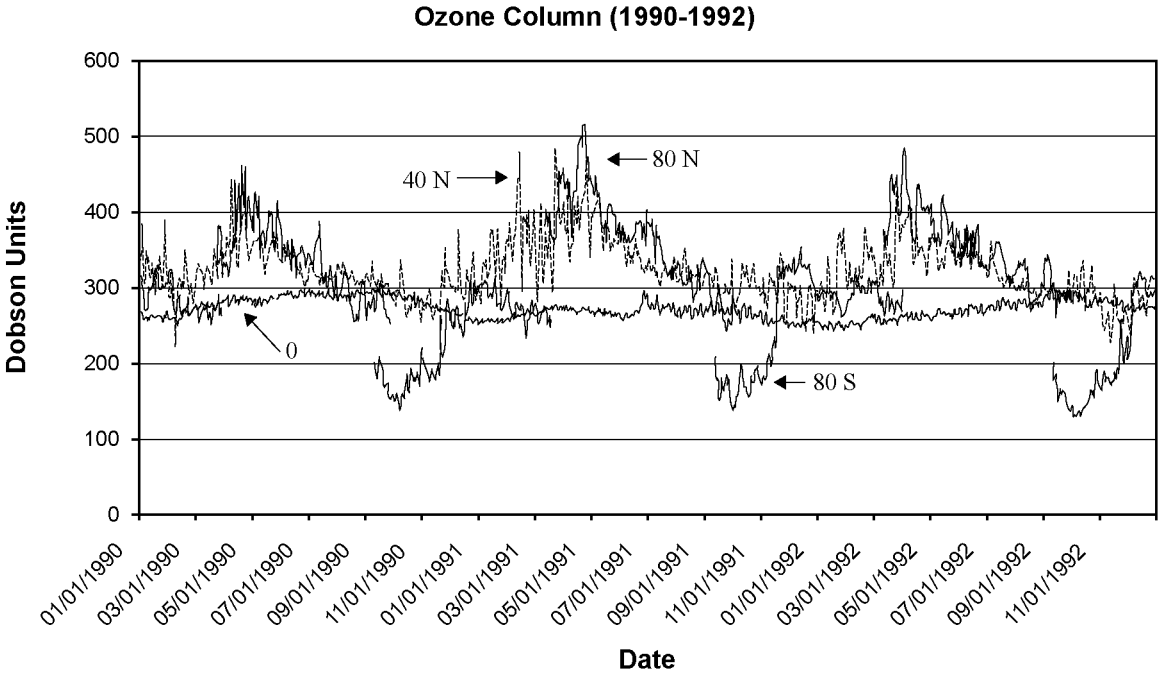


Figure 10-3. Ozone column abundances from the years 1990 to 1992 for 0, 40, and 80° N as well as 80° S. The data for 80° S are incomplete, but the graph shows the effects of the Antarctic O_3 hole on total column abundances at this latitude. The data for the Northern Hemisphere illustrate the natural variations in the O_3 column over time. The data are taken from the TOMS (Total Ozone Monitoring Satellite) data set (1979 to 1993).

Source: Cockell (2001).

Nacreous and polar stratospheric clouds, and aerosols, such as those injected into the stratosphere by explosive volcanic eruptions, both absorb and scatter radiation. Relative to the troposphere, the stratosphere is low in atmospheric pressure. Stratospheric clouds and aerosols are also more dispersed than those in the troposphere. Consequently, UV radiation can traverse the stratosphere with a substantially lower probability of encountering a gas molecule or cloud or an aerosol particle than it would in the troposphere. In the radiative transfer literature, the

stratosphere is described as a "single scattering" regime for UV radiation, and UV that has penetrated the stratosphere is referred to as "direct beam UV." The troposphere, due to its high gas and particle concentrations is referred to as a "multiple scattering" regime.

Radiative Interactions in the Troposphere: Solar Irradiance Versus Actinic Flux

The troposphere contains ≤10% of the total column O_3 but ~78% of the total atmospheric mass (including clouds, and gas- and particle-phase radiation scatterers and absorbers), making it a "multiple scattering" regime for UV radiation. These scattering processes increase the mean-free path a photon must travel before reaching the Earth's surface, transforming the direct beam UV solar irradiance that has penetrated the stratosphere into diffuse, or actinic, UV irradiance (Brühl and Crutzen, 1988).

Rayleigh and Mie scattering of solar radiation are sensitive to molecular and particle size. In Rayleigh scattering, gas molecules that are smaller than the wavelength of the incident photon isotropically deflect incoming photons. Conversely, aerosol and cloud droplets scatter incoming radiation with distinctive forward- and backscattering tendencies, i.e., Mie scattering. Actinic flux, especially at the Earth's surface, is directly proportional to surface albedo (Wendisch and Mayer, 2003). Surface albedo is very strongly wavelength dependent. For example, fresh and wet snow reflect 60 to 90% of incident violet light, while soil and grass surfaces reflect <5% (Xenopoulos and Schindler, 2001). In their in situ measurement and modeling study of the vertical distribution of solar irradiance, Wendisch and Mayer (2003) found that surface albedos must be measured in order to accurately simulate solar flux, due to the large variations in albedo that may occur within a given surface type. Snow cover, even many kilometers from measurement sites is known to increase detected UV irradiances. Complicated interactions result when radiation is scattered by snow (or other bright surfaces) and backscattered or absorbed by atmospheric particles and clouds in the same vicinity (WMO/UNEP, 2002).

Variation in Solar Flux with Altitude

Solar flux increases with altitude above sea level, due to the decreased presence of clouds and declining concentrations of scattering and absorbing atmospheric pollutants. Rayleigh scattering, also lessens with decreasing atmospheric pressure. A number of measurements of UV radiation have been taken at various altitudes and are reviewed by Xenopoulos and Schindler

(2001). Increases in flux as a function of altitude are given as percent irradiance enhancement per 1000 m relative to sea level. The effect can range from 9 to 24% /1000 m as function of the altitude at which the measurement was taken (Xenopoulos and Schindler, 2001). The effect corresponds to the relative pathlength traveled by the solar photon: flux is strongest when the photon is not impeded by atmospheric scattering or absorbing agents. Similarly, this effect is seen as a function of solar zenith angle, i.e., flux is at its maximum when the atmospheric depth through which the photon must pass is at its shallowest.

Clouds

In principle, clouds have the largest influence on surface-level UV irradiance, but their effects are difficult to quantify. The depth and composition of a cloud determine, in part, the amount and wavelengths of radiation that it will scatter or absorb. Geometry is an especially important factor, as the reduction in irradiance may be small with scattered or broken clouds – or may be enhanced by scattering between clouds, increasing surface flux (WMO/UNEP, 2002). Quantifying the effect of clouds on surface UV flux, therefore, requires detailed information on cloud composition, geometry, altitude, and the position of the sun relative to the cloud and the underlying surface as a function of time. Provided that all of this information is available, a three-dimensional model is then required to calculate surface-level reductions or enhancements in UV flux.

Particulate Matter

On a zonally averaged basis, PM does not contribute significantly to lower tropospheric absorption of UV radiation. However, in urban areas or other areas subject to high smog levels (areas of significant biomass combustion), PM may be the most important determinant of ground-level erythemal UV flux, second only to cloud cover (U.S. Environmental Protection Agency, 2004; WMO/UNEP, 2002). Model-to-measurement comparisons of ground-level flux for Greece and Toronto, Canada, have shown 20 and 5-10% reductions, respectively (McKenzie et al., 2003). Increases over the past 20 to 30 years in combustion-associated PM and black carbon may account for the inability to detect a surface trend in UV-B radiation caused by a known decrease in stratospheric O_3 over the Northern Hemisphere (Barnard et al., 2003).

Gases

In the upper troposphere, the UV-absorbing gases O_3 and, of lesser importance, formaldehyde (CH_2O) and SO_2 are vented or diffuse from the surface. Stratospheric intrusions force O_3-rich air into the troposphere where it mixes, increasing regional background O_3 levels (see Chapter 3). Tropospheric O_3 data are typically expressed on a concentration basis, e.g., parts per billion by volume (ppbv), where 1 ppbv tropospheric $O_3 \equiv 0.65$ DU (IPCC, 2001a). Ozone concentrations decrease with increasing altitude from the surface up to, roughly, the mid-troposphere, then increase up into the stratosphere. Figure 10-4 shows a series of O_3 vertical profiles for 4 sites within the continental U.S., i.e., plots of O_3 concentrations as a function of atmospheric pressure (correlating to altitude). The mean values of O_3 in the free troposphere reported in the literature range from ~50 to ~80 ppbv, with higher values occurring at the tropopause. For example, a series of ozonesonde soundings over France from 1976 to 1995 showed an O_3 increase from 48.9 ppbv in the 2.5 to 3.5 km layer to 56.5 ppbv in the 6.5 to 7.5 km layer, although the data revealed no statistically significant increasing trend over time (Ancellet and Beekmann, 1997).

Photochemistry produces a diurnal rise and fall in O_3 and PM concentrations in polluted urban settings. Temperature inversions that often occur in these settings prevent the upward mixing and dilution of ground-level O_3, also trapping primary and secondary PM within the boundary layer. A recent study of the concurrence of O_3 and PM is provided by Koloutsou-Vakakis et al. (2001). No measurement technique is currently available that can distinguish between absorption of incident UV radiation by O_3 versus absorption by PM.

Ultraviolet absorption by gases becomes significant under aerosol- and cloud-free conditions. Figure 10-5 shows a calculation by Krotkov et al. (1998) of the sensitivity, as a function of wavelength, of ground-level UV flux to a 1-DU decrease in total column O_3 under cloud- and aerosol-free conditions. A 1991 to 1992 study in Chicago in which ambient O_3, broadband UV irradiance, and total sunlight were monitored (Frederick et al., 1993) found a significant negative correlation between the UV irradiance and ambient O_3 when the atmosphere was relatively free of clouds and haze. Although Frederick et al. (1993) estimated that a 10-ppbv reduction in O_3 was associated with a 1.3% increase in erythemally-weighted UV-B, they cautioned that this value had a comparatively large uncertainty (±1.2%, or nearly 100% of the predicted increase).

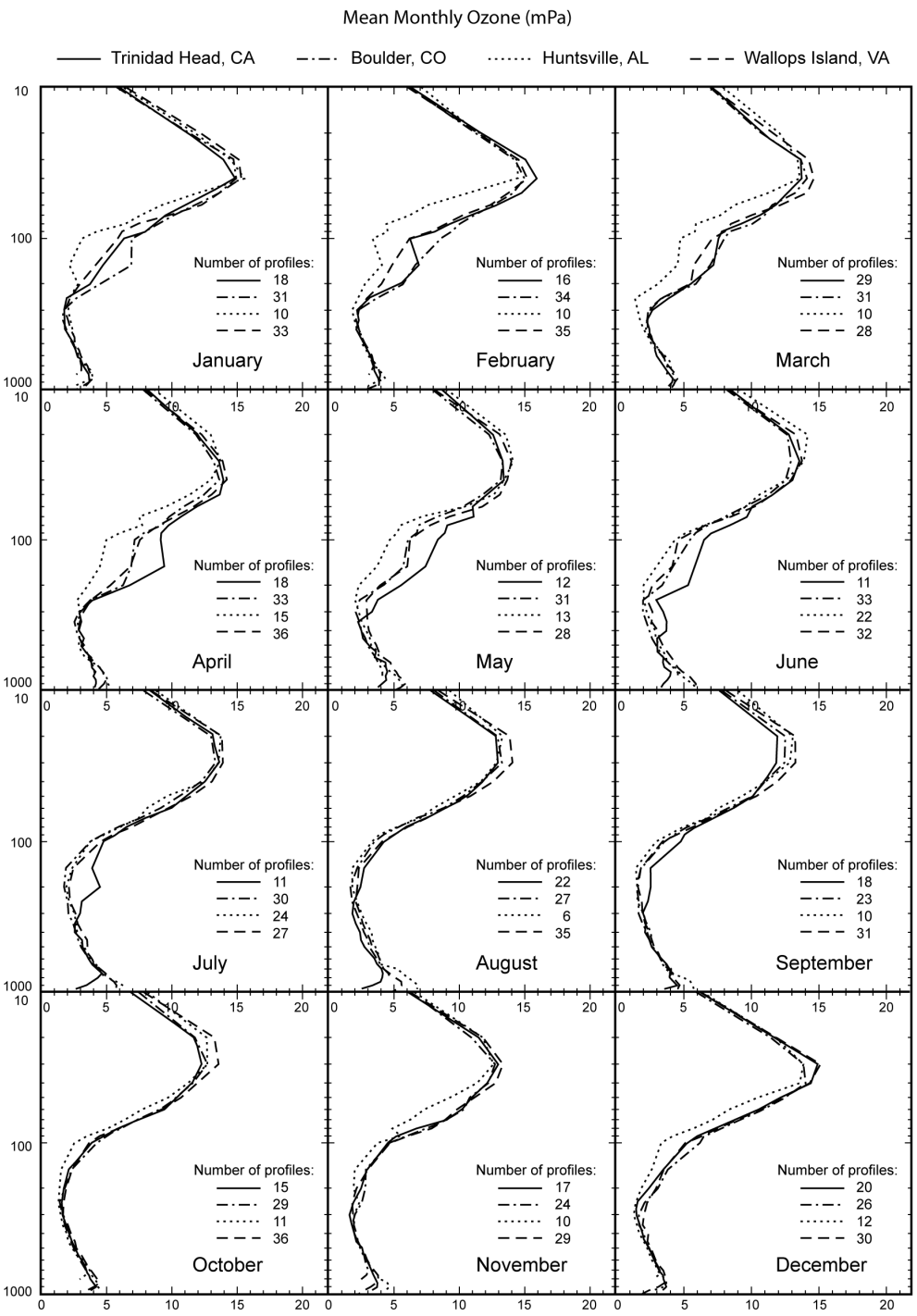


Figure 10-4. Monthly averaged vertical O_3 profiles (partial pressure in mPa) as a function of atmospheric pressure (in mBar) for Trinidad Head, CA (solid line); Boulder, CO (dot-dashed line); Huntsville, AL (dotted line); and Wallops Island, VA (dashed line). The number of launches at each site for each month are indicated on the charts.

Source: Newchurch et al. (2003).

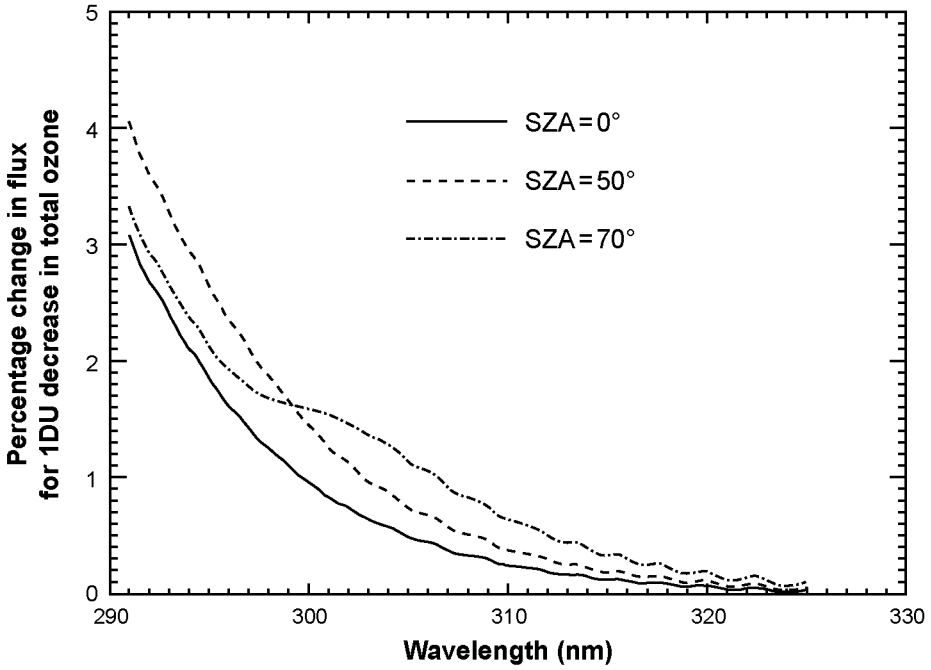


Figure 10-5. The sensitivity of ground-level UV flux to a 1 DU change in total column O_3, under clear sky conditions, as a function of solar zenith angle (SZA).

Source: Krotkov et al. (1998).

Note that when attempting to apply the results of studies such as Krotkov et al. (1998) in an analysis of the importance of surface O_3 to protecting humans from UVB-related diseases, is that flux is ordinarily determined as a function of total column O_3 density, which includes the stratospheric and upper tropospheric O_3, not simply surface-level pollutant O_3. Nearly all of the routine data on tropospheric O_3 concentrations in the U.S. is from ground-level O_3 monitors, such as those used to determine the attainment of the O_3 air quality standards. Such measurements, alone, are not sufficient information for making reliable estimates of ambient O_3 concentrations above the boundary layer.

10.2.1.4 Data Requirements for a Surface UV-B Climatology

A means of establishing the range of variability in UV-B at ground level would be in the development of a map of flux levels under typical seasonal conditions based on historical

records. In the atmospheric sciences community, a map of this type is referred to as a "climatology."

The WMO/UNEP (2002) stated that, in principle, if the spatial distribution of all UV absorbers and scatterers were fully known, the wavelength and angular distribution of the UV irradiance at the Earth's surface could be determined with model calculations. However, the very limited information available on the distribution of the primary components (i.e., clouds, particles, O_3, and surface albedo) makes detailed predictions impossible. In an earlier assessment of the environmental effects of stratospheric O_3 depletion (WMO/UNEP, 1999), the UNEP concluded that, in view of the high spatial and temporal variability of surface UV radiation and the difficulty in maintaining calibration within networks of UV monitoring instruments, satellite-based observations are necessary to develop a satisfactory UV climatology. Furthermore, satellite-derived estimates of surface UV are limited by the availability of instruments in orbit, with available datasets comprising interpolations based upon a single satellite overpass per day for a given region. Complete assessment of the uncertainties in predictions of UV surface flux would require comparisons between sparse available ground-level observations and satellite data over longer periods of time and for different geographical locations. No such assessment has been reported in the scientific literature.

10.2.2 Factors Governing Human Exposure to Ultraviolet Radiation

An assessment of public health benefits due to the attenuation of UV-B radiation by surface-level O_3 requires appropriate consideration of: (1) the multiple factors that alter the flux of UV-B radiation at ground-level, as described above; (2) the factors that influence the extent of human exposure to UV-B radiation, particularly behavioral decisions; and (3) the effects of UV-B radiation exposure on human health. Consideration must also be given to the public health benefits from exposure to UV-B radiation. The present section outlines the most recent information on the determinants of exposure to UV-B radiation in human populations. Quantitative evaluation of human exposure to UV-B radiation is scientifically necessary to perform health risk assessment and to define subpopulations at risk for UV-B-related health effects.

10.2.2.1 Outdoor Activities

Exposure to solar UV radiation is related to one predominating factor: time spent outdoors during daylight hours. A large U.S. study was conducted using the EPA National Human Activity Pattern Survey (NHAPS) to assess UV radiation dose in Americans (Godar, 2001; Godar et al., 2001, 2003). The EPA NHAPS recorded the activity profiles of 9,386 Americans (age 0 to 60+ years) over a 24-month period to assess their exposure to various environmental pollutants, including UV radiation. Available UV radiation was assessed using the EPA UV-monitoring program. Solar radiation in the UV-A and UV-B waveband regions were measured daily at a monitoring site in each quadrant of the U.S. There is considerable error associated with quantifying UV radiation dose from exposure surveys and four UV-monitoring sites across the country; however, the qualitative information regarding factors that increase human exposure to UV radiation is still of relevance. The EPA-UV monitoring network has since expanded to 21 sites, located in 14 U.S. national parks and 7 urban areas across the U.S. (http://www.epa.gov/uvnet/). A UV-B monitoring network by the U.S. Department of Agriculture is also available for the quantitative assessment of UV radiation exposure (http://uvb.nrel.colostate.edu/UVB/). This monitoring network has 30+ monitoring sites across the U.S. and three additional sites in Canada and New Zealand.

Godar et al. (2001) observed a strong seasonal preference for outdoor activities, with people spending the most time outdoors during the summer followed by spring, fall, and, lastly, winter. Because the solar erythemal (i.e., skin reddening) UV radiation dose is also highest during the summer, the estimated UV radiation dose of Americans was more than 10-fold greater in the summer compared to the winter season (Godar et al., 2001).

Vacationing at the beach in the summer was associated with higher UV radiation exposures (Godar et al., 2001; Thieden et al., 2001). Even after accounting for sunscreen use at the beach, the erythemal UV radiation doses were more than 40% higher during a 3-week beach vacation compared to a 3-week stay at home (Godar et al., 2001). Danish children and adolescents were found to receive >50% of their annual UV radiation dose while vacationing at European beaches (Thieden et al., 2004a). Sunbathing also was associated with increased annual UV radiation dose in the Canadian National Survey on Sun Exposure and Protective Behaviours (Shoveller et al., 1998). Among the 3,449 adults (age 25+ years) who completed the telephone household survey, 21% stated that they spent time actively sunbathing. In a Danish study with 164

participants, all children (age 1 to 12 years) and teenagers (age 13 to 19 years) as well as 94% of adults (age 20 to 76 years) had days with risk behavior (Thieden et al., 2004b). Teenagers, who had the highest number of risk-behaviors days, were found to have the highest annual UV radiation doses. Among teenagers, 76% (95% CI: 41, 98) of their UV radiation dose during the measurement period was received on risk-behavior days, as determined using personal electronic UV dosimeters and exposure diaries (Thieden et al., 2004b).

An Australian study examining time profiles of daily UV radiation exposure among 8th grade students observed that up to 47% of the daily UV radiation dose fell within the time periods when students were outdoors during school hours, sitting under shaded structures during lunch breaks and participating in routine outdoors or sports activities (Moise et al., 1999). Other studies also have found that participation in outdoor sports (e.g., basketball, soccer, golfing, swimming, cycling) significantly increased UV radiation exposure (Moehrle, 2001; Moehrle et al., 2000; Thieden et al., 2004a,b).

10.2.2.2 Occupation

Of the various factors that affect human exposure to UV radiation, occupation is also important. Approximately 5% of the American workforce work outdoors, as determined by the EPA NHAPS (Godar et al., 2001). On average, American indoor workers spend ~10% of their day outdoors. During their time outdoors, they are exposed to ~30% of the total ground-level UV flux, as measured by the EPA UV-monitoring program (Godar et al., 2001). Compared to indoor or in-home workers, outdoor workers are exposed to much higher levels of UV radiation (Kimlin et al., 1998a; Thieden et al., 2004a), frequently at levels that are above current exposure limits set by the International Commission on Non-Ionizing Radiation Protection (ICNIRP, 2004). For example, Thieden et al. (2004a) observed that the annual UV radiation dose, estimated using personal electronic UV dosimeters and exposure diaries, was ~70% higher for gardeners than indoor workers. The gardeners received the majority (55%) of their UV radiation dose on working days (Thieden et al., 2004a). Another study found that outdoor workers received three to four times the annual UV radiation exposure of indoor workers (Diffey, 1990). At-risk working populations include farmers (Airey et al., 1997; Schenker et al., 2002), fishermen (Rosenthal et al., 1988), landscapers (Rosenthal et al., 1988), building and construction workers (Gies and Wright, 2003), physical education teachers (Vishvakarman et al.,

2001), mail delivery personnel (Vishvakarman et al., 2001), and various other workers who spend the majority of their day outdoors during peak UV radiation hours.

10.2.2.3 Age

Age may be a factor that influences human exposure to UV radiation. In a large U.S. study using the EPA NHAPS, the average UV radiation dose among American children (age <12 years) was estimated to be slightly higher (~20%) than that of adolescents (age 13 to 19 years) (Godar, 2001). A large Canadian survey found that 89% of children (age <12 years) had 30 minutes or more of daily UV exposure compared to 51% for both adults (age 25+ years) and youth (age 15 to 24 years) (Lovato et al., 1998a, 1998b; Shoveller et al., 1998). In an English study (Diffey et al., 1996), UV radiation exposure was estimated in 180 children (age 9 to 10 years) and adolescents (age 14 to 15 years) using personal film badges and exposure records. Once again, children were found to have received higher UV radiation exposure compared to adolescents (Diffey et al., 1996). However, as discussed earlier, a Danish study found that the annual UV radiation dose in teenagers (age 13 to 19 years) was 14-24% higher compared to children (age 1 to 12 years) and adults (Thieden et al., 2004b). This increase in UV radiation dose in the Danish teenagers was attributed to their increased risk-behavior days. Therefore, age may affect human exposure to UV radiation by influencing other factors of exposure, such as outdoor activity and risk behavior.

Two studies examined lifetime UV radiation exposure among persons in the U.S. (Godar et al., 2003) and Denmark (Thieden et al., 2004b). Both studies observed that while there are slight differences in UV radiation dose by age, generally people receive fairly consistent UV doses at different age intervals throughout their lives.

10.2.2.4 Gender

Studies have indicated that females generally spend less time outdoors and, consequently, have lower UV radiation exposure compared to males (Gies et al., 1998; Godar et al., 2001; Shoveller et al., 1998). The U.S. study by Godar et al. (2001) observed that while both males and females had relatively consistent erythemal UV radiation doses throughout their lives, males consistently received higher overall UV doses compared to females at all age groups. Among all Americans, the lowest exposure to UV radiation was received in females during their child-

raising years (age 22 to 40 years) (Godar et al., 2001). The highest exposure was observed in males aged 41 to 59 years in the U.S. study (Godar et al., 2001). A similar Canadian survey found that younger adult males had the greatest exposures to UV radiation (Shoveller et al., 1998).

10.2.2.5 Geography

In the U.S. study by Godar et al. (2001), erythemal UV radiation doses were examined in persons living in northern and southern regions. Northerners and southerners were found to spend an equal amount of time outdoors; however, the higher solar flux at lower latitudes significantly increased the annual UV radiation dose for southerners (Godar et al., 2001). The annual UV radiation doses in southerners were 25 and 40% higher in females and males, respectively, compared to northerners (Godar et al., 2001). Other studies also have shown that altitude and latitude influence personal exposure to UV radiation (Kimlin et al., 1998b; Rigel et al., 1999).

10.2.2.6 Protective Behavior

Protective behaviors such as using sunscreen (e.g., Nole and Johnson, 2004), wearing protective clothing (e.g., Rosenthal et al., 1988; Sarkar, 2004; Wong et al., 1996), and spending time in shaded areas (Moise et al., 1999; Parisi et al., 1999) have been shown to reduce exposure to UV radiation. In one study, the use of sunscreen was associated with extended intentional UV radiation exposure (Autier et al., 1999); however, a follow-up study indicated that sunscreen use increased duration of exposures to doses of UV radiation that were below the threshold level for erythema (Autier et al., 2000).

In a national study of U.S. youths aged 11 to 18 years, the most prevalent protective behavior was sunscreen use (39.2%) followed by use of a baseball hat (4.5%) (Davis et al., 2002). There were significant differences in the use of sunscreen by age group and gender, with the younger age group (age 11 to 13 years) and girls having greater likelihood (47.4 and 48.4%, respectively) of using sunscreen (Davis et al., 2002). The Canadian National Survey on Sun Exposure and Protective Behaviours observed that less than half of the adults (age 25+ years, n = 3,449) surveyed took adequate protective actions (Shoveller et al., 1998). Once again, children (age <12 years, n = 1,051) were most protected from exposure to UV radiation, with

76% using sunscreen and 36% avoiding the sun, as reported by their parents (Lovato et al., 1998a). However, the protection level was still not adequate, as indicated by the high 45% rate of erythema in children. Among Canadian youth (age 15 to 24 years, n = 574), protective actions from UV radiation exposure included wearing a hat (38%) and seeking shade and avoiding the sun between the peak hours of 11:00 a.m. to 4:00 p.m. (26%) (Lovato et al., 1998b). The lowest prevalence of protective behavior among the youth was likely responsible for the highest proportion of erythema (68%) experienced in this age group. A Danish study observed that both children and teenagers applied sunscreen on more days than adults, but teenagers had the most days with erythema, due to their increased risk behavior (Thieden et al., 2004b). A survey in Switzerland of 1,285 individuals, including children and parents, indicated that sunscreen use was the protective action most commonly used, but only at the beach and not in routine daily exposure (Berret et al., 2002). In general, protective clothing and avoiding the sun were not highly used among these individuals to protect against UV-related health effects.

10.2.2.7 Summary of Factors that Affect Human Exposures to Ultraviolet Radiation

The factors that potentially influence UV radiation doses were discussed in the previous sections and include outdoor activities, occupation, age, gender, geography, and protective behavior. Results from the various studies indicate that the following subpopulations may be at risk for higher exposures to UV radiation:

- Individuals who engage in high-risk behavior, viz., sunbathing;
- Individuals who participate in outdoor sports and activities;
- Individuals who work outdoors with inadequate shade, e.g., farmers, fishermen, gardeners, landscapers, building and construction workers; and
- Individuals living in geographic areas with higher solar flux (i.e., lower latitudes [e.g., Honolulu, HI] and higher altitudes [e.g., Denver, CO]).

10.2.3 Factors Governing Human Health Effects due to Ultraviolet Radiation

Ultraviolet radiation occupies a specific region of the electromagnetic spectrum of wavelengths and can be further subdivided into three parts, UV-A (320 to 400 nm), UV-B (280 to 320 nm), and UV-C (200 to 280 nm). Most of the health risks associated with UV radiation exposure are wavelength dependent. Wavelengths <180 nm are of little practical

biological significance as they are almost completely absorbed by the stratosphere (ICNIRP, 2004).

"Action spectra" of a given biological response to UV radiation across its spectral range are used to estimate exposure by weighting individual wavelength intensities according to the associated response. The overall effectiveness of the incident flux at inducing the biological response of interest is computed by means of the relationship:

$$effective\ irradiance = \int_{\lambda} I_{\lambda} E_{\lambda} d\lambda \qquad (10\text{-}4)$$

where I_{λ} and E_{λ} are, respectively, the irradiance and its relative effectiveness at wavelength λ.

Until 1980, it was generally thought that wavelengths <315 nm were responsible for the most significant adverse UV radiation health effects; however, recent studies have found that the longer wavelengths in the UV-A range also may produce adverse responses at substantially higher doses (ICNIRP, 2004). As UV-A radiation is not absorbed by O_3, health effects solely induced by UV-A exposure are not relevant in an analysis of public health risks/benefits associated with O_3-related UV attenuation. Therefore, this section focuses on the latest available information on the various adverse health effects associated with acute and chronic UV-B radiation exposure.

10.2.3.1 Erythema

Association Between Ultraviolet Radiation Exposure and Erythema

The most conspicuous and well-recognized acute response to UV radiation is erythema, or the reddening of the skin, which is likely caused by direct damage to DNA by UV-B and UV-A radiation (Matsumura and Ananthaswamy, 2004). Indirect oxidative damage also may occur at longer wavelengths (Matsumura and Ananthaswamy, 2004). Skin type appears to play a large role in the sensitivity to UV radiation-induced erythema. The Fitzpatrick classifications for skin types are: (1) skin type I – individuals with extremely sensitive skin that sunburns easily and severely, and is not likely to tan (e.g., very fair skin, blue eyes, freckles); (2) skin type II – individuals with very sensitive skin that usually sunburns easily and severely, and tans minimally (e.g., fair skin, red or blond hair, blue, hazel or brown eyes); (3) skin type

III – individuals with sensitive skin that sunburns moderately and tans slowly (e.g., white skin, dark hair); (4) skin type IV – individuals with moderately sensitive skin that sunburns minimally and usually tans well (e.g., white or light brown skin, dark hair, dark eyes); (5) skin type V – individuals with minimally sensitive skin that rarely sunburns and tans deeply (e.g., brown skin); and (6) skin type VI – individuals with nonsensitive skin that never sunburns and tans profusely (e.g., dark skin). Harrison and Young (2002) found that the perceptible minimal erythemal dose was approximately twofold greater for individuals with skin type IV compared to skin type I, although there was considerable overlap in the minimal erythemal dose among the four skin types. Waterston et al. (2004) further observed that within an individual, erythemal response differed by body site (e.g., abdomen, chest, front upper arm, back of thigh). These differences were likely attributable to body site-specific variations in melanin pigmentation.

Kollias et al. (2001) investigated the change in erythemal response following a previous exposure to UV radiation. Body sites that received a second exposure to UV radiation always showed a reduced erythemal response compared to body sites with a single exposure, especially when the first exposure was at levels greater than the minimal erythemal dose. The suppression of erythema was more pronounced when the second exposure was given 48 hours after the first. These findings support the well established notion that repeated exposures to UV radiation results in adaptation (e.g., stimulation of melanogenesis). Kaidbey and Kligman (1981) examined individuals with skin types I, II, and III, and found that multiple exposures to subthreshold doses of UV radiation at 24-hour intervals resulted in cumulative injury to the skin, as indicated by a lowering of the minimal erythemal dose. These results suggest that a longer time period than 24 hours may be necessary to repair damage from a single exposure to UV radiation. Henriksen et al. (2004) also observed a lowering of the minimal erythemal dose with repeated exposure at 24-hour intervals in 49 healthy volunteers with skin types II, III, IV, and V. However, adaptation was reached after the 4th consecutive exposure. Henriksen et al. further found that the change in threshold depended on skin type. After 4 days of repeated UV radiation, there was little change (10 to 20%) in the erythemal threshold dose with repeated exposure to UV radiation in the fair-skinned individuals. Among the darker-skinned individuals, the minimal erythemal dose was lowered by 40 to 50%. However, both the initial UV dose and the dose to erythema after four days of exposure was still higher in the dark-skinned persons.

A reference erythema action spectrum was adopted by the Commission Internationale de l'Eclairage (International Commission on Illumination, CIE) in 1987 (McKinlay and Diffey, 1987). The CIE erythema action spectrum indicates that UV-B radiation is orders of magnitude more effective per unit dose than UV-A radiation.

Risk of Erythema from Changes in Tropospheric O_3 Levels

There is no literature examining the risk of erythema associated with changes specifically in tropospheric or ground-level O_3 levels. The scientific studies, available to date, focus on the effects of a reduction in stratospheric ozone. One such study has assessed the effects of stratospheric O_3 depletion on the risk of erythema (Longstreth et al., 1998). The analysis by Longstreth et al. (1998) concluded that the risk of erythema would not appreciably increase with depletion of the stratospheric O_3 layer. This is due to the powerful adaptation of the skin to different levels of UV radiation, as evidenced by its ability to cope with changes in UV radiation by season (van der Leun and de Gruijl, 1993). Gradual exposure to increasing UV radiation from the winter to summer leads to decreased sensitivity of the skin. In midlatitudes, the UV-B radiation in the summer is 10-fold greater than in the winter. In contrast, the steady depletion of the O_3 layer has been estimated to result in an approximately 20% increase in UV-B over 10 years (Longstreth et al., 1998). The comparatively small increase in UV radiation throughout the years, therefore, would not significantly increase the risk of erythema. Tropospheric O_3 constitutes no more than 10% of total atmospheric O_3. Given that stratospheric O_3 depletion was unlikely to increase the risk of erythema, one could reasonably conclude that small changes in ground-level O_3 that take place with attainment of the O_3 NAAQS would also not result in increased risk.

10.2.3.2 Skin Cancer

According to the American Academy of Dermatology, one in five Americans develop skin cancer during their lifetime. The three main forms of skin cancer include basal cell carcinoma and squamous cell carcinoma, which are both nonmelanoma skin cancers, and malignant melanoma. Nonmelanoma skin cancers constitute more than one-third of all cancers in the U.S. and ~90% of all skin cancers, with basal cell carcinoma being approximately four times as common as squamous cell carcinoma (Diepgen and Mahler, 2002; ICNIRP, 2004). The incidence of malignant melanoma is much lower than nonmelanoma skin cancers. In 2004 more

than one million cases of basal and squamous cell skin cancer are expected to be newly diagnosed, compared to 40,780 cases of melanoma (Jemal et al., 2004). However, melanoma has great metastatic potential and accounts for the majority of skin cancer deaths.

Exposure to UV radiation is considered to be a major risk factor for all three forms of skin cancer (Gloster and Brodland,1996; Diepgen and Mahler, 2002; IARC, 1992). Ultraviolet radiation is especially effective in inducing genetic mutations and acts as both a tumor initiator and promoter. Keratinocytes have evolved DNA repair mechanisms to correct the damage induced by UV; however, mutations can occur, leading to skin cancers that are appearing with increasing frequency (Hildesheim and Fornace, 2004). The relationship between skin cancer and chronic exposure to UV radiation is further explored below, followed by discussion of the influence of O_3 on the incidence of skin cancer.

10.2.3.3 Ultraviolet Radiation Exposure and the Incidence of Nonmelanoma Skin Cancers

The incidence of all three types of cancers has been shown to rise with increasing UV radiation concentrations across the U.S. (de Gruijl, 1999); however, the most convincing evidence for a causal relationship exists between UV radiation and squamous cell carcinoma. Squamous cell carcinoma occurs almost exclusively on skin that is regularly exposed to the sun, such as the face, neck, arms, and hands. The incidence is higher among whites in areas of lower latitudes, where solar flux is greater (Kricker et al., 1994). The risk of squamous cell carcinoma was shown to increase with life-long accumulated exposure to UV radiation in one cross-sectional study (Vitasa et al., 1990); however, increased risk was found to be associated only with exposure 10 years prior to diagnosis in a case-control study (Gallagher et al., 1995a). One of the major concerns with both types of studies is the potential for recall bias in reporting past UV radiation exposure by individuals already aware of their disease status.

Ultraviolet radiation also has been linked to basal cell carcinoma. Basal cell carcinoma is common on the face and neck (80-90%) but rarely occurs on the back of the hands (de Gruijl, 1999). While cumulative UV radiation exposure was not associated with an increased risk of basal cell carcinoma (Vitasa et al., 1990), increased risk was observed in individuals with greater recreational UV radiation exposure in adolescence and childhood (age <19 years) and individuals with a history of severe erythema in childhood (Gallagher et al., 1995b). Once again, consideration must be given to potential recall bias in assessing these results. Thus, there is

suggestive evidence that UV radiation also plays a role in the development of basal cell carcinoma, but the etiologic mechanisms for squamous cell carcinoma and basal cell carcinoma likely differ. In an Australian study conducted in a subtropical community, the factors of having fair skin, a history of repeated sunburns, and nonmalignant solar skin damage diagnosed by dermatologists were strongly associated with both types of nonmelanoma skin cancer (Green et al., 1996). The authors attributed the finding that outdoor occupation was not associated with nonmelanoma skin cancer to self-selection. Individuals with fair or medium complexions and a tendency to sunburn accounted for more than 80% of the community study sample; however, they were systematically underrepresented among outdoor workers (Green et al., 1996). Such self-selection bias might partly explain the lack of consistent quantitative evidence of a causal link between UV radiation and skin cancer in humans.

De Gruijl et al. (1993) assessed the action spectrum for nonmelanoma skin cancers using hairless albino mice. Human data are not available regarding wavelength dependence of the carcinogenicity of UV radiation. After adjusting for species differences, the Skin Cancer Utrecht-Philadelphia action spectrum indicated the highest effectiveness in the UV-B range with a maximum at 293 nm, which dropped to 10^{-4} of this maximum at the UV-A range above 340 nm (de Gruijl et al., 1993). The mutations commonly present in the *p53* tumor suppressor gene in individuals with squamous cell carcinoma and basal cell carcinoma are called the "signature" mutations of UV-B radiation (de Gruijl, 2002). UV-B radiation is highly mutagenic, because DNA is a chromophore for UV-B, but not for UV-A radiation (Ichihashi et al., 2003). Nevertheless, other studies have found that UV-A radiation, in addition to UV-B radiation, can induce DNA damage (Persson et al, 2002; Rünger et al., 2000). DNA damage by UV-A is mediated by reactive oxygen species, making it indistinguishable from damage caused by other agents that generate reactive oxygen species (de Gruijl, 2002). Epidemiologic evidence of a carcinogenic effect of UV-A was found in a study of psoriasis patients receiving oral psoralen and UV-A radiation treatment (Stern et al., 1998). High-dose exposure to oral psoralen and UV-A radiation was associated with a persistent, dose-related increase in the risk of squamous cell cancer. Risk of basal cell cancer also was increased in those patients exposed to very high levels of UV-A radiation. Therefore, although UV-B radiation has long been considered the main culprit for nonmelanoma skin cancer, limited evidence suggest that UV-A radiation may also play a role.

Susceptible populations for nonmelanoma skin cancers include individuals with reduced capacity for nucleotide excision repair, the primary repair mechanism for UV radiation-induced DNA lesions (Ichihashi et al., 2003). At particular risk are individuals with xeroderma pigmentosum, as they have defective nucleotide excision repair in all tissues (Kraemer, 1997; Sarasin, 1999). Skin type also largely affects susceptibility to skin cancer. Of the six skin phenotypes, the most sensitive individuals are those with skin types I and II, who have a fair complexion, blue or green eyes, and red or blond hair (Diepgen and Mahler, 2002). These individuals tend to sunburn easily, tan poorly, and freckle with sun exposure. A history of repeated sunburns also appears to increase the risk of both cancers, while sunburns during childhood are more associated with increased basal cell carcinoma (Gallagher et al., 1995b; Green et al., 1996).

Ultraviolet Radiation and the Incidence of Cutaneous Malignant Melanoma

From 1973 to 1994, the incidence rate of melanoma increased 120.5% along with an increased mortality rate of 38.9% among whites in the U.S. (Hall et al., 1999). The ICNIRP (2004) states that during the past 40 years or so, each decade has seen a twofold increase in the incidence of malignant melanoma in white populations, with increased incidence observed more prominently in individuals living in lower latitudes. Cutaneous malignant melanoma has a mutifactorial etiology with environmental, genetic, and host factors (Lens and Dawes, 2004). The major environmental factor of malignant melanoma has been identified as UV radiation exposure (Diepgen and Mahler, 2002); therefore, the increased incidence of melanoma throughout the years might be partially attributable to changes in human activity patterns (e.g., increased outdoor activity) that influence UV exposure or increased UV radiation at the ground level. The risk of melanoma appears to depend on the interaction between the nature of the exposure and skin type (Lens and Dawes, 2004).

Fears et al. (2002) examined the association between invasive cutaneous melanoma and UV radiation in non-Hispanic whites using a case-control study design. Lifetime residential history was coupled with mid-range UV-B radiation flux measurements to reduce exposure misclassification and recall bias. A 10% increase in the average annual UV-B flux was significantly associated with a 19% (95% CI: 5, 35) increase in individual odds for melanoma in men and a 16% (95% CI: 2, 32) increase in women. Whiteman et al. (2001) conducted a

systematic review of studies that examined the association between childhood UV radiation exposure and risk of melanoma. Researchers found that ecological studies assessing ambient sun exposure consistently reported higher risks of melanoma among people who resided in an environment with high UV radiation during their childhood (Whiteman et al., 2001). The lack of consistency among the case-control studies was likely due to the varying methods used to assess UV radiation dose.

While the evidence is generally suggestive of a causal relationship between UV radiation and malignant melanoma, possibly conflicting data also has been observed. For example, the highest occurrence of malignant melanoma is on men's backs and women's legs, areas that do not have prolonged exposure to the sun (Rivers, 2004). This indicates that, unlike nonmelamona skin cancers, malignant melanoma tends to occur in sites of intermittent, intense sun exposure (trunk and legs), rather than in areas of cumulative sun damage (head, neck, and arms) (Swetter, 2003). A study by Whiteman et al. (2003) observed that individuals with melanomas of the trunk had more melanocyte nevi and less solar keratoses compared to individuals with head and neck melanomas and, suggesting that cutaneous melanomas may arise through two pathways, one associated with melanocyte proliferation and the other with chronic exposure to sunlight. Green et al. (1999) also found that melanomas of the soles and palms resembled other cutaneous melanomas in their association with sun exposure, but were distinguished from them by their strong positive associations with nevi on the soles.

The available data conflict with regard to the relative importance of UV-A versus UV-B in inducing melanomas. UV-A has a much higher flux rate at the Earth's surface, as it is not absorbed by O_3 and it is able to penetrate more deeply into the skin surface due to its longer wavelength. However, UV-B, as mentioned earlier, is much more energetic and, therefore, more effective in photochemically altering DNA. The individual roles of UV-A and UV-B in the development of cutaneous malignant melanoma have been examined in several studies. A case-control study of 571 patients and 913 matched controls found an elevated odds ratio of 1.8 (95% CI: 1.2, 2.7), after adjusting for skin type, hair color, raised nevi, and number of sunburns, for developing malignant melanoma in individuals who regularly used tanning beds, which typically are UV-A sources (Westerdahl et al., 2000). In a study by Setlow et al. (1993), an action spectrum using the tropical fish *Xiphophorus* indicated that UV-A range wavelengths were especially important in malignant melanoma induction. However, an action spectrum

using the opossum *Monodelphis domestica* found that the potency of UV-A for melanoma induction was extremely low compared to that of UV-B (Robinson et al., 2000). A recent study by De Fabo et al. (2004) examined the differences in wavelength effectiveness using a hepatocyte growth factor/scatter factor-transgenic mouse model. The epidermal tissue of these transgenic mice behaves similar to the human epidermis in response to UV exposure. Given the absence of a mammalian melanoma action spectrum, the standardized CIE erythema action spectrum was used to deliver identical erythemally effective doses. Only UV-B radiation was found to initiate mammalian cutaneous malignant melanoma. UV-A radiation, even at doses considered physiologically relevant, were ineffective at inducing melanoma (De Fabo et al., 2004). Overall, current evidence suggests that UV-B, and not UV-A, is the primary risk factor for malignant melanoma (ICNIRP, 2004).

The populations susceptible for malignant melanoma are similar to those for nonmelanoma skin cancers. Once again, individuals with xeroderma pigmentosum or a reduced capacity for nucleotide excision repair are at increased risk (Tomescu et al., 2001; Wei et al., 2003). Individuals with skin types I and II, or the fair-skin phenotype (blue or green eyes; blond or red hair; skin that freckles, sunburns easily, and does not tan), have increased susceptibility to malignant melanoma (Evans et al., 1988; Swetter, 2003; Veierød et al., 2003). However, the incidence of melanoma was also positively associated with UV radiation in Hispanics and blacks (Hu et al., 2004). Although the incidence of melanoma is much lower in Hispanics and blacks compared to whites, melanomas in these populations are more likely to metastasize and have a poorer prognosis (Black et al., 1987; Bellows et al., 2001). Among children, malignant melanoma appears to have similar epidemiologic characteristics to the adult form of the disease (Whiteman et al., 1997). Individuals with intermittent, intense sun exposure, particularly during childhood, were found to have increased risk of melanoma (Whiteman et al., 2001), in contrast to the association between cumulative exposure and increased risk of squamous cell carcinoma. One study found that a personal history of nonmelanoma skin cancer or precancer, higher socioeconomic status, and increased numbers of nevocytic nevi also were associated with increased incidence of melanoma (Evans et al., 1988).

Effect of Changes in Tropospheric O_3 Levels on Skin Cancer Incidence

The current evidence strongly suggests a causal link between exposure to UV radiation and the incidence of both nonmelanoma and melanoma skin cancer. Genetic factors, including skin phenotype and ability to repair DNA, affect an individual's susceptibility to skin cancer. Quantifying the relationship between UV radiation and skin cancer is complicated by the uncertainties involved in the selection of an action spectrum and appropriate characterization of dose (e.g., peak or cumulative levels of exposure, childhood or lifetime exposures). In addition, there are multiple complexities in attempting to quantify the effect of tropospheric O_3 levels on UV-radiation exposure, as described in Section 10.2. The absence of published studies that critically examine increased incidence of skin cancer attributable to decreased tropospheric O_3 levels reflects the significant challenges in determining ground-level O_3-related changes in UV radiation exposure. An analysis by Lutter and Wolz (1997) attempted to examine the effects of a nationwide 10 ppb reduction in seasonal average tropospheric O_3 on the incidence of nonmelanoma and melanoma skin cancers and cataracts. Their estimate, however, depended upon several simplifying assumptions, ranging from an assumed generalized 10 ppb reduction in O_3 column density, national annual average incidence rates for the two types of skin cancer, and simple, linear biological amplification factors. Further, the methodologies used in this analysis inherently have ignored area-specific factors that are important in estimating the extent to which small, variable changes in ground-level O_3 mediate long-term exposures to UV-B radiation. More reasonable estimates of the human health impacts of enhanced UV-B penetration following reduced surface O_3 concentrations require both a solid understanding of the multiple factors that define the extent of human exposure to UV-B at present, and well-defined and quantifiable links between human disease and UV-B exposure. The reader is referred to the U.S. EPA 2002 Final Response to Court Remand (Federal Register, 2003) for detailed discussions of the data and scientific issues associated with the determination of public health benefits resulting from the attenuation of UV-B by surface-level O_3.

In the absence of studies specifically addressing the reduction of tropospheric O_3 (by assuming that the key variable is total column O_3 density), inferences could be made concerning the effects of reduced tropospheric O_3-related increases in UV-B exposure on the basis of studies focused on stratospheric O_3 depletion. Several studies have examined the potential effect of stratospheric O_3 depletion on the incidence of skin cancer (de Gruijl, 1995; Longstreth et al.,

1995; Madronich and de Gruijl, 1993; Slaper et al., 1996; Urbach, 1997). Note that several of the concerns expressed for Lutter and Wolz (1997) are relevant here as well. Stratospheric O_3 depletion is likely to increase the ground-level UV-B flux, as O_3 absorbs radiation in that wavelength range with high efficiency. Because UV-B radiation is primarily implicated in the induction of skin cancer, especially among persons with skin phenotypes I and II, there is concern that the depletion of the O_3 layer would result in significantly increased incidence of skin cancers.

Estimation of the increased risk in melanoma associated with stratospheric O_3 depletion cannot be done adequately due to the lack of a mammalian action spectrum for melanoma. Furthermore, the complexity of the UV-related induction mechanism of melanoma adds an additional layer of uncertainty to the calculations. The excess risk in nonmelanoma skin cancers associated with a decrease in stratospheric O_3 was estimated using the Skin Cancer Utrecht-Philadelphia action spectrum based on hairless albino mice (Longstreth et al., 1995). Quantification of how much more UV radiation would reach ground level with each percentage decrease in O_3 required several assumptions: (1) annual doses were an appropriate measure; (2) personal doses were proportional to ambient doses; and, most notably, (3) each percentage decrease in O_3 was associated with a 1.2% increase in UV radiation. Next, the relationship between UV radiation and nonmelanoma skin cancer incidence was determined: each percent increase in annual UV radiation dose was estimated to cause a 2.5% increase in squamous cell carcinoma and 1.4% increase in basal cell carcinoma over a human lifetime. Incorporating all these factors, Longstreth et al. (1995) calculated that a sustained 10% decrease in stratospheric O_3 concentration would result in 250,000 additional nonmelanoma skin cancer cases per year. Madronich and de Gruijl (1993) noted that the largest percent of O_3-induced nonmelanoma skin cancer increases would be at high latitudes, where baseline incidence of skin cancer is usually small. Assuming a phaseout of primary O_3-depleting substances by 1996, as established by the Copenhagen Amendments in 1992, Slaper et al. (1996) estimated that the number of excess nonmelanoma skin cancers in the U.S. caused by O_3 depletion would exceed 33,000 per year (or approximately 7 per 100,000) around the year 2050.

However, estimating the increase in nonmelanoma skin cancer incidence attributable to the depletion of the stratospheric O_3 layer is marred by uncertainty. The following statement by

Madronich and de Gruijl (1994) describes the uncertainty of estimating the effect of stratospheric O_3 depletion on the incidence of skin cancer:

> Extrapolating trends and effects of UV into the future is very hypothetical due to uncertainties that arise from atmospheric chemistry, epidemiology, and related disciplines. The values that we calculated are one plausible measure of the magnitude of the O_3-UV effects….The timescales for atmospheric change and skin-cancer development are still far from certain: O_3 reductions are expected to continue well into next century, and the time between UV exposure and development of skin cancer is essentially unknown.

Therefore, much caution is necessary when assessing and interpreting the quantitative results of excess nonmelanoma skin cancer incidence due to stratospheric O_3 depletion. Although the effect of reductions in tropospheric or ground-level O_3 concentrations on skin cancer incidence has not been assessed, it would be expected to be much less compared to the effect from the depletion of the stratospheric O_3 layer, given that tropospheric O_3 makes up $\leq$10% of the total atmospheric O_3.

10.2.3.4 Ocular Effects of Ultraviolet Radiation Exposure

Ultraviolet Radiation Exposure and Risk of Ocular Damage

Ocular damage from UV radiation exposures includes effects on the cornea, lens, iris, and associated epithelial and conjunctival tissues. Absorption of UV radiation differs by wavelength, with short wavelengths (<300 nm) being almost completely absorbed by the cornea, whereas longer wavelengths are transmitted through the cornea and absorbed by the lens (McCarty and Taylor, 2002). The most common acute ocular effect of environmental UV exposure is photokeratitis, also known as snowblindness, caused by absorption of short wavelength UV radiation by the cornea. The action spectrum indicated that maximum sensitivity of the human eye was found to occur at 270 nm (ICNIRP, 2004; Pitts, 1993). The threshold for photokeratitis in humans varied from 4 to 14 mJ/cm^2 for wavelengths 220 to 310 nm.

Exposure to longer wavelengths has been shown to cause both transient and permanent opacities of the lens, or cataracts. Extensive toxicologic and epidemiologic evidence supports the causal association between UV radiation and cataracts (Hockwin et al., 1999; McCarty and Taylor, 2002). Ultraviolet radiation-induced cataracts are hypothesized to be caused by

oxidative stress leading to increased reactive species in the lens, which then causes damage to lens DNA and cross-linking of proteins. Exposure time to low-dose UV radiation was found to strongly influence cataract formation (Ayala et al., 2000). An action spectrum determined using young female rats indicated that the rat lens was most sensitive to 300 nm, correcting for corneal transmittance (Merriam et al., 2000). Oriowo et al. (2001) examined the action spectrum for cataract formation using whole cultured lens from pigs. As pigs lens are similar in shape and size to the human lens, some inferences may be made. Results indicated that the 270 to 315 nm waveband was most effective in producing UV-induced cataracts in vitro. However, the threshold values varied widely within that range, from 0.02 J/cm^2 for 285 nm to 0.74 J/cm^2 for 315 nm (Oriowo et al., 2001). At wavelengths >325 nm, the threshold levels were orders of magnitude larger, with a minimum threshold value of 18.7 J/cm^2.

An epidemiologic study examined the effects of UV radiation on cataract formation in watermen (e.g., commericial fishermen, boat workers) who worked on Chesapeake Bay, MD (Taylor et al., 1988). Among the 838 individuals surveyed in this study, 111 had cortical cataracts and 229 had nuclear cataracts. Results indicated that UV-B radiation was significantly associated with cortical, but not nuclear, cataract formation. For a given age, a doubling of cumulative UV-B exposure was associated with a 60% excess risk (95% CI: 1, 164) of cortical cataracts. No association was observed between cataracts and UV-A radiation in this outdoor-working population.

Risk of Ocular Damage from Changes in Tropospheric Ozone Levels

Cataracts are the most common cause of blindness in the world. McCarty et al. (2000) calculated that ocular UV radiation exposure accounted for 10% of the cortical cataracts in an Australian cohort of 4,744 individuals from both urban and rural areas. A study by Javitt and Taylor (1994-1995) found that the probability of cataract surgery in the U.S. increased by 3% for each 1° decrease in latitude. These results suggest that depletion of the stratospheric O_3 layer may increase UV radiation-induced cataract formation. After assuming a certain wavelength dependency along with several additional assumptions, every 1% decrease in the stratospheric O_3 layer was estimated to be associated with a 0.3 to 0.6% increase in cataracts (Longstreth et al., 1995). Longstreth et al. (1995) noted that this estimate has a high degree of uncertainty due to inadequate information on the action spectrum and dose-response relationships. Quantitative

estimates have not been possible for photokeratitis, pterygium, or other UV-related ocular effects due to lack of epidemiologic and experimental data.

As is the case for all of the other UV-related health outcomes, there is no published information on the potential effects on cataract formation due to any changes in surface-level UV flux resulting from decreases in tropospheric O_3.

10.2.3.5 Ultraviolet Radiation and Immune System Suppression

Experimental studies have suggested that exposure to UV radiation may suppress local and systemic immune responses to a variety of antigens (Clydesdale et al., 2001; Garssen and van Loveren, 2001; Selgrade et al., 1997). In rodent models, these effects have been shown to worsen the course and outcome of some infectious diseases and cancers (Granstein and Matsui, 2004; Norval et al., 1999). Granstein and Matsui (2004) stated that exposure to UV-B radiation caused immunosuppression in mice ultimately by releasing cytokines that prevent antigen-presenting cells from performing their normal functions and causing direct damage to epidermal Langerhans cells. Noonan et al. (2003) investigated UV skin cancer induction in two strains of reciprocal F1 hybrid mice and found that genetically determined differences in susceptibility to UV-induced immunosuppression was a risk factor for skin cancer. At high UV radiation doses, mice with greater susceptibility to immune suppression had a larger proportion of skin tumors compared to those with lower susceptibility (Noonan et al., 2003). In a study by Yoshikawa et al. (1990), development of contact hypersensitivity to dinitrochlorobenzene on irradiated buttock skin was examined. Individuals who failed to develop contact hypersensitivity were considered to be susceptible to UV-B radiation. Virtually all skin cancer patients (92%) were susceptibility to UV-B radiation-induced suppression of contact hypersensitivity, compared to approximately 40% of healthy volunteers. Others studies have observed increased skin cancer in immune suppressed organ transplant patients (Caforio et al., 2000; Lindelöf et al., 2000). Collectively, results from these studies suggest that immune suppression induced by UV radiation may be a risk factor for skin cancer induction (Ullrich, 2005).

There is also some evidence that UV radiation has indirect involvement in viral oncogenesis through the human papillomavirus (Pfister, 2003). Additional evidence of UV-related immunosuppression comes from an epidemiologic study of 919 patients with rare autoimmune muscle diseases from 15 cities on four continents with variable UV radiation

intensity (Okada et al., 2003). Ultraviolet radiation was strongly associated with the prevalence of dermatomyositis, an autoimmune disease distinguished by the presence of photosensitive pathognomonic rashes (Okada et al., 2003). In patients with the human immunodeficiency virus, UV-B radiation lead to activation of the virus in their skin through the release of cytoplasmic nuclear factor kappa B (Breuher-McHam et al., 2001). In a study by Selgrade et al. (2001), UV-induced immunosuppression was examined in 185 subjects with different skin pigmentations. To assess immune suppression, dinitrochlorobenzene was applied to irradiated buttock skin 72 hours after irradiation. Differences in sensitivity were unrelated to skin type based on the Fitzpatrick classification or minimal erythemal dose (Selgrade et al., 2001). However, erythemal reactivity, assessed by the steepness of the erythemal dose-response curve, was shown to be significantly associated with UV-induced immunosuppression. Only subjects with steep erythemal responses, which included individuals with skin types I through V, showed a dose-response relationship between UV exposure and immune suppression (Selgrade et al., 2001).

In other studies, UV radiation was associated with decreased autoimmune diseases. Several ecologic studies observed a decreased prevalence of multiple sclerosis, insulin-dependent diabetes mellitus, and rheumatoid arthritis in regions with lower latitude (i.e., higher UV radiation exposure) (Ponsonby et al., 2002). These results may be attributable to UV radiation-induced immunosuppression and UV-B-related production of vitamin D, which has immunomodulatory effects (Cantorna et al., 2000). The protective effects of UV radiation resulting from its active role in vitamin D production are further discussed in the next section.

Most action spectrum investigations have concluded that immunosuppression is caused most effectively by the UV-B waveband (Garssen and van Loveren, 2001). The effects of UV-A on local and systemic immunosuppression have been unclear and inconsistent. There is some evidence that high doses of UV-A is protective of immunosuppression induced by UV-B exposure (Halliday et al., 2004). Given the variety of outcomes of immune suppression and possible mechanisms of effect, little detailed information exists on UV radiation action spectrums and dose-response relationships. The available data are insufficient to conduct a critical risk assessment of UV radiation-induced immunosuppression in humans.

10.2.3.6 Protective Effects of Ultraviolet Radiation – Production of Vitamin D

Any risk assessment that attempts to quantify the consequences of increased UV-B exposure on humans due to reduced ground-level O_3 must include consideration of both negative and positive effects. A potential health benefit of increased UV-B exposure relates to the production of vitamin D in humans. Most humans depend on sun exposure to satisfy their requirements for vitamin D (Holick, 2004). UV-B photons are absorbed by 7-dehydrocholesterol in the skin, leading to its transformation to previtamin D_3, which is rapidly converted to vitamin D_3. Vitamin D_3 is metabolized in the liver, then in the kidney to its biologically active form of 1.25-dihydroxyvitamin D_3. One minimal erythemal dose produces vitamin D equivalent to an oral dose of 20,000 IU vitamin D, which is 100 times the recommended dietary allowance for adults under 50 years of age (Giovannucci, 2005; Holick, 2004).

Vitamin D deficiency can cause metabolic bone disease among children and adults, and also may increase the risk of many common chronic diseases, including type I diabetes mellitus and rheumatoid arthritis (Holick, 2004). Substantial in vitro and toxicologic evidence also support a role for vitamin D activity against the incidence or progression of various cancers (Giovannucci, 2005; Studzinski and Moore, 1995). Large geographical gradients in mortality rates for a number of cancers in the U.S. are not explained by dietary or other risk factors; therefore, it has been hypothesized that some carcinomas are due to insufficient UV-B radiation.

Published literature indicates that solar UV-B radiation, by increasing vitamin D production, is associated with a reduced risk of cancer. Most of these studies used an ecologic study design, in which latitude gradient was examined in relation to cancer rates. Kimlin and Schallhorn (2004) observed that latitude was a valid predictor of vitamin D-producing UV radiation. The strongest evidence exists for an association between UV radiation and reduced risk of colorectal cancer (Giovannucci, 2005; Grant and Garland, 2004; Freedman et al., 2002). Several other studies also have found an inverse relationship between UV radiation and various other cancers, including cancer of the breast (Freedman et al., 2002; Garland et al., 1990; Gorham et al., 1990; Grant, 2002a; John et al., 1999), ovary (Freedman et al., 2002; Lefkowitz and Garland, 1994), and prostate (Freedman et al., 2002; Hanchette and Schwartz, 1992), as well as non-Hodgkin lymphoma (Hughes et al, 2004; Hartge et al., 1996). Eight other cancers (i.e.,

bladder, esophageal, kidney, lung, pancreatic, rectal, stomach, and corpus uteri) have been found to exhibit an inverse correlation between mortality rates and UV-B radiation (Grant, 2002b).

Using UV-B data from July 1992 and U.S. cancer mortality rates from 1970 to 1994, premature cancer deaths attributable to insufficient UV-B exposure were analyzed in an ecologic study (Grant, 2002b). The minimum mortality rate, which was determined as the value corresponding to the maximum UV-B dose, was used to calculate the number of premature deaths. This analysis observed that the annual number of premature deaths from various cancers due to inadequate UV-B exposures was 21,700 (95% CI: 20,400, 23,400) for white Americans; 1,400 (95% CI: 1,100, 1,600) for black Americans; and 500 (95% CI: 400, 600) for Asian Americans and other minorities. Uncertainty in the estimations of UV-B exposure limits the confidence for the estimates of excess cancer deaths attributable to insufficient exposure. Caution is required in interpreting results from ecologic data; however, no strong alternative explanation is indicated in the association observed between UV radiation and the decreased risk of cancer (Giovannucci, 2005). No study has assessed the decreased risk of cancer mortality resulting from increased UV radiation attributable to decreased tropospheric O_3 levels, but the change in risk is expected to be unappreciable.

In establishing guidelines on limits of exposure to UV radiation, the ICNIRP agreed that some low-level exposure to UV radiation has health benefits (ICNIRP, 2004). However, the adverse health effects of higher UV exposures necessitated the development of exposure limits for UV radiation. The ICNIRP recognized the challenge in establishing exposure limits that would achieve a realistic balance between beneficial and adverse health effects.

10.2.4 Summary and Conclusions for Ozone Effects on UV-B Flux

Latitude and altitude are primary variables in defining UV-B flux at the Earth's surface, immediately followed in importance by clouds, surface albedo, particulate matter concentration and composition, and then by gas phase pollution. Of all of these, only latitude and altitude can be defined with small uncertainty in any effort to develop a UV climatology for use in a public health benefits analysis relevant to the areas not presently attaining the NAAQS for O_3. Cloud cover, and its effect on surface UV flux, continues to be extremely difficult to define and predict. Particulate matter and gas-phase tropospheric pollutants are subject to similarly high degrees of uncertainty in predicting their relative concentration distributions. Land cover and,

consequently, surface albedos are highly variable at the geographic scales relevant to NAAQS attainment.

Within the uncertain context of presently available information on UV-B surface fluxes, a risk assessment of UV-B-related health effects would need to factor in human habits (e.g., daily activities, recreation, dress, and skin care) in order to adequately estimate UV-B exposure levels. Little is known about the impact of variability in these human factors on individual exposure to UV radiation. Furthermore, detailed information does not exist regarding the relevant type (e.g., peak or cumulative) and time period (e.g., childhood, lifetime, or current) of exposure, wavelength dependency of biological responses, and interindividual variability in UV resistance. Recent reports of the necessity of UV-B in the production of vitamin D – a vitamin in which many individuals are deficient – suggests that increased risks of human disease due to a slight excess in UV-B radiation exposure may be offset by the benefits of enhanced vitamin D production. However, as with other impacts of UV-B on human health, this beneficial effect of UV-B has not been studied in sufficient detail to allow for a credible health benefits assessment. In conclusion, the effect of changes in surface-level O_3 concentrations on UV-induced health outcomes cannot yet be critically assessed within reasonable uncertainty.

10.3 TROPOSPHERIC OZONE AND CLIMATE CHANGE

Water vapor, CO_2, O_3, N_2O, CH_4, CFCs, and other polyatomic gases present in the Earth's troposphere, trap infrared radiation emitted by the Earth's surface, leading to surface warming. This phenomenon is widely known as the "Greenhouse Effect" (Arrhenius, 1896), and the gases involved are known as "greenhouse gases" (GHGs). The term used for the role a particular atmospheric component, or any other component of the greater climate system, plays in altering the Earth's radiative balance is "forcing." In the past decade, the global atmospheric sciences and climate communities have made significant progress in determining the specific role O_3 plays in forcing climate.

The Intergovernmental Panel on Climate Change (IPCC) was founded in 1988 by the World Meteorological Society (WMO) and the United Nations Environmental Program (UNEP) to support the work of the Conference of Parties (COP) to the United Nations Framework Convention on Climate Change (UNFCCC). Drawing from the global climate and atmospheric

sciences community for its authors and reviewers, the IPCC produces reports containing thorough assessments of the available peer-reviewed science regarding the physical climate system, past and present climate, and evidence of human-induced climate change. This section will summarize the reviews of the available information on the forcing properties of tropospheric O_3 as provided by the IPCC Third Assessment Report (IPCC, 2001a), and will also describe some of the more recent developments on the subject.

The projected effects of global climate change will be briefly explained to provide the context within which O_3 serves as a regional, and possibly global, anthropogenic pollutant. The concept of climate forcing is also explained, along with the factors that influence the extent of climate forcing by O_3. The section concludes with a summary of the various estimates that have been placed on the amount of globally averaged forcing due to O_3.

10.3.1 The Projected Impacts of Global Climate Change

The study of the atmospheric processes involved in global climate change, and its potential consequences for human health and global ecosystems, is an area of active research. The IPCC Third Assessment Report (TAR) is the most thorough evaluation available of the science concerning climate change. In addition to the first and second IPCC assessments in 1990 and 1995, along with other IPCC reports, earlier assessments included those conducted by the UNEP (1986), the WMO (1988), the U.S. Environmental Protection Agency (1987), and others (e.g., Patz et al., 2000a,b). The reader is referred to those documents for a complete discussion of climate change science. An abbreviated list of the IPCC conclusions to date and a short discussion of the potential impacts of climate change on human health and welfare is provided here to serve as the context for the discussion of the role of the increasing tropospheric O_3 concentration in climate change.

According to various historic and modern measurement records, atmospheric GHG concentrations have increased dramatically in the past century, and have been attributed to human activities. The IPCC TAR describes the scientific theory and evidence linking increases in GHGs to human activities (IPCC, 2001a).

An increasing body of geophysical observations shows that the Earth is warming and that other climate changes are underway. These observations include the global surface temperature record assembled since the year 1860, the satellite temperature record begun in 1979, recorded

changes in snow and ice cover since the 1950s, sea level measurements taken throughout the 20th century, and sea surface temperature observations recorded since the 1950s.

Observations (Levitus et al., 2005) show that ~84% of the total heating of the Earth System (oceans, atmosphere, continents, and cryosphere) over the last 40 years has gone into warming the oceans**.** Barnett et al. (2005) have reported the emergence of a clear pattern of ocean surface warming associated with anthropogenic GHGs. The authors constructed a model-based fingerprint (i.e., a map of predicted changes in the vertical temperature profile of the Earth's six major oceans), and compared this map to the newly upgraded and expanded ocean temperature data set (Levitus et al., 2005)**.** They concluded that the warming signal far exceeds what would be expected from natural variability, a finding that was in compelling agreement with GHG-forced model profiles. Other evidence of ocean warming includes a marked increase in the frequency, intensity, and persistence of the zonal atmospheric circulation shifts known as the El Niño-Southern Oscillation (ENSO) over the past 100 years. ENSO events occur when the tropical Pacific Ocean has accumulated a large, localized mass of warm water that interrupts cold surface currents along South America, altering precipitation and temperature patterns in the tropics, subtropics, and the midlatitudes.

IPCC (1998, 2001a) reports also describe the results of general circulation model (GCM) studies predicting that human activities will alter the climate system in a manner likely to lead to marked global and regional changes in temperature, precipitation, and other climate properties. These changes are expected to increase the global mean sea level as well as increase the number of extreme weather events such as floods and droughts, increased wind speeds and precipitation intensity of tropical cyclones, and changes in soil moisture. These predicted changes can be expected to directly impact human health, ecosystems, and global economic sectors (e.g., hydrology and water resources, food and fiber production) (IPCC, 1998, 2001b). Table 10-1 summarizes these projected impacts.

Wide variations in the course and net impacts of climate change in different geographic areas are expected. In general, the projected changes constitute additional stressors on natural ecosystems and human societal systems already impacted by increasing resource demands, unsustainable resource management practices, and pollution. Some of the predicted changes include alterations in ecological balances; in the availability of adequate food, water, clean air;

Table 10-1. Examples of Impacts Resulting From Projected Changes in Extreme Climate Events

Projected changes during the 21st Century in Extreme Climate Phenomena and their Likelihood[a]	**Representative Examples of Projected Impacts[b]** ***(all high confidence of occurrence in some areas[c])***
Simple Extremes	
Higher maximum temperatures; more hot days and heat waves[d] over nearly all land areas (*very likely*[a])	• Increased incidence of death and serious illness in older age groups and urban poor • Increased heat stress in livestock and wildlife • Shift in tourist destinations • Increased risk of damage to a number of crops • Increased electric cooling demand and reduced energy supply reliability
Higher (increasing) minimum temperatures; fewer cold days, frost days, and cold waves[d] over nearly all land areas (*very likely*[a])	• Decreased cold-related human morbidity and mortality • Decreased risk of damage to a number of crops, and increased risk to others • Extended range and activity of some pest and disease vectors • Reduced heating energy demand
More intense precipitation events (*very likely*[a] over many years)	• Increased flood, landslide, avalanche, and mudslide damage • Increased soil erosion • Increased flood runoff could increase recharge of some floodplain aquifers • Increased pressure on government and private flood insurance systems and disaster relief
Complex Extremes	
Increased summer drying over most midlatitude continental interiors and associated risk of drought (*likely*[a])	• Decreased crop yields • Increased damage to building foundations caused by ground shrinkage • Decreased water resource quantity and quality • Increased risk of forest fire
Increase in tropical cyclone peak wind intensities, mean and peak precipitation intensities (*likely*[a] over some areas)[e]	• Increased risk to human life, risk of infections, disease epidemics, and many other risks • Increased coastal erosion and damage to coastal buildings and infrastructure • Increased damage to coastal ecosystems such as coral reefs and mangroves
Intensified droughts and floods associated with El Niño events in many different regions (*likely*[a]) (see also under droughts and intense precipitation events)	• Decreased agricultural and rangeland productivity in drought- and flood-prone regions • Decreased hydropower potential in drought-prone regions
Increased Asian summer monsoon precipitation variability (*likely*[a])	• Increased flood and drought magnitude and damages in temperate and tropical Asia

Table 10-1 (cont'd). Examples of Impacts Resulting From Projected Changes in Extreme Climate Events

Projected changes during the 21st Century in Extreme Climate Phenomena and their Likelihood[a]	Representative Examples of Projected Impacts[b] *(all high confidence of occurrence in some areas[c])*
Complex Extremes (cont'd)	
Increased intensity of midlatitude storms (little agreement between current models)[d]	• Increased risks to human life and health • Increased property and infrastructure losses • Increased damage to coastal ecosystems

[a]Likelihood refers to judgmental estimates of confidence used by TAR WGI: *very likely* (90-99% chance); *likely* (66-90% chance). Unless otherwise stated, information on climate phenomena is taken from the Summary for Policymakers, TAR WGI. TAR WGI = Third Assessment Report of Working Group 1 (IPCC, 2001a).
[b]These impacts can be lessened by appropriate response measures.
[c]High confidence refers to probabilities between 67 and 95%.
[d]Information from TAR WGI, Technical Summary.
[e]Changes in regional distribution of tropical cyclones are possible but have not been established.

Source: IPCC (2001b).

and in human health and safety. Poorer nations can be expected to suffer the most, given their limited adaptive capabilities.

Although many regions are predicted to experience severe, possibly irreversible, adverse effects due to climate change, beneficial changes may also take place. For example, certain regions may benefit from warmer temperatures or increased CO_2 fertilization, e.g., U.S. West Coast coniferous forests, and some Western rangelands. Specific benefits may include reduced energy costs for heating, reduced road salting and snow-clearance costs, longer open-water seasons in northern channels and ports, and improved agricultural opportunities in the northern latitudes as well as in the Western interior and coastal areas. For further details about the projected effects of climate change on a U.S.-regional scale, the reader is also referred to several regionally-focused reports (MARAT, 2000; Yarnal et al., 2000; NERAG, 2001; GLRAG, 2000), as well as a report on potential impacts of climate change on human health (Bernard et al., 2001a,b). The IPCC report, "The Regional Impacts of Climate Change," (IPCC, 1998) describes the projected effects of human-induced climate change on various regions of the globe including Africa, the Arctic and Antarctic, the Middle East and arid Asia, Australasia, Europe, Latin America, North America, the small island nations, temperate Asia, and tropical Asia.

While current climate models can successfully simulate the present-day annual mean global climate and the seasonal cycles on a continental scale, they have been less successful on a regional scale. Clouds and humidity, essential factors in defining local and regional (sub-grid scale) climate, are significantly uncertain (IPCC, 2001a). Due to modeling uncertainties, both in reproducing regional climate and in predicting the future economic activity that will govern future GHG emissions, the projected impacts discussed above are also uncertain.

Findings from the U.S. Global Change Research Program (USGCRP) (NAST, 2000) and related reports illustrate the considerable uncertainties and difficulties in projecting likely climate change impacts at the regional or local scale. The USGCRP findings also reflect the mixed nature of projected potential climate change impacts, i.e., combinations of deleterious and beneficial effects, for U.S. regions and the variation of projected impacts across different regions. Difficulties in projecting region-specific climate change impacts are complicated by the need to evaluate the potential effects of regional- or local-scale changes in key air pollutants not only on global-scale temperature trends, but also on regional- or local-scale temperature and precipitation patterns. The EPA is currently leading a research effort that uses regional-scale climate models with the goal of identifying changes to O_3 and PM concentrations that may occur in a warming climate. An assessment of the results of this effort will be available by the next review of the O_3 NAAQS. This focused effort to determine the impact of a warming climate on criteria air pollution requires regional-scale models with improved skill in reproducing climate history and predicting change. Among the innovations being employed in this effort is the downscaling of global circulation model outputs to provide boundary conditions for model calculations at the regional scale (Liang et al., 2005). While focusing on projecting the impact of a warming climate on regional O_3 concentrations, the effort applied to improving regional-scale modeling will also lead to improved estimates of current and projected future impacts of tropospheric O_3 on climate.

10.3.2 Solar Energy Transformation and the Components of the Earth's Climate System

Mass, in any form, has the capacity to interact with solar and terrestrial radiation, but the manner in which it interacts with radiation is governed by its particular physical form and/or molecular properties. Water provides one of the most interesting examples of how physical form

affects radiative properties. In its gaseous form, water is the most important GHG present in the climate system, due to its ability to absorb long-wave terrestrial radiation. Conversely, in its frozen form as snow or sea ice, water plays a very important role in the climate system by scattering UV and visible solar radiation back to space, i.e., decreasing the Earth's net solar radiation receipts by increasing the Earth's reflective properties (albedo). In its liquid aerosol form as clouds, water also greatly increases the Earth's albedo. In its bulk liquid form as ocean water, it absorbs terrestrial radiation, and represents the Earth's most important reservoir of heat energy.

The atomic composition and molecular structure of a gas determines the wavelengths of light it can absorb and, therefore, its role in defining the heat capacity of the atmosphere. Ozone and O_2 provide examples of the relative importance of these molecular properties. While these molecules are both composed solely of oxygen atoms, their bond structures are distinct. Ozone has a three-atom, bent molecular structure, giving it the capacity to absorb terrestrial (infrared) wavelengths – making it a GHG. At any altitude, i.e., in the stratosphere or troposphere, O_3 has the capacity to absorb UV radiation of 320 nm and shorter, further increasing the energy-absorbing capacity of the troposphere. Conversely, O_2, due to its diatomic, linear structure, is limited to absorbing very short-wave UV light – and does so at altitudes too high to influence the climate system significantly.

Each component of the climate system plays a role in absorbing, transforming, storing, dispersing, or scattering solar radiation. Weather is a tangible consequence of the transformation and dispersion of terrestrial radiation within the atmosphere. The term "weather" refers to the condition of the Earth's atmosphere at a specific time and place. It is defined by several specific variables: the air temperature, air pressure, humidity, clouds, precipitation, visibility, and wind speed. The "climate" for a given place on the Earth's surface is a long-term average of these variables accounting for daily and seasonal weather events. The frequency of extreme weather events is used to distinguish among climates that have similar averages (Ahrens, 1994).

Climate components, including GHGs, land, oceans, sea ice, land ice and snow, atmospheric particles, vegetation, clouds, etc., all contribute to the Earth's heat capacity, i.e., its ability to absorb and retain solar energy. Changes in the properties (or mass) of these components will "force" the climate system in one direction or the other, i.e., warmer versus cooler. The transformation of atmospheric O_2 into O_3 by way of air pollution chemistry,

enhances the heat capacity of the atmosphere. The principles behind the important concept of climate forcing are further described, below.

10.3.3 The Composition of the Atmosphere and the Earth's Radiative Equilibrium

The Greenhouse Effect is the term given to the decreased rate of reemission of absorbed solar energy due to the heat-retaining properties of the Earth's atmosphere. According to simple radiative transfer theory, at thermal equilibrium, the Earth's temperature should be near –15 °C. This is the temperature of a theoretical "black body" that is receiving and then reemitting 342.5 Wm^{-2}, i.e., the globally averaged amount of full-spectrum solar energy absorbed and then reemitted by the Earth as infrared terrestrial radiation per square meter. In fact, satellite observations well above the atmosphere indicate that the Earth's average *planetary* temperature is remarkably close to its theoretical black body value at –18 °C, a temperature at which liquid water ordinarily does not exist.

At its *surface*, however, the Earth's average temperature is +15 °C. The +33 °C temperature differential between the Earth's planetary and surface temperatures is due to the presence of infrared (IR) radiation-absorbing components in the atmosphere such as water vapor, CO_2, CH_4, several other trace gases, and some types of particles and clouds.

The atmosphere, when cloud-free, is largely transparent in the solar wavelength range. A small fraction of this radiation is absorbed and reemitted as black body radiation by dark atmospheric particles (IPCC, 2001a). However, the majority of clouds and particles, in part, offset the greenhouse effect by increasing the Earth's albedo, thereby decreasing the overall amount of solar radiation absorbed by the Earth system.

Ozone, SO_2 and NO_2 also absorb ultraviolet and near ultraviolet wavelengths, in addition to infrared radiation. Once absorbed by a gas molecule, the energy introduced by a photon may induce a photochemical reaction with the residual energy thermally exciting (heating) the products of the reaction. Alternatively, the energy introduced into the molecule by the photon may be dispersed amongst neighboring molecules via intermolecular collisions, or reemitted in part as a lower energy (i.e., IR) photon.

Radiation from the sun or the Earth's surface that is absorbed by gases and particles is reemitted isotropically, i.e., it is equally likely to be emitted in all directions. Therefore, to a

first approximation, half of the radiation trapped by the Earth's atmosphere is reflected back to its surface. A portion of this radiation is transformed into the heat energy that drives the atmospheric processes that form the basis of weather and climate. Radiation that is not absorbed by gases and aerosols reaches the Earth's surface where it is scattered (reflected) or absorbed, depending on the surface albedo.

Successful modeling of the Earth's climate and, therefore, the assessment of the extent of human-induced climate change and development of appropriate policy depend on the quality of available information on the relative efficiencies, amounts, and spatial and temporal distributions of the various radiatively active components of the atmosphere that absorb and/or reflect solar and terrestrial radiation, along with all the other nonatmospheric components of the Earth system.

10.3.3.1 Forcing of the Earth's Radiative Balance

As mentioned earlier, the commonly used measure of the relative influence of a given component of the climate system on the Earth's radiative balance is its radiative forcing (IPCC, 2001a; Houghton et al., 1990). Radiative forcing, in Wm^{-2}, is a quantity that was developed by the climate modeling community as a first-order-only means of estimating relative effects of individual anthropogenic and natural processes on the energy balance within the climate system.

When the effect of a particular component of the climate system is to reduce the amount of solar energy absorbed, usually by increasing the Earth's albedo, this component is said to provide a "negative" forcing, measured in Wm^{-2}. The convention assigns a positive value to the forcing induced by climate system components that enhance the Greenhouse Effect or otherwise act to increase the heat-absorbing capacity of the Earth system. Purely reflective atmospheric aerosol, clouds, white rooftops, snow-covered land surfaces, and dense sea ice provide a negative forcing, while highly absorbing dark-colored atmospheric aerosols, GHGs, and increases dark ocean surface area, due to the melting of sea ice sheets, positively force the climate system.

Global and regional climate are roughly defined by the balance between the large number of positive and negative forcings induced by the many different components of the Earth system and any changes in the properties of those components due to natural processes or anthropogenic activities. Following a perturbation or added forcing, such as an increase in GHG concentrations

or modification the Earth's albedo through changes in land use, this balance is re-established via a complex redistribution of energy within the Earth system. Feedback mechanisms that are theorized but difficult to resolve at the quantitative level further complicate the prediction of the sensitivity of variables, such as surface temperature, to changes in forcing.

A simple example of a positive feedback would be melting sea ice. As sea ice melts with increasing ocean temperatures, the dark ocean surface that is revealed is more efficient at absorbing IR radiation, further increasing the rate of warming. A negative feedback would be the formation of clouds over a moist, warming surface. As clouds form, less radiation is available to warm the surface, leading to cooling. The role of feedbacks in determining the sensitivity of climate to changes in radiative forcing is described in detail in the IPCC TAR (IPCC, 2001a).

Discussions are presently underway within the climate community regarding a metric to replace forcing as the standard measure of climate impact – one that will account for more of the factors that determine the effectiveness of a specific change in altering climate. However, forcing remains the current standard (NRC, 2005).

The IPCC has reported estimated values for forcing by individual radiatively active gases, and by particle-phase components of the atmosphere that were derived primarily through expert judgment incorporating the results of peer-reviewed modeling studies. The forcing estimates, shown in Figure 10-6, are global averages attributed to known GHGs, including O_3, particles, anthropogenic cirrus clouds, land-use change, and natural solar flux variations. Uncertainty ranges are assigned to reflect the range of modeled values reported in those studies. The current estimate of forcing due to long-lived, well-mixed, GHGs accumulated in the atmosphere from the preindustrial era (ca., 1750) through the year 2000 is +2.4 Wm^{-2} ± 10% (IPCC, 2001a). An indication of the level of confidence in each of these estimates is given along the bottom of this figure, again reflecting the expert judgment of the IPCC.

The IPCC reported a global average forcing value of 0.35 ± 0.15 Wm^{-2} for tropospheric O_3, based on model calculations constrained by climatological observations. The considerations and studies used to estimate this value will be outlined below. Hansen and Sato (2001), accounting for uncertainties in pre-industrial emissions levels, more recently estimated a value of 0.5 ± 0.2 Wm^{-2} for forcing by O_3.

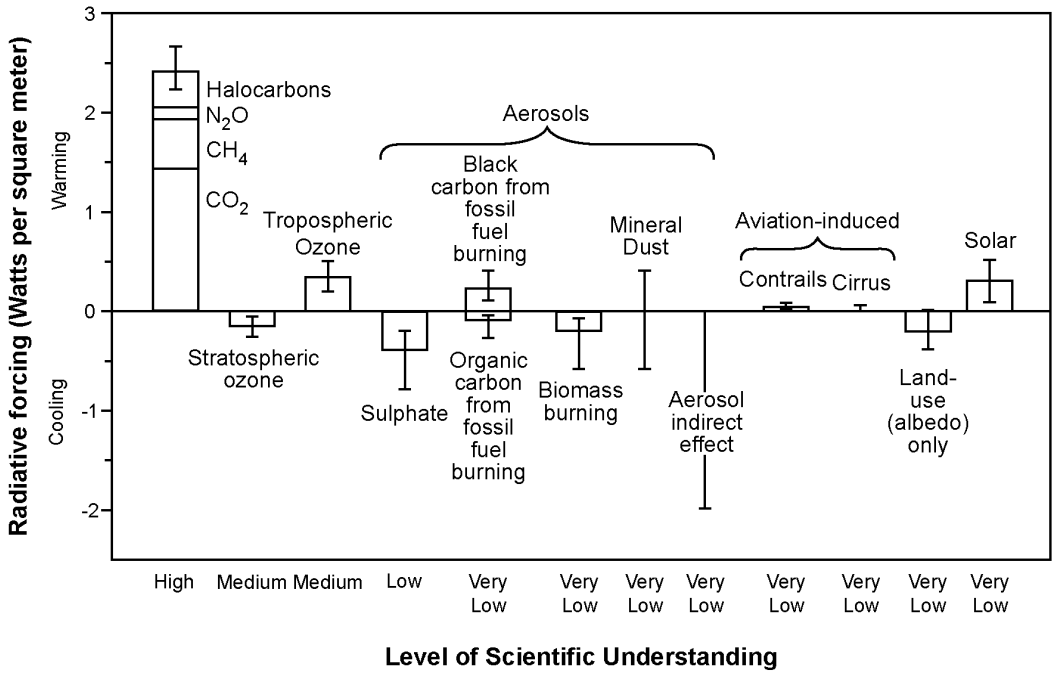


Figure 10-6. Estimated global mean radiative forcing exerted by gas and various particle phase species for the year 2000, relative to 1750.

Source: IPCC (2001a).

10.3.4 Factors Affecting the Magnitude of Climate Forcing by Ozone

The radiative properties of O_3 are distinct from those of other important GHGs in that it is capable of absorbing both UV and IR radiation. Furthermore, it is able to absorb long-wave radiation in a portion of the IR spectrum where water vapor does not absorb, i.e., the 9 to10 m wavelength range, meaning that its ability to trap heat and force climate are unchanged by variations in humidity. Given its relatively short atmospheric lifetime in comparison to other GHGs, the distribution of tropospheric O_3 is highly variable in geographic extent and time. These properties enhance the prospect of attributing a unique geographic and time-dependent pattern or fingerprint to forcing by O_3.

Due to human activities, tropospheric O_3 is estimated to have provided the third largest increase in direct forcing since preindustrial times. It may also play a role in indirect forcing through its participation in the oxidative removal of other radiatively active trace gases, such as CH_4 and the HCFCs. The direct and indirect forcing that tropospheric O_3 imposes on the climate

system depends upon its geospatial and temporal distribution, but it also depends upon the albedo of the underlying surface and its vertical position (altitude) in the atmosphere.

10.3.4.1 The Global Burden of Tropospheric Ozone

Little historical data exist that may be used to estimate the global O_3 burden prior to industrialization, although a few late 19th-century measurements suggest that O_3 has more than doubled in Europe during the 20th century. The insufficient data record on preindustrial tropospheric O_3 distributions introduces a major uncertainty in the estimation of the change in O_3-induced forcing since that period (IPCC, 2001a; Mickley et al., 2004a; Mickley et al., 2001; Shindell and Faluvegi, 2002).

Ozone reacts photochemically at time scales that are generally shorter than those for large-scale mixing processes in the atmosphere. Concentrated O_3 plumes evolve downwind of strong sources of its precursor pollutants: reactive nitrogen, CO, and non-methane hydrocarbons (NMHCs). The most important of these sources are midlatitude industrialized areas and tropical biomass burning. When viewed from above the atmosphere by satellite-borne spectrometers, O_3 enhancements appear as relatively localized air masses or regional-scale plumes, usually originating from industrialized areas or areas in which active biomass burning is underway. The IPCC (2001a) describes the efforts of several research teams who have analyzed data supplied by the satellite-borne Total Ozone Mapping Spectrometer (TOMS) and other remote-sensing instruments to map the global distribution of tropospheric O_3 and to attempt to identify processes that influence the global tropospheric O_3 budget (IPCC, 2001a). More recently, coincident observations of total O_3 by TOMS and the Solar Backscattered UV (SBUV) instrument were used by Fishman et al. (2003) to construct well-resolved spatial and temporal maps of the regional distribution of tropospheric O_3. Their results were consistent with those reported by others, but with higher regional-scale resolution. They reported large O_3 enhancements in the southern tropics in austral spring, and in the northern temperate latitudes in the summer. The regional nature of high O_3 concentrations was clearly visible in northeastern India, the eastern United States, eastern China, and west and southern Africa, each coincident with high population densities. Fishman et al. (2003) noted, as have the other groups cited above, significant interannual variability in the concentrations observed over these regions. *In situ* measurements of tropospheric O_3 concentrations range from 10 ppb over remote oceans to 100 ppb in both the

upper troposphere and in plumes downwind from polluted metropolitan regions (IPPC, 2001a). Ground-level concentrations in urban areas are often >100 ppb. In the Southern Hemisphere, one of the largest sources of O_3 precursors is biomass burning. Biomass burning elevates O_3 on large spatial scales, particularly in the tropical Atlantic west of the coast of Africa and in Indonesia.

In its third assessment report, the IPPC estimates placed the global burden of tropospheric O_3 at a highly uncertain 370 Tg, equivalent to an average column density of 34 Dobson Units (1 DU = 2.687×10^{16} molecules/cm^{-2}) or a mean concentration of 50 ppb (IPCC, 2001a). Accounting for differences in levels of industrialization between the hemispheres, the average column burden in the Northern Hemisphere is estimated to be 36 DU, with the Southern Hemisphere estimated to average 32 DU. Due to its rapid photochemistry, individual surface measurements of tropospheric O_3 cannot capture large-scale concentrations, nor will they represent the higher altitude concentrations. Dense surface and vertical measurements (ozonesondes) would be required to supplement available output from remote sensing instruments to provide the complete set of observations needed to derive a credible global O_3 budget. Such a measurement program appears, at present, to be impractical.

10.3.4.2 Background Concentrations versus Regionally-Oriented Ozone Enhancements

Vingarzan (2004) surveyed the air quality literature and reported that annual average background O_3 concentrations at ground level in the Northern Hemisphere appear to range between 20 and 45 ppb, depending upon geographic location, elevation, and the influence of local sources. Fiore et al. (2003) modeled the U.S. continental O_3 concentrations and found that surface background levels overlap the lower end of the range reported by Vingarzan (2004), e.g., 15 to 35 ppb, with higher levels (40 to 50 ppb) arising at high-elevation sites due to the influence of the upper troposphere (See Chapter 3 and its associated annexes for a complete discussion of "policy relevant background [PRB]). Local- and regional-scale enhancements in O_3 may be thought of as roughly superimposed upon these background levels, with the exception of longer stagnation events in which preexisting background O_3 reacts away or is deposited as fresh O_3 is produced from local precursors.

Lin et al. (2001) analyzed the EPA AIRS database for the 1980-1998 period and noted that O_3 concentrations have declined at the high end of the probability distribution, consistent

with the effects of emissions controls, but had increased at the low end of the distribution by 3-5 ppb. They divided the monitoring data for the continental U.S. into 4 quadrants by geography, and noted a pattern of increase for the Western states that might be attributed to the long-range transport of O_3 precursors from Asia. They found, however, that the Northeastern quadrant had the highest increase in the low end of the concentration probability distribution, which could not be reasonably attributed to transboundary transport of O_3 precursors.

While not representing an ideal source of information for assessing the climatic effects of O_3 within the continental United States, data from the large air-quality-focused ground-based monitoring network may be used to identify boundary-layer geospatial and temporal patterns in O_3 concentrations for comparison to regional-scale chemistry/climate models. Extensive analysis of data available within the EPA AQS database can be found in Chapter 3 of this document, including an analysis showing the diurnal O_3 concentration patterns for several large metropolitan areas with peak values ranging up to160 ppb (Los Angeles). Lehman et al. (2004) analyzed the AQS database of daily 8-h maximum O_3 concentrations collected for 1,090 stations in the eastern half of the United States for the 1993 to 2002 period. They applied a rotated principle component analysis to a reasonably complete, spatially representative, nonurban subset of the database in order to identify coherent, regionally oriented patterns in O_3 concentrations. Five spatially homogenous regions were identified: the U.S. Northeast, Great Lakes, Mid-Atlantic, Southwest (including Alabama, Louisiana, Texas, Oklahoma), and Florida. The Mid-Atlantic region displayed the highest mean concentration (52 ppb) of all of the regions analyzed, with the Great Lakes, Southwest, and Northeast regions following with around 47 ppb. The average concentration derived for Florida was 41 ppb. The authors found strong correlations in measured concentrations among stations within the same region, suggesting that the geospatial patterns of pollutant emissions and meteorological activity may also have a regional orientation. These results that these regions may define natural domains for regional scale modeling studies of the influence of O_3 (as well as PM) on climate.

10.3.4.3 Ozone Trends: Globally and in North America

For the Northern Hemisphere, weekly continuous data are available from 1970 for only nine stations in the latitude range 36° N to 59° N (IPCC, 2001a). Available tropospheric O_3 measurements do not reveal a clear trend in concentration, while trends in the stratosphere are

more readily identified. Different trends are seen at different locations for different periods, consistent with regional changes in pollutant emission, especially NO_x. Logan et al. (1999) analyzed the composite record of mid-tropospheric O_3 abundance from the nine-station network. A plot of data is shown in Figure 10-7. While no clear trend appeared for 1980 through 1996, the average level for second half of this record (about 57 ppb) is clearly greater than for the first half (about 53 ppb). The trend may be consistent with changes in regional NO_x emission rates occurring due to pollution reduction efforts in developed countries and increasing emissions in rapidly growing economies in Asia. The measurements shown in Figure 10-7 are for surface concentrations only. Fewer locations have measured changes in the concentrations of O_3 as a function of altitude. Fewer still are locations that have collected and maintained data records prior to 1970. The absence of historical data on the vertical distribution of O_3 adds to the difficulty in estimating historical atmospheric burdens and trends in O_3-related climate forcing.

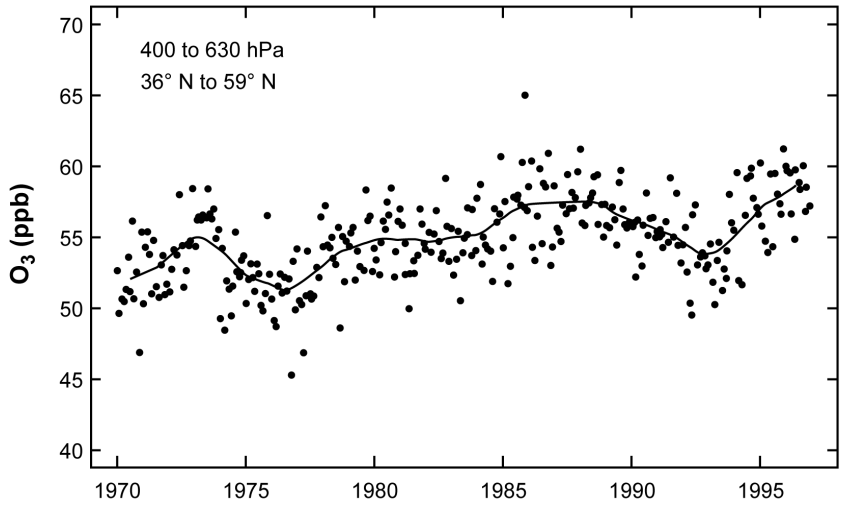


Figure 10-7. Mid-tropospheric O_3 abundance (ppb) in northern midlatitudes (36 °N-59 °N) for the years 1970 to 1996. Observations between 630 and 400 hPa are averaged from nine ozonesonde stations (four in North America, three in Europe, two in Japan), following the data analysis of Logan et al. (1999). Values are derived from the residuals of the trend fit, with the trend added back to allow for discontinuities in the instruments. Monthly data (points) are shown with a smoothed 12-month mean (line).

Source: IPCC (2001a).

The IPCC (2001a) surveyed the results of published chemistry transport model (CTM) modelling studies (see Table 10-2) that estimated the global average increases in total column O_3 since the preindustrial era. Model estimates ranged from +7 to +12 DU. On the basis of these estimates, available measurements, and other analyses, the IPCC estimated that total column O_3 has increased by 9 DU with a 67% confidence range of +6 to +13 DU. In some of the modelling studies, emissions scenarios predicted a further increase in column O_3 due to growing emissions of O_3 precursors. Fusco and Logan (2003) stated that, according to models, increased NO_x emissions from fossil fuel combustion have had the greatest effect on O_3 in the lower troposphere since the 1970s. In addition, increases in background CH_4 have also contributed as much as 20% to the increase in tropospheric O_3, in the northern latitudes. Given its longer atmospheric residence time, CH_4 can serve as an O_3 precursor at much longer distances from its source than can other O_3 precursors, and, therefore, has a more uniform effect across the globe.

Table 10-2. CTM Studies Assessed by the IPCC for its Estimate of the Change in Global and Total Column O_3 Since the Preindustrial Era

Estimated Change in Column O_3 in DU	Model Used	References
7.9	*GFDL*	Haywood et al. (1998)
8.9	*MOZART-1*	Hauglustaine et al. (1998)
8.4	*NCAR/2D*	Kiehl et al. (1999)
9.5	*GFDL-scaled*	Levy et al. (1997)
12	*Harvard/GISS*	Mickley et al. (1999)
7.2	*ECHAM4*	Roelofs et al. (1997)
8.7	*UKMO*	Stevenson et al. (2000)
9.6	*UIO*	Berntsen et al. (1999)
8	*MOGUNTIA*	VanDorland et al. (1997)

Source: IPCC (2001a)

Fusco and Logan (2003) found a 10% increase in O_3 concentrations year-round over Canada, Europe, and Japan and a 20% increase for Japan and Europe during spring and summer. It was expected — but not found — that O_3 concentrations over Japan would increase in line

with emissions from China. The authors suggested that convective activity over Asia is stronger than that seen over other industrialized areas of the global. Such a meteorological characteristic would result in an injection of pollutants into the free troposphere, allowing long-range transport to North America. Their suggestion is supported by evidence of increasing background concentrations within the United States (Fiore et al., 1998).

NARSTO (2000), in its assessment of the available information on O_3 pollution in North America, stated that no single pattern for trends in O_3 over North America can be found in the available monitoring data. In the United States, the average 1-h concentration at surface monitoring sites decreased by 15% between 1986 and 1996, with most of the observed declines occurring in urban and urban-influenced locations. The largest declines occurred in Los Angeles, New York, and Chicago. Free tropospheric O_3 concentrations appeared to hold steady, or only declined slightly, from the 1980s, forward.

In preparation for the IPCC TAR (IPCC, 2001a), research groups engaged in modeling global-scale tropospheric chemistry were invited to participate in a model intercomparison focusing on potential changes in the oxidative capacity of the atmosphere (OxComp), which included O_3 concentrations, for the 2000 to 2100 period. Participating groups employed the IPCC A2p scenario, i.e., including the highest emissions levels, to calculate the geospatial distribution of O_3 up to 20 km. The predicted spatial distributions of O_3 where quite variable, but the predictions for total column O_3 density change fell within 9 DU of each other (11.4 to 20.5 DU) in all cases and that was considered to be encouraging by the authors. Fusco and Logan (2003) pointed out that several unresolved issues may limit the ability of models in reproducing observed trends in tropospheric O_3. Among these are the use of different meteorological inputs, photochemical reaction schemes, and predicted cloud cover — each contributing to different predictions in O_3 production and loss rates.

10.3.4.4 The Sensitivity of Ozone-Related Forcing Surface to Albedo

The characteristics of the surface underlying an O_3 enhancement play a role in the O_3 forcing effect. Highly reflective surfaces, such as light-colored deserts, sea ice and snow, scatter solar short wave (UV and visible) radiation. UV and visible radiation can then be absorbed, transformed into long-wave radiation, and reemitted in part back to the surface by tropospheric O_3. Studies by two groups, Hauglustaine et al. (1998) and Mickley et al. (1999), have shown

that industrial pollution that has been transported to the Arctic induces a high, regional O_3-related forcing due to the highly reflective underlying ice and snow surface.

Liao et al. (2004) calculated that the maximum change in O_3-related top-of-the-atmosphere forcing occurs over high albedo regions in high northern latitudes. Surface forcing was calculated to be greatest at high northern latitudes as well as at dust-source regions, which also tend to have high surface albedos.

10.3.4.5 The Altitude Dependence of Forcing by Tropospheric Ozone

Altitude plays an important role in the forcing effect of tropospheric O_3 (IPCC, 1992; Gauss et al., 2003). The efficiency of IR absorption by O_3 depends upon its temperature – at atmospheric temperatures that are low, relative to the Earth's surface, it has the capacity to absorb more IR radiation than O_3 at temperatures close to that of the surface. While this temperature effect applies to all GHGs, it introduces a complication for estimating forcing by O_3, because O_3 is not homogeneously mixed within the troposphere. Ozone forcing estimates must account for these difficult-to-predict vertical inhomogeneities. However, as part of the OxComp modeling intercomparison, Gauss et al. (2003) found that the overall forcing by O_3 can be calculated within reasonable uncertainty simply on the basis of total column density.

10.3.4.6 Co-occurrence of Ozone with Particulate Matter

Analysis of the 2001 data from the AQS database showed infrequent co-occurrence of high $PM_{2.5}$ and O_3 concentrations (Chapter 3 of this document). For those cases when O_3 production is high, in combination with high PM concentrations, there is a suggestion in the literature that heterogeneous chemistry on PM surfaces may lead to reduced gas-phase O_3. Liao et al. (2004) modeled heterogeneous chemistry taking place on PM, and found a significant titration of O_3 and its NO_x precursors. The importance of this titration effect remains an open question, given the difficulty in obtaining in situ measurements to validate model calculations.

Liao et al. (2004) also estimated that forcing by BC, mineral dust, and organic carbon aerosols substantially offsets forcing by tropospheric O_3, yielding an overall negative globally averaged forcing at both the top of the atmosphere and at the Earth's surface. However, such estimates neglect the regional aspects of forcing by these individual pollutants. Elevated concentrations of these very different types of pollutants often appear independently of the

others, such as with biomass burning plumes, Saharan dust, and organic aerosols associated with biogenic terpene emissions by forests. It is unlikely that a global average of the forcing effects of these individual pollutants will adequately capture their impacts on climate at the regional scale.

10.3.5 Estimated Forcing by Tropospheric Ozone

10.3.5.1 Direct Climate Forcing Due to Ozone

The inhomogeneous distribution of O_3 within the troposphere, coupled with the large uncertainty in the global O_3 budget, significantly complicates the matter of estimating the global average direct forcing due to O_3. The IPCC TAR (2001a) lists the results of several modeling studies that estimated the annual change in the relative forcing by O_3 from preindustrial times. It was noted that the differences among the estimates were most likely due to differences in predicted O_3 chemistry, including the emissions inventories used and the chemical process and transport mechanisms incorporated into the models, rather than by factors relating to radiative transfer. The IPCC intercomparison of the models and their results indicate that the uncertainties in estimated forcings due to O_3 have decreased since the IPCC Second Assessment Report (1996).

The O_3-related forcings estimated by studies considered by the IPCC (2001a) are listed in Table 10-3. Ten of the listed estimates are based on global CTM calculations. One study was constrained by a climatology derived from observations. Given the differences in calculated total column O_3 among the models, a normalized forcing (Wm^{-2} per Dobson Unit of tropospheric O_3 change) is listed in addition to the absolute forcing (Wm^{-2}) estimated by each model. Both clear sky (cloud-free) and total sky (including clouds) forcing estimates are listed.

The largest O_3-related forcings coincide with the strongest sources of tropospheric O_3, which the models predict occur in the northern midlatitude regions (40° to 50° N) and reach as much as 1 Wm^{-2} in the summer as well as in the tropics, and are related to biomass burning. In general, the estimates are comparable in magnitude and show similarity in geographic distribution. For total sky conditions, the range in globally and annually averaged tropospheric O_3 forcing from all of these models is from 0.28 to 0.43 Wm^{-2}, while the normalized forcing is 0.033 to 0.056 Wm^{-2} per DU. As expected, they are opposite in sign to the forcing estimated for sulfate aerosols, which scatter radiation. The range in normalized

Table 10-3. Tropospheric O_3 Change (O_3) in Dobson Units (DU) Since Preindustrial Times, and the Accompanying Net (SW plus LW) Radiative Forcings (Wm^{-2}), After Accounting for Stratospheric Temperature Adjustment (using the Fixed Dynamical Heating Method). Estimates are Taken From the Published Literature. Normalized Forcings (norm.) Refer to Radiative Forcing per O_3 Change (Wm^{-2} per DU)

Estimated Global Average Forcing Due to Tropospheric Ozone

		Clear sky conditions		Total sky conditions	
Reference	**ΔO_3**	**Net**	**Net (norm.)**	**Net**	**Net (norm.)**
Berntsen et al. (1997) – [Reading model]	7.600	0.310	0.041	0.280	0.037
Stevenson et al. (1998)	8.700	0.391	0.045	0.289	0.033
Berntsen et al. (1997) – [Oslo model]	7.600	0.390	0.051	0.310	0.041
Haywood et al. (1998)	7.900	0.380	0.048	0.310	0.039
Kiehl et al. (1999)	8.400	0.379	0.045	0.320	0.038
Berntsen et al. (2000)	9.600	0.428	0.045	0.342	0.036
Brasseur et al. (1998)	—	——	—	0.370	—
van Dorland et al. (1997)	8.070	0.443	0.055	0.380	0.047
Roelofs et al. (1997)	7.200	0.397	0.055	0.404	0.056
Lelieveld and Dentener (2000)	—	—	—	0.420	—
Hauglustaine et al. (1998)	8.940	0.511	0.057	0.426	0.048
Mean	8.224	0.403	0.049	0.343	0.042

Source: IPCC (2001a).

forcings emphasizes the differences in assumptions used by the different models. The tropospheric O_3 forcing constrained by the observational climatology is 0.32 Wm^{-2} for globally averaged, total sky conditions. As shown in Figure 10-6, the IPCC (2001a) concluded that 0.35 ± 0.15 Wm^{-2} represents the most likely value for annually and globally-averaged forcing by tropospheric O_3. Not included here is the study by Hansen and Sato (2001), that evaluated forcing by O_3 with corrections made to the assumptions concerning pre-industrial O_3 concentrations and the effects of natural O_3 precursors, especially NOx generated by lightning. Hansen and Sato (2001) concluded that a more likely range for globally averaged forcing by O_3 is 0.4 to 0.8 Wm^{-2}, with 0.5 Wm^{-2} as their best estimate.

Since the publication of the IPCC TAR (2001a), new studies have been published that illuminate some of the regionally-relevant details associated with direct forcing by O_3 (Mickley et al., 2004a; Liao et al., 2004). Forcing by O_3, due to its capacity for absorbing solar UV as well as solar and terrestrial IR radiation, can be divided into "shortwave" forcing and "long-wave" forcing. These forcings occur under different conditions. Shortwave forcing can only take place during daytime, while long-wave forcing can occur at all hours as a function of the diurnally varying concentration of atmospheric O_3. As noted, earlier, unlike CO_2, the absorption spectrum for O_3 is distinct from that of water vapor — meaning that O_3 will absorb and reemit long-wave radiation without interference by water under high humidity conditions. Mickley et al. (2004a) reported that, according to their modeling study, surface temperature response to the predicted O_3 enhancement since the preindustrial period differs greatly from that of the CO_2 response, and that this difference can only be explained by the geographical distribution and absorption properties of O_3. Liao et al. (2004) estimated globally averaged top-of-the-atmosphere separate short- and long-wave forcings to O_3 to be 0.21 W/m^2 and 0.32 W/m^2, respectively.

10.3.5.2 Indirect Forcing Due to Ozone

Ozone has an indirect climate forcing effect due to its role in the oxidative removal of other reactive GHGs, including CH_4, hydrofluorocarbons (HFCs), and other reactive NMHCs. The primary actor in this effect is a second generation product of the photolysis of O_3, the hydroxyl (OH) radical. Hydroxyl radicals are produced by way of a pair of reactions that start with the photodissociation of O_3 by solar UV.

$$O_3 + h\nu \rightarrow O(^1D) + O_2 \qquad (10\text{-}5)$$

$$O(^1D) + H_2O \rightarrow OH + OH \qquad (10\text{-}6)$$

Reactions with OH are the primary removal mechanism for CH_4 and NMHCs as well as the pollutants NO_x and CO. Methane and CO are in especially high abundance in the global atmosphere. OH is estimated to react with these two gases within 1 second of its formation.

In addition to CH_4, NO_x, CO, and the NMHCs, OH concentrations are controlled by local concentrations of H_2O (i.e., humidity) and the intensity of solar UV. Different atmospheric concentrations of the required precursors suggest that preindustrial OH concentrations were likely to have been different from present-day concentrations, but there is no consensus on the magnitude of this difference. Observations of global atmospheric concentrations of methylchloroform (CH_3CCl_3), a well-mixed tropospheric species that also reacts with OH, have been used to estimate OH abundances. Independent studies have shown overlapping trends for the period 1978 to 1994, but none of the trends are outside the given uncertainty ranges (0.5 ± 0.6%/year) (Prinn et al., 1995; Krol et al., 1998). The IPCC (2001a) reported a range of +5% to −20% for predicted changes in global OH abundances.

Given the difficulty in estimating global OH abundances in the past, present, and future, estimates of indirect forcing due to O_3 have been difficult to obtain and are highly uncertain.

10.3.5.3 Predictions for Future Climate Forcing by Anthropogenic Ozone

The rate of increase in surface O_3 in Europe and North America since 1980 appears to be slowing, likely due to control measures intended to improve urban air quality. Not surprisingly, CTM modeling attempts to predict future precursor emissions and resulting O_3 abundances indicate that the largest future O_3-related forcings will be related to population growth and economic development in Asia (van Dorland et al., 1997; Brasseur et al., l998). The results of these modeling studies predict that the globally averaged total radiative forcing due to O_3 from preindustrial times 0.66 Wm^{-2} will rise to 0.63 Wm^{-2} by 2050. Chalita et al. (1996) predicted a change in the globally averaged radiative forcing from preindustrial times to 2050 of 0.43 Wm^{-2}. Stevenson et al. (l998) predicted an O_3-related forcing of 0.48 Wm^{-2} in 2100. Applying the SRES scenario projecting the highest emissions out to the year 2100 (IPCC, 2000), the OxComp model intercomparison study yielded a projected O_3-induced forcing ranging from 0.40 to 0.78 Wm^{-2}. The authors concluded, given their prediction for forcing by well-mixed GHGs of 5.6 Wm^{-2}, that O_3 would remain an important contributor to overall anthropogenic forcing well into the future. However, all of these predictions must be viewed with caution given the considerable uncertainties associated with the long-term economic activity projections required for such estimates.

10.3.6 The Impact of a Warming Climate on Atmospheric Ozone Concentrations

Evaluation of the potential impact of climate warming on U.S. air quality is currently underway. Initial modeling results reported by Mickley et al. (2004b) suggest that reduced cyclone frequency in a warmer climate will lead to increases in the severity of summertime pollution episodes. Cyclonic weather patterns are known to play an important role in ventilating pollution away from the surface. They note that compelling evidence is accumulating that the frequency of these cyclones has decreased over the past few decades. An early study by Jacob et al. (1993) found a correlation between O_3 concentrations and temperature was due to the effect of O_3 on atmospheric chemistry, biogenic emissions, and stagnation.

10.3.7 Conclusion

The general consensus within the atmospheric sciences community, as represented by the United Nations Intergovernmental Panel on Climate Change (IPCC), is that human activities have a discernable effect on the Earth's climate. However, quantifying the extent of human-induced forcing on climate requires detailed information about human-induced change on the components of the Earth System that govern climate. Tropospheric O_3 is a well-known GHG, but information regarding its historical trends in concentration, its current and future atmospheric burden, and other critical details needed for estimating its direct and indirect forcing effects on the climate system are highly uncertain.

The IPCC has estimated that the globally averaged forcing due to O_3 is approximately 0.35 ± 0.15 Wm^{-2}, with an updated value of 0.5 ± 0.2 Wm^{-2} provided by Hansen and Sato (2001). However, the most important role of O_3 in climate is likely to be at the regional scale, adjacent to the sources of its chemical precursors. This expectation is consistent with satellite observations of high regional scale column O_3 densities near large urban areas and large-scale biomass burning activity. Modeling studies evaluated by the IPCC have estimated that regional-scale forcing due to O_3 can approach 1 Wm^{-2}, or as much as 40% of the globally averaged forcing due to the well-mixed GHGs. While more certain estimates of the overall importance of global-scale forcing due to tropospheric O_3 await further advances in monitoring and chemical transport modeling, the overall body of scientific evidence suggests that high concentrations of O_3 on the regional scale could have a discernable influence on climate, leading to surface

temperature and hydrological cycle changes. Confirming this effect requires improvement in regional-scale modeling — an activity that is currently underway.

REFERENCES

Ahrens, D. C. (1994) Meteorology today: an introduction to weather, climate and the environment. Minneapolis, MN: West Publishing Co., p. 17.

Airey, D. K.; Wong, J. C.; Fleming, R. A.; Meldrum, L. R. (1997) An estimate of the UV-B exposure for outdoor workers during a south-east Queensland summer. Health Phys. 72: 544-549.

Ancellet, G.; Beekmann, M. (1997) Evidence for changes in the ozone concentrations in the free troposphere over southern France from 1976 to 1995. Atmos. Environ. 31: 2835-2851.

Arrhenius, S. (1896) On the influence of carbonic acid in the air upon the temperature of the ground. Philos. Mag. 41: 237-276.

Autier, P.; Doré, J. F.; Négrier, S.; Liénard, D.; Panizzon, R.; Lejeune, F. J.; Guggisberg, D.; Eggermont, A. M. (1999) Sunscreen use and duration of sun exposure: a double-blind, randomized trial. J. Natl. Cancer Inst. 91: 1304-1309.

Autier, P.; Doré, J.-F.; Reis, A. C.; Grivegnée, A.; Ollivaud, L.; Truchetet, F.; Chamoun, E.; Rotmensz, N.; Severi, G.; Césarini, J. P.; EORTC Melanoma Co-operative Group. (2000) Sunscreen use and intentional exposure to ultraviolet A and B radiation: a double blind randomized trial using personal dosimeters. Br. J. Cancer 83: 1243-1248.

Ayala, M. N.; Michael, R.; Söderberg, P. G. (2000) Influence of exposure time for UV radiation-induced cataract. Invest. Ophthalmol. Visual Sci. 41: 3539-3543.

Barnard, W. F.; Saxena, V. K.; Wenny, B. N.; DeLuisi, J. J. (2003) Daily surface UV exposure and its relationship to surface pollutant measurements. J. Air Waste Manage. Assoc. 53: 237-245.

Barnett, T. P.; Pierce, D. W.; AchutaRao, K. M.; Gleckler, P. J.; Santer, B. D.; Gregory, J. M.; Washington, W. M. (2005) Penetration of human-induced warming into the world's oceans. Science 309: 284-287.

Bellows, C. F.; Belafsky, P.; Fortgang, I. S.; Beech, D. J. (2001) Melanoma in African-Americans: trends in biological behavior and clinical characteristics over two decades. J. Surg. Oncol. 78: 10-16.

Bernard, S. M.; McGeehin, M. G.; Patz, J. A., eds. (2001a) Health: the potential consequences of climate variability and change. Environ. Health Perspect. 109(suppl. 2): 175-233.

Bernard, S. M.; Samet, J. M.; Grambsch, A.; Ebi, K. L.; Romieu, I. (2001b) The potential impacts of climate variability and change on air pollution-related health effects in the United States. Environ. Health Perspect. 109 [suppl. 2]: 199-209.

Berntsen, T. K.; Isaksen, I. S. A. (1999) Effects of lightning and convection on changes in tropospheric ozone due to NO_x emissions from aircraft. Tellus 51B: 766-788.

Berntsen, T. K.; Isaksen, I. S. A.; Myhre, G.; Fuglestvedt, J. S.; Stordal, F.; Larsen, T. A.; Freckleton, R. S.; Shine, K. P. (1997) Effects of anthropogenic emissions on tropospheric ozone and its radiative forcing. J. Geophys. Res. [Atmos.] 102: 28,101-28,126.

Berntsen, T. K.; Myhre, G.; Stordal, F.; Isaksen, I. S. A. (2000) Time evolution of tropospheric ozone and its radiative forcing. J. Geophys. Res. [Atmos.] 105: 8915-8930.

Berret, J.; Liardet, S.; Scaletta, C.; Panizzon, R.; Hohlfeld, P.; Applegate, L. A. (2002) Use of sunscreens in families living in Switzerland. Dermatology 204: 202-208.

Black, W. C.; Goldhahn, R. T., Jr.; Wiggins, C. (1987) Melanoma within a southwestern Hispanic population. Arch. Dermatol. 123: 1331-1334.

Brasseur, G. P.; Kiehl, J. T.; Müller, J.-F.; Schneider, T.; Granier, C.; Tie, X.-X.; Hauglustaine, D. (1998) Past and future changes in global tropospheric ozone: impact on radiative forcing. Geophys. Res. Lett. 25: 3807-3810.

Breuer-McHam, J.; Simpson, E.; Dougherty, I.; Bonkobara, M.; Ariizumi, K.; Lewis, D. E.; Dawson, D. B.; Duvic, M.; Cruz, P. D., Jr. (2001) Activation of HIV in human skin by ultraviolet B radiation and its inhibition by NFKB blocking agents. Photochem. Photobiol. 74: 805-810.

Brühl, C.; Crutzen, P. J. (1988) On the disproportionate role of tropospheric ozone as a filter against solar UV-B radiation. Geophys. Res. Lett. 16: 703-706.

Caforio, A. L. P.; Fortina, A. B.; piaserico, S.; Alaibac, M.; Tona, F.; Feltrin, G.; Pompei, E.; Testolin, L.; Gambino, A.; Volta, S. D.; Thiene, G.; Casarotto, D.; Peserico, A. (2000) Skin cancer in heart transplant recipients: risk factor analysis and relevance of immunosuppressive therapy. Circulation 102(suppl. III): III-222 - III-227.

Cantorna, M. T. (2000) Vitamin D and autoimmunity: is vitamin D status an environmental factor affecting autoimmune disease prevalence? Proc. Soc. Exp. Biol. Med. 223: 230-233.

Chalita, S.; Hauglustaine, D. A.; Le Treut, H.; Müller, J. F.; Penkett, S., eds. (1996) Radiative forcing due to increased tropospheric ozone concentrations. Atmos. Environ. 30: 1641-1646.

Clydesdale, G. J.; Dandie, G. W.; Muller, H. K. (2001) Ultraviolet light induced injury: immunological and inflammatory effects. Immunol. Cell Biol. 79: 547-568.

Cockell, C. S. (2001) A photobiological history of Earth. In: Cockell, C. S.; Blaustein, A. R., eds. UV radiation flux into ecosystems. New York, NY: Springer-Verlag.
Cutchis, P. (1974) Stratospheric ozone depletion and solar ultraviolet radiation on Earth. Science (Washington, DC) 184: 13-19.
Davis, K. J.; Cokkinides, V. E.; Weinstock, M. A.; O'Connell, M. C.; Wingo, P. A. (2002) Summer sunburn and sun exposure among US youths ages 11 to 18: national prevalence and associated factors. Pediatrics 110: 27-35.
De Fabo, E. C.; Noonan, F. P.; Fears, T.; Merlino, G. (2004) Ultraviolet B but not ultraviolet A radiation initiates melanoma. Cancer Res. 64: 6372-6376.
De Gruijl, F. R. (1995) Action spectrum for photocarcinogenesis. Recent Results Cancer Res. 139: 21-30.
De Gruijl, F. R. (1999) Skin cancer and solar UV radiation. Eur. J. Cancer 35: 2003-2009.
De Gruijl, F. R. (2002) Photocarcinogenesis: UVA vs. UVB radiation. Skin Pharmacol. Appl. Skin Physiol. 15: 316-320.
De Gruijl, F. R.; Henricus, J. C. M.; Sterenborg, H.; Forbes, P. D.; Davies, R. E>; Cole, C.; Kelfkens, G.; Van Weelden, H.; Slaper, H.; Van der Leun, J. C. (1993) Wavelength dependence of skin cancer induction by ultraviolet irradiation of albino hairless mice. Cancer Res. 53: 53-60.
Diepgen, T. L.; Mahler, V. (2002) The epidemiology of skin cancer. Br. J. Dermatol. 146(suppl. 61): 1-6.
Diffey, B. L. (1990) Human exposure to ultraviolet radiation. Semin. Dermatol. 9: 2-10.
Diffey, B. L. (1994) Observed and predicted minimal erythema doses: a comparative study. Photochem. Photobiol. 60: 380-382.
Diffey, B. L.; Gibson, C. J.; Haylock, R.; McKinlay, A. F. (1996) Outdoor ultraviolet exposure of children and adolescents. Br. J. Dermatol. 134: 1030-1034.
Evans, R. D.; Kopf, A. W.; Lew, R. A.; Rigel, D. S.; Bart, R. S.; Friedman, R. J.; Rivers, J. K. (1988) Risk factors for the development of malignant melanoma--I: review of case-control studies. J. Dermatol. Surg. Oncol. 14: 393-408.
Fears, T. R.; Bird, C. C.; Guerry, D., IV; Sagebiel, R. W.; Gail, M. H.; Elder, D. E.; Halpern, A.; Holly, E. A.; Hartge, P.; Tucker M. A. (2002) Average midrange ultraviolet radiation flux and time outdoors predict melanoma risk. Cancer Res. 62: 3992-3996.
Federal Register. (2003) National ambient air quality standards for ozone: final response to remand; final rule. F. R. (January 6) 68: 614-645.
Fiore, A. M.; Jacob, D. J.; Logan, J. A.; Yin, J. H. (1998) Long-term trends in ground level ozone over the contiguous United States, 1980-1995. J. Geophys. Res. (Atmos.) 103: 1471-1480.
Fiore, A.; Jacob, D. J.; Liu, H.; Yantosca, R. M.; Fairlie, T. D.; Li, Q. (2003) Variability in surface ozone background over the United States: implications for air quality policy. J. Geophys. Res. (Atmos.) 108(D24): 10.1029/2003JD003855.
Fishman, J.; Wozniak, A. E.; Creilson, J. K. (2003) Global distribution of tropospheric ozone from satellite measurements using the empirically corrected tropospheric ozone residual technique: identification of the regional aspects of air pollution. Atmos. Chem. Phys. 3: 893-907.
Frederick, J. E.; Koob, A. E.; Weatherhead, E. C. (1993) Empirical studies of tropospheric transmission in the ultraviolet: roadband measurements. J. Appl. Meteorol. 32: 1883-1892.
Freedman, D. M.; Dosemeci, M.; McGlynn, K. (2002) Sunlight and mortality from breast, ovarian, colon, prostate, and non-melanoma skin cancer: a composite death certificate based case-control study. Occup. Environ. Med. 59: 257-262.
Fusco, A. C.; Logan, J. A. (2003) Analysis of 1970-1995 trends in tropospheric ozone at Northern Hemisphere midlatitudes with the GEOS-CHEM model. J. Geophys. Res. (Atmos.) 108(D15): 10.1029/2002JD002742.
Gallagher, R. P.; Hill, G. B.; Bajdik, C. D.; Coldman, A. J.; Fincham, S.; McLean, D. I.; Threlfall, W. J. (1995a) Sunlight exposure, pigmentation factors and risk of nonmelanocytic skin cancer. II. Squamous cell carcinoma. Arch. Dermatol. 131: 164-169.
Gallagher, R. P.; Hill, G. B.; Bajdik, C. D.; Fincham, S.; Coldman, A. J.; McLean, D. I.; Threlfall, W. J. (1995b) Sunlight exposure, pigmentary factors, and risk of nonmelanocytic skin cancer. I. Basal cell carcinoma. Arch. Dermatol. 131: 157-163.
Garland, F. C.; Garland, C. F.; Gorham, E. D.; Young, J. F. (1990) Geographic variation in breast cancer mortality in the United States: a hypothesis involving exposure to solar radiation. Prev. Med. 19: 614-622.
Garssen, J.; Van Loveren, H. (2001) Effects of ultraviolet exposure on the immune system. Crit. Rev. Immunol. 21: 359-397.
Gauss, M.; Myhre, G.; Pitari, G.; Prather, M. J.; Isaksen, I. S. A.; Berntsen, T. K.; Brasseur, G. P.; Dentener, F. J.; Derwent, R. G.; Hauglustaine, D. A.; Horowitz, L. W.; Jacob, D. J.; Johnson, M.; Law, K. S.; Mickley, L. J.;

Müller, J.-F.; Plantevin, P. H.; Pyle, J. A.; Rogers, H. L.; Stevenson, D. S.; Sundet, J. K.; Van Weele, M.; Wild, O. (2003) Radiative forcing in the 21st century due to ozone changes in the troposphere and the lower stratosphere. J. Geophys. Res. (Atmos.) 108(D9): 10.1029/2002JD002624.

Gies, P.; Wright, J. (2003) Measured solar ultraviolet radiation exposures of outdoor workers in Queensland in the building and construction industry. Photochem. Photobiol. 68: 78-83.

Gies, P.; Roy, C.; Toomey, S.; MacLennan, R.; Watson, M. (1998) Solar UVR exposures of primary school children at three locations in Queensland. Photochem. Photobiol. 68: 78-83.

Giovannucci, E. (2005) The epidemiology of vitamin D and cancer incidence and mortality: a review (United States). Cancer Causes Control 16: 83-95.

Gloster, H. M., Jr.; Brodland, D. G. (1996) The epidemiology of skin cancer. Dermatol. Surg. 22: 217-226.

Godar, D. E. (2001) UV doses of American children and adolescents. Photochem. Photobiol. 74: 787-793.

Godar, D. E.; Wengraitis, S. P.; Shreffler, J.; Sliney, D. H. (2001) UV doses of Americans. Photochem. Photobiol. 73: 621-629.

Godar, D. E.; Urbach, F.; Gasparro, F. P.; Van Der Leun, J. C. (2003) UV doses of young adults. Photochem. Photobiol. 77: 453-457.

Gorham, E. D.; Garland, F. C.; Garland, C. F. (1990) Sunlight and breast cancer incidence in the USSR. Int. J. Epidemiol. 19: 820-824.

Granstein, R. D.; Matsui, M. S. (2004) UV radiation-induced immunosuppression and skin cancer. Cutis 74(suppl. 5): 4-9.

Grant, W. B. (2002a) An ecologic study of dietary and solar ultraviolet-B links to breast carcinoma mortality rates. Cancer 94: 272-281.

Grant, W. B. (2002b) An estimate of premature cancer mortality in the U.S. due to inadequate doses of solar ultraviolet-B radiation. Cancer 94: 1867-1875.

Grant, W. B.; Garland, C. F. (2004) A critical review of studies on vitamin D in relation to colorectal cancer. Nutr. Cancer 48: 115-123.

Great Lakes Regional Assessment Group (GLRAG). (2000) Preparing for a changing climate: the potential consequences of climate variability and change. Great Lakes overview. Washington, DC: U.S. Environmental Protection Agency; Office of Research and Development; U.S. Global Change Research Program (USGCRP) and Ann Arbor, MI: University of Michigan; Atmospheric, Oceanic and Space Sciences Department. Available: http://www.geo.msu.edu/glra/assessment/assessment.html [17 April 2002].

Green, A.; Battistutta, D.; Hart, V.; Leslie, D.; Weedon, D. (1996) Skin cancer in a subtropical Australian population: incidence and lack of association with occupation. The Nambour Study Group. Am. J. Epidemiol. 144: 1034-1040.

Green, A.; McCredie, M.; MacKie, R.; Giles, G.; Young, P.; Morton, C.; Jackman, L.; Thursfield, V. (1999) A case-control study of melanomas of the soles and palms (Australia and Scotland). Cancer Causes Control 10: 21-25.

Hall, H. I.; Miller, D. R.; Rogers, J. D.; Bewerse, B. J. (1999) Update on the incidence and mortality from melanoma in the United States. J. Am. Acad. Dermatol. 40: 35-42.

Halliday, G. M.; Byrne, S. N.; Kuchel, J. M.; Poon, T. S.; Barnetson, R. S. (2004) The suppression of immunity by ultraviolet radiation: UVA, nitric oxide and DNA damage. Photochem. Photobiol. Sci. 3: 736-740.

Hanchette, C. L.; Schwartz, G. G. (1992) Geographic patterns of prostate cancer mortality. Cancer 70: 2861-2869.

Hansen, J. E.; Sato, M. (2001) Trends of measured climate forcing agents. Proc. Natl. Acad. Sci. 98: 14778-14783.

Harrison, G. I.; Young, A. R. (2002) Ultraviolet radiation-induced erythema in human skin. Methods (San Diego, CA, U.S.) 28: 14-19.

Hartge, P.; Devesa, S. S.; Grauman, D.; Fears, T. R.; Fraumeni, J. F., Jr. (1996) Non-Hodgkin's lymphoma and sunlight. J. Natl. Cancer Inst. 88: 298-300.

Hartmann, D. L. (1994) Global physical climatology. New York, NY: Academic Press. (International Geophysics: v. 56).

Hauglustaine, D. A.; Brasseur, G. P.; Walters, S.; Rasch, P. J.; Müller, J.-F.; Emmons, L. K.; Carroll, M. A. (1998) MOZART, a global chemical transport model for ozone and related chemical tracers 2. model results and evaluation. J. Geophys. Res. (Atmos.) 103: 28291-28335.

Haywood, J. M.; Schwarzkopf, M. D.; Ramaswamy, V. (1998) Estimates of radiative forcing due to modeled increases in tropospheric ozone. J. Geophys. Res. [Atmos.] 103: 16,999-17,007.

Henriksen, M.; Na, R.; Agren, M. S.; Wulf, H. C. (2004) Minimal erythema dose after umltiple UV exposures depends on pre-exposure skin pigmentation. Photodermatol. Photoimmunol. Photomed. 20: 163-169.

Hildesheim, J.; Fornace, A. J., Jr. (2004) The dark side of light: the damaging effects of UV rays and the protective efforts of MAP kinase signaling in the epidermis. DNA Repair 3: 567-580.
Hockwin, O.; Kojima, M.; Sakamoto, Y.; Wegener, A.; Shui, Y. B.; Sasaki, K. (1999) UV damage to the eye lens: further results from animal model studies: a review. J. Epidemiol. 9(suppl. 6): S39-S47.
Holick, M. F. (2004) Sunlight and vitamin D for bone health and prevention of autoimmune diseases, cancers, and cardiovascular disease. Am. J. Clin. Nutr. 80(6 suppl.): 1678S-1688S.
Houghton, J. T.; Jenkins, G. J.; Ephraums, J. J., eds. (1990) Climate change: the IPCC scientific assessment. Cambridge, MA: Cambridge University Press; p. 55.
Hu, S.; Ma, F.; Collado-Mesa, F.; Kirsner, R. S. (2004) UV radiation, latitude, and melanoma in US Hispanics and blacks. Arch. Dermatol. 140: 819-824.
Hughes, A. M.; Armstrong, B. K.; Vajdic, C. M.; Turner, J.; Grulich, A. E.; Fritschi, L.; Milliken, S.; Kaldor, J.; Benke, G.; Kricker, A. (2004) Sun exposure may protect against non-Hodgkin lymphoma: a case-control study. Int. J. Cancer 112: 865-871.
Ichihashi, M.; Ueda, M.; Budiyanto, A.; Bito, T.; Oka, M.; Fukunaga, M.; Tsuru, K.; Horikawa, T. (2003) UV-induced skin damage. Toxicology 189: 21-39.
Intergovernmental Panel on Climate Change (IPCC). (1992) Climate change 1992 - the supplementary report to the IPCC scientific assessment. Cambridge, United Kingdom: Cambridge University Press.
Intergovernmental Panel on Climate Change. (1996) Climate change 1995: the science of climate change. Summary for policymakers and technical summary of the working group I report. Presented at: the fifth session of the IPCC Working Group I; November; Madrid, Spain. Geneva, Switzerland: World Meteorological Association.
Intergovernmental Panel on Climate Change (IPCC). (1998) The regional impacts of climate change: an assessment of vulnerability, a special report of IPCC Working Group II. Watson, R. T.; Zinyowera, M. C.; Moss, R. H., eds. Cambridge, United Kingdom: Cambridge University Press.
Intergovernmental Panel on Climate Change (IPCC). (2000) Emissions scenarios: a special report of working group III of the Intergovernmental Panel on Climate Change. Cambridge, United Kingdom: Cambridge University Press. Available: http://www.grida.no/climate/ipcc/emission/ [26 August, 2005].
Intergovernmental Panel on Climate Change (IPCC). (2001a) Climate change 2001: the scientific basis. Contribution of working group I to the third assessment report of the Intergovernmental Panel on Climate Change. Cambridge, United Kingdom: Cambridge University Press.
Intergovernmental Panel on Climate Change (IPCC). (2001b) Climate change 2001: impacts, adaptation, and vulnerability. Contribution of working group II to the third assessment report of the Intergovernmental Panel on Climate Change. Cambridge, United Kingdom: Cambridge University Press.
International Agency for Research on Cancer. (1992) Solar and ultraviolet radiation. Lyon, France: International Agency for Research on Cancer. (IARC monographs on the evaluation of carcinogenic risks to humans: v. 55).
International Commission on Non-Ionizing Radiation Protection. (2004) Guidelines on limits of exposure to ultraviolet radiation of wavelengths between 180 nm and 400 nm (incoherent optical radiation). Health Phys. 87: 171-186.
Jacob, D. J.; Logan, J. A.; Yevich, R. M.; Gardner, G. M.; Spivakovsky, C. M.; Wofsy, S. C.; Munger, J. W.; Sillman, S.; Prather, M. J.; Rodgers, M. O.; Westberg, H.; Zimmerman, P. R. (1993) Simulation of summertime ozone over North America. J. Geophys. Res. [Atmos.] 98: 14,797-14,816.
Javitt, J. C.; Taylor, H. R. (1994-95) Cataract and latitude. Doc. Ophthalmol. 88: 307-325.
Jemal, A.; Tiwari, R. C.; Murray, T.; Ghafoor, A.; Samuels, A.; Ward, E.; Feuer, E. J.; Thun, M. J. (2004) Cancer statistics, 2004. CA-Cancer J. Clin. 54: 8-29.
John, E. M.; Schwartz, G. G.; Dreon, D. M.; Koo, J. (1999) Vitamin D and breast cancer risk: the NHANES I Epidemiologic Follow-up Study, 1971-1975 to 1992. Cancer Epidemiol. Biomarkers Prev. 8: 399-406.
Kaidbey, K. H.; Kligman, A. M. (1981) Cumulative effects from repeated exposures to ultraviolet radiation. J. Invest. Dermatol. 76: 352-355.
Kerr, J. B.; McElroy, C. T. (1993) Evidence for large upward trends of ultraviolet-B radiation linked to ozone depletion. Science (Washington, DC) 262: 1032-1034.
Kiehl, J. T.; Schneider, T. L.; Portmann, R. W.; Solomon, S. (1999) Climate forcing due to tropospheric and stratospheric ozone. J. Geophys. Res. [Atmos.] 104: 31,239-31,254.
Kimlin, M. G.; Schallhorn, K. A. (2004) Estimations of the human 'vitamin D' UV exposure in the USA. Photochem. Photobiol. Sci. 3: 1067-1070.

Kimlin, M. G.; Parisi, A. V.; Wong, J. C. (1998a) Quantification of personal solar UV exposure of outdoor workers, indoor workers and adolescents at two locations in Southeast Queensland. Photodermatol. Photoimmunol. Photomed. 14: 7-11.

Kimlin, M. G.; Wong, J. C.; Parisi, A. V. (1998b) Simultaneous comparison of the personal UV exposure of two human groups at different altitudes. Health Phys. 74: 429-434.

Kollias, N.; Stamatas, G. N.; Youn, J. I. (2001) Suppression of UVB-induced cutaneous erythema by a previous UVB exposure. Photochem. Photobiol. 74: 471-476.

Koloutsou-Vakakis, S.; Helmis, C. G.; Assimakopoulos, V.; Güsten, H. (2001) Middle and lower troposphere aerosol characteristics and ozone concentrations over northwestern Greece during STAAARTE 1997. Atmos. Environ. 35: 1517-1526.

Kraemer, K. H. (1997) Sunlight and skin cancer: another link revealed. Proc. Natl. Acad. Sci. U. S. A. 94: 11-14.

Kricker, A.; Armstrong, B. K.; English, D. R. (1994) Sun exposure and non-melanocytic skin cancer. Cancer Causes Control 5: 367-392.

Krol, M.; Van Leeuwen, P. J.; Lelieveld, J. (1998) Global OH trend inferred from methylchloroform measurements. J. Geophys. Res. [Atmos.] 103: 10,697-10,711.

Krotkov, N. A.; Bhartia, P. K.; Herman, J. R.; Fioletov, V.; Kerr, J. (1998) Satellite estimation of spectral surface UV irradiance in the presence of tropospheric aerosols 1. cloud-free case. J. Geophys. Res. (Atmos.) 103: 8779-8793.

Lehman, J.; Swinton, K.; Bortnick, S.; Hamilton, C.; Baldridge, E.; Ender, B.; Cox, B. (2004) Spatio-temporal characterization of tropospheric ozone across the eastern United States. Atmos. Environ. 38: 4357-4369.

Lefkowitz, E. S.; Garland, C. F. (1994) Sunlight, vitamin D, and ovarian cancer mortality rates in U.S. women. Int. J. Epidemiol. 23: 1133-1136.

Lelieveld, J.; Dentener, F. J. (2000) What controls tropospheric ozone? J. Geophys. Res. [Atmos.] 105: 3531-3551.

Lens, M. B.; Dawes, M. (2004) Global perspectives of contemporary epidemiological trends of cutaneous malignant melanoma. Br. J. Dermatol. 150: 179-185.

Levitus, S.; Antonov, J.; Boyer, T. (2005) Warming of the world ocean, 1955-2003. Geophys. Res. Lett. 32(L02604): 10.1029/2004GL021592.

Levy, H., II; Kasibhatla, P. S.; Moxim, W. J.; Klonecki, A. A.; Hirsch, A. I.; Oltmans, S. J.; Chameides, W. L. (1997) The global impact of human activity on tropospheric ozone. Geophys. Res. Lett. 24: 791-794.

Liang, X.-Z.; Pan, J. P.; Zhu, J. H.; Kunkel, K. E.; Dai, A.; Meehl, J. (2005) Regional climate model downscaling of U.S. climate. J. Geophys. Res.: submitted.

Liao, H.; Seinfeld, J. H.; Adams, P. J.; Mickley, L. J. (2004) Global radiative forcing of coupled tropospheric ozone and aerosols in a unified general circulation model. J. Geophys. Res. [Atmos.] 109(D16): 10.1029/2003JD004456.

Lin, C.-Y. C.; Jacob, D. J.; Fiore, A. M. (2001) Trends in exceedances of the ozone air quality standard in the continental United States, 1980-1998. Atmos. Environ. 35: 3217-3228.

Lindelöf, B.; Sigurgeirsson, B.; Gäbel, H.; Stern, R. S. (2000) Incidence of skin cancer in 5356 patients following organ transplantation. Br. J. Dermatol. 143: 513-519.

Logan, J. A. (1999) An analysis of ozonesonde data for the troposphere: recommendations for testing 3-D models and development of a gridded climatology for tropospheric ozone. J. Geophys. Res. 104: 16,115-16,149.

Longstreth, J. D.; De Gruijl; Kripke, M. L.; Takizawa, Y.; Van der Leun, J. C. (1995) Effects of increased solar ultraviolet radiation on human health. Ambio 24: 153-165.

Longstreth, J.; De Gruijl, F. R.; Kripke, M. L.; Abseck, S.; Arnold, F.; Slaper, H. I.; Velders, G.; Takizawa, Y.; Van Der Leun, J. C. (1998) Health risks. J. Photochem. Photobiol. B 46: 20-39.

Lovato, C. Y.; Shoveller, J. A.; Peters, L.; Rivers, J. K. (1998a) Canadian National Survey on Sun Exposure & Protective Behaviours: parent's reports on children. Cancer Prev. Control. 2: 123-128.

Lovato, C. Y.; Shoveller, J. A.; Peters, L.; Rivers, J. K. (1998b) Canadian National Survey on Sun Exposure & Protective Behaviours: youth at leisure. Cancer Prev. Control 2: 117-122.

Lutter, R.; Wolz, C. (1997) UV-B screening by tropospheric ozone: implications for the national ambient air quality standard. Environ. Sci. Technol. 31: 142A-146A.

Madronich, S.; De Gruijl, F. (1993) Skin cancer and UV radiation. Nature (London, UK) 366(6450): 23.

Madronich, S.; De Gruijl, F. (1994) Reply to "Kricker, A.; Armstrong, B. K.; McMichael, A. J. (1994) Skin cancer and ultraviolet. Nature (London, UK) 368: 594." Nature (London, UK) 368(6472): 594.

Matsumura, Y.; Ananthaswamy, H. N. (2004) Toxic effects of ultraviolet radiation on the skin. Toxicol. Appl. Pharmacol. 195: 298-308.

McCarty, C. A.; Nanjan, M. B.; Taylor, H. R. (2000) Attributable risk estimates for cataract to prioritize medical and public health action. Invest. Ophthalmol. Vis. Sci. 41: 3720-3725.
McCarty, C. A.; Taylor, H. R. (2002) A review of the epidemiologic evidence linking ultraviolet radiation and cataracts. Dev. Ophthalmol. 35: 21-31.
McKenzie, R. L.; Seckmeyer, G.; Bais, A. F.; Kerr, J. B.; Madronich, S. (2001) Satellite retrievals of erythemal UV dose compared with ground-based measurements at northern and southern midlatitudes. J. Geophys. Res. [Atmos.] 106: 24,051-24,062.
McKenzie, R.; Smale, D.; Bodeker, G.; Claude, H. (2003) Ozone profile differences between Europe and New Zealand: effects on surface UV irradiance and its estimation from satellite sensors. J. Geophys. Res. [Atmos.] 108(D6): 10.1029/2002JD002770.
McKinlay, A. F.; Diffey, B. L. (1987) A reference action spectrum for ultraviolet induced erythema in human skin. In: Passchier, W. F.; Bosnijakovic, B. F. M., eds. Human exposure to ultraviolet radiation: risks and regulations. Amsterdam, The Netherlands: Elsevier Science Publishers; pp. 83-87.
Merriam, J. C.; Lofgren, S.; Michael, R.; Soderberg, P.; Dillon, J.; Zheng, L.; Ayala, M. (2000) An action spectrum for UV-B radiation and the rat lens. Invest. Ophthalmol. Vis. Sci. 41: 2642-2647.
Mickley, L. J.; Murti, P. P.; Jacob, D. J.; Logan, J. A.; Koch, D. M.; Rind, D. (1999) Radiative forcing from tropospheric ozone calculated with a unified chemistry-climate model. J. Geophys. Res. [Atmos.] 104: 30,153-30,172.
Mickley, L. J.; Jacob, D. J.; Rind, D. (2001) Uncertainty in preindustrial abundance of tropospheric ozone: implications for radiative forcing calculations. J. Geophys. Res. (Atmos.) 106(D4): 3389-3399.
Mickley, L. J.; Jacob, D. J.; Field, B. D.; Rind, D. (2004a) Climate response to the increase in tropospheric ozone since preindustrial times: a comparison between ozone and equivalent CO_2 forcings. J. Geophys. Res. [Atmos.] 109(D5): 10.1029/2003JD003653.
Mickley, L. J.; Jacob, D. J.; Field, B. D.; Rind, D. (2004b) Effects of future climate change on regional air pollution episodes in the United States. Geophys. Res. Lett. 31(L24103): 10.1029/2004GL021216.
Mid-Atlantic Regional Assessment (MARA) Team. (2000) Preparing for a changing climate: the potential consequences of climate variability and change. Mid-Atlantic overview. Washington, DC: U.S. Environmental Protection Agency; Office of Research and Development; U.S. Global Change Research Program (USGCRP) and University Park, PA: Pennsylvania State University. Available: http://www.usgcrp.gov/usgcrp/nacc/midatlantic.htm [17 April 2002].
Moehrle, M. (2001) Ultraviolet exposure in the Ironman triathlon. Med. Sci. Sports Exercise 33: 1385-1386.
Moehrle, M.; Heinrich, L.; Schmid, A.; Garbe, C. (2000) Extreme UV exposure of professional cyclists. Dermatology 201: 44-45.
Moise, A. F.; Buttner, P. G.; Harrison, S. L. (1999) Sun exposure at school. Photochem. Photobiol. 70: 269-274.
National Assessment Synthesis Team (NAST). (2000) Climate change impacts on the United States: the potential consequences of climate variability and change. Overview. Washington, DC: U.S. Global Change Research Program. Available at: http://www.usgcrp.gov/usgcrp/Library/nationalassessment/overview.htm (17 January 2003).
National Research Council. (2005) Radiative forcing of climate change: expanding the concept and addressing uncertainties. Washington, DC: The National Academies Press.
New England Regional Assessment Group (NERAG). (2001) Preparing for a changing climate: the potential consequences of climate variability and change. New England regional overview. Washington, DC: U.S. Environmental Protection Agency; Office of Research and Development; U.S. Global Change Research Program (USGCRP) and Durham, NH: University of New Hampshire; Institute for the Study of Earth, Oceans, and Space. Available: http://www.necci.sr.unh.edu/2001-NERA-report.html [17 April 2002].
Newchurch, M. J.; Ayoub, M. A.; Oltmans, S.; Johnson, B.; Schmidlin, F. J. (2003) Vertical distribution of ozone at four sites in the United States. J. Geophys. Res. 108(D1): 10.1029/2002JD002059.
Nole, G.; Johnson, A. W. (2004) An analysis of cumulative lifetime solar ultraviolet radiation exposure and the benefits of daily sun protection. Dermatol. Ther. 17(suppl. 1): 57-62.
Noonan, F. P.; Muller, H. K.; Fears, T. R.; Kusewitt, D. F.; Johnson, T. M.; De Fabo, E. D. (2003) Mice with genetically determined high susceptibility to ultraviolet (UV)-induced immunosuppression show enhanced UV carcinogenesis. J. Invest. Dermatol. 121: 1175-1181.
North American Research Strategy for Tropospheric Ozone (NARSTO) Synthesis Team. (2000) An assessment of tropospheric ozone pollution: a North American perspective. Palo Alto, CA: Electric Power Research Institute.

Norval, M.; Garssen, J.; Van Loveren, H.; El-Ghorr, A. A. (1999) UV-induced changes in the immune response to microbial infections in human subjects and animal models. J. Epidemiol. 9(suppl. 6): S84-S92.
Okada, S.; Weatherhead, E.; Targoff, I. N.; Wesley, R.; Miller, F. W.; International Myositis Collaborative Study Group. (2003) Global surface ultraviolet radiation intensity may modulate the clinical and imunologic expression of autoimmune muscle disease. Arthritis Rheum. 48: 2285-2293.
Oriowo, O. M.; Cullen, A. P.; Chou, B. R.; Sivak, J. G. (2001) Action spectrum and recovery for in vitro UV-induced cataract using whole lenses. Invest. Ophthalmol. Vis. Sci. 42: 2596-2602.
Parisi, A. V.; Willey, A.; Kimlin, M. G.; Wong, J. C. (1999) Penetration of solar erythemal UV radiation in the shade of two common Australian trees. Health Phys. 76: 682-686.
Patz, J. A.; Engelberg, D.; Last, J. (2000a) The effects of changing weather on public health. Annu. Rev. Public Health 21: 271-307.
Patz, J. A.; McGeehin, M. A.; Bernard, S. M.; Ebi, K. L.; Epstein, P. R.; Grambsch, A.; Gubler, D. J.; Reiter, P.; Romieu, I.; Rose, J. B.; Samet, J. M.; Trtanj, J. (2000b) The potential health impacts of climate variability and change for the United States: executive summary of the report of the health sector of the U.S. national assessment. Environ. Health Perspect. 108: 367-376.
Persson, A. E.; Edstrom, D. W.; Backvall, H.; Lundeberg, J.; Ponten, F.; Ros, A. M.; Williams, C. (2002) The mutagenic effect of ultraviolet-A1 on human skin demonstrated by sequencing the p53 gene in single keratinocytes. Photodermatol. Photoimmunol. Photomed. 18: 287-293.
Pfister, H. (2003) Human papillomavirus and skin cancer. J. Natl. Cancer Inst. Monogr. 31: 52-56.
Pitts, D. G. (1993) Ocular effects of radiant energy. In: Pitts, D. G.; Kleinstein, R. N., eds. Environmental Vision. Stoneham, MA: Butterworth-Heinemann, pp. 151-220.
Ponsonby, A.-L.; McMichael, A.; Van der Mei, I. (2002) Ultraviolet radiation and autoimmune disease: insights from epidemiological research. Toxicology 181/182: 71-78.
Prinn, R. G.; Weiss, R. F.; Miller, B. R.; Huang, J.; Alyea, F. N.; Cunnold, D. M.; Fraser, P. J.; Hartley, D. E.; Simmonds, P. G. (1995) Atmospheric trends and lifetime of CH_3CCl_3 and global OH concentrations. Science (Washington, DC) 269: 187-192.
Rigel, D. S.; Rigel, E. G.; Rigel, A. C. (1999) Effects of altitude and latitude on ambient UVB radiation. J. Am. Acad. Dermatol. 40: 114-116.
Rivers, J. K. (2004) Is there more than one road to melanoma? Lancet 363: 728-730.
Robinson, E. S.; Hill, R. H., Jr.; Kripke, M. L.; Setlow, R. B. (2000) The Monodelphis melanoma model: initial report on large ultraviolet A exposures of suckling young. Photochem. Photobiol. 71: 743-746.
Roelofs, G. J.; Lelieveld, J.; Van Dorland, R. (1997) A three-dimensional chemistry/general circulation model simulation of anthropogenically derived ozone in the troposphere and its radiative climate forcing. J. Geophys. Res. [Atmos.] 102: 23,389-23,401.
Rosenthal, F. S.; Phoon, C.; Bakalian, A. E.; Taylor, H. R. (1988) The ocular dose of ultraviolet radiation to outdoor workers. Invest. Ophthalmol. Vis. Sci. 29: 649-656.
Rozema, J.; Manetas, Y.; Bjorn, L. O., eds. (2001) Responses of plants to UV-B radiation. Dordrecht, The Netherlands: Kluwer Academic. [Advances in Vegetation Science, v. 18].
Rünger, T. M.; Möller, K.; Jung, T.; Dekant, B. (2000) DNA damage formation, DNA repair, and survival after exposure of DNA repair-proficient and nucleotide excision repair-deficient human lymphoblasts to UVA1 and UVB. Int. J. Radiat. Biol. 76: 789-797.
Salby, M. L. (1996) Fundamentals of atmospheric physics. New York, NY: Academic Press. (The International Geophysics Series, Monographs and Textbooks, v. 61).
Sarasin, A. (1999) The molecular pathways of ultraviolet-induced carcinogenesis. Mutat. Res. 428: 5-10.
Sarkar, A. K. (2004) An evaluation of UV protection imparted by cotton fabrics dyed with natural colorants. BMC Dermatol. 4: 15.
Schenker, M. B.; Orenstein, M. R.; Samuels, S. J. (2002) Use of protective equipment among California farmers. Am. J. Ind. Med. 42: 455-464.
Selgrade, M. K.; Repacholi, M. H.; Koren, H. S. (1997) Ultraviolet radiation-induced immune modulation: potential consequences for infectious, allergic, and autoimmune disease. Environ. Health Perspect. 105: 332-334.
Selgrade, M. K.; Smith, M. V.; Oberhelman-Bragg, L. J.; LeVee, G. J.; Koren, H. S.; Cooper, K. D. (2001) Dose response for UV-induced immune suppression in people of color: differences based on erythemal reactivity rather than skin pigmentation. Photochem. Photobiol. 74: 88-95.
Setlow, R. B.; Grist, E.; Thompson, K.; Woodhead, A. D. (1993) Wavelengths effective in induction of malignant melanoma. Proc. Natl. Acad. Sci. U. S. A. 90: 6666.6670.

Shindell, D. T.; Faluvegi, G. (2002) An exploration of ozone changes and their radiative forcing prior to the chlorofluorocarbon era. Atmos. Chem. Phys. 2: 363-374.
Shoveller, J. A.; Lovato, C. Y.; Peters, L.; Rivers, J. K. (1998) Canadian National Survey on Sun Exposure & Protective Behaviours: adults at leisure. Cancer Prev. Control 2: 111-116.
Slaper, H.; Velders, G. J. M.; Daniel, J. S.; de Gruijl, F. R.; Van der Leun, J. C. (1996) Estimates of ozone depletion and skin cancer incidence to examine the Vienna Convention achievements. Nature (London, U.K.) 384: 256-258.
Solanki, S. K.; Fligge, M. (2000) Reconstruction of past solar irradiance. Space Sci. Rev. 94: 127-138.
Stern, R. S.; Liebman, E. J.; Vakeva, L. (1998) Oral psoralen and ultraviolet-A light (PUVA) treatment of psoriasis and persistent risk of nonmelanoma skin cancer. PUVA Follow-up Study. J. Natl. Cancer Inst. 90: 1278-1284.
Stevenson, D. S.; Johnson, C. E.; Collins, W. J.; Derwent, R. G.; Shine, K. P.; Edwards, J. M. (1998) Evolution of tropospheric ozone radiative forcing. Geophys. Res. Lett. 25: 3819-3822.
Stevenson, D. S.; Johnson, C. E.; Collins, W. J.; Derwent, R. G.; Edwards, J. M. (2000) Future estimates of tropospheric ozone radiative forcing and methane turnover - the impact of climate change. Geophys. Res. Lett. 27: 2073-2076.
Studzinski, G. P.; Moore, D. C. (1995) Sunlight--can it prevent as well as cause cancer? Cancer Res. 55: 4014-4022.
Swetter, S. M. (2003) Dermatological perspectives of malignant melanoma. Surg. Clin. N. Am. 83: 77-95.
Taylor, H. R.; West, S. K.; Rosenthal, F. S.; Munoz, B.; Newland, H. S.; Abbey, H.; Emmett, E. A. (1988) Effect of ultraviolet radiation on cataract formation. N. Engl. J. Med. 319: 1429-1433.
Thieden, E.; Agren, M. S.; Wulf, H. C. (2001) Solar UVR exposures of indoor workers in a working and a holiday period assessed by personal dosimeters and sun exposure diaries. Photodermatol. Photoimmunol. Photomed. 17: 249-255.
Thieden, E.; Philipsen, P. A.; Heydenreich, J.; Wulf, H. C. (2004a) UV radiation exposure related to age, sex, occupation, and sun behavior based on time-stamped personal dosimeter readings. Arch. Dermatol. 140: 197-203.
Thieden, E.; Philipsen, P. A.; Sandby-Moller, J.; Heydenreich, J.; Wulf, H. C. (2004b) Proportion of lifetime UV dose received by children, teenagers and adults based on time-stamped personal dosimetry. J. Invest. Dermatol. 123: 1147-1150.
Tomescu, D.; Kavanagh, G.; Ha, T.; Campbell, H.; Melton, D. W. (2001) Nucleotide excision repair gene *XPD* polymorphisms and genetic predisposition to melanoma. Carcinogenesis 22: 403-408.
Ullrich, S. E. (2005) Mechanisms underlying UV-induced immune suppression. Mutat. Res. 571: 185-205.
U.S. Environmental Protection Agency. (1987) Assessing the risks of trace gases that can modify the stratosphere, v. 1-8. Washington, DC: Office of Air and Radiation; report no. EPA 400/1-87/001A-H.
U.S. Environmental Protection Agency. (2004) Air quality criteria for particulate matter. Research Triangle Park, NC: National Center for Environmental Assessment; report no. EPA/600/P-99/002aF-bF. 2v. Available: http://cfpub.epa.gov/ncea/ [9 November, 2004].
United Nations Environment Programme (UNEP). (1986) Report of the international conference on the assessment of the role of carbon dioxide and of other greenhouse gases in climate variations and associated impacts; October 1985; Villach, Austria. Geneva, Switzerland: World Meteorological Organization; WMO no. 661.
Urbach, F. (1997) Ultraviolet radiation and skin cancer of humans. J. Photochem. Photobiol. B 40: 3-7.
Van der Leun, J. C.; De Gruijl, F. R. (1993) Influences of ozone depletion on human and animal health. In: Tevini, M., ed. UV-B radiation and ozone depletion: Effects on humans, animals, plants, microorganisms, and materials. Ann Arbor: Lewis Publisher; pp. 95-123.
Van Dorland, R.; Dentener, F. J.; Lelieveld, J. (1997) Radiative forcing due to tropospheric ozone and sulfate aerosols. J. Geophys. Res. [Atmos.] 102: 28,079-28,100.
Veierød, M. B.; Weiderpass, E.; Thörn, M.; Hansson, J.; Lund, E.; Armstrong, B.; Adami, H.-O. (2003) A prospective study of pigmentation, sun exposure, and risk of cutaneous malignant melanoma in women. J. Natl. Cancer Inst. 95: 1530-1538.
Vingarzan, R. (2004) A review of surface ozone background levels and trends. Atmos. Environ. 38: 3431-3442.
Vishvakarman, D.; Wong, J. C.; Boreham, B. W. (2001) Annual occupational exposure to ultraviolet radiation in central Queensland. Health Phys. 81: 536-544.
Vitasa, B. C.; Taylor, H. R.; Strickland, P. T.; Rosenthal, F. S.; West, S.; Abbey, H.; Ng, S. K.; Munoz, B.; Emmett, E. A. (1990) Association on nonmelanoma skin cancer and actinic keratosis with cumulative solar ultraviolet exposure in Maryland watermen. Cancer 65: 2811-2817.
Waterston, K.; Naysmith, L.; Rees. J. L. (2004) Physiological variation in the erythemal response to ultraviolet radiation and photoadaptation. J. Invest. Dermatol. 123: 958-964.

Wayne, R. P. (2000) Chemistry of Atmospheres: an introduction to the chemistry of the atmospheres of Earth, the planets, and their satellites. 3rd ed. New York, NY: Oxford University Press, Inc.

Wei, Q.; Lee, J. E.; Gershenwald, J. E.; Ross, M. I.; Mansfield, P. F.; Strom, S. S.; Wang, L. E.; Guo, Z.; Qiao, Y.; Amos, C. I.; Spitz, M. R.; Duvic, M. (2003) Repair of UV light-induced DNA damage and risk of cutaneous malignant melanoma. J. Natl. Cancer Inst. 95: 308-315.

Wendisch, M.; Mayer, B. (2003) Vertical distribution of spectral solar irradiance in the cloudless sky: a case study. Geophys. Res. Lett. 30: 10.1029/2002GL016529.

Westerdahl, J.; Ingvar, C.; Måsbäck, A.; Jonsson, N.; Olsson, H. (2000) Risk of cutaneous malignant melanoma in relation to use of sunbeds: further evidence for UV-A carcinogenicity. Br. J. Cancer 82: 1593-1599.

Whiteman, D. C.; Valery, P.; McWhirter, W.; Green, A. C. (1997) Risk factors for childhood malanoma in Queensland, Australia. Int. J. Cancer 70: 26-31.

Whiteman, D. C.; Whiteman, C. A.; Green, A. C. (2001) Childhood sun exposure as a risk factor for melanoma: a systematic review of epidemiologic studies. Cancer Causes Control 12: 69-82.

Whiteman, D. C.; Watt, P.; Purdie, D. M.; Hughes, M. C.; Hayward, N. K.; Green, A. C. (2003) Melanocytic nevi, solar keratoses, and divergent pathways to cutaneous melanoma. J. Natl. Cancer Inst.

Wong, J. C.; Airey, D. K.; Fleming, R. A. (1996) Annual reduction of solar UV exposure to the facial aea of outdoor workers in southeast Queensland by wearing a hat. Photodermatol. Photoimmunol. Photomed. 12: 131-135.

World Meteorological Organization. (1988) Developing policies for responding to climatic change: a summary of the discussions and recommendations of workshops; September-October 1987; Villach, Austria; and November 1987; Bellagio, Austria. Geneva, Switzerland: World Meteorological Organization; report no. WMO/TD; no. 225. [World Climate Impact Programme series report no. WCIP-1].

World Meteorological Organization (WMO). (1999) Scientific assessment of ozone depletion: 1998. Geneva, Switzerland: World Meteorological Organization, Global Ozone and Monitoring Project; report no. 44.

World Meteorological Organization (WMO). (2002) Scientific assessment of ozone depletion: 2002. Geneva, Switzerland: United Nations Environment Programme. [Global Ozone and Monitoring Project; report no. 47].

Xenopoulos, M. A.; Schindler, D. W. (2001) Physical factors determining ultraviolet radiation flux into ecosystems. In: Cockell, C.; Blaustein, A. R. Ecosystems, evolution, and ultraviolet radiation. New York, NY: Springer; pp. 36-62.

Yarnal, B.; Kalkstein, L. S.; Scheraga, J. D. (2000) Mid-Atlantic regional assessment of climate change impacts. Clim. Res. (CR Special 7) 14: 153-269.

Yoshikawa, T.; Rae, V.; Bruins-Slot, W.; Van den Berg, J. W.; Taylor, J. R.; Streilein, J. W. (1990) Susceptibility to effects of UVB radiation on induction of contact hypersensitivity as a risk factor for skin cancer in humans. J. Invest. Dermatol. 95: 530-536.

11. EFFECT OF OZONE ON MAN-MADE MATERIALS

Ozone (O_3) and other photochemical oxidants react with many economically important man-made materials, decreasing their useful life and aesthetic appearance. Some materials known to be damaged by ozone include elastomers, fibers, dyes, and paints. This chapter provides a brief discussion of O_3 effects on man-made materials, including denoting of damage mechanisms and, where possible, concentration-response relationships. Much of what is known about ozone effects on man-made materials is derived from research conducted in the 1970's, 1980's, and early 1990's, with very little new research on the subject having been conducted since then. Since only very limited new information has been published on effects of ozone on materials, this chapter mainly summarizes key information assessed in the previous 1996 Air Quality Criteria Document for Ozone and other photochemical oxidants (1996 O_3 AQCD) (U.S. Environmental Protection Agency, 1996) and provides detailed discussion of the very limited new information that has become available since then. In the ensuing sections, discussion is focused on ozone effects on: elastomers (Sect 11.1); textiles and fabrics (11.2); dyes, pigments, and inks (11.3); artist's pigments (11.4); and surface coatings (11.5). Evaluation of the relevance and economic importance of O_3 materials damage information, as it affects productivity or cultural resources (such as museums), is beyond the scope of this chapter. The reader is referred to the previous criteria document (1996 O_3 AQCD) for a more detailed discussion of the earlier studies summarized below.

11.1 ELASTOMERS

The elastomeric compounds, natural rubber and synthetic polymers and copolymers of butadiene, isoprene, and styrene, are particularly susceptible to even low levels of ozone. Elastomeric compounds are long chain unsaturated organic molecules. Ozone damages these compounds by breaking the molecular chain at the carbon-carbon double bond; a chain of three oxygen atoms is added directly across the double bond, forming a five-membered ring structure (Mueller and Stickney, 1970). The change in structure promotes the characteristic cracking of stressed/stretched rubber called "weathering." A 5% tensile strain will produce cracks on the

surface of the rubber that increase in number with increased stress/stretching. The rate of crack growth is dependent on the degree of stress, the type of rubber compound, concentration, time of exposure, velocity, and temperature (Bradley and Haagen-Smit, 1951; Lake and Mente, 1992) (Gent and McGrath, 1965). Once cracking occurs, there is further penetration, additional cracking, and eventually mechanical weakening or stress relaxation (U.S. Environmental Protection Agency, 1996). Razumovskii et al. (1988) demonstrated the effect of ozone on stress relaxation of polyisoprene vulcanizates. A decrease in stress (stress relaxation) is caused by ozone-induced cracks in exposed elastomers resulting in irreversible changes in the elastomer dimensions and decreased tensile strength.

To counteract ozone effects on elastomers, antiozonants and wax are often added to the elastomeric formulations during processing. An antiozonant is an additive used to protect a polymer against the effects of ozone-induced degradation and, hence, is used mainly in diene rubbers. Antiozonant protection works either (a) by providing a physical barrier to ozone penetration via forming a thin surface film of an ozone-resisting wax or (b) by chemically reacting with ozone or polymer ozonolysis products, as do aromatic diamines such as p-phenylene diamine derivatives. The antiozonant diffuses to the surface of the elastomeric material, where it reacts with ozone faster than ozone reacts to break the molecular chain and the carbon-carbon double bond, or the antiozonant diffuses to the surface of the material but is not reactive with ozone and serves as a protective coating against ozone attack. The antiozonant may also serve to scavenge ozone while also providing protective film against ozone attack (Andries et al., 1979; Lattimer et al., 1984).

Most of the studies on ozone effects on elastomers were designed to evaluate the effectiveness of antiozonants in counteracting the rubber cracking produced by ozone exposure. Consequently, many of the studies were conducted using ozone concentrations higher than those typically found in the ambient air. Natural rubber strips exposed to high concentrations of ozone (20,000 ppm) under stressed conditions cracked almost instantaneously and were broken within 1 sec. When the ozone concentration was lowered (0.02 to 0.46 ppm), the time to required to produce cracks in the exposed rubber material was increased (Bradley and Haagen-Smit, 1951). Lake and Mente (1992) studied the effect of temperature on ozone-induced elastomer cracking and antiozonant protection on natural rubber, epoxidised natural rubber, and two acrylonitrile-butadiene copolymers under constant strain. Temperatures ranged from -20 °C to +70 °C. The

elastomers were exposed to 0.05 to 1,000 ppm ozone for 16 h. Ozone cracking decreased at lower ambient temperatures; however, diffusing of both chemical and wax antiozonants also were slowed at the lower temperatures. Cracking was slightly increased at the higher temperatures but the antiozonants offered more protection.

Serrano et al. (1993) evaluated the appropriateness of using ozone-induced elastomer cracking to estimate ambient ozone concentrations. Two vulcanized natural rubber compounds were exposed for 24 h to varying ozone concentrations under stressed conditions. Ozone concentrations were 60, 80, 90, 100, and 120 ppb for durations of 2, 4, or 6 h. The 24 h average ozone concentrations ranged from 31 to 57.5 ppb. There was a clear relationship between the 24-h average ozone concentration and the distribution of crack length frequencies on the rubber surface. Table 11-1 gives the average 24-h ozone concentration and lengths for two vulcanized natural rubber strips.

Table 11-1. Average 24-h Ozone Concentrations Producing the Highest Frequency of Cracks of a Certain Length in the Middle and Central Zones of the Rubber Test Strips

	1% Antiozonant 4010NA #		0.5% Antiozonant 4010NA	
Crack Length (mm)	**Middle Zones**	**Central Zones**	**Middle Zones**	**Central Zones**
0.05 - 0.10	37.5	37.5	40.0	42.5
0.10 - 0.15	45.0	48.0	48.0	53.0
0.15 - 0.20	48.0	≥57.5	≥57.5	≥57.5
0.20 - 0.40	≥57.5	≥57.5	≥57.5	≥57.5

Ozone concentrations given in ppb.

Adapted from Serrano et al. (1993).

11.2 TEXTILES AND FABRICS

Ozone can damage textiles and fabrics by methods similar to those associated with elastomers. Generally, synthetic fibers are less affected by ozone than natural fibers; however, ozone contribution to the degradation of textiles and fabrics is not considered significant (U.S.

Environmental Protection Agency, 1996). A study reported by Bogaty et al. (1952) showed that ozone affects moistened cloth more than dry cloth. Scoured cotton duck cloth and commercially bleached cotton print cloth were exposed to 20 to 60 ppb for 1,200 h (50 days). The rate of deterioration was measured by the changes in cuprammonium fluidity values and the fabric breaking strength. At the end of the 1,200-h exposure, there was a 20% loss in breaking strength. Table 11-2 list the changes in cuprammonium fluidity values for both fabrics.

Table 11-2. Cuprammonium Fluidity of Moist Cotton Cloth Exposed to 20 to 60 ppb Ozone

	Duration of Exposure (h)	Cuprammonium Fluidity (rhes)
Duck Cloth	0	2.6
	200	2.8
	680	4.0
	960	6.8
	1200	9.5
Bleached Print Cloth	0	8.2
	200	8.7
	510	9.4
	650	12.0
	865	12.7
	1500	16.5

Adapted from Bogaty et al. (1952).

11.3 DYES, PIGMENTS, AND INKS

Ozone fading of textile dyes is diffusion-controlled; the rate of fading is controlled by the diffusion of the dye to the fiber surface. Many textile dyes react with ozone; however, the rate and severity of the ozone attack is influenced by the chemical nature of the textile fiber and the manner in which the dye is applied. Ozone molecules break the aromatic ring portion of the dye molecule, oxidizing the dye (U.S. Environmental Protection Agency, 1996). In case of aromatic azo dyes, ozone attacks the aromatic rings and electron rich nitrogen atoms (Matsui et al., 1988). Grosjean et al. (1987; 1988a,b) proposed a mechanism for reactions of ozone with indigo, thioindigo, and dibromoindigo, alazarin, and curcumin dyes under dark conditions. Ozone

attaches to the dye molecule at the unsaturated carbon = carbon bond. An ozone adduct is formed (1,2,3-trioxolane), followed by scission of the carbon–carbon bond and the subsequent formation of the corresponding Criegee biradical. A similar mechanism was proposed for the reaction of ozone with triphenylmethane colorant Basic Violet 14. Ozone attacked Basic Violet 14 at the carbon=carbon unsaturated bond and at the carbon–nitrogen unsaturated bond under dark conditions. Other members of the group of triphenylmethane colorants with unsaturated carbon–carbon bonds also are expected to be subject to ozone fading. Tripheylmethane colorants that are expected to be ozone-fugitive include the amino-substituted cationic dyes (Malachite Green, Brilliant Green, Crystal Violet, Pararosaniline Chloride, Methyl Green, and others) (Grosjean et al., 1989).

An indication that ozone caused textile dye fading was first reported by Salvin and Walker (1955). The researchers found that the fading was primarily the result of the destruction of the blue dye molecule. Drapes made of acetate, Arnel, and Dacron and dyed with anthraquinone blue dye exhibited a decrease in shade that was not accompanied by the characteristic reddening caused by NO_x. Figures 11-1 and 11-2 demonstrate the effect of ozone exposure on nylon 6 yarn colored with several blue dyes. Nylon samples inside the home were located on a wall away from sunlight. Outside nylon samples were placed on a covered patio or under the eaves of the house to minimize exposure to sunlight and rain. Ozone concentrations ranged from 2 to 5 ppb outside and 0 to 2 ppb inside. The percent change in dye color was determined monthly by extraction and analysis of the remaining dye or by instrumental measurement of the color change (Haylock and Rush, 1978).

11.4 ARTISTS' PIGMENTS

Several artists' pigments are sensitive to fading and oxidation by ozone when exposed to concentrations found in urban areas (Shaver et al., 1983; Drisko et al., 1985; Whitmore et al., 1987; Whitmore and Cass, 1988; Grosjean et al., 1993). The organic pigments that are ozone fugitive include alizarin red pigments containing lakes of the polycyclic aromatic compound 1,2-dihydroxyanthraquinone, blue-violet pigments containing substituted triphenylmethane lakes, indigo, and yellow coloring agents containing polyfunctional, polyunsaturated compounds such as curcumin (Grosjean et al., 1987). Because of the potential of ozone to damage works of

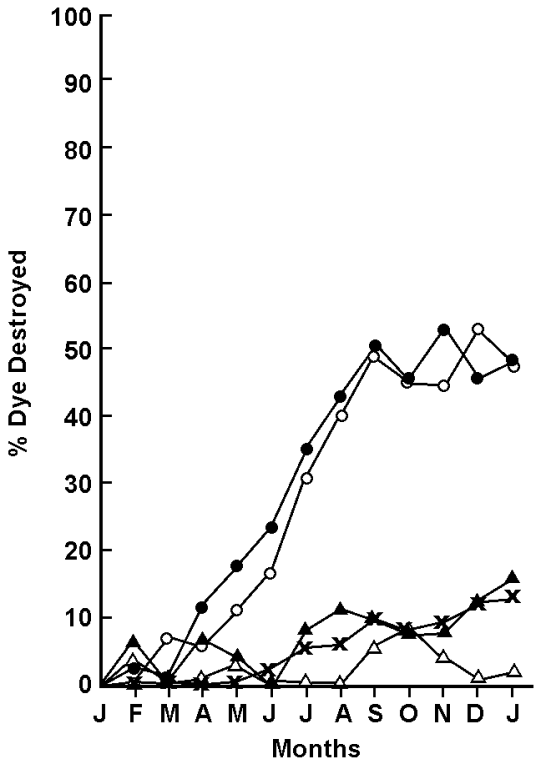


Figure 11-1. In-service fading of nylon 6 yarn inside house. ● = C.I. Disperse Blue 3; O = C.I. Basic Blue 22; ▲ = C.I. Acid Blue 27; x = C.I. Disperse Blue 56; △ = C.I. Acid Blue 232.

Source: Haylock and Rush (1978).

art, recommended limits on ozone concentrations in museums, libraries, and archives are relatively low, ranging from 0.013 to 0.01 ppm.

Experimental studies demonstrate a concentration × time (C × T) relationship between ozone concentration, exposure time, and pigment fading. Cass et al. (1991) summarized some of the earlier research on the effects of ozone on artists' pigments. In studies evaluating the effect of ozone on organic and inorganic watercolors and traditional organic pigments, only the traditional organic pigments showed measurable fading from ozone exposure. Of the inorganic pigments tested, only the arsenic sulfides showed ozone-related changes. The pigments were exposed to 0.3 to 0.4 ppm ozone for 3 mo in the absence of light, at 22 °C and 50% RH. The authors equated this exposure to a C × T of 6 to 8 years inside a Los Angeles museum with air conditioning but without a pollutant removal system.

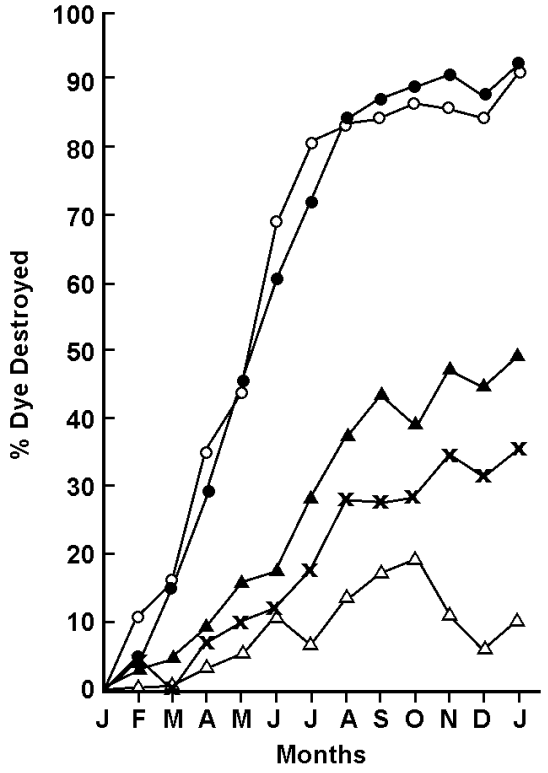


Figure 11-2. In-service fading of nylon 6 yarn outside house. ● = C.I. Disperse Blue 3; O = C.I. Basic Blue 22; ▲ = C.I. Acid Blue 27; x = C.I. Disperse Blue 56; △ = C.I. Acid Blue 232.

Source: Haylock and Rush (1978).

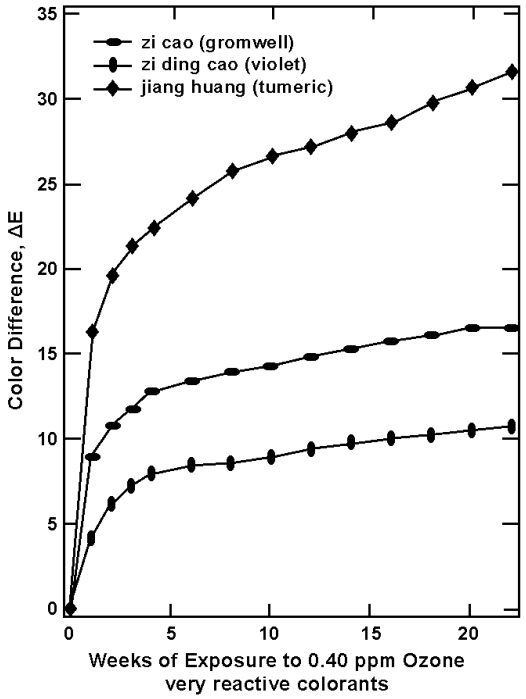
Whitmore and Cass (1988) studied the effect of ozone on traditional Japanese colorants. Most of these compounds are insoluble metal salts that are stable in light and air. Suspensions or solutions of the colorants were airbrushed on hot-pressed watercolor paper or silk cloths. A sample of Japanese woodblock print also was included in the analysis. Samples were exposed to 0.4 ppm ozone at 22 °C, 50% relative humidity, in the absence of light for 12 wk. Changes in reflectance spectra were used to evaluate the effect of ozone exposure on colorant fading. Among the colorants tested on paper, curmin, indigo, madder lake, and lac lake were the most sensitive to ozone exposure. Gamboge was relatively insensitive to ozone. The blue and green areas of the sample from the woodblock print was very reactive due to the indigo dye ozone sensitivity. The other colorants, red, yellow, and purple, showed very little sensitivity to ozone.

The textiles dyes that reacted with ozone were indigo, alone or in combination with several yellow dyes.

Ye et al. (2000) reported the rate of ozone fading of traditional Chinese plant dyes. Twelve different colorants were applied to watercolor paper and silk and exposed to 0.4 ppm ozone at 25 °C, at 50% RH, in the absence of light for 22 wks. Dye fading was greater when the colorant was applied to the watercolor paper compared to the silk cloth due to the darker initial depth of the shade, the greater saturation of the colorant throughout the cloth. Tumeric, gromwell, and violet on paper was particularly reactive. Tangerine peel was moderately reactive and sappan wood, dalbergia wood, Chinese gall, indigo, and Chinese yellow cork tree were slightly reactive to ozone. Black tea was not reactive to ozone. The colorants on silk samples showing color changes were gromwell, sappan wood, gardenia, tummeric, and violet. Figures 11-3 and 11-4 demonstrate the color change of the various colorants on watercolor paper and silk.

Artists' pigments also have exhibited fading when exposed to a mixture of photochemical oxidants. Grosjean et al. (1993) exposed 35 artists' pigments to a mixture of photochemical oxidants consisting of ozone, nitrogen dioxide (NO_2), and peroxyacetyl nitrate (PAN) for 12 wks. Weekly average photochemical concentrations were 200 ppb for ozone, 56 ± 12 to 99 ± 24 for NO_2, and 11 ± 3 to 18 ± 2 for PAN. All exposures were carried out at room temperature in the absence of light. To determine the effect of humidity on pigment fading, the relative humidity was increased from 46% after 8 weeks of exposure to 83% for a 2 week period and then returned to 46% for the remainder of the exposure.

Table 11-3 lists the artists' pigment and degree of fading. Eleven of the pigments exhibited negligible color change, 12 had small color changes, 3 had modest color changes, and 9 exhibited substantial color changes. Fading of Disperse Blue 3 and Reactive Blue 2 were likely the result of NO_2 exposure, the fading of triphenylmethanes is consistent with exposure to nitric acid formed under high humidity conditions. Fading of the indigos was dominated by ozone exposure and curcumin was faded by all of the photochemicals studied. Increasing the relative humidity resulted in a substantial color change for all of the pigments, with the exception of curcumin and indigo.



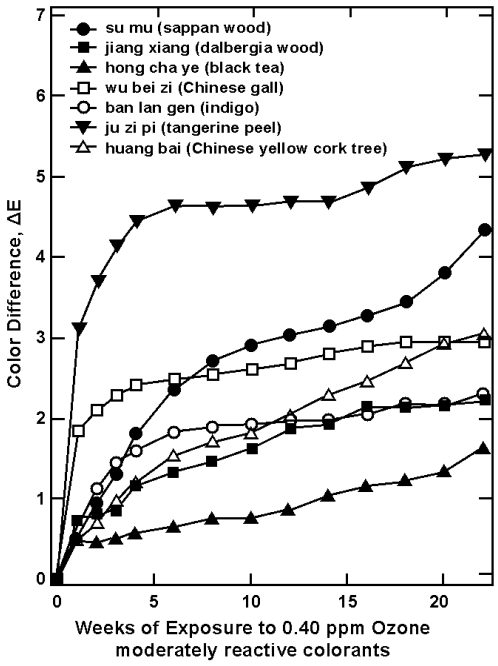


Figure 11-3. Observed color changes for natural colorant-on-paper systems during exposure to 0.40 ppm ozone at 25 °C ± 1 °C, 50% RH, in the absence of light.

Source: Ye et al. (2000).

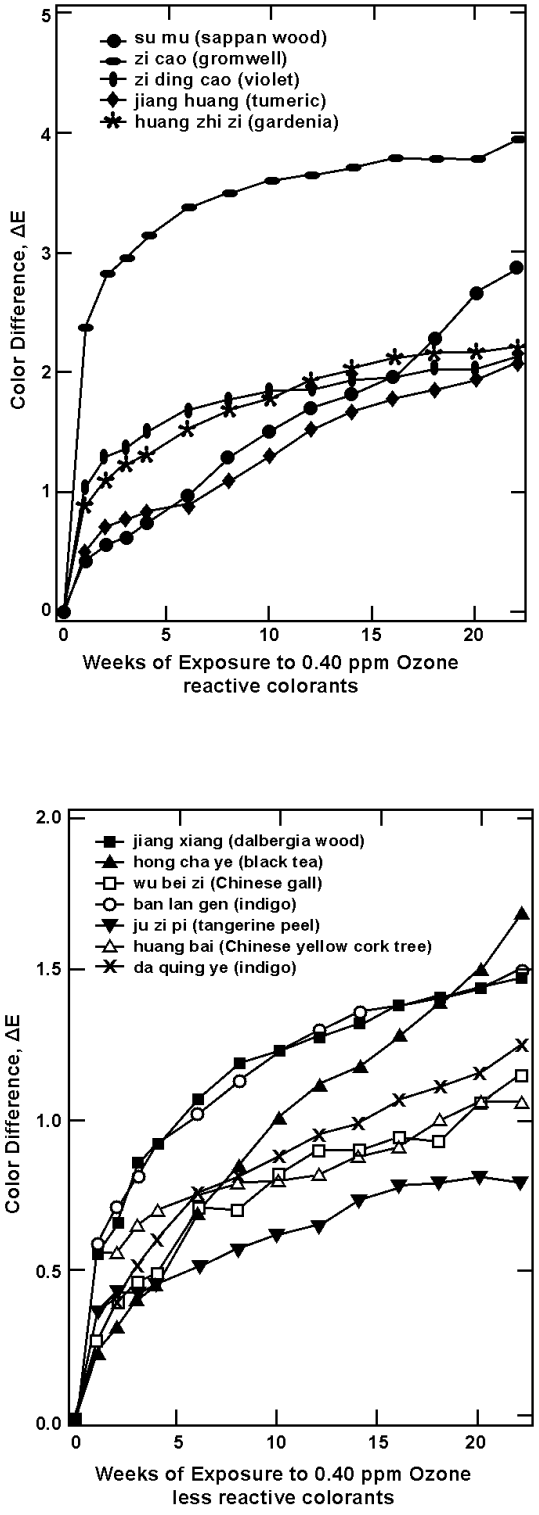


Figure 11-4. Observed color changes for natural colorant-on-site during exposure to 0.40 ppm ozone at 25 °C ± 1 °C, 50% RH, in the absence of light.

Source: Ye et al. (2000).

Table 11-3. Color Change After 12 Weeks of Exposure to a Mixture of Photochemical Oxidants

Colorant*	Color Change (ΔE units)†	Chemical Functionality or Chemical Composition
Acid Red 37 (17045)‡	11.7 ± 0.5	Aminophenyl-substituted azo dye, sulfonate salt
Acid Yellow 65 ‡	1.8 ± 0.5	Nitro- and phenyl-substituted azo dye, sulfonate salt
Alizarin Carmine	1.8 ± 0.2	Alizarin lake
Alizarin Crimson (Pigment Red 83)	1.4 ± 0.2	Alizarin lake
Aurora Yellow (77199)	0.5 ± 0.1	Cadmium sulfide
Basic Fuschin (42510)‡	33.4 ± 3.0	Amino-substituted triphenylmethane
Brilliant Green (42040)‡	20.6 ± 2.1	Amino-substituted triphenylmethane
Brown Madder	1.7 ± 0.1	Alizarin lake
Cadmium Yellow (77199)	0.4 ± 0.1	Cadmium sulfide
Carmine	1.8 ± 0.2	Lake of cochineal (substituted anthraquinone)
Chrome Yellow (77600)‡	1.7 ± 1.2	Lead chromate
Copper phthalocyamne (Pigment Blue 15)	1.0 ± 0.1	Copper phthalocyanine
Crimson Lake	3.5 ± 0.3	Alizarin lake
Curcumin (Natural Yellow 3)	15.2 ± 2.6	1,7 bis (4-hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione
Disperse Blue 3	10.8 ± 0.1	Amino-substituted anthraquinone
French Ultramarine Blue	0.8 ± 0.3	
Gamboge (Natural Yellow 24)	0.4 ± 0.1	Gambogic acid
Hooker's Green Light	1.5 ± 0.4	Chlorinated copper phthalocyanine plus ferrous beta naphthol derivative
Indigo (a formulation)	1.1 ± 0.1	Alizarin lake plus lampblack plus copper plthalocyanine
Indigo carmine ‡	14.0 ± 1.9	5,5-indigo disulfonic acid, sodium salt
Indigo (73000)‡	64.1 ± 4.5	
Mauve	3.6 ± 0.5	Lake of triphenyl methane (basic fuschin) plus copper phthalyocyanine
New Gamboge	0.9 ± 0.1	Arylamide yellow (CI 11680) plus toluidine red

Table 11-3 (cont'd). Color Change After 12 Weeks of Exposure to a Mixture of Photochemical Oxidants

Colorant*	Color Change (ΔE units)†	Chemical Functionality or Chemical Composition
Pararosaniline base (42500)‡	25.6 ± 4.7	Amino-substituted triphenylmethane
Payne's Grey	1.0 ± 0.1	Alizarin lake plus prussian blue plus lampblack plus ultramarine blue
Permanent Magenta	1.1 ± 0.1	Quinacridone
Permanent Rose	2.0 ± 0.1	Quinacridone
Prussian Blue	0.7 ± 0.2 1.6 ± 0.3	Ferric ferrocyanide
Prussian Green	0.9 ± 0.2	Arylamide yellow plus prussian blue
Purple Lake	2.3 ± 0.3	Alizarin lake
Reactive Blue 2 (61211)‡	14.4 ± 1.1	Amino-substituted anthraquinone, sulfonate salt
Rose Carthane (12467)	0.8 ± 0.2	Arylamide (Pigment Red 10) plus xanthene (Pigment Red 90)
Rose Doré	2.0 ± 0.2	Quinacridone plus Yellow 3
Thioindigo Violet (73312)‡	1.9 ± 1.2	Chlorinated thioindigo
Winsor Yellow (11680)	0.5 ± 0.2	Arylamide yellow

* On watercolor paper unless otherwise indicated. Color Index (CI) names or CI numbers are given in parentheses.
† Mean ± one standard deviation for triplicate samples calculated from the parameters L^*, a^*, and b^* measured with the color analyzer.
‡ On Whatman 41 paper.

Source: Grosjean et al. (1993).

11.5 SURFACE COATINGS

Ozone will act to erode some surface coatings (paints, varnishes, and lacquers). However, many of the available studies on ozone degradation of surface coatings do not separate the effects of ozone from other pollutants or environmental factors such as weather, humidity, and temperature. Campbell et al. (1974) attempted to demonstrate an ozone related effect on oil house paint, acrylic latex coating, alkyd industrial maintenance coating, urea alkyd coil coating, and nitrocellulose/acrylic automotive paint. Painted test panels were exposed to 100 and

1,000 ppb ozone in a xenon arc accelerated weathering chamber for up to 1,000 h. Using weight loss as a measure of ozone-induced erosion the researchers concluded that all of the paints tested suffered degradation in the presence of ozone and that the automotive finish suffered the most ozone-induce degradation. When ozone degradation was measured using scanning electron microscopy, the oil house paint and latex coating samples showed erosion above that seen with clean air but only at the highest exposure level. No effects were noted for the automotive paint. The other painted surfaces were not evaluated.

Spence et al. (1975) studied the effect of air pollutants and relative humidity on oil based house paint, acrylic latex house paint, acrylic coil coating, and vinyl coil coating under laboratory conditions. Test panels were exposed in weathering chambers equipped with a xenon are light for simulating sunlight to low and high levels of ozone (0.08 and 0.5 ppm), sulfur dioxide (0.03 and 0.5 ppm), and nitrogen dioxide (0.05 and 0.5 ppm) and relative humidity (50 and 90%). Samples were exposed for a total of 1000 h. The exposure cycle consisted of 20 min of dew and 20 min of light. The effects of the exposure on the painted surfaces were measured by weight loss and loss in film thickness. The acrylic coil coating had the lowest erosion rate of the surface coatings tested. However, ozone was the only pollutant that had a significant effect on the surface erosion. Sulfur dioxide and relative humidity were significant factors in the erosion of oil base house paints and vinyl coil coating. The findings for acrylic latex house paint were not reported.

11.6 CONCLUSIONS

Ozone and other photochemical oxidants react with many economically important man-made materials, decreasing their useful life and aesthetic appearance. Some materials known to be damaged by ozone include elastomers, fibers and dyes, and paints. Most studies have been on single compounds rather than complex materials.

The elastomeric compounds, natural rubber and synthetic polymers and copolymers of butadiene, isoprene, and styrene, are particularly susceptible to even low concentrations of ozone. Ozone damages these compounds by breaking the molecular chain at the carbon-carbon double bond; a chain of three oxygen atoms is added directly across the double bond. The change in structure promotes the characteristic cracking of stressed/stretched rubber called

"weathering." Tensile strain produces cracks on the surface of the rubber that increase in number with increased stress/stretching.

The rate of crack growth is dependent on the degree of stress, the type of rubber compound, ozone concentration, time of exposure, ozone velocity, and temperature. After initial cracking, there is further ozone penetration, resulting in additional cracking and, eventually, mechanical weakening or stress relaxation.

Ozone can damage textiles and fabrics by methods similar to those associated with elastomers. Generally, synthetic fibers are less affected by ozone than natural fibers; however, ozone contribution to the degradation of textiles and fabrics is not considered significant .

Ozone fading of textile dyes is a diffusion-controlled process; the rate of fading is controlled by the diffusion of the dye to the fiber surface. Many textile dyes react with ozone. The rate and severity of the ozone attack is influenced by the chemical nature of the textile fiber and the manner in which the dye is applied.

Several artists' pigments are also sensitive to fading and oxidation by ozone when exposed to concentrations found in urban areas.

REFERENCES

Andries, J. C.; Rhee, C. K.; Smith, R. W.; Ross, D. B.; Diem, H. E. (1979) A surface study of ozone attack and antiozonant protection of carbon black loaded natural rubber compounds. Rubber Chem. Technol. 52: 823-837.

Bogaty, H.; Campbell, K. S.; Appel, W. D. (1952) The oxidation of cellulose by ozone in small concentrations. Text. Res. J. 22: 81-83.

Bradley, C. E.; Haagen-Smit, A. J. (1951) The application of rubber in the quantitative determination of ozone. Rubber Chem. Technol. 24: 750-755.

Campbell, G. G.; Schurr, G. G.; Slawikowski, D. E.; Spence, J. W. (1974) Assessing air pollution damage to coatings. J. Paint Technol. 46: 59-71.

Cass, G. R.; Nazaroff, W. W.; Tiller, C.; Whitmore, P. M. (1991) Protection of works of art from damage due to atmospheric ozone. Atmos. Environ. Part A 25: 441-451.

Drisko, K.; Cass, G. R.; Whitmore, P. M.; Druzik, J. R. (1985) Fading of artists' pigments due to atmospheric ozone. In: Vendl, A.; Pichler, B.; Weber, J.; Banik, G., eds. Wiener Berichte über Naturwissenschaft in der Kunst: Doppelband 2/3. Vienna, Austria: Verlag ORAC; pp. 66-87.

Gent, A. N.; McGrath, J. E. (1965) Effect of temperature on ozone cracking of rubbers. J. Polymer Sci. A 3: 1473-1482.

Grosjean, D.; Whitmore, P. M.; De Moor, C. P.; Cass, G. R.; Druzik, J. R. (1987) Fading of alizarin and related artists' pigments by atmospheric ozone: reaction products and mechanisms. Environ. Sci. Technol. 21: 635-643.

Grosjean, D.; Whitmore, P. M.; Cass, G. R.; Druzik, J. R. (1988a) Ozone fading of natural organic colorants: mechanisms and products of the reaction of ozone with indigos. Environ. Sci. Technol. 22: 292-298.

Grosjean, D.; Whitmore, P. M.; De Moor, C. P.; Cass, G. R.; Druzik, J. R. (1988b) Ozone fading of organic colorants: products and mechanism of the reaction of ozone with curcumin. Environ. Sci. Technol. 22: 1357-1361.

Grosjean, D.; Whitmore, P. M.; Cass, G. R.; Druzik, J. R. (1989) Ozone fading of triphenylmethane colorants: reaction products and mechanisms. Environ. Sci. Technol. 23: 1164-1167.

Grosjean, D.; Grosjean, E.; Williams, E. L., II. (1993) Fading of artists' colorants by a mixture of photochemical oxidants. Atmos. Environ. Part A 27: 765-772.

Haylock, J. C.; Rush, J. L. (1978) Studies on the ozone fading of anthraquinone dyes on nylon fibers. Part II: In-service performance. Text. Res. J. 48: 143-149.

Lake, G. J.; Mente, P. G. (1992) Ozone cracking and protection of elastomers at high and low temperatures. J. Nat. Rubber Res. 7: 1-13.

Lattimer, R. P.; Layer, R. W.; Rhee, C. K. (1984) Mechanisms of antiozonant protection: antiozonant-rubber reactions during ozone exposure. In: 32nd annual conference on mass spectrometry and allied topics. May/June; San Antonio, TX. Bethesda, MD: American Society for Mass Spectrometry; pp. 357-358.

Matsui, M.; Koike, T.; Shibata, K. (1988) Ozone fading of phenolphthalein and aurin. J. Soc. Dyers Colour 104: 482-486.

Mueller, W. J.; Stickney, P. B. (1970) A survey and economic assessment of the effects of air pollution on elastomers: final report. Columbus, OH: Battelle Memorial Institute, Columbus Laboratories; National Air Pollution Control Administration contract no. CPA-22-69-146.

Razumovskii, S. D.; Podmasteriev, V. V.; Zaikov, G. E. (1988) Kinetics and mechanism of stress relaxation of polyisoprene vulcanizates under ozone ageing. Polym. Degrad. Stab. 20: 37-47.

Salvin, V. S.; Walker, R. A. (1955) Service fading of disperse dyestuffs by chemical agents other than the oxides of nitrogen. Text. Res. J. 25: 571-585.

Serrano, E.; Castro, M.; Macías, A. (1993) An improved direct method of rubber cracking analysis for estimating 24-hour ozone levels. Atmos. Environ. Part A 27: 431-442.

Shaver, C. L.; Cass, G. R.; Druzik, J. R. (1983) Ozone and the deterioration of works of art. Environ. Sci. Technol. 17: 748-752.

Spence, J. W.; Haynie, F.; Upham, J. B. (1975) Effects of gaseous pollutants on paints: a chamber study. J. Paint Technol. 47: 57-63.

U.S. Environmental Protection Agency. (1996) Air quality criteria for ozone and related photochemical oxidants. Research Triangle Park, NC: Office of Research and Development; report nos. EPA/600/AP-93/004aF-cF. 3v. Available from: NTIS, Springfield, VA; PB96-185582, PB96-185590, and PB96-185608. Available: http://cfpub2.epa.gov/ncea/.

Whitmore, P. M.; Cass, G. R. (1988) The ozone fading of traditional Japanese colorants. Stud. Conserv. 33: 29-40.
Whitmore, P. M.; Cass, G. R.; Druzik, J. R. (1987) The ozone fading of traditional natural organic colorants on paper. J. Am. Inst. Conserv. 26: 45-58.
Ye, Y.; Salmon, L. G.; Cass, G. R. (2000) The ozone fading of traditional Chinese plant dyes. J. Am. Inst. Conserv. 39: 245-257.

United States
Environmental Protection
Agency

Please make all necessary changes in the below label, detach copy or copy, and return to the address in the upper left-hand corner.

If you do not wish to receive these reports CHECK HERE ☐; detach copy or copy, and return to the address in the upper left-hand corner.

PRESORTED STANDARD
POSTAGE & FEES PAID
EPA
PERMIT No. G-35

National Center for
Environmental Assessment
Research Triangle Park, NC 27711

Official Business
Penalty for Private Use
$300

EPA/600/R-05/004bB
August 2005