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CHROMOSOMAL ABNORMALITIES AMONG WELDER TRAINEES

by

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FOREWORD

The many benefits of our modern, developing, industrial society are accompanied by certain hazards. Careful assessment of the relative risk of existing and new man-made environmental hazards is necessary for the establishment of sound regulatory policy. These regulations serve to enhance the quality of our environment in order to promote the public health and welfare and the productive capacity of our Nation's population.

The Health Effects Research Laboratory, Research Triangle Park, conducts a coordinated environmental health research program in toxicology, epidemiology, and clinical studies using volunteer subjects. These studies address problems in air pollution, non-ionizing radiation, environmental carcinogenesis and the toxicology of pesticides as well as other chemical pollutants. The Laboratory participates in the development and revision of air quality criteria documents on pollutants for which national ambient air quality standards exist or are proposed, provides the data for registration of new pesticides or proposed suspension of those already in use, conducts research on hazardous and toxic materials, and is primarily responsible for providing the health basis for non-ionizing radiation standards. Direct support to the regulatory function of the Agency is provided in the form of expert testimony and preparation of affidavits as well as expert advice to the Administrator to assure the adequacy of health care and surveillance of persons having suffered imminent and substantial endangerment of their health.

This report presents the results of a study of welder students who, during their welding training, it was thought would be exposed each day to ozone generated by the welding process. Mutagenicity of this frequently encountered air pollutant had been suggested by several previous studies. Thus, as a biological indicator of ambient air pollution health effects, chromosomal change offered a heretofore little-used, objective measurement. Demonstration of its feasibility and efficacy as an indicator could provide a new method of determining the effects of community air pollution.

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ABSTRACT

Serial cytogenetic observations were made on a group of 273 military recruits who were being trained as welders at Aberdeen, Maryland. The trainees were being exposed to presumably increased levels of ozone in the course of their welding school experience, and it was the purpose of this study to determine whether or not ozone, at low to moderate doses, is capable of inducing chromosomal aberrations in peripheral blood lymphocytes.

Previous exposures and the past medical experiences of the trainees were determined by questionnaire, enabling us to obtain a profile on the medical-social characteristics of the study sample. Each welder was to serve as his own control, having a blood sample drawn at the beginning of his twelve week training program, prior to ozone exposure, with two post-exposure bloods being obtained at six and twelve weeks after the start of the program.

Ozone levels and the levels of the oxides of nitrogen were determined in the immediate area of the welding. The ozone levels were repeatedly found to be negligible, with the nitrogen oxides appearing to be the primary toxic agents involved. For those 165 subjects on whom two blood samples were obtained, and for those 86 on whom all three blood samples were obtained, no statistically significant increases in chromosomal aberrations were found.

ACKNOWLEDGEMENTS

The cooperation of the U.S. Army welder trainees at Aberdeen formed the basis of this study, and the leadership at the Base and at the welding school in particular, helped us throughout.

Numerous persons helped directly in the generation of the data included in this Report. Among these were Dr. S. Neriishi, Mr. K. Ohki, Ms. J. Beaird, Ms. D. Campos, and Ms. Z. Pyatt. We are particularly indebted to Dr. Neriishi, who aided in the laboratory supervision, and Ms. Pyatt, who was invaluable in the laboratory and in the on-site work at the Aberdeen Proving Grounds in Maryland. Dr. Granville Sewell and his colleagues of the School of Public Health at Columbia University were responsible for the measurements of ozone and the oxides of nitrogen.

James Stebbings of EPA was the original Project Officer. Dorothy Calafiore was the EPA Project Officer in the later stages of the study, and we here wish to express our gratitude to her for her considerable assistance in the data analyses and for her guidance on this Report. Ms. Rhoda Serra, Administrative Assistant to the author, is owed a particular note of thanks for her high quality work in preparation of this Final Report.

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INTRODUCTION

A. SUMMARY BACKGROUND

Considerable evidence exists for the induction by ozone of both point and chromosomal mutations. Zelac et al.[1] exposed Chinese hamsters to 0.2 parts per million of ozone for five hours, and demonstrated an increase in chromosomal aberrations in their peripheral blood lymphocytes. The major types of aberrations reported were chromosomal exchanges and deletions. Sachsenamier et al. [2] had earlier found ozone-induced chromosomal aberrations in chick embryo fibroblasts, and Fetner [3] had demonstrated chromosomal aberrations produced by ozone in Vicia fava. Furthermore, ozone is known to induce point mutations. Prat et al. [4] demonstrated that ozone alters the pyrimidine bases thymine and uracil in E. coli DNA, and Davis [5] demonstrated the induction of mutant colonies of E. coli after mutagenesis with ozonated water.

Human cell studies of ozone effects have been few. Merz et al. [6] showed that in six subjects exposed to 0.5 ppm ozone for six and ten hours, as part of an assessment of the effects of ozone on pulmonary function, neither chromatid nor chromosome-type aberrations were produced in peripheral blood lymphocytes; but achromatic lesions, interpreted as representing unrepaired single strand breaks, and chromatid deletions were seen. The peak frequencies of aberrations were seen at two weeks post-exposure, with a return to lower aberration frequencies by six weeks.

B. SCOPE OF WORK

In addition to exposure of humans to ozone in areas of heavy air pollution, occupational exposure, particularly among welders, is known to occur. The mounting evidence for genic and cytogenetic effects of ozone warranted a more exhaustive *in vivo* study of human chromosome response to ozone and to the oxides of nitrogen than has been available to the present time. For this reason, we undertook a study, on behalf of the U.S. Environmental Protection Agency, to determine whether the peripheral blood lymphocytes of military welder trainees would have increased numbers of chromosomal aberrations after six to twelve weeks of welding experience. The study consisted of two major aspects: 1) the cytogenetic laboratory program; and 2) the environmental monitoring program.

MATERIALS AND METHODS

A. SELECTION OF SUBJECTS

The target population for this study was a group of military recruits (U.S. Army) who were training as welders at Aberdeen, Maryland. Each welder trainee served as his own control, having a blood sample, for peripheral blood cell chromosome studies, drawn at the beginning of the twelve-week training period, prior to ozone exposure. This blood sample was to establish the baseline frequency of chromosomal aberrations for that individual. Each recruit in the twelve-week, overlapping programs was asked to volunteer for this study and sign a consent form (see Appendix A). Confidential questionnaires (see Appendix B) were then administered to those who agreed to participate. Information regarding x-ray exposure within the preceding six to twelve months, viral infections and immunizations, and drug histories were elicited at the time of the initial blood drawing, with interval histories being obtained at the time of the post-exposure blood drawings, at six and twelve weeks after the start of the program. Thus, the ideal situation was to have one blood sample prior to ozone exposure, with two after exposure, at six week intervals.

B. BLOOD PROCESSING AND MICROSCOPY

Sterile heparinized vacutainers were used for transportation of blood samples from Aberdeen to New York City. Each blood sample was assigned a code number by the blood drawers in Aberdeen, with the code not having been broken until after the chromosomes were analyzed. Bloods were drawn once or twice per week, as required, and the time interval between blood drawing and culture initiation did not exceed six hours. The bloods were chilled, but not frozen, in a portable container, and sent by commercial bus.

Leukocytes were then separated by low-speed centrifugation and duplicate cultures were initiated from each blood sample, as back up. Short-term, leukocyte cultures were established and cells were harvested after a fifty hour culture time, according to the method of Bloom and Iida [7]. Non-banded Giemsa staining methods were used.

Slides were coded by subject number and read blindly, without knowledge of the exposure status of the subject. An

attempt was made to read 100 cells from each blood culture. The following definitions [8] were applied for the major aberrations: Single chromatid and isochromatid breaks were scored as such if there was displacement of the distal portion of the chromatid(s). When the chromosome of origin of an acentric fragment could not be determined, it was called a free fragment. Multicentric chromosomes were scored in accordance with the number of centromeres, while ring chromosomes were classified as either centric or acentric. Fragments accompanying multicentrics or rings were not scored separately. Translocations were generally detectable by the elongated long or short arms of the affected chromosome. Inversions showed displacement of the centromere. Other aberrations scored were quadriradials (chromatid exchanges), breaks at the centromere, and deletions, which were usually terminal. The presence of an aberration was confirmed by at least two experienced microscopists.

C. OZONE DETERMINATIONS

Using an AeroChem Chemoluminescence Analyzer Model AA-5 for NO, NO_x, and O_3, a mobile air sampling unit was assembled during early September, 1974, in the welding school facility at Aberdeen. About a week was spent in testing and refining the operation of the equipment, and a program of testing air intensively initially and then on a monthly basis over an 18-month period, was subsequently conducted. For readings during the welding operations, a teflon tube with dust filter was used to collect air samples within the welding booths at six positions (described below) around the welder's head, so measurements represented the gas mixture that was probably inhaled. The readings were later verified by field tests performed, as confirmation, by the staff of the U.S. Environmental Protection Agency.

Test Sequence	Code	Position
1	F (Front)	Front of welder over the welding or cutting activity. This represented the position of highest gas concentration. Some of this gas was drawn directly under the bottom edge of the face shield for respiration. Proportion appeared to vary according to the physique of the individual and his work habits.
2	R (Right)	Next to welder's right ear where respiratory air was being drawn into the face shield. This represented one of the four zones - top, bottom (above), and two sides - that acted

Test Sequence	Code	Position
		as sources of respiratory air.
3	B (Back)	Behind head of welder. Since he was usually bending over his work, this test registered respiratory air quality entering the top of his face shield, usually the first or second most important point of air entry.
4	L (Left)	Next to welder's left ear.
5	T (Top)	Top of booth at exhaust duct.
6	O (Outside)	Ambient air quality approximately two feet from front of booth at nose level.

Operation of the instrumentation was supervised by Granville H. Sewell, Ph.D., Associate Professor of Public Health (Environmental Health Sciences) and Acting Head, Division of Environmental Health Sciences, and Dhun B. Patel, Ph.D., Assistant Professor of Public Health (Environmental Health Sciences).

D. STATISTICAL ANALYSIS OF CYTOGENETIC DATA

All statistical analyses were carried out by John P. Creason, Ph.D., Chief, Statistics Staff, Environmental Protection Agency.

The aberration types analyzed were:

1. Single chromatid gaps
2. Single chromatid breaks
3. Isochromatid gaps
4. Isochromatid breaks
5. Multicentric-dicentric rings
6. Acentric fragments
7. Translocations
8. "Others"

Analysis of the counts was done using the Grizzle-Starmer-Koch analysis procedure for categorical data. In this analysis, functions of the proportions, $F(\pi)$, in the cells are estimated for the model under consideration. The tests appropriate to this problem are formulated as $F(\pi) = 0$. A test statistic is generated which has asymptotically a central χ^2 distribution if the hypothesized model is true.

RESULTS

A. OF THE QUESTIONNAIRE ON POPULATION CHARACTERISTICS

The questionnaire, developed by us with J. Stebbings of the EPA, was administered to all of the 273 trainees from whom blood was originally drawn. However, we have analyzed only those bloods yielding 75 cells or more, or a total of 253. Thus, for the sake of consistency, we wanted to include only the data from the questionnaires of that group. However, six of these people failed to answer the questions, so the following data is based on a total of 247 questionnaires.

One hundred eighty-four (74.0%) of the trainees were between the ages of 18-21, eight being only slightly younger, and 55 ranging in age from 22 to 57. (Note that the 57 year old was not a military recruit but a Civilian attending the school).

One hundred seventy-five (70.9%) of the trainees were White; 42 (17.0%), Black; 1 (0.4%), Oriental, and 3 (1.2%), "other American." Six made no reply.

The majority (210 trainees, or 85%) had had a high school education, while 13 (5.3%) had completed grade 8, and 3 (1.2%) had completed four years of college. Fifty-eight (23.5%) had attended a trade school.

All entered the welding school in 1975, and 91.9% entered active duty either the year of the study or the year immediately preceding it.

Of the total 247 trainees, 122 (49.4%) stated they had had previous school or job welding experience. However, 70 trainees (28.4%) who had had previous experience, had it more than one year before the first blood was taken.

Two hundred nineteen trainees (88.7%) reported that they were in generally good health, with no significant physical or mental condition. One hundred sixty-nine (68.4%) were cigarette smokers at the time of the study.

Information regarding various illnesses the trainees had had within six months prior to the drawing of the first blood is summarized in the following table:

Disease	No. affected
Rash, non-specific	14
Infectious Mononucleosis	4
Hepatitis	1
Flu	19
Alcoholism	1
Anemia	1
Blood Poisoning	1
Nerves	1
Measles	3
Respiratory Infections	19
Seizures	1
Unknown Illness	2

In addition, the following information regarding health problems six months or more prior to the initial blood sampling was ascertained: 3 trainees reported tuberculosis; 4, a psychogenic illness such as a nervous condition or migraine; 1, recovered drug addiction and alcoholism; 1, ulcer; 1, liver and kidney problem; 1, abdominal pain of unknown origin; 1, asthma; and 2, orthopedic problems.

51.0% of the trainees reported taking no medicines or drugs within 6 months prior to blood #1. 23.1%, however, had taken over-the-counter cold remedies; 6.1% had taken antibiotics; 2.8% - analgesics; 2.0% - allergy medication; 2.4% - prescription drugs such as Mellaril, Emprazil, Parafon Forte, Antabuse, and Amyl Nitrate; 5.3% reported taking unknown drugs.

4.5% had taken the drug before entering active duty; 19.8%, after entering active duty; 12.1%, both before and after; while 12.6% made no reply.

63.5% of the trainees reported that they had been x-rayed within the six months preceding blood #1; the largest percentage (17.8%) having had dental x-rays, followed by 17.0% who had had a chest or back x-ray. 45% of those x-rayed had been done so either before, after, or both before and after entering active duty. No-one reported receiving radiation therapy, although one man stated he had been given a radioactive substance for pain and dizziness, but couldn't remember when. X-ray exposures six months or more prior to the initial blood sampling were varied, as expected, with most (41 persons, or 15%) of these exposures having been for studies of the upper or lower extremities.

B. OF THE CYTOGENETIC ANALYSES

As presented in Table 1, a total of 610 blood samples were drawn from 273 welder trainees. The culture failure rate

was 1.6%, with an additional 9.2% of cultures providing less than 75 readable metaphases. In the following discussions of these results, we will focus primarily on the data obtained from the 89.2% of cultures which yielded at least 75 quality cells.

It should be pointed out that while compliance was reasonably high for the first and second blood drawings (75.5%), it fell off markedly for the third blood sample, with only 131 of the original 273 subjects (48.0%) providing the final, third blood sample. The major reason for this decline was that many of the recruits did not complete the entire twelve weeks of the training program, with approximately 25% leaving prior to the final sampling date.

Examination of the data provided in Table 2, in which we pooled all cases having 75 or more cells analyzed, revealed that small increments, not statistically significant, were seen in the frequencies of cells with isochromatid gaps, with dicentrics and accompanying acentric fragments, and in translocations and quadriradials. These minimal increases were generally seen in the six week blood sample, much less impressively or not at all, in the twelve week, or third, blood sample.

However, analysis of the data from those 86 subjects a) who contributed three blood samples, and b) each of whose three blood samples provided more than 75 cells, suggests that chromatid-type aberrations were more generally increased, particularly, again, in the six week sample. The percentage of cells with single chromatid gaps rose from 0.84 in the 86 control bloods, to 1.40 at six weeks, and declined to 1.10 by twelve weeks. The proportion of cells with isochromatid gaps rose from 0.04 in the controls, to 0.11 at six weeks, and to 0.18 at twelve weeks. The isochromatid break frequency showed a rise from 0.01% of cells to 0.06% at six and twelve weeks. Among the chromosome-type aberrations, no statistically significant increases were seen in any category of aberration, though a few more dicentrics with fragments, fragments alone, and quadriradials were detected at six and twelve weeks than in the control bloods (see Table 3).

C. OF THE O_3, NO, AND NO_x MONITORING PROGRAM

The trainees were exposed to ozone levels ranging from below the sensitivity of the instruments to a maximum of 0.015 ppm in both the ambient air of the welding sheds and the respiratory zone within individual welding booths. Normally, however, the instrument registered "zero" ozone levels.

The NO levels varied from 6.0 ppm to 0.2 ppm, while the NO_x levels were found to register from 8.0 ppm to 0.5 ppm.

Highest exposures occurred for each trainee only during periods of active welding, which totaled considerably less than one hour each day. Typical maximum ambient exposure levels were 1.0 ppm of NO and 1.5 of NO_x. Exposure varied from booth to booth, area to area, and time to time.

Averaging the readings did not appear feasible for three reasons: 1) the readings were typically close to the sensitivity limits of the instrument, 2) general atmospheric pollution conditions were so severe that the instrument deteriorated rapidly and later readings did not have the reliability of the initial ones, and 3) the readings at specific locations were not sufficient in number and comparability to provide statistical reliability. On any one day, the school may have had up to fifty trainees welding in various parts of the facility. However, the locations and type of work being performed by these trainees varied from day to day. Furthermore, the intensity of exposure for each trainee varied constantly during welding.

During operation of the instruments, the teflon filters in the sampling line had to be replaced frequently, usually twice a day, due to accumulation of gritty particles. Towards the end of the monitoring period, the instrument began to increasingly lose sensitivity despite the filter changes. When the instrument case was removed, severe corrosion was found to have occurred on many surfaces, particularly at electrical contacts and soldered points. This was interpreted as further evidence of the corrosive nature of the emissions from the welding.

Table 4 provides data from two days during the early stage of the project when the standard testing procedure was rigorously applied.

D. OF STATISTICAL ANALYSIS OF CYTOGENETIC DATA

The previously described statistical approach to the data was used by Dr. John Creason of EPA and us to interpret the findings. A number of data analyses were done, some of which are not presented here but are available on request. These include summaries of aberration types by week; and analysis of the final counts used for subjects with one and two bloods and for subjects with all three bloods.

A summary of the analyses for those subjects with blood samples #1 and #2 is given in Table 5, while a summary of the results for those subjects with all three blood samples is given in Table 6. One should remember that all the subjects with three blood samples also were included in the analysis of those with the first two blood samples, so that these analyses

are not independent of one another and should show similar results when examining the difference between blood samples #1 and #2. It is clear that the magnitude of the differences in aberration frequencies between bloods #1 and bloods #2 and/or #3 are small and, for the most part, not statistically significant.

DISCUSSION

When this project was originally designed (1974), there was legitimate concern over the possible cytogenetic effects of ozone and of the oxides of nitrogen. (We have referred to some of the most relevant papers on the subject of genetic effects of ozone in the Background section above). This project was one of several initiated by the EPA to study the effects of these gases specifically on human chromosomes, with our population being the occupationally exposed group.

As we discuss in the next several paragraphs, and as presented under Results, the ozone levels were, in fact, very low, and we were dealing here primarily with the nitrogen oxides.

In interpreting the monitoring data, three circumstances should be considered:

1.) The atmospheric conditions during welding were extremely adverse. In addition to the gases that were monitored, the trainees were believed exposed to a wide variety of other gases and particulates - many with corrosive or toxic properties - formed during the melting or combustion of welding rods and flux coatings. Coatings can contain a deoxidizer, such as ferromanganese or silicomanganese, and a slag former, such as titanium dioxide, magnesium carbonate, feldspar asbestos, or silica [9]. The steel of a welding rod can contain various alloys including chromium, cobalt, molybdenum, tungsten, boron, and other elements. Coatings on materials being welded can include cadmium, lead, beryllium, fluorides, and other materials [10]. Each of these can form primary and secondary pollutants.

2.) Trainees were exposed to the welding atmosphere only a small fraction of the time, generally on the order of 25%, when they were assigned to the welding shops. Since they also attended classes and other activities, they presumably received the intense exposure for some time period of less than 10% of the total manhours of their tenure at the Aberdeen Proving Grounds.

Even when welding, the intensity of exposure varied. Trainees were required to master the welding procedures with the work in three positions: a) horizontal on the table at waist height, b) vertical on an extension rod that elevated

the work from the table to approximately shoulder height, and c) above the head. Because most of the gases rose while heavier particulates fell, each position had its hazards, with position (b) probably being the one with least exposure.

Identifying the passage of the inhaled air was complicated by the welding face shield that extended from the chest to above the head. When the head was tilted downwards, the primary passages appeared to be from above the head and over the shoulders since the bottom of the shield would fit tightly against the chest. When the head tilted upwards, the passage at the bottom of the shield would open.

3.) Trainees were required to gain experience with a wide variety of welding materials, including brass, stainless steel, and aluminum. Each had a special rod and set of hazards. Some also involved gases such as acetylene.

When the trainees were not welding but were in the welding shops, they were exposed to an ambient level of NO and NO_x. This was somewhat surprising because of the extensive ventilation system. However, this system was primarily focused on the welding booths, which were three-sided enclosures having a canvas curtain that could be drawn over the open entrance side. The NO and NO_x levels could have had three sources: a) gases that "escaped" the ventilation system, b) gases formed in secondary reactions between the materials released in welding and the air environment, and c) electrical equipment, particularly the heavy transformers, that was on the shop floor. Because heavy concentrations were not registered in the vicinity of the equipment, this is not considered a probable source.

Because of the intense ionization of the air during welding, ozone was presumably formed but it could not be registered in significant amounts. This phenomenon has also been noted in the technical literature. The most likely explanation was that the ozone rapidly oxidized the NO and other gases formed during the welding, thus disappearing. This hypothesis was supported by the relatively high ozone values registered in readings immediately over the welding arc and the rapid disappearance of ozone as readings at higher elevations were taken.

Our cytogenetic findings indicated that for virtually all types of scored aberrations, little difference in frequency was seen between the control blood samples (bloods #1) and the follow-up bloods (#'s 2 and 3), obtained when the subjects were well into their welder training program. The most impressive increases, such as they were, were seen in the chromatid-types of lesions. The isochromatid breaks were found to be minimally increased in follow-up bloods, as were the single and isochromatid gaps which we well recognize to be far

less a reliable indicator of chromosomal damage than any other "aberration" type.

It is important to note here that, while many of the welder trainees had had some previous welding experience, the frequency of chromosomal aberrations in the control blood samples was compatible with that found in other putatively normal human populations. Further, there were no subjects among those studied in whom the follow-up aberration frequencies increased consistently and strikingly.

Our basically negative findings with respect to ozone and the oxides of nitrogen are consistent with the findings of McKenzie et al. [11], who examined the peripheral blood lymphocyte chromosomes of 30 men exposed to 0.4 ppm ozone for four hours. If no increase in aberrations is to be expected after a known, acute, relatively high dose exposure, it is possible, but unlikely, that repeated exposure to minimal doses over a maximum of twelve weeks will generate chromosomal aberrations. We say this because the expectation is that repair of some damage will doubtless take place in non-repair-deficient persons, interfering with our ability to detect damaged cells which may be present in very small numbers to begin with.

CONCLUSION

While this study was set up primarily to evaluate the cytogenetic effects of ozone in human somatic cells, there appeared to be rapid conversion of ozone to the oxides of nitrogen, providing measurable amounts of the latter with little residual ozone. The frequency of chromosomal aberrations in the six and twelve week bloods was not significantly different from that in the pre-exposure bloods, leading inevitably to the conclusion that at these exposure levels for these gases, little, if any, chromosomal damage is induced.

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TABLE 1.

Bloods drawn from welder trainees exposed to Ozone, NO_1 and NO_x

	Number of bloods drawn	> 75 cells analyzed	< 75 cells analyzed	No. growth
Pre-exposure sample	273	253 (92.7%)	16 (5.9%)	4 (1.5%)
Post-exposure sample #1 (6 weeks)	206	184 (89.3%)	19 (9.2%)	3 (1.5%)
Post-exposure sample #2 (12 weeks)	131	107 (81.7%)	21 (16.0%)	3 (2.3%)
TOTAL	610	544 (89.2%)	56 (9.2%)	10 (1.6%)

TABLE 2

Aberrations seen in cases with 75 or more cells analyzed

	No. of Cases	No. of Cells Observed	% of Cells c Modal No.
Pre-exposure 1st Blood sample	253	25,263	97.0
Post-exposure #1 2nd Blood (6 wks)	184	18,358	96.8
Post-exposure #2 3rd Blood (12 wks)	107	10,675	96.1

TABLE 2 - CONTINUED

	No. of chromatid aberrations (% of cells with)					No of chromosome aberrations (% of cells with)				
	SCG	SCB	ICG	ICB	dic c ace	dic	ace	t	del	QR
Pre-exposure 1st Blood sample	336 (1.33)	43 (0.17)	27 (0.11)	4 (0.02)	2 (0.01)	6 (0.02)	6 (0.02)	0 (0.00)	2 (0.01)	4 (0.02)
Post-exposure #1 - 2nd Blood (6 wks)	219 (1.19)	17 (0.09)	18 (0.10)	9 (0.05)	7 (0.04)	4 (0.02)	8 (0.04)	2 (0.01)	0 (0.00)	9 (0.05)
Post-exposure #2 - 3rd Blood (12 wks)	133 (1.25)	10 (0.09)	18 (0.17)	5 (0.05)	3 (0.03)	2 (0.02)	4 (0.04)	0 (0.00)	0 (0.00)	0 (0.02)

SCG = single chromatid gap
SCB = single chromatid break
ICG = isochromatid gap
ICB = isochromatid break

dic = dicentric
ace = acentric fragment
t = translocation
QR = quadriradial

TABLE 3

Aberrations in 86 subjects on whom all 3 blood samples were obtained and at least 75 cells analyzed

	No. of cells Obsvd.	% of cells c modal no.
Pre-exposure Sample	8,578	97.0
Post-exposure Sample (6 wks.)	8,580	96.4
Post-exposure Sample (12 wks.)	8,575	96.5

TABLE 3 - CONTINUED

	No. of chromatid aberrations (% cells with)					No. of chromosome aberrations (% cells with)				
	SCG	SCB	ICG	ICB	dic $\bar{c}$ ace	dic	ace	t	del	QR
Pre-exposure Sample	72 (0.84)	8 (0.09)	3 (0.04)	1 (0.01)	0 (0.00)	3 (0.04)	0 (0.00)	0 (0.00)	0 (0.00)	1 (0.01)
Post-exposure Sample (6 wks)	120 (1.40)	7 (0.08)	9 (0.11)	5 (0.06)	1 (0.01)	3 (0.04)	1 (0.01)	2 (0.02)	0 (0.00)	2 (0.02)
Post-exposure Sample (12 wks)	94 (1.10)	10 (0.12)	15 (0.18)	5 (0.06)	2 (0.02)	2 (0.02)	2 (0.02)	0 (0.00)	0 (0.00)	3 (0.04)

TABLE 4.

Sample Levels of O_3, NO_x and NO
in the
Welding School, Aberdeen Proving Ground
1975

Ozone levels in ppm

Booth	Date	Front	Right	Back	Left	Top	Ambient
4 (MIG/ TIG)	9/17	0.006	0.006	0.007	0.007	0.007	0.006
12 "	9/17	8.0**	0.006	0.006	0.006	0.007	0.006
62 "	9/17	0.01	0.015	0.005	0.004	0	0
57(ARC)	9/17	0.012	0.012	0.012	0.012	0.012	0.012
9 (TIG)	9/26	off scale (over oil)	0.042	0.004	0.008	0.001	0.001
Arc Rm.	9/26		ALL READINGS ZERO				
Body Shop	9/26		WORK NOT OCCURRING				0.002
30(MIG)	9/26		ALL READINGS ZERO				
3 (TIG)	9/26						0.005*

NO levels in ppm

Booth	Date	Front	Right	Back	Left	Top	Ambient
4 (MIG/ TIG)	9/17	1.8	1.3	1.0	1.0	6.0-8.0	1.0
12 "	9/17	0.1-1.0	0.1	1.2	1.2	1.5	0.4-1.0
65 "	9/17	2.3					1.3
62 "	9/17	1.0	2.5	1.0	1.0	2.0	1.0
57 (Arc)	9/17	1.0	0.5	0.4	0.6	0.8	0.4
9 (TIG)	9/26	0.8-3.2	0.1-0.8	0.2-0.6	0.2-0.4	1.5-3.4	0.1-0.3
Arc Rm.	9/26	3.5-5.4	4.0	3.6	2.0	2.0	2.0-2.5
30 (MIG)	9/26	5.0-6.0	1.2-1.4	2.0	4.8-5.3	1.3	0.5
3 (TIG)	9/26						0.004*

* Background. Work had not started for day.
** Believed to be caused by malfunction of equipment

TABLE 4 (CONTINUED)

NO_x levels in ppm

Booth	Date	Front	Right	Back	Left	Top	Ambient
4 (MIG/ TIG)	9/17	1.5	1.4	1.2	2.8	4.7	1.2
12 "	9/17	6.0-8.0	1.1	1.6	1.7	2.2-3.0	1.5
60 "	9/17	1.5	1.0	1.0	0.8	1.3	0.5
57 (Arc)	9/17	6.5-7.0	0.8	0.7	1.4	1.1	0.7
9 (TIG)	9/26	1.0-6.3	2.7	0.7-1.1	0.5	1.5	0.6
Arc Rm.	9/26	2.0-2.7	2.2-5.2	1.5	1.5-1.7	1.2-1.6	1.5-1.7
30 (MIG)	9/26	4.4-7.1	1.6-3.7	1.0-1.8	1.6-3.0	1.5-2.2	0.5
3 (TIG)	9/26						0.0125*

* Background. Work had not started for day.

TABLE 5.

Incidence of Chromosomal Aberrations:
Analysis of Subjects with Blood Samples 1 and 2

Aberration Type	Percent Yes Blood 1	Percent Yes Blood 2	Difference	p-value for Difference
Single chromatid gaps	51.5	62.4	10.9	0.05
Single chromatid breaks	11.5	9.1	2.4	0.48
Isochromatid gaps	5.4	9.7	4.3	0.12
Isochromatid breaks	1.8	3.6	1.8	0.32
Multicentric-dicentric rings	2.4	6.7	4.3	0.07
Acentric fragments	4.2	5.5	1.3	0.62
Others	1.2	4.8	3.6	0.06

TABLE 6.

Incidence of Chromosomal Aberrations:
Analysis of Subjects with Three Blood Samples

Aberration Type	% Yes Blood 1	% Yes Blood 2	% Yes Blood 3	Difference 1 vs 2	Difference 1 vs 3	P_0 Overall	P_1 1 vs 2	P_2 1 vs 3
Single chromatid gaps	48.1	62.7	60.2	14.6	12.1	0.13	--	--
Single chromatid breaks	9.6	7.2	12.0	2.4	2.4	0.60	--	--
Isochromatid gaps	3.6	12.1	12.1	8.4	8.4	0.03	0.03	0.05
Isochromatid breaks	1.2	4.8	4.8	3.6	3.6	0.21	--	--
Multicentric-dicentric rings	2.4	7.2	4.8	4.8	2.4	0.33	--	--
Acentric fragments	1.2	2.4	6.0	1.2	4.8	0.25	--	--
Others	1.2	3.6	3.6	2.4	2.4	0.51	--	--

APPENDIX A

ID Number

STATEMENT OF PERMISSION

I agree to participate voluntarily in this chromosome study of welder trainees. The purpose of the study has been explained to me. I have been informed that this study is being conducted by doctors at Columbia University, in New York, for the U.S. Environmental Protection Agency. I understand that the study has been approved by the Department of the Army and by the Army personnel in charge of this welder training program at Aberdeen, Maryland.

I understand that a 10 cubic centimeter venous blood sample (a small tube) will be requested of me on three occasions: at the beginning of my participation in the study, and twice thereafter, but that I have the right of refusal each time.

I understand that I may refuse to answer any question in this interview, or the brief interview to be given when I leave the Welding School. I give permission for my Army medical and dental records to be reviewed for illnesses, x-rays, or medicines which might affect my blood.

I understand that no information collected from me will become part of my military record, nor will my items of information collected be released to any individual not working in this study.

_____
Signature

_____
Name (Printed)

APPENDIX B

☐ ☐ ☐
ID Number

CHROMOSOME STUDY IN WELDER TRAINEES
QUESTIONNAIRE (INITIAL)

1. Name _____
(last) (first) (middle)

2a. Date of Birth _____ 2b. Serial Number _____
(day) (month) (year)

3a. Date entered Welding School _____ 3b. Date of Interview _____
(day) (month) (year) (day) (month) (year)

4. On what date did you enter active duty?

_____ _____ _____
Day Month Year

IF GREATER THAN __ MONTHS BEFORE ENTRY INTO WELDING SCHOOL, EXCUSE TRAINEE FROM STUDY

Rejected for length of service ☐

5. Have you had past experience in a welding shop or in a welding course in school?

☐ No ☐ YES, High School
☐ YES, Trade School
☐ YES, Job

Rejected Previous Welding Experience ☐

IF YES, EXCUSE FROM STUDY

_____
Duration

6. Date of first blood sample _____ OR Participation Refused ☐
(day) (month) (year)

YOUR ANSWERS TO THIS QUESTIONNAIRE WILL BE HELD IN STRICT CONFIDENCE. THEY WILL NOT BECOME PART OF YOUR MILITARY RECORD. RESULTS WILL BE SUMMARIZED FOR GROUPS OF PERSONS. NO INFORMATION ON INDIVIDUAL RESPONSES WILL BE RELEASED.

THANK YOU FOR YOUR COOPERATION.

☐ ☐ ☐
ID Number

7. ILLNESS HISTORY

Have you had one of the following illnesses within the last six months:

a. Rash YES ☐ NO ☐
IF YES:
Cause _____

Approximate Dates _____

Was that Before After entering active duty?

b. Infectious Mononucleosis YES ☐ NO ☐
IF YES:
Approximate Dates _____

Was that Before After entering active duty?

c. Hepatitis YES ☐ NO ☐
IF YES:
Approximate Dates _____

Was that Before After entering active duty?

d. Flu YES ☐ NO ☐
IF YES:

Approximate Dates _____

Was that Before After entering active duty?

e. Any other illnesses in the last six months? YES ☐ NO ☐
IF YES:
i. Nature of illness _____

Approximate Dates _____

Was that Before After entering active duty?

ii. Nature of illness _____

Approximate Dates _____

Was that Before After entering active duty?

(Continue on rear of page as necessary)

☐ ☐ ☐
ID Number

8. MEDICATION HISTORY

We need to know about any medicines or drugs that you have taken in the last 6 months, such as aspirin, penicillin, hay fever medicine, or any other drugs. Start with the most recent. NONE ☐

IF YES: Name or type of medication	Approximate Dates	Time in relation to enetering active duty (Circle)
_____	_____	Before After Both
_____	_____	Before After Both
_____	_____	Before After Both

(Continue on rear if necessary)

9a. RECENT X-RAY HISTORY

We also need to know about any x-rays you have had within the last six months, again starting with the most recent. NONE ☐

IF YES:

Part of body x-rayed	Approximate Dates	Time in relation to entering active duty (Circle)
_____	_____	Before After Both
_____	_____	Before After Both
_____	_____	Before After Both

9b-d. PAST RADIATION EXPOSURE

b. Have you ever had radiation therapy? YES ☐ NO ☐

IF YES: Describe suspected medical conditions and test substances if known, and dates of therapy.

_____

_____

_____

_____

☐☐☐
ID Number

c. Have you ever been given radioactive substances for diagnosis or treatment? YES ☐ NO ☐

IF YES: Describe suspected medical conditions and test substances if known, and dates of diagnosis or treatment.

_____

_____

_____

_____

d. Have you ever had x-rays other than routine chest and dental x-rays? YES ☐ NO ☐

IF YES: Describe medical reason, if known, part of body x-rayed, and dates of x-rays.

_____

_____

_____

_____

10. Are there any comments on your general health that you would like to make? Anything you think we should know? NO ☐

Describe: _____

_____

_____

_____

_____

_____

_____

☐☐☐
ID Number

11. How would you describe your ethnic background?

Black/Afro-American ☐

Mexican/Puerto Rican/ Other Latin American ☐

White/Caucasian American ☐

Oriental American ☐

Other American ☐

12. EDUCATION

a. What is the highest grade in school that you have completed? (Exclude any college) _____

b. Have you attended college? YES ☐ NO ☐

IF YES: For how many years? _____

Did you receive your Batchelor's degree? YES ☐ NO ☐

13. Have you attended any trade school after your regular schooling?

YES ☐ NO ☐

IF YES: Describe course, and length of enrollment.

_____

_____

_____

14. What is your age (last birthday) _____

15. a. Do you now regularly smoke cigarettes? YES ☐ NO ☐

IF YES:
b. About how many cigarettes do you smoke per day?

_____

(A pack = 20; record frequency if respondent does not smoke every day)

☐ ☐ ☐
ID Number

c. At what age did you begin smoking cigarettes regularly? (Record age; if respondent has smoked less than a year, also record number of months of smoking to date)

_____

SKIP TO QUESTION 18

16. a. Have you ever regularly smoked cigarettes? YES ☐ NO ☐

IF YES:
b. How long ago did you give up smoking cigarettes?

_____
(Record months if less than 1 year ago, years if longer)

c. How many months or years did you smoke cigarettes?

_____

d. When you were smoking what was the average number of cigarettes you smoked per day?

_____

17. APPLYING THE QUESTIONS ABOVE TO PIPE AND CIGAR SMOKING, CODE:

a. Cigars ☐ Nonsmoker

☐ Smoker _____ No. per day _____ How long smoked

☐ Exsmoker _____ No. per day _____ How long smoked

_____ How long since stopped

b. Pipe ☐ Nonsmoker

☐ Smoker _____ No. per day _____ How long smoked

☐ Exsmoker _____ No. per day _____ How long smoked _____ stopped

TECHNICAL REPORT DATA
(Please read Instructions on the reverse before completing)

EPORT NO. PA-600/1-79-011	2.	3. RECIPIENT'S ACCESSION NO.
ITLE AND SUBTITLE Chromosomal Abnormalities Among Welder Trainees		5. REPORT DATE February 1979
		6. PERFORMING ORGANIZATION CODE
UTHOR(S) Arthur D. Bloom		8. PERFORMING ORGANIZATION REPORT NO.
EPFORMING ORGANIZATION NAME AND ADDRESS Columbia University New York, New York		10. PROGRAM ELEMENT NO. 1AA817
		11. CONTRACT/GRANT NO. Contract No. 68-02-1738
SPONSORING AGENCY NAME AND ADDRESS Health Effects Research Laboratory Office of Research and Development U.S. Environmental Protection Agency Research Triangle Park, NC 27711	RTP,NC	13. TYPE OF REPORT AND PERIOD COVERED
		14. SPONSORING AGENCY CODE EPA 600/11

SUPPLEMENTARY NOTES

ABSTRACT

Serial cytogenetic observations were made on a group of 273 military recruits who ere being trained as welders at Aberdeen, Maryland. The trainees were being exposed to esumably increased levels of ozone in the course of their welding school experience, nd it was the purpose of this study to determine whether or not ozone, at low to mod-ate doses, is capable of inducing chromosomal aberrations in peripheral blood lympho-ytes.

Previous exposures and the past medical experiences of the trainees were determined y questionnaire, enabling us to obtain a profile on the medical-social characteristics f the study sample. Each welder was to serve as his own control, having a blood sample rawn at the beginning of his twelve week training program, prior to ozone exposure, with wo post-exposure bloods being obtained at six and twelve weeks after the start of the rogram.

Ozone levels and the levels of the oxides of nitrogen were determined in the mmediate area of the welding. The ozone levels were repeatedly found to be negligible, ith the nitrogen oxides appearing to be the primary toxic agents involved. For those 65 subjects on whom two blood samples were obtained, and for those 86 on whom all three lood samples were obtained, no statistically significant increases in chromosomal berrations were found.

KEY WORDS AND DOCUMENT ANALYSIS

DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Chromosomal Cytogentic Ozone	Welders Blood samples	06,F T

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