United States
Environmental Protection
Agency

EPA-600/7-82-038b
May 1982

EPA

Research and Development

ENVIRONMENTAL ASSESSMENT OF
A LOW-EMISSION OIL-FIRED
RESIDENTIAL HOT WATER
CONDENSING HEATING SYSTEM
Volume II. Data Supplement

Prepared for

Office of Air Quality Planning and Standards

Prepared by

Industrial Environmental Research
Laboratory
Research Triangle Park NC 27711

RESEARCH REPORTING SERIES

Research reports of the Office of Research and Development, U.S. Environmental Protection Agency, have been grouped into nine series. These nine broad categories were established to facilitate further development and application of environmental technology. Elimination of traditional grouping was consciously planned to foster technology transfer and a maximum interface in related fields. The nine series are:

1. Environmental Health Effects Research
2. Environmental Protection Technology
3. Ecological Research
4. Environmental Monitoring
5. Socioeconomic Environmental Studies
6. Scientific and Technical Assessment Reports (STAR)
7. Interagency Energy-Environment Research and Development
8. "Special" Reports
9. Miscellaneous Reports

This report has been assigned to the INTERAGENCY ENERGY-ENVIRONMENT RESEARCH AND DEVELOPMENT series. Reports in this series result from the effort funded under the 17-agency Federal Energy/Environment Research and Development Program. These studies relate to EPA's mission to protect the public health and welfare from adverse effects of pollutants associated with energy systems. The goal of the Program is to assure the rapid development of domestic energy supplies in an environmentally-compatible manner by providing the necessary environmental data and control technology. Investigations include analyses of the transport of energy-related pollutants and their health and ecological effects; assessments of, and development of, control technologies for energy systems; and integrated assessments of a wide range of energy-related environmental issues.

EPA REVIEW NOTICE

This report has been reviewed by the participating Federal Agencies, and approved for publication. Approval does not signify that the contents necessarily reflect the views and policies of the Government, nor does mention of trade names or commercial products constitute endorsement or recommendation for use.

This document is available to the public through the National Technical Information Service, Springfield, Virginia 22161.

ACUREX TECHNICAL REPORT TR-81-78/EE

ENVIRONMENTAL ASSESSMENT OF A LOW EMISSION OIL-FIRED RESIDENTIAL HOT WATER CONDENSING HEATING SYSTEM

VOLUME II: DATA SUPPLEMENT

April 1982

Acurex Project 7600

Contract 68-02-3188

For

EPA Project Officer - R. E. Hall
Combustion Research Branch
Energy Assessment and Control Division
Industrial Environmenta Research Laboratory
Research Triangle Park, North Carolina 27711

By

C. Castaldini
Acurex Corporation
Energy & Environmental Division
485 Clyde Avenue
Mountain View, California 94042

CONTENTS

SECTION 1

INTRODUCTION

The purpose of this Data Supplement is to document data in greater detail than was practical in the final report (reference 1-1). It is intended to provide sufficient detail for researchers to perform their own analysis of the data. Readers are referred to the final report for objectives, description of the emission results, interpretations, and conclusions.

The data supplement contains the following information:

- Preliminary equipment calibration -- calibration data for the dry gas meters used to sample flue gas for both EPA Method 5 and Source Assessment Sampling System (SASS) trains
- Complete furnace operating data -- furnace water and flue gas temperatures throughout each burner-on/burner-off cycle; cooling water flowrate, and thermostat settings
- Sampling data sheets -- emission data obtained with continuous monitoring instrumentation: operating data tables for EPA Method 5 (for particulate mass emissions), EPA Method 8 (for SO_2 and SO_3 sampling), and SASS (for particulate, trace element, and organic sampling)
- Analytical laboratory results -- ultimate analysis of distillate fuel oil used in the test program: laboratory

analysis reports on particulate levels by gravimetric analysis; sulfur by turbimetric analysis; trace elements by Spark Source Mass Spectrometry (SSMS) analysis and Atomic Absorption (AA) analysis; anions by ion chromatography; Total Chromatographable Organic (TCO) emissions by onsite Gas Chromatography (GC); speciation of organic emissions by Gas Chromatography/Mass Spectrometry (GC/MS); biological assays of the extract and water discharge samples

- SAM IA work sheets -- Source Assessment Model IA evaluation of emission and effluent data to determine the potential hazard posed by both flue gas and water discharge samples

REFERENCE FOR SECTION 1

1-1. Castaldini, C., "Environmental Assessment of a Low-NO_x Residential Hot Water Condensing Heating System Burning Distillate Oil, Volume 1: Technical Results."

SECTION 2

PRELIMINARY EQUIPMENT CALIBRATION

Date 5-7-80

Time 1341

Barometric Pressure 30.16

Ambient Temperature 70°

Orifice Meter

Orifice Magnehelic 70221 9m28

Primary Calibration Meter 433583

Control Module #043

Operators BRANAM

Wet Bulb Temperature

METER CALIBRATION DATA

Orifice Manometer ΔH_{ref} (in.wg.)	Orifice Magnehelic ΔH_0 (in.wg.)	Primary Meter ΔH_p (in.wg.)	Dry Test Meter P_{dg} (in. wg.)	Gas Volume Primary Meter V_p (ft.³)	Gas Volume Dry Test Meter V_d (ft.³)	Temperature: Primary Meter: Inlet, T_{pi} (°F)	Outlet, T_{po} (°F)	Avg., T_{pa} (°F)	Temperature: Dry Test Meter: Inlet, T_{di} (°F)	Outlet, T_{do} (°F)	Avg., T_{da} (°F)	Time t (min.)	α	K_0
	0.2													
	0.4													
	0.6													
	0.8			663.147 679.420	962.379 978.339	74 86	76 76	78	72 72	72 72	72.0	6.0	.994	3.685
	1.0			16.273	15.96									
	1.2													
	1.4													
	1.6													
	1.8			679.420 699.999	978.329 997.547	80 97	76 78	82.75	72 73	72 71	72.0	5.0	.955	3.431
	2.0			20.579	A.208									
	2.2													
	2.4													
	2.6													
	2.8			699.999 718.174	997.548 016.430	87 102	79 81	86.0	73 74	72 72	72.75	4.0	1.072	3.477
	3.0			18.175	18.882									
	3.2													
	3.4													
	3.6													
	3.8			718.174 734.576	016.430 032.699	91 101	82 84	89.5	73 73	73 75	73.5	3.0	1.029	3.994
	4.0			16.402	16.269									
	4.2													
	4.4													
	4.6													
	4.8													
	5.0													

Average	
1.013	3.722

Date 5-7-80 | Orifice Meter | Operators BCANAM

Time 1200 | Orifice Magnehelic 702819 M28 | Wet Bulb Temperature

Barometric Pressure 30.16 | Primary Calibration Meter 433583

Ambient Temperature 70° | Control Module # 043

METER CALIBRATION DATA

Orifice Manometer ΔH_{ref} (in.wg.)	Orifice Magnehelic ΔH_0 (in.wg.)	Primary Meter ΔH_p (in.wg.)	Dry Test Meter P_{dg} (in. wg.)	Gas Volume Primary Meter V_p (ft.³)	Gas Volume Dry Test Meter V_d (ft.³)	Temperature: Primary Meter Inlet, T_{pi} (°F)	Primary Meter Outlet, T_{po} (°F)	Primary Meter Avg., T_{pa} (°F)	Dry Test Meter Inlet, T_{di} (°F)	Dry Test Meter Outlet, T_{do} (°F)	Dry Test Meter Avg., T_{da} (°F)	Time t (min.)	α	K_0
	0.2													
	0.4													
	0.6													
	(0.8)			599.163 614.863	900.115 915.426	66 68	66 68	67.0	68 71	70 71	70.0	31.0	1.029	.699
	1.0			15.1	15.311									
	1.2													
	1.4													
	1.6													
	(1.8)			614.863 630.899	915.426 931.049	70 72	70 72	71.0	68 69	68 69	68.5	21.0	.964	.731
	2.0			16.036	15.623									
	2.2													
	2.4													
	2.6													
	(2.8)			630.899 646.840	931.049 946.554	72 78	72 72	73.5	70 70	70 70	70.0	17.0	.437	.719
	3.0			15.941	15.505									
	3.2													
	3.4													
	3.6													
	(3.8)			646.840 663.148	946.554 962.379	73 82	73 74	75.5	70 70	70 70	70.0	15.0	.991	.716
	4.0			16.308	15.825									
	4.2													
	4.4													
	4.6													
	4.8													
	5.0													
													Average	
													.993	.716

A-7274-1

Date 5-7-80
Time 921
Barometric Pressure 30.02
Ambient Temperature 70°

Orifice Meter
Orifice Magnehelic 5052.3PN54
Primary Calibration Meter 433683
Control Module 081

Operators Branam
Wet Bulb Temperature

METER CALIBRATION DATA

Orifice Manometer ΔH_{ref} (in.wg.)	Orifice Magnehelic ΔH_0 (in.wg.)	Primary Meter ΔH_p (in.wg.)	Dry Test Meter P_{dg} (in. wg.)	Gas Volume Primary Meter V_p (ft.³)	Gas Volume Dry Test Meter V_d (ft.³)	Temperature: Primary Meter: Inlet, T_{pi} (°F)	Primary Meter: Outlet, T_{po} (°F)	Primary Meter: Avg., T_{pa} (°F)	Dry Test Meter: Inlet, T_{di} (°F)	Dry Test Meter: Outlet, T_{do} (°F)	Dry Test Meter: Avg., T_{da} (°F)	Time t (min.)	α	K_0
	0.2													
	0.4													
	0.6													
	0.8 (circled)			531.811 547.623	184.100 799.711	65 65	65 65	65.0	66 74	65 66	67.75	6.0		
	1.0			15.812	15.611									
	1.2													
	1.4													
	1.6													
	1.8 (circled)			547.623 563.126	199.711 215.065	65 66	65 65	65.25	70 74	66 68	69.5	4.0		
	2.0			15.503	15.354									
	2.2													
	2.4													
	2.6													
	2.8 (circled)			563.126 581.891	215.065 233.770	65 66	65 66	65.5	72 78	68 70	72	4.0		
	3.0			18.765	18.705									
	3.2													
	3.4													
	3.6													
	3.8 (circled)			581.891 597.713	233.770 249.665	65 65	65 65	65.0	74 73	66 69	70	3.0	1.023	
	4.0			15.822	15.895									
	4.2													
	4.4													
	4.6													
	4.8													
	5.0													
													Average	

A-21391

SECTION 3

COMPLETE FURNACE OPERATING DATA

FURNACE OPERATING DATA

* EACH ENTRY REPRESENTS ONE MINUTE INTERVAL

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
							1
1	1.7	53	56	68	97	61	76
	↓	↓	56	71	101	63	77
			56	76	107	68	74
			56	80	113	73	75
			56	84	119	76	74
			56	87	124	78	77
			56	90	129	79	77
			56	92	133	80	77
			56	94	136	82	80
			56	97	140	82	79
			56	98	144	83	80
			56	100	147	83	80
			56	103	151	83	77
			56	103	154	83	90
			56	100	153	81	99
			56	95	149	80	98
			56	93	145	78	96
			56	90	141	77	97
			56	88	138	77	97
			56	87	134	77	93
			56	84	132	76	93
			56	83	129	75	93
	↓	↓	56	81	126	74	92

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
6	1.7	53	56	80	124	74	92
			56	79	121	72	90
			56	78	119	72	90
			56	77	117	72	89
			56	75	115	72	89
			56	74	113	72	88
			56	74	111	72	87
			56	73	109	72	87
			56	72	107	72	87
			56	72	106	73	83
			56	71	104	72	85
			56	70	103	73	83
			56	70	101	73	82
			56	69	100	73	84
			56	68	98	73	85
			56	68	97	61	76
2	1.7	53	56	71	100	63	73
			56	77	106	68	71
			56	81	112	73	75
			56	84	118	76	77
			56	87	123	78	75
			56	90	128	79	74
			56	92	132	80	74

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53	56	94	136	81	72
			56	97	140	81	73
			56	98	144	83	69
			55	100	147	83	72
			55	102	151	83	72
			55	104	153	84	72
			55	105	156	85	71
			55	102	156	79	82
			55	98	153	77	80
			55	94	149	73	75
			54	90	145	72	78
			55	88	141	72	74
			55	87	138	71	72
			55	86	135	70	79
			55	83	132	69	73
			54	81	129	68	72
			54	80	126	68	70
			55	78	123	68	69
			54	78	121	67	69
			54	77	119	67	69
			55	76	117	67	69
			54	75	114	67	69
			55	73	112	67	68

FURNACE OPERATING DATA

4

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53	55	72	110	67	69
			55	71	108	67	67
			55	71	107	66	65
			55	70	105	59	65
3	1.7	53.5	54	73	107	62	64
			55	78	113	68	65
			55	83	118	73	66
			55	87	123	77	65
			55	89	128	79	67
			55	91	133	80	65
			55	93	137	81	68
			54	96	141	81	69
			55	98	145	82	68
			55	101	148	83	67
			55	102	151	84	71
			55	104	154	85	68
			55	106	157	86	69
			55	108	160	86	69
			55	109	162	87	75
			56	105	162	83	83
			55	100	157	81	85
			55	97	153	78	84
			55	94	149	76	84

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	55	91	146	74	83
	↓	↓	55	90	142	72	86
			56	88	138	72	79
			55	85	135	70	79
			55	84	132	69	75
			55	81	129	68	76
			55	80	126	68	75
			54	80	124	67	77
			55	77	121	67	76
			55	77	119	66	74
			55	76	117	66	77
			55	74	114	67	77
			55	74	113	67	75
			55	73	111	66	72
			55	72	109	66	70
			55	72	107	66	72
			55	71	106	66	73
	↓	↓	55	70	104	61	68
4	1.7	53.5	55	74	107	64	67
	↓	↓	55	79	113	70	68
			55	83	117	74	68
			55	87	123	77	70
	↓	↓	55	89	128	79	69

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	55	92	132	80	70
	↓	↓	55	94	136	80	71
			55	96	140	82	68
			55	99	145	83	70
			56	100	147	83	72
			56	102	150	84	72
			55	104	153	85	72
			55	106	156	86	71
			55	107	159	87	73
			56	107	160	83	84
			55	101	157	81	88
			56	97	153	79	83
			55	94	149	77	84
			56	92	145	74	80
			55	90	142	73	84
			56	88	138	72	86
			56	86	135	71	86
			55	84	132	71	83
			55	83	129	71	76
			56	82	127	71	80
			56	80	124	71	82
			56	80	121	71	80
	↓	↓	56	79	119	70	79

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	55	77	117	70	78
	↓	↓	56	76	115	70	77
			55	76	113	70	77
			56	74	111	70	79
			55	73	109	70	75
			56	72	107	71	77
			56	72	106	72	74
			56	72	104	71	74
	↓	↓	56	71	103	62	71
5	1.7	53.5	56	75	107	65	70
	↓	↓	56	80	112	71	72
			56	84	117	74	70
			56	87	122	77	70
			56	90	127	79	71
			56	92	132	80	73
			56	94	136	81	75
			56	97	139	82	72
			56	99	143	83	72
			56	100	147	83	74
			56	103	150	85	74
			56	104	153	85	74
			56	107	156	86	77
	↓	↓	56	108	158	86	81

FURNACE OPERATING DATA

8

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	56	104	158	82	87
			56	99	154	79	90
			56	96	150	77	91
			56	93	146	75	92
			56	91	142	74	85
			56	89	139	74	90
			56	87	135	74	89
			56	85	133	73	86
			56	84	130	73	84
			56	83	127	72	84
			56	81	125	71	81
			56	80	122	70	84
			56	79	119	71	81
			56	78	117	69	82
			56	77	115	69	80
			56	76	114	71	82
			56	75	111	72	79
			56	74	110	72	79
			56	73	108	72	77
			56	73	106	71	77
			56	72	104	71	77
			56	72	103	71	78
	↓	↓	56	71	102	62	73

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
6	1.7	53.5	56	73	103	63	73
			56	78	108	68	73
			56	81	113	72	72
			56	85	118	77	73
			56	88	124	78	73
			56	91	128	80	72
			56	93	133	80	74
			56	96	137	82	75
			56	97	141	82	75
			56	100	145	83	77
			56	102	148	83	77
			56	104	151	84	77
			56	105	154	84	75
			56	107	157	85	80
			56	102	156	81	84
			56	99	152	79	90
			56	94	148	77	92
			56	92	145	76	88
			56	90	141	75	79
			55	88	138	74	83
			56	86	134	73	89
			56	84	132	74	88
	↓	↓	56	83	129	74	87

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	56	82	126	73	80
	↓	↓	56	80	124	74	80
			56	80	121	74	82
			56	79	118	74	79
			56	77	117	73	83
			56	77	114	73	80
			56	76	113	73	83
			56	75	110	72	80
			56	74	109	72	80
			56	73	107	72	81
			56	73	106	72	80
			56	72	104	72	81
			56	72	102	73	82
			56	71	101	63	78
	↓	↓	56	72	101	63	77
7	1.7	53.5	57	77	106	68	78
	↓	↓	56	82	111	73	75
			56	86	117	77	73
			57	87	122	79	75
			57	90	127	80	74
			56	93	131	81	75
			56	95	135	82	75
	↓	↓	56	98	140	83	73

FURNACE OPERATING DATA

11

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	56	99	144	83	75
	↓	↓	56	101	146	83	76
			57	103	150	84	76
			57	105	153	85	77
			56	107	155	85	83
			56	103	155	83	89
			56	99	152	81	94
			57	96	149	81	95
			57	93	145	80	93
			57	90	141	80	90
			57	89	137	78	90
			56	87	134	77	92
			56	86	131	75	94
			56	84	129	73	91
			57	83	125	74	89
			56	82	124	73	87
			56	81	121	74	89
			57	80	119	73	91
			56	78	116	73	88
			56	77	114	74	85
			56	77	113	73	84
			56	76	110	72	87
	↓	↓	56	75	109	72	83

FURNACE OPERATING DATA

12

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	57	74	107	72	84
			56	73	105	74	85
			56	72	104	75	83
			57	72	102	75	79
			56	72	101	74	81
			56	71	100	73	80
			56	70	98	63	77
8	1.7	53.5	56	72	100	64	76
			56	77	106	69	80
			56	81	111	73	79
			56	84	117	77	78
			56	87	122	79	77
			57	90	126	80	78
			57	93	131	80	79
			57	95	135	81	78
			56	97	139	82	79
			56	99	143	82	77
			56	101	146	82	78
			56	103	149	83	80
			56	105	152	83	80
			56	103	154	83	91
			56	98	151	81	92
			57	94	147	80	86

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	53.5	56	92	143	78	91
			56	90	139	77	96
			57	88	136	77	90
			56	87	133	77	94
			57	85	131	75	88
			56	84	128	76	87
			57	83	125	77	87
			56	81	123	76	77
			57	80	120	74	93
			56	79	118	73	85
			56	77	116	72	86
			56	77	114	74	88
			57	76	112	73	88
			57	76	110	72	83
			57	75	109	73	83
			57	74	107	75	86
			57	73	105	74	77
			57	72	104	73	79
			56	72	103	72	81
			57	71	101	73	81
			56	70	100	67	80
	↓	↓	57	71	99	63	80
9	1.7	55	57	75	103	66	82

FURNACE OPERATING DATA

14

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	80	108	71	80
	↓	↓	57	83	114	76	80
			56	87	119	77	82
			56	89	124	79	79
			56	92	129	80	80
			57	94	133	81	79
			56	96	137	81	79
			56	99	141	81	78
			57	100	144	82	79
			56	103	148	83	81
			57	105	150	84	84
			57	101	151	82	87
			57	97	148	81	93
			56	94	145	80	90
			57	91	140	79	91
			57	90	137	78	90
			57	88	134	77	93
			57	87	131	77	93
			57	85	129	77	85
			57	83	126	77	90
			56	81	123	77	83
			56	80	121	75	80
	↓	↓	56	79	119	75	79

FURNACE OPERATING DATA

15

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	79	117	74	85
			56	77	114	75	82
			57	76	112	74	85
			57	76	110	74	85
			57	75	109	74	88
			57	74	107	76	77
			56	73	106	76	77
			56	72	104	76	78
			56	72	102	74	78
			56	71	101	74	78
			56	71	100	75	82
	↓	↓	57	70	99	63	77
10	1.7	55	56	72	100	64	75
			56	77	106	69	77
			57	82	112	73	77
			57	85	117	76	78
			56	88	122	77	78
			56	90	126	79	78
			57	92	131	80	78
			56	95	134	81	77
			57	97	138	81	77
			56	94	142	81	78
	↓	↓	56	101	145	82	80

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	103	148	83	80
			56	104	152	84	78
			56	105	154	82	84
			56	100	153	80	82
			56	96	149	79	84
			56	93	144	79	89
			56	90	141	78	83
			56	90	141	78	84
			56	90	141	78	82
			56	90	140	77	82
			56	90	140	77	82
			56	90	138	77	83
			56	87	135	77	82
			56	86	132	76	80
			56	83	130	75	77
			55	82	127	75	77
			56				
			56 TAPE	80 PROBLEM			
			56	79			
			56	78			
			56	77	115	73	77
			56	77	114	72	76
	↓	↓	56	76	113	72	77

FURNACE OPERATING DATA

17

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	75	110	72	77
	↓	↓	56	74	109	72	75
			56	73	107	73	74
			56	72	105	72	74
			56	72	104	72	78
			56	71	103	73	76
	↓	↓	56	70	101	62	74
11	1.7	55	56	74	105	64	74
	↓	↓	56	79	110	68	75
			56	83	114	72	74
			56	86	120	75	75
			56	88	124	76	76
			56	90	128	77	77
			56	93	133	78	75
			56	95	137	79	77
			56	98	141	80	76
			56	100	145	81	77
			56	102	148	81	79
			57	104	150	82	78
			56	105	153	83	80
			56	104	155	81	81
			56	99	153	80	81
	↓	↓	56	95	149	78	83

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (^{0}C)	Inlet Water Temperature ^{0}F	Outlet Water Temperature ^{0}F	Tank Water Temperature ^{0}F	Exhaust Duct Temperature ^{0}F	Ambient Temperature ^{0}F
	1.7	55	56	93	145	77	80
			56	90	141	77	82
			56	89	138	77	87
			56	87	135	76	81
			56	85	132	75	80
			56	84	129	75	80
			56	83	127	75	80
			56	81	124	75	78
			56	80	121	74	78
			56	79	119	75	83
			56	78	117	75	83
			56	77	115	75	85
			56	77	113	76	84
			56	75	111	77	83
			56	74	109	77	86
			56	74	108	77	86
			56	74	107	78	87
			57	73	105	79	84
			56	72	103	76	85
			56	72	101	79	84
			57	71	100	78	87
			57	71	99	72	84
	↓	↓	57	71	98	63	83

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
12	1.7	55	57	74	101	67	80
			56	80	107	71	82
			56	83	112	74	82
			57	87	117	77	84
			57	89	122	78	83
			57	91	127	79	84
			56	93	131	79	86
			57	95	134	80	86
			57	97	138	80	87
			57	100	142	81	84
			57	99	145	82	91
			57	94	143	81	95
			57	91	140	79	91
			57	89	137	80	92
			57	87	133	79	94
			57	86	130	79	93
			56	84	128	80	90
			57	83	124	81	91
			56	81	122	78	93
			57	80	120	77	91
			57	79	118	80	90
			57	78	116	80	90
	↓	↓	57	77	114	80	91

FURNACE OPERATING DATA

20

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	57	76	112	80	90
	↓	↓	56	76	110	77	87
			57	75	108	80	89
			57	74	107	78	86
			57	74	105	80	86
			57	72	104	78	87
			57	72	102	80	85
			57	72	100	79	85
			57	71	100	80	85
			57	71	98	77	83
			57	70	97	79	84
			57	70	96	77	83
			56	69	95	79	86
			57	69	93	69	83
	↓	↓	57	69	93	64	82
13	1.7	55	57	74	98	67	80
	↓	↓	57	78	104	70	83
			56	81	110	73	82
			57	85	115	76	80
			57	87	119	77	80
			57	89	124	77	80
			57	91	129	79	83
	↓	↓	57	93	133	79	81

27

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	96	136	80	81
	↓	↓	56	98	140	80	83
			57	100	144	81	83
			56	102	147	82	87
			56	102	149	83	100
			56	97	148	83	104
			57	93	144	83	102
			57	91	140	81	99
			56	89	137	82	100
			56	87	134	82	100
			56	85	131	82	98
			57	83	128	81	89
			57	82	125	81	88
			56	81	123	81	89
			57	80	121	81	92
			56	79	119	80	87
			57	77	116	79	86
			56	77	114	77	86
			57	76	112	79	86
			57	75	110	75	87
			57	75	109	78	85
			56	73	107	78	84
	↓	↓	56	73	105	79	84

FURNACE OPERATING DATA

22

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	72	104	76	86
	↓	↓	57	71	102	78	86
			57	71	101	80	86
			56	70	100	80	87
			56	70	98	80	86
			56	69	97	80	87
			56	69	96	80	87
	↓	↓	56	68	95	65	82
14	1.7	55	57	71	97	67	81
	↓	↓	56	76	103	70	81
			57	80	109	73	81
			57	83	113	76	80
			56	86	119	77	81
			57	87	124	78	83
			56	90	128	79	84
			56	92	132	80	84
			56	94	135	80	83
			57	96	139	81	84
			56	99	143	82	85
			57	101	147	82	83
			57	102	149	83	93
			57	97	148	83	93
	↓	↓	57	94	144	83	93

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	57	91	141	81	89
			57	88	137	81	90
			56	87	134	80	86
			56	85	131	80	89
			56	83	128	78	87
			57	83	126	78	92
			56	81	123	79	89
			57	80	121	77	86
			56	79	118	77	86
			56	78	116	76	87
			57	77	114	76	87
			57	76	112	76	85
			57	75	110	76	87
			57	75	109	77	86
			57	73	107	80	87
			57	73	106	81	87
			57	72	104	81	88
			56	72	103	82	88
			57	71	101	80	87
			56	71	100	80	85
			57	70	98	81	85
	↓	↓	57	69	97	71	81
15	1.7	55	56	71	99	69	80

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	76	104	72	81
			56	80	109	74	81
			57	84	114	77	82
			57	87	120	78	81
			56	88	124	78	82
			56	91	128	79	82
			56	93	133	80	82
			57	95	136	81	81
			57	97	140	82	83
			57	99	144	82	82
			56	100	146	82	82
			56	100	149	83	90
			56	96	147	83	98
			56	93	144	82	94
			57	90	141	81	93
			56	88	137	80	92
			57	87	133	82	93
			57	84	131	79	93
			56	83	128	79	90
			56	82	125	79	91
			57	81	123	80	91
			57	80	121	79	87
	V	V	56	78	118	80	86

FURNACE OPERATING DATA

25

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	79	116	80	91
			56	77	114	80	91
			57	76	112	80	89
			57	75	110	81	89
			56	75	108	82	87
			56	74	107	82	90
			56	73	105	81	91
			56	72	104	82	88
			57	72	103	82	87
			57	71	101	81	90
			57	71	100	71	83
16	1.7	55	57	73	102	71	81
			57	78	107	73	81
			57	83	112	76	81
			56	85	117	78	83
			56	88	122	79	83
			56	90	128	80	83
			56	93	131	80	83
			56	94	135	80	83
			57	96	138	81	84
			57	98	142	82	84
			57	100	145	82	85
			56	103	148	83	84

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	101	151	83	95
			57	97	148	83	91
			57	93	146	81	92
			56	91	141	81	89
			57	89	138	81	90
			56	87	135	80	87
			56	86	132	79	87
			56	84	128	80	87
			57	83	127	80	85
			56	82	124	80	89
			57	80	121	80	87
			57	80	119	81	86
			57	78	117	82	88
			57	77	115	82	87
			56	77	113	82	87
			56	76	111	83	86
			57	75	109	82	85
			56	74	107	82	88
			56	73	106	82	87
			56	72	104	83	87
			56	72	103	82	86
			57	71	102	82	87
	↓	↓	57	71	100	82	86

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	57	70	99	72	83
17	1.7	55	57	73	100	70	82
			56	78	106	72	83
			57	82	110	75	83
			56	85	116	77	84
			57	87	121	79	83
			57	90	125	80	83
			57	92	129	80	82
			56	93	133	80	84
			56	96	138	81	83
			57	97	141	81	83
			57	99	144	82	83
			57	102	148	82	83
			57	102	150	83	93
			56	97	148	83	92
			57	94	145	81	92
			57	91	141	80	91
			56	90	138	80	92
			57	88	135	79	90
			56	87	132	79	91
			57	84	129	80	89
			56	83	127	82	87
	V	V	57	81	124	83	89

FURNACE OPERATING DATA

21

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	57	81	122	83	89
			56	79	119	83	87
			57	78	117	82	87
			56	77	115	81	85
			57	77	113	81	84
			57	76	111	81	83
			56	74	109	81	82
			56	73	107	81	82
			56	73	105	81	80
			56	72	104	80	82
			56	72	103	80	80
	↓	↓	56	71	101	70	79
18	1.7	55	56	73	103	68	78
			56	78	108	72	77
			56	82	113	75	77
			56	85	118	76	77
			56	87	123	78	78
			56	90	128	78	78
			56	92	131	79	78
			56	94	136	80	79
			55	96	138	80	79
			56	98	143	80	77
	↓	↓	56	100	146	80	77

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	101	149	81	82
			56	104	151	82	81
			56	105	155	83	80
			56	106	157	83	83
			56	105	158	83	90
			55	103	158	83	91
			56	99	154	81	92
			56	96	150	80	90
			56	93	146	78	88
			56	91	143	77	89
			56	89	139	77	91
			56	87	136	77	83
			56	86	133	77	86
			56	83	130	77	85
			56	83	128	77	86
			56	81	125	77	86
			56	80	122	77	83
			56	79	120	77	81
			56	78	118	77	82
			56	77	116	77	79
			56	76	114	77	80
			56	76	112	77	82
	↓	↓	56	74	110	78	82

FURNACE OPERATING DATA

30

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (°C)	Inlet Water Temperature °F	Outlet Water Temperature °F	Tank Water Temperature °F	Exhaust Duct Temperature °F	Ambient Temperature °F
	1.7	55	56	74	108	78	82
	↓	↓	56	73	107	75	80
	↓	↓	56	73	105	68	76
19	1.7	55	56	76	109	69	76
	↓	↓	56	81	114	74	78
	↓	↓	56	86	120	77	78
	↓	↓	56	89	124	80	78
	↓	↓	56	92	130	81	77
	↓	↓	56	95	134	82	80
	↓	↓	56	98	138	83	79
	↓	↓	56	100	142	84	80
	↓	↓	56	101	146	83	88
	↓	↓	56	96	145	82	90
	↓	↓	56	92	142	81	89
	↓	↓	56	90	138	80	87
	↓	↓	56	88	135	79	90
	↓	↓	56	86	131	79	87
	↓	↓	56	84	128	78	88
	↓	↓	56	83	126	77	85
	↓	↓	56	82	124	77	86
	↓	↓	56	80	121	77	86
	↓	↓	56	79	118	77	84
	↓	↓	56	78	117	76	82

FURNACE OPERATING DATA

Cycle No.	Cooling Water Flowrate (gpm)	Thermostat Setting (^{0}C)	Inlet Water Temperature ^{0}F	Outlet Water Temperature ^{0}F	Tank Water Temperature ^{0}F	Exhaust Duct Temperature ^{0}F	Ambient Temperature ^{0}F
	1.7	55	56	77	115	75	83
	↓	↓	56	76	112	75	80
			56	75	111	77	82
			56	74	109	75	80
			56	74	107	75	83
			57	72	105	74	79
			57	72	104	74	79
			56	72	103	75	79
			56	72	102	76	78
			56	71	100	76	78
			57	70	99	76	78
			57	70	98	76	77
			56	69	96	76	77
			57	69	95	76	77
			56	68	94	68	76
			57	68	93	66	75
			57	68	92	65	76
			57	68	91	64	75
	↓	↓	57	67	91	63	75
			39801	59881			

SECTION 4

SAMPLING DATA SHEETS

FIELD DATA SHEET

ARIREX Aerotherm

Plant RESIDENTIAL WATER HEATER WITH M.A.N. Burner
Location ACUREX COMBUSTION LABORATORY
Operator CASTALDINI
Date 5-21-80
Load Range CAPACITY
Thermostat setting at 53-55 °C
Water flow rate ~ 1.7 gpm

Ambient Temperature 73-75 °F Room air close to Burner
Barometric Pressure
Static Pressure Duct
Fuel DISTILLATE OIL

Sheet No. 1/8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	CO_2 %	CO ppm	SO_2 ppm	NO ppm	NO_x ppm	NO_2 ppm	CORRECTED TO 0% O_2 DRY NO ppm	NO_x ppm	NO_2 ppm	HYDROCARBONS (HC) Total HC	Methane	Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
6:18	Exhaust			2.1	12.4	165	—	68	68	0				Not				60.7	Start of test - cycle No 1
6:20	Duct			2.0	12.5	20		70	70	0				Working				63.0	
6:22				1.9	12.7	15		72	72	0								73.3	Burner on time = 13 min
6:24				1.85	12.7	15		72	72	0								78.3	
6:28				1.9	12.7	10		72	72	0								81.7	
6:32				1.85	12.7	10		72	72	0								83.4	Burner off
6:58				2.05		150		66	66	0								68	Start of Cycle No. 2
7:00				2.0		35		68	68	0								72.9	
7:02				2.0		35		70	70	0								76.3	Burner on time = 14 min
7:04				2.0		35		70	70	0								78.6	
7:06				2.0		30		72	72	0								84.3	
7:10				2.1		30		72	72	0								84.3	
7:12				1.95		25		72.5	72.5	0								84.7	Burner off
7:32				2.2	12.3	150		68	68	0				1.5				62.1	Start of Cycle No. 3
7:36				2.13	12.7	25.2		68	68	0				1.2				78.7	
7:37				2.08	13.0	23.9		67.5	67.5	0				0.8				79.6	
7:39				2.05	13.3	24.1		70	70	0				1.0				80.8	Burner on time = 5 min
7:40	▼			2.07	12.5	24.4		69	69	0				0.9				82.1	
7:41				2.09	13.1	22.9		69	69	0				0.8				82.5	
7:42				2.02	12.7	24.8		70	70	0				0.7				83.8	
																			AVERAGE

a - As C_3H_8

Form EED-063 12/78

FIELD DATA SHEET

AUREX Aerotherm

Plant RESIDENTIAL WATER HEATER WITH H.A.N. Burner
Location Aurex Combustion Laboratory
Operator CASTALDINI
Date 5-21-80
Load Range Capacity
Thermostat setting at 53-55°C
WATER Flow = 1.7 gpm

Ambient Temperature 74-87 °F Room air
Barometric Pressure
Static Pressure Duct
Fuel Distillate oil

Sheet No. 2/3

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	DRY UNCORRECTED CO_2 %	DRY UNCORRECTED CO ppm	DRY UNCORRECTED SO_2 ppm	DRY UNCORRECTED NO ppm	DRY UNCORRECTED NO_x ppm	DRY UNCORRECTED NO_2 ppmn	CORRECTED TO 3% O2 DRY NO ppm	CORRECTED TO 3% O2 DRY NO_x ppm	CORRECTED TO 3% O2 DRY NO_2 ppm	HYDROCARBONS (HC) Total HC[2]	Methane	Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
7:47	Exhaust Duct			2.0	12.7	25		71	71	0				0.5				87.3	Burner off
8:09				2.5	12.6	150		60	60	0				3				63.9	Start of Cycle No 4
8:10				2.47	12.6	30		59	59	0				3				69.7	
8:11				2.21	12.8	28		65	65					2.6				74.2	
8:12				2.18	12.6	23		63	63					2.0				76.9	Burner on
8:13				2.16	13.1	21		68	68					1.5				78.2	time = 15 min
8:14				2.14	12.9	23.4		70	70					1.5				80.1	
8:15				2.11	12.6	22		70	70					1.4				80.4	
8:16				2.07	13.0	23		70	70					1.2				81.7	
8:17				2.05	13.0	20		71	71					0.9				83.0	
8:24				2.0	13.0	20		72	72					0.8				83.4	Burner off
8:46				2.25	12.9	150		66.5	66.5	0				—				65.2	Start of Cycle No 5
8:50				2.2	12.7	25		67	67					—				78.2	
8:52				2.15	12.7	20		68	68					—				80.8	Burner on
8:56				2.05	12.8	20		69	69					—				84.7	time = 14 min
8:58				2.1	12.9	20		69	69					—				85.6	
9:00				1.95	12.9	20		69.5	69.5					—				85.6	Burner off
														—					
																			AVERAGE

[2] As C_3H_8

Form EED-063 12/78

FIELD DATA SHEET

UREX Aerotherm

Plant: Residential Water Heater with M.A.N. Burner
Location: Aerotherm Combustion Laboratory
Operator: Castaldini
Date: 5-21-80
Load Range: Capacity
Thermostat setting at 53-55°C (127-131)
Water flow = 1.7 gpm

Ambient Temperature: 73-83 Room Air
Barometric Pressure:
Static Pressure Duct:
Fuel: Distillate 0.1

Sheet No. 3 /8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	DRY UNCORRECTED CO_2 %	DRY UNCORRECTED CO ppm	DRY UNCORRECTED SO_2 ppm	DRY UNCORRECTED NO ppm	DRY UNCORRECTED NO_x ppm	DRY UNCORRECTED NO_2 ppm	CORRECTED TO 0% Stk O2 DRY NO ppm	CORRECTED TO 0% Stk O2 DRY NO_x ppm	CORRECTED TO 0% Stk O2 DRY NO_2 ppm	HYDROCARBONS (HC) Total HC[2]	HYDROCARBONS (HC) Methane	HYDROCARBONS (HC) Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
9:24	Exhaust			2.20	12.6	165		65	65	0								63.4	Start of Cycle No. 6
9:25	Duct			2.07	12.7	41.7		65.6	65.6	0				5.1				67.7	
9:26				2.03	12.9	42.8		67	67					5.0				68	
9:27				2.06	12.7	42.4		68.7	68.7					4.4				72.4	
9:28				2.07	12.8	42		68.6	68.6					4.1				76.5	Burner
9:29				2.01	12.6	41		67.3	67.3					3.3				78.3	on
9:30				2.07	12.6	39.7		69.7	69.7					3.1				78.6	time = 13 min
9:31				1.97	13.4	42.2		70.4	70.4					2.6				80.4	
9:32				1.97	12.7	40.4		70.6	70.6					2.9				81.7	
9:33				2.01	12.8	40.3		69.2	69.2					2.7				83	
9:37				1.95	12.8	40		71	71					-				83.8	Burner off
10:01				2.25	12.6	165		60	60									68.4	Start of Cycle No. 7
10:04				2.23	12.9	40.6		63	63					3.6				76.5	
10:05				2.12	12.6	40.0		65.2	65					2.9				79.2	Burner on
10:07				2.07	13.0	38.9		65.3	65					2.2				80.8	time = 13 min
10:10				2.06	13.0	40.5		67.6	68					1.6				83.0	
10:11				2.01	12.8	42.5		67.0	67					1.4				83.0	
10:14				1.95	12.9	42.5		67.5	67.5					-				85.2	Burner off
																			AVERAGE

a - As C_3H_8

Form EED-063 12/78

FIELD DATA SHEET

AUREX Aerotherm

Plant RESIDENTIAL WATER HEATER WITH M.A.N. BURNER
Location AUREX COMBUSTION LABORATORY
Operator CASTALDINI
Date 5-21-80
Load Range CAPACITY
Thermostat Setting at 53-55°C
Water Flow Rate = 1.7 gpm

Ambient Temperature 76-86 °F ROOM AIR
Barometric Pressure
Static Pressure Duct
Fuel DISTILLATE OIL

Sheet No. 4 /8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	DRY UNCORRECTED CO_2 %	DRY UNCORRECTED CO ppm	DRY UNCORRECTED SO_2 ppm	DRY UNCORRECTED NO ppm	DRY UNCORRECTED NO_x ppm	DRY UNCORRECTED NO_2 ppm	CORRECTED TO 0% O2 DRY NO ppm	CORRECTED TO 0% O2 DRY NO_x ppm	CORRECTED TO 0% O2 DRY NO_2 ppm	HYDROCARBONS (HC) Total HC[2]	HYDROCARBONS (HC) Methane	HYDROCARBONS (HC) Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
10:39	Exhaust			1.75	12.6	200		65	65	0				-				64	Start Cycle No.8
10:43	Duct			1.75	12.6	42.5		67	67					1.8				77	Burner on time = 14 min
10:44				1.75	12.7	41.3		70	70					1.7				79	
10:47				1.67	13.4	41.8		69.7	69.7					1.0				80.4	
10:53				1.65	13.4	42		72	72					-				83.4	Burner off
11:17				2.1	12.6	165		64.2	64.2					-				66	Start Cycle No.9
11:18				2.1	12.6	48		64	64					4.1				71.1	
11:19				1.98	13.2	48		68.6	68.6					3.1				75.6	
11:20				2.01	12.9	44		-	67.5					3.4				77.4	Burner on time = 12 min
11:21				1.98	13.0	44		-	70.6					2.0				79.2	
11:22				1.97	12.9	42		-	70.9					1.7				80.1	
11:23				1.91	12.9	44.7		-	70.6					1.4				80.8	
11:24				1.86	13.1	44.2		-	69.8					1.2				81.2	
11:25				1.84	13.2	44.2		-	72.2					0.4				81.2	
11:26				1.98	13.3	42.8		-	71.1					0.3				82.1	
11:29				1.75	13.6	43		-	72.0					0.2				82.1	Burner off
																			AVERAGE

[2] As C_3H_8

Form EED-083 12/78

FIELD DATA SHEET

ACUREX Aerotherm

Plant: Residential Water Heater with M.A.N Burner

Location: Acurex Combustion Laboratory

Operator: Castaldini

Date: 5-21-80

Load Range: Capacity

Thermostat Setting at 53-55 °C

Water flow rate = 1.7 gpm

Ambient Temperature: 77-83 °F Room Air close to Burner

Barometric Pressure:

Static Pressure Duct:

Fuel: Distillate oil

Sheet No. 5/8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	Dry Uncorrected							Corrected to 0% O2 Dry			Hydrocarbons (HC)			H_2O %	Stack Gas Temp °F	Comments
				O_2 %	CO_2 %	CO ppm	SO_2 ppm	NO ppm	NO_x ppm	NO_2 ppm	NO ppm	NO_x ppm	NO_2 ppm	Total HC[a]	Methane	Non-Methane HC			
11:53	Exhaust			2.2	13.0	165		–	65	0				–				63.9	Start Cycle No. 10
11:56	Duct			1.99	13.4	46.4		–	67.8	↓				2.4				76	
11:58	↓			1.96	13.3	44.7		–	70.3					1.0				79	Burner on
12:00				1.99	13.0	42.9		–	71.3					0.7				81	time = 13 min
12:02				1.84	13.2	45.3		–	71.9					0.4				81.2	
12:06														–				82	Burner off
12:29				2.0	12.2	155		–	66					–				64.3	Start Cycle No 11
12:30				1.97	12.5	38.6		–	67.6					2.0				68.4	Burner on
12:32				1.72	12.6	38.8		–	69.1					2.0				75.1	time = 13 min
12:39				1.65	12.9	36.3		–	73					0.9				80.8	
12:42				1.50	13.0	75		–	74					–				83	Burner off
13:08				1.95	12.5	150		–	66					–				66.6	Start Cycle No. 12
13:09				1.91	12.9	40.3		–	65					5.5				70.6	
13:10				1.91	13.2	36		–	67.4					3.8				73.8	Burner on
13:11				1.83	12.9	34.8		–	69.4					2.6				76.5	time = 10 min
13:12				1.82	12.7	33.8		–	71.5					2.0				77.8	
13:13				1.73	13.0	34.5		–	72.7					1.7				78.7	
13:14				1.72	13.2	34.3		–	72.8					2.3				79.2	
13:15				1.63	12.8	34.7		–	73.8					2.5				79.0	
13:18				1.45	13.0	38		–	75					–				81.7	Burner off
	↓									↓									
																			AVERAGE

a - As C_3H_8

Form EED-083 12/78

FIELD DATA SHEET

AUREX Aerotherm

Plant: RES. DEST. GAS WATER HEATER WITH MAN BURNER
Location: Aurex Corporate Laboratory
Operator: CASTAGNOLI
Date: 5-21-80
Load Range: Capacity
Thermostat Setting at 53-55 °C
Water flow rate = 1.7 gpm

Ambient Temperature: 82-90 °F Room Air close to Burner
Barometric Pressure:
Static Pressure Duct:
Fuel: Distillate oil

Sheet No. 6 /8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	CO_2 %	CO ppm	SO_2 ppm	NO ppm	NO_x ppm	NO_2 ppm@	CORRECTED TO 0% 2lb O2 DRY NO ppm	NO_x ppm	NO_2 ppm	HYDROCARBONS (HC) Total HC[3]	Methane	Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
13:46	Exhaust			1.65	12.5	145		-	69	O				-				66.6	Start Cycle No 13
13:50	Duct			1.81	12.9	34.1		-	69.5					4.9				76.9	Burner on time = 13 min
13:54				1.43	13.5	42.5		-	73.4					5.5				80.1	
13:56				1.28	14.0	46.5		-	73.2					5.7				81.2	
13:59				1.62	13.3	45.3		-	70.1					5.5				83	Burner off
14:25				2.0	12.5	145			62.5					-				66.6	Start Cycle No 14
14:27				1.98	12.8	38.9		-	63.6					8				73.3	Burner on time = 12 min
14:29				1.86	12.7	37.3		-	66.6					6.7				77.4	
14:33				1.60	13.0	40.5		-	67.1					3.8				80.1	
14:37				-	-	-			-					-				83	Burner off
15:02														-				69.3	Start Cycle No 15
15:04				1.71	12.7	-		-	65					4.9				71.5	Burner on time = 12
15:06				1.74	12.6	-		-	67.9					3.8				78.3	
15:08				1.67	13.2	-		-	70.1					3.2				79.2	
15:11				1.41	13.2	135	?	-	73.6					1.6				82.1	
15:13				1.45	13.0	131	!	-	72.0					2.6				82.1	
15:14														-				83.4	Burner off
																			AVERAGE

2- As C_3H_8

Form EED-083 12/78

FIELD DATA SHEET

AUREX Aerotherm

Plant Residential Water Heater with MAN Burner
Location Aerotherm Combustion Laboratory
Operator CALIFANO
Date 5-21-80
Load Range Capacity ~ 0.51 gph
Thermostat Setting 54°C
Water flow rate = 1.7 gpm

Ambient Temperature 81 - 83 °F Room Air closed to Burner
Barometric Pressure
Static Pressure Duct
Fuel Distillate Oil

Sheet No. 7/8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	CO_2 %	CO % ppm	SO_2 ppm	NO ppm	NO_x ppm	NO_2 ppm	CORRECTED TO 0% O2 DRY NO ppm	NO_x ppm	NO_2 ppm	HYDROCARBONS (HC) Total HC[2]	Methane	Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
15:37	Exhaust			2.39	12.8	135		-	62.7	0				5.2				71	Start Cycle No. 16
15:40	Duct			2.02	13.0	29.8		-	68.0					3.7				78	
15:42				1.97	12.6	28.8		-	67.7					3.2				80.1	Burner on time
15:44				1.86	13.0	24.7		-	71.0					1.9				82.4	= 12 min
15:46				1.81	13.0	23.5		-	71.8					2.3				81.7	
15:48				1.63	13.2	25		-	68.5					-				82.5	
15:49				1.50	13.4	25		-	71.0					-				83.7	Burner off
16:13				2.0	12.5	135			67.5					-				69.7	Start Cycle No. 17
16:15				1.78	12.7	15.2		-	66.2					6.1				75.1	
16:17				1.70	13.2	14.3		-	69.9					5.7				78.7	Burner on
16:19				1.84	13.1	10.7		-	71.1					4.6				78.6	time = 12
16:21				1.70	13.1	11.0		-	73.2					3.9				80.1	
16:23				1.58	12.9	12.5		-	74.5					3.2				81.2	CO calibration off
16:25				1.25	13.6	18			75.0					-				82.5	
16:47				1.95	12.5	155		-	68					-				68.4	Start Cycle No 18
16:50				1.87	12.6	38		-	68.4					4.4				76	Burner on
16:52				1.90	12.8	376		-	72					3.1				78.3	time = 16 min
16:54				1.64	13.0	38.4		-	72.7					2.8				78.6	
16:56				1.44	13.2	37.5		-	74					2.3				80.4	
17:03	↓			1.45	13.2	3.0		-	76	↓				-				83.4	Burner off
																			AVERAGE

[2] As C_3H_8

Form EED-083 12/78

FIELD DATA SHEET

UREX Aerotherm

Plant Residential Water Heater with Miten Burner
Location Aerotherm Combustion Laboratory
Operator Castaldini
Date 5-21-80
Load Range Capacity ~ 0.5 gph
Thermostat setting = 54°C
Water flow rate = 1.7 gpm

Ambient Temperature 81-83 °F Room Temp
Barometric Pressure close to burner
Static Pressure Duct
Fuel Distillate oil

Sheet No 8/8

Time	Sample Point	Load MW	Steam Flow lb/hr x1000	DRY UNCORRECTED O_2 %	CO_2 %	CO ppm	SO_2 ppm	NO ppm	NO_x ppm	NO_2 ppm	CORRECTED TO 3% O2 DRY NO ppm	NO_x ppm	NO_2 ppm	HYDROCARBONS (HC) Total HC[2]	Methane	Non-Methane HC	H_2O %	Stack Gas Temp °F	COMMENTS
17:23	Exhaust			0.5		165		-	82	0									Start Cycle No 19
17:25	Duct			0.5		35		-	82.5	0									Burner on time = 6 min
17:27				0.25		31,640		-	42.9	↓									Burner off
17:29				0.00		16,643		-	86	↓									Problem with burner shut off because of explosion operation
		During cycle No. 19 fuel pressure was reduced to																	
		135 psig causing a reduction in CO but																	
		burner operation determinated then test was terminated																	
	Oil	weight at start of test = 38.2 lbs																	
		at end of test = 23.5 lbs																	
		Oil used = 14.7 lbs																	
		Total Minutes burned = 242 min																	
		Burning rate = 0.0607 lbs/min = 3.645 lb/hr																	
																			AVERAGE

2 - As C_3H_8

Form EED-083 12/78

ACUREX Corporation

PARTICULATE TEST FIELD DATA SHEET

Plant ACUREX — Barometric Pressure 29.87

Date 5-17-80 — Static Pressure +1.25 in. H2O

Test Location FURNACE MAIN RES' HEATER — Stack Pressure 29.89

Run Number #1 HVSS — Probe Number 3' glass

Stack Diameter inches 4" — Pitot Coefficient —

Duct Dimensions in. x in. — — Pitot Number —

Start Time 0621 — Meter Box Number O

Operator JARMAN — Orifice Coefficient K° 3.722/

Nozzle Size & Number 1.493

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.
B-5(mb)		

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
250	277	+27			
250	314	+64			
0	40	+40			
		1st & 2nd	Silica gels 68.9	gain	
SILICA GEL					

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ΔP in. wg. from	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT	PUMP VACUUM in. Hg	√ΔP	BEGIN LEAK RATE .003 @ 15hg.
0621	START	0	210	.78	347.831	63	247			245	74	74	2.5		
		5	206	.76	360.0	78	251			247	77	75	3		
		10	205	.76	372.13	78	254			249	77	75	3		
	STOP	12.9			379.881										
0659	START	12.9	215	.78	379.581	64	251			252	76	76	3		
		17.9	200	.77	392.35	77	251			251	82	76	3		
		22.9	205	.76	405.16	82	255			251	90	77	3		
	STOP	27.3			416.015										
0734	START	27.3	205	.78	416.015	62	261			252	78	77	3		
		32.3	215	.76	428.79	79	255			251	84	77	3		
		37.3	210	.77	441.48	83	260			254	92	79	3		
	STOP	42.0			453.141										
0810	START	42.0	210	.79	453.141	62	277			252	80	79	3		
		47.0	200	.77	465.85	77	260			253	85	79	3		
		52.0	200	.77	478.41	82	254			251	93	80	3		
		56.7			490.384	85					97	81			
0846	START	56.7	200	.78	490.384	66	269			251	80	80	3		
		61.7	200	.77	503.14	77	253			252	86	79	3		
		66.7	210	.77	515.77	82	250			251	92	80	3		
	STOP	71.4			527.752										
	AVG/TOTAL														

ACUREX Corporation

PARTICULATE TEST FIELD DATA SHEET

Plant ACUREX

Date 5-17-80

Test Location FURNACE MAIN RES HEATER

Run Number

Stack Diameter inches

Duct Dimensions in. x in.

Start Time

Operator

Barometric Pressure

Static Pressure

Stack Pressure

Probe Number

Pitot Coefficient

Pitot Number

Meter Box Number

Orifice Coefficient

Nozzle Size & Number

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.

IMPINGER VOLUMES	TIME	CO_2	O_2	CO
SILICA GEL				

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ΔP in. wg.	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F: STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER: IN	GAS METER: OUT	PUMP VACUUM in. Hg	√ΔP	
0925	START	71.4	210	.78	527.752	68	278			252	80	81	3		
		76.4	200	.77	540.40	78	264			255	86	81	3		
		81.4	200	.77	553.1	82	259			257	93	81	3		
	STOP	85.2			562.678										CHANGE OF SILICA GEL
1002	START	85.2	200	.77	562.678	72	269			257	81	81	3		
		90.2	205	.78	576.29	70	259			258	88	81	3		
		95.2	205	.77	588.68	83	258			251	96	82	3		
	STOP	98.9			598.062										
1040	START	98.9	200	.78	598.062	70	259			251	82	82	3		
		103.9	190	.77	610.92	77	263			252	85	82	3		
		108.9	190	.77	623.51	82	259			253	95	82	3		
	STOP	112.2			632.972										
1118	START	112.2	190	.79	632.972	65	270			252	81	82	3		
		117.2	190	.77	644.72	78	258			252	88	82	3		
		122.2	180	.77	657.42	83	254			253	95	83	3		
	STOP	124.7			663.809										
1154	START	124.7	200	.79	663.809	65	264			252	82	82	3		
		129.7	190	.77	—	79	266			253	91	82	3		
		134.7	200	.77	—	82	268			252	98	83	3		
	STOP	138.3			698.348										END LEAK RATE .003 @ 13 Hg
	AVG/TOTAL			.77	350.517	75					83				

ACUREX Corporation

SASS ~~PARTICULATE~~ TEST FIELD DATA SHEET

Plant ACUREX | Barometric Pressure 29.87

Date 5-21-80 | Static Pressure +.25 in H_2O

Test Location FURNACE (MAN REED) | Stack Pressure 29.89

Run Number 1 | Probe Number 3' glass

Stack Diameter inches 4 | Pilot Coefficient —

Duct Dimensions in. x in. | Pilot Number

Start Time 0621 | Meter Box Number 81

Operator MARKOJA | Orifice Coefficient 3.536

Nozzle Size & Number 1.5

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.
B26 MG		

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
500	520				
500	485				
500	485				
SILICA GEL					
750					

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ~~ΔP in. wg.~~ mm	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F: STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT	PUMP VACUUM in. Hg	$\sqrt{\Delta P}$	LEAK RATE .008 @ 20 in Hg.
0621	START	0	210	3.8	524.133	63	386		54	401	76	78	13		
		5	206	3.9	545.00	78	376		55	397	77	RUN 78	13		
	STOP	12.9			578.442										
0659	START	12.9	215	4.0	578.442	64	388		56	399	75	76	13		
		17.9	200	4.0	599.3	77	378		56	397	79	76	13		
		22.9	205	4.0	620.4	82	377		57	397	86	77	13		
	STOP	27.3			639.053										
0734	START	27.3	205	4.0	639.053	62	384		55	395	77	77	13		
		32.3	215	4.0	660.3	79	375		54	395	81	77	13		
		37.3	210	4.0	681.7	83	376		54	394	88	79	13		.59 ml cond
0810	STOP	42.0			701.465										
	START	42.0	210	4.0	701.465	62 RUN	406		55	399	79	79	14		
		47.0	200	4.0	722.9	77	377		56	397	84	79	14		
		52.0	200	4.0	744.2	82	379		59	397	92	81	14		
	STOP	56.7			764.178	85	380		60	397	96	84			
0846	START	56.7	200	4.0	764.178	66	390		50	399	79	79	14		
		61.7	200	4.0	785.7	76	382		51	397	84	80	14		
		66.7	210	4.0	807.2	81	380		51	396	89	81	14		
	STOP	71.4			827.414										
0925	START	71.4	210	4.0	827.414	65	411 RUN		54	400	81	81	14		
	AVG/TOTAL														

ACUREX Corporation

PAGE 2 OF 4

SASS ~~PARTICULATE~~ TEST FIELD DATA SHEET

Plant ACUREX

Date 5-21-80

Test Location FURNACE (MAIN RESID)

Run Number

Stack Diameter inches

Duct Dimensions in. x in.

Start Time

Operator

Barometric Pressure

Static Pressure

Stack Pressure

Probe Number

Pitot Coefficient

Pitot Number

Meter Box Number

Orifice Coefficient

Nozzle Size & Number

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
SILICA GEL					

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ~~ΔP in. wg.~~ FAN	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT	PUMP VACUUM in. Hg	√ΔP	
		76.4	200	4.0	848.9	78	384		55	397	85	81	14		CHANGE SIL. GEL
		81.4	200	4.0	870.0 RUN	82	380		54	396	91	82	14		77 ml condensate
	STOP	86.2			886.273										
1002	START	86.2	200	4.0	886.273	72	414		55	401	81	81	13		
		90.2	205	4.0	907.5	70	383		51	397	90	82	13		
		95.2	205	4.0	929.1	83	382		51	397	102	85	13		
	STOP	98.9			944.715										
1040	START	98.9	200	4.0	944.715	70	414		55	400	84	84	12		
		103.9	190	4.0	966.2	78	381		51	398	84	83	12		
		108.9	190	4.0	987.8	82	378		53	397	92	84	12		
	STOP	112.2			1001.873										
1118	START	112.2	190	4.0	1001.873	65	394		52	401	81	83	13		
		117.2	190	4.0	1023.3	78	383		51	399	84	83	13		
		122.2	180	4.0	1044.8	83	382		53	397	91	84	13		
	STOP	124.7			1055.494										
1154	START	124.7	200	4.0	1055.494	65	413		59	399	82	82	13		
		129.7	190	4.0	1078.	79	380		56	398	89	83	13		
		134.7	200	4.0	1098.3 RUN	82	378		57	396	93	84	13		
	STOP	138.3			1113.818										
	START	138.3	185	4.0	1113.818	68	385		62	396	83	83	13		
	AVG/TOTAL														

ACUREX Corporation

SASS ~~PARTICULATE~~ TEST FIELD DATA SHEET

Plant ACUREX — Barometric Pressure ____

Date 5-21-80 — Static Pressure ____

Test Location FURNACE (COMM. RES. D) — Stack Pressure ____

Run Number ____ — Probe Number ____

Stack Diameter inches ____ — Pitot Coefficient ____

Duct Dimensions in. x in. ____ — Pitot Number ____

Start Time ____ — Meter Box Number ____

Operator ____ — Orifice Coefficient ____

Nozzle Size & Number ____

Molecular Weight ____

BWO ____

FILTER DATA

NUMBER	TARE	FINAL WT.

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
SILICA GEL					

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ~~ΔP in. wg.~~ FPM	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F: STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT	PUMP VACUUM in. Hg	√ΔP	
		1143.3	185	4.0	1136.	77	378		63	396	87	84	13		
		1148.3	185	4.0	1156.2 RUN	81	374		64	396	93	85	13		
	STOP	151.6			1170.295										
1908	START	151.6	180	4.0	1170.295	67	384		69	401	83	84	13		REMOVE IMPINGERS
		156.6	180	4.0	1191.5	76	377		61	399	85	84	13		
		161.6	180	4.0	1213.4	81	380		57	398	92	85	13		119ml CONDENSATE
	STOP	162.9			1218.916										
1347	START	162.9	190	4.0	1218.916	66	391		52	401	82	84	10		
		167.9	180	4.0	1240.3	76	379		50	399	84	84	10		
		172.9	185	4.0	1261.8	81	379		51	398	90	84	10		
	STOP	176.0			1274.944										
1426	START	176.0	190	4.0	1274.944	73	390		52	400	81	83	10		
		181.0	180	4.0	1296.6	78	378		50	399	84	83	10		
		186.0	185	4.0	1319.	82	380		50	398	90	84	10		
	STOP	188.7			1329.388										
	START	188.7	195	4.0	1329.388	73	395		51	401	81	82	10		
		193.7	175	4.0	1350.9	77	385		49	398	83	82	10		
		198.7	165	4.0	372.4	82	383		50	399	90	83	10		
	STOP	201.0			1382.355										
1538	START	201.0	180	4.0	1382.355	76	401		52	392	81	82	10		
	AVG/TOTAL														

ACUREX Corporation

PAGE 4 OF 4 SASS ~~PARTICULATE~~ TEST FIELD DATA SHEET

Plant ACUREX — Barometric Pressure

Date FURNACE (MAIN RESID) — Static Pressure

Test Location 5-21-80 — Stack Pressure

Run Number — Probe Number

Stack Diameter inches — Pitot Coefficient

Duct Dimensions in. x in. — Pitot Number

Start Time — Meter Box Number

Operator — Orifice Coefficient

Nozzle Size & Number

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
SILICA GEL					

SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ΔP in. wg. FPM	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F							PUMP VACUUM in. Hg	$\sqrt{\Delta P}$	
					STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT			
	206.0	155	4.0	1403.7	79	384		50	399	85	82	10		
	211.0	145	4.0	1427.	82	381		50	398	90	84	10		
STOP	213.6			1436.314										
START	213.6	160	4.0	1436.314	76	388		52	401	83	83	10		
	218.6	150	4.0	1457.6	78	378		51	399	86	83	10		
	223.6	145	4.0	1479.4	82	375		52	398	92	84	10		
STOP	226.1			1489.954										
1648 START	226.1	145	4.0	1489.954	73	393		53	399	83	83	10		
	231.1	135	4.0	1511.2	77	378		52	398	87	84	10		
	236.6	125	4.0	1535.2	81	375		53	397	94	85	10		
STOP	241.5			1555.997										LEAK CHECK .018@ 14-in Hg
AVG/TOTAL				1031.864	76					84				

Form EED-021 1 79

ACUREX Corporation

PAGE 1 OF 2

PARTICULATE TEST FIELD DATA SHEET

Plant ACUREX — Barometric Pressure 29.87

Date 5-17-80 — Static Pressure

Test Location FURNACE MAN RES. — Stack Pressure +.25 in H_2O

Run Number #1 HEATER METHOD 8 — Probe Number 3' GLASS

Stack Diameter inches 44" — Pitot Coefficient —

Duct Dimensions in. x in. — Pitot Number —

Start Time — Meter Box Number 04-3

Operator UNEMAN — Orifice Coefficient .716

Nozzle Size & Number PN1 · 8713

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
102	55				
100	145				
100	115				
0	3				
45.8					
SILICA GEL					
250	276.8				

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ΔP in. wg.	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F: STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT	PUMP VACUUM in. Hg	$\sqrt{\Delta P}$	LEAK RATE .015@ 15 hg
1347	START	0	190	1.97	698.902	67	250				81	81	5		
		5	180	1.94	702.59	76	269				81	81	5		
		10	185	1.924	706.31	81	260				81	81	5		
	STOP	13.0			708.562										
1426	START	13.0	190	1.95	708.562	73	253				81	81	5		
		18.0	180	1.93	712.25	78	261				81	81	5		
		23.0	185	1.92	715.88	82	275				81	81	5		
	STOP	25.7			717.964										
1504	START	25.7	195	1.95	717.964	73	268				80	80	5		
		30.7	175	1.95	—	80	267				80	81	5		
		35.7	165	1.92		82	273				81	81	5		
	STOP	38.0			727.145										
1538	START	38.0	180	1.94	727.145	76	261				81	81	5		
		43.0	155	1.93	729.58	79	274				81	81	5		
		48.0	145	1.92	—	82	273				81	81	5		
		50.6			736.487										
1615	START	50.6	160	1.94	736.487	76	278				81	81	5		
		55.6	150	1.94	740.15	77	274				81	80	5		
		60.6	145	1.92	743.91	82	275				81	80	5		
	STOP	63.2			745.815										
	AVG/TOTAL														

Form EED-021 1 79

ACUREX Corporation

Page 2 of 2

PARTICULATE TEST FIELD DATA SHEET

Plant ACUREX

Date 5-17-80

Test Location FURNACE MAIN RES HEATER

Run Number

Stack Diameter inches

Duct Dimensions in. x in.

Start Time

Operator

Barometric Pressure

Static Pressure

Stack Pressure

Probe Number

Pitot Coefficient

Pitot Number

Meter Box Number

Orifice Coefficient

Nozzle Size & Number

Molecular Weight

BWO

FILTER DATA

NUMBER	TARE	FINAL WT.

IMPINGER VOLUMES		TIME	CO_2	O_2	CO
SILICA GEL					

	SAMPLE POINT	CLOCK TIME	VELOCITY HEAD ΔP in. wg.	ORIFICE METER ΔH in. wg.	GAS METER VOLUME FT³	TEMPERATURES °F STACK	PROBE	IMPINGER	ORGANIC MODULE	OVEN	GAS METER IN	GAS METER OUT	PUMP VACUUM in. Hg	√ΔP	
1648	START	63.2	145	1.95	745.815	74	275				81	81	5		
		68.2	135	1.94	—	77	271				81	81	5		
		73.5	125	1.92	753.48	81	273				81	81	5		
	STOP	76.5			757.249										
1702	START	76.5	130	1.94	757.249	77	261				81	81	5		
		81.5	135	1.94	759.36	80	266				81	81	5		
		86.5	135	1.92	763.12	82	267				81	81	5		
	STOP	87.5			763.913										END LEAK RATE .018 @ 8 Hg
	AVG/TOTAL			1.94	65.011	78					81				

Form EED-021 1 79

55

SECTION 5

ANALYTICAL LABORATORY RESULTS

5.1 FUEL ANALYSIS REPORT

LABORATORY CERTIFICATE

ACUREX

CURTIS & TOMPKINS, LTD.

ESTABLISHED 1878

ANALYTICAL · CHEMISTS · CONSULTING

SAMPLERS — INSPECTORS

MEMBERS OFFICIAL CHEMISTS AND/OR SAMPLERS FOR MANY COMMODITY AND TRADE ORGANIZATIONS

CORPORATE OR STAFF MEMBERSHIP IN PRINCIPAL SCIENTIFIC SOCIETIES

290 DIVISION STREET
SAN FRANCISCO, CALIF. 94103
U.S.A.

CABLE ADDRESS: ANALYST

REFEREE ANALYSES
RESEARCH — INVESTIGATIONS
VITAMIN ASSAYS — BIOCHEMISTRY

SPECIALISTS IN BULK COMMODITIES

Laboratory No. 80k10
Preliminary No. 4132

Reported 10/6/80
Sampled -------
Received 9/8/80

For ACUREX CORPORATION

Report on 5 samples of Oil

Mark Sample # Sample Type
(5) 80-1562, Fuel Oil
(All) Project No.: 7601.22, Customer No.: RB 59186A, rel. 01, Prime Contract: 68-02, 3188, Acurex Project: 7601, Date: September 5, 1980, Subcontract No.: RB59186A.

	(5)		
	1st Test	2nd Test	3rd Test
Carbon (C),% ------	86.94	----	----
Hydrogen (H), % ---	12.90	----	----
Oxygen (O), by difference,% -----	0.01*	----	----
Nitrogen (N), % ---	0.03	0.04	0.05
Sulfur (S),% ------	0.18	0.20	0.20
Heating Value: BTU/Pound --------	19,190	----	----
Gravity, °API @ 60°F ------	33.75	----	----

*Less Than

Curtis & Tompkins Ltd.

5.2 PARTICULATE GRAVIMETRIC ANALYSES AND MASS EMISSION CALCULATIONS

ACUREX Corporation

PARTICULATE CALCULATIONS

For High Volume Stack Sampler - Method 5

1. Volume of dry gas sampled at standard conditions, 68° F, 29.92 inch Hg (scf)

$$Vm_{std} = 17.64 \frac{Vm}{\alpha}\left(\frac{P_b + \frac{\Delta H}{13.6}}{T_{m,avg} + 460}\right) = 17.64\left(\frac{350.517}{1.013}\right)\left(\frac{29.87 + 0.77/13.6}{543}\right)$$

or $Vm_{stack} = \left(\frac{Vm}{\alpha} + Vw_{std}\right)\left(\frac{T_{s,avg} + 460}{T_{m,avg} + 460}\right)\left(\frac{P_m}{P_s}\right)$ (acf at stack conditions)

$= 336.4$

2. Stack gas moisture condensed at standard conditions (scf)

$$Vw_{std} = 0.04707\ Vl_c$$
$$= 0.04707\ (199.9) = 9.409$$

3. Stack gas proportion of water vapor, by volume

$$B_{wo} = \frac{Vw_{std}}{Vw_{std} + Vm_{std}}$$
$$= \frac{9.409}{9.409 + 336.4} = 2.7\%$$

4. Stack gas dry molecular weight (lb/lb-mole)

$$MW_d = 0.44\ (\%CO_2) + 0.32\ (\%O_2) + 0.28\ (\%N_2 + \%CO)$$
$$= 0.44(12.9) + 0.32(1.9) + 0.28(85.2) = 30.14$$

5. Stack gas molecular weight (lb/lb-mole)

$$MW_s = MW_d\ (1 - B_{wo}) + 18\ (B_{wo})$$
$$= 30.14(1 - 0.027) + 18(0.027) = 29.81$$

6. Pressure stack, in. Hg

$$P_s = P_b + \frac{P_{st}}{13.6}$$
$$= 29.89$$

7. Stack gas velocity at stack conditions (ft/sec)

$$V_s = 85.49\ (C_p)\ (\sqrt{\Delta P})_{avg} \sqrt{\frac{T_{s,avg} + 460}{P_s\ MW_s}}$$

$= 200$ ft/min $= 3.3$ ft/s

Velocity was not detectable with pitot tube or inclined water manometer. Stack gas velocity was measured with hot wire anemometer

8. Stack gas volume at standard conditions (scfm)

$$Q_s = 60\ (1 - B_{wo})\ Vs_{avg}\ A_s \left(\frac{528}{T_{s,avg} + 460}\right)\left(\frac{P_s}{29.92}\right) =$$

or $Q_a = 60\ Vs_{avg}\ A_s$ (acfm) $= 60(1-0.027)(3.3)\frac{4^2\pi}{4(144)}\left(\frac{528}{75+460}\right)\frac{29.89}{29.92} = 16.72\ Q_s$

9. Test percent isokinetic

$$\%I = \frac{17.33\ (T_{s,avg} + 460)\ \ [0.04707\ (W_{lc}) + Vm_{std}]}{\theta\ V_s\ P_s\ D_n^2}$$

$= 104.4\ \%$

10. Particulate matter concentration, gr/scf

$$C_s = 15.432\ \frac{M_p}{Vm_{std}} = \frac{15.432\ (0.0175\ \text{grams})}{336.4} = 8.028 \times 10^{-4}$$

or $C_a = 15.432\ \frac{M_p}{Vm_{stack}}$ (gr/acf) =

11. Emission rate of particulate matter, lb/hr

$$ER = 0.00857\ (Q_s)\ C_s$$

$= 0.00857\ (16.72)\ (8.028 \times 10^{-4}) = 1.15 \times 10^{-4}$ lb/hr

$= 0.0063$ lb/10^6 Btu

$= 2.70$ ng/J

12. Percent excess air at sampling point

$$\%\ EA = \frac{100\ [\%O_2 - 0.5\ (\%CO)]}{0.264\ \ \%N_2 - (\%O_2 - 0.5 \times \%CO)} =$$

13. Emission rate of particulate matter, lb/10^6 Btu

$$E = 2.679 \times 10^6 \left(\frac{M_p}{Vm_{std}}\right) F \left(\frac{20.9}{20.9 - \%O_2}\right)$$

ACUREX Corporation

PARTICULATE CALCULATIONS

1. Volume of dry gas sampled at standard conditions, 68° F, 29.92 inch Hg (scf)

$$Vm_{std} \quad 17.64 \frac{Vm}{\alpha}\left(\frac{P_b + \frac{\Delta H}{13.6}}{T_{m,avg} + 460}\right) = 17.64 \quad \frac{1031.864}{1.003}\left(\frac{29.87 + \frac{4}{13.6}}{544}\right) = 1006.263$$

$$\text{or} \quad Vm_{stack} = \left(\frac{Vm}{\alpha} + Vw_{std}\right)\left(\frac{T_{s,avg} + 460}{T_{m,avg} + 460}\right)\left(\frac{P_m}{P_s}\right) \quad \text{(acf at stack conditions)}$$

=

2. Stack gas moisture condensed at standard conditions (scf)

$$Vw_{std} = 0.04707\ Vl_c$$

$$= 31.311$$

3. Stack gas proportion of water vapor, by volume

$$B_{wo} = \frac{Vw_{std}}{Vw_{std} + Vm_{std}}$$

$$= \frac{31.311}{1006.263 + 31.311} = 3.0\%$$

4. Stack gas dry molecular weight (lb/lb-mole)

$$MW_d = 0.44\ (\%CO_2) + 0.32\ (\%O_2) + 0.28\ (\%N_2 + \%CO)$$

$$= 30.14$$

5. Stack gas molecular weight (lb/lb-mole)

$$MW_s = MW_d\ (1 - B_{wo}) + 18\ (B_{wo})$$

$$= 30.14\ (1 - 3.0) + 18\ (3.0)$$

6. Pressure stack, in. Hg

$$P_s = P_b + \frac{P_{st}}{13.6}$$

$$= 29.89$$

7. Stack gas velocity at stack conditions (ft/sec)

$$V_s = 85.49\ (C_p)\ (\sqrt{\Delta P})_{avg} \sqrt{\frac{T_{s,avg} + 460}{P_s\ MW_s}}$$

$= \sim 200$ ft/min ≈ 3.3 ft/s

Velocity was to small to be detected with pitot tube or inclined water manometer. Stack gas velocity was measured with hot wire anemometer.

8. Stack gas volume at standard conditions (scfm)

$$Q_s = 60\ (1 - B_{wo})\ Vs_{avg}\ A_s \left(\frac{528}{T_{s,avg} + 460}\right)\left(\frac{P_s}{29.92}\right) =$$

or $Q_a = 60\ Vs_{avg}\ A_s$ (acfm) $= 60(1-0.03)3.3 \frac{\pi 4^2}{4(144)} \frac{528}{75+460} \frac{27.89}{27.72} = 16.53$

9. Test percent isokinetic

$$\%I = \frac{17.33\ (T_{s,avg} + 460)\ [0.04707\ (W_{lc}) + Vm_{std}]}{\theta\ V_s\ P_s\ D_n^2}$$

$= 178\ \%$

10. Particulate matter concentration, gr/scf

$$C_s = 15.432 \frac{M_p}{Vm_{std}} = 15.432 \frac{0.024c}{1006.26} = 0.0004$$

or $C_a = 15.432 \frac{M_p}{Vm_{stack}}$ (gr/acf) =

11. Emission rate of particulate matter, lb/hr

$$ER = 0.00857\ (Q_s)\ C_s$$

$= 0.00857(16.53)(0.0004) = 0.0001$ lb/hr

$= 0.0029$ lb/10^6 Btu

$= 1.22$ ng/J

12. Percent excess air at sampling point

$$\%\ EA = \frac{100\ [\%O_2 - 0.5\ (\%CO)]}{0.264\ \%N_2 - (\%O_2 - 0.5 \times \%CO)} =$$

13. Emission rate of particulate matter, lb/10^6 Btu

$$E = 2.679 \times 10^{-6} \left(\frac{M_p}{Vm_{std}}\right) F \left(\frac{20.9}{20.9 - \%O_2}\right)$$

=

5.3 SULFUR ANALYSIS REPORT AND EMISSION CALCULATION

H_2SO_4 Mist and SO_2 Clean-Up and Analysis

Plant M.A.N. Residential Furnace Date 5-21-80

Project No. CMEA 7605.21 Recorded by G. Nicoll

H_2SO_4 Mist and SO_2 Train Description and Recovery Data

Impinger Sequence	1	2	3	4	5	6
Impinger Number						
G-S Standard ( )						
G-S Modified ( )						
Contents	IPA	H_2O_2	H_2O_2	Empty	Si. Gel	
Concentration (%)	80	3	3	–	–	
Initial Volume (ml)	100	100	100	0	250.0g	
Final Volume (ml)	55	145	115	3	276.8g	
Dilution Volume (ml)	250	500				
Container Number	102	103	103			

Location of glass wool in train Between impingers 1 & 2

Location and type of filter in train glass filter

Total Condensate (No. 8-7) 45 ml

$$C_{SO_2} = \frac{7.05\times10^{-5}(19.4-0.05)0.011\left(\frac{500}{5}\right)}{64.187} = 2.34\times10^{-5}\ \frac{lb}{ft^3}$$

H_2O_2 Sample Blank No. 104

IPA Sample Blank No. 101 $C_{SO_4} = 0$

H_2SO_4 and SO_2 Analysis

Sample Number	102	103				
Volume of Titrant for Sample (ml)	0.1	19.4				
Volume of Titrant for Blank (ml)	0.1	0.05				
Normality of Titrant	0.011	0.011				
Ttl. Vol. of Soln. (ml)	250	250				
Volume of Sample Aliquot Titrated (ml)	100	5.0				
Performed by	Gardena	Gardena				

Spike and recovery at start Interference 103% Solution past end point

5.4 TRACE ELEMENT ANALYSIS REPORT

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 330 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 23, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 80-1535
XAD - BLANK

IAD No.: 97-E697-116-28

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN PPM WEIGHT

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.3
Thorium		Gadolinium		Molybdenum	0.4	Titanium	2
Bismuth	<0.1	Europium		Niobium		Scandium	<0.1
Lead	0.6	Samarium		Zirconium	1	Calcium	170
Thallium		Neodymium		Yttrium	<0.1	Potassium	24
Mercury	NR	Praseodymium	0.2	Strontium	0.2	Chlorine	1
Gold		Cerium	0.9	Rubidium	<0.1	Sulfur	9
Platinum		Lanthanum	2	Bromine	0.4	Phosphorus	26
Iridium		Barium	1	Selenium	0.2	Silicon	*MC
Osmium		Cesium		Arsenic	<0.1	Aluminum	13
Rhenium		Iodine	<0.1	Germanium	0.2	Magnesium	17
Tungsten		Tellurium		Gallium	0.1	Sodium	98
Tantalum	≤0.6	Antimony	0.2	Zinc	6	Fluorine	≈3
Hafnium		Tin	0.1	Copper	13	Oxygen	NR
Lutetium		Indium	STD	Nickel	MC	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	3	Carbon	NR
Thulium		Silver		Iron	120	Boron	<0.1
Erbium		Palladium		Manganese	39	Beryllium	
Holmium		Rhodium		Chromium	29	Lithium	0.1
Dysprosium						Hydrogen	NR

* Heterogeneous

STD — Internal Standard
NR — Not Reported
All elements not detected < 0.1 ppm
MC — Major Component
INT — Interference

Approved: M Speed

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 230 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Avenue
Mountain View, California 94042

Date: September 26, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 1475
MAN Test
1st Impinger Blank

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN µg/ml

IAD No.: 97-E697-116-28

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.002
Thorium		Gadolinium		Molybdenum	0.05	Titanium	0.01
Bismuth		Europium		Niobium		Scandium	0.001
Lead	≤0.02	Samarium		Zirconium	0.01	Calcium	6
Thallium		Neodymium		Yttrium		Potassium	2
Mercury	NR	Praseodymium		Strontium	0.006	Chlorine	0.3
Gold		Cerium		Rubidium		Sulfur	0.2
Platinum		Lanthanum		Bromine	0.02	Phosphorus	3
Iridium		Barium	* 1	Selenium		Silicon	0.4
Osmium		Cesium		Arsenic		Aluminum	0.3
Rhenium		Iodine	0.01	Germanium		Magnesium	0.08
Tungsten		Tellurium		Gallium		Sodium	* 6
Tantalum		Antimony		Zinc	0.07	Fluorine	≈0.1
Hafnium		Tin	0.2	Copper	0.07	Oxygen	NR
Lutetium		Indium	STD	Nickel	0.03	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	0.004	Carbon	NR
Thulium		Silver	0.5	Iron	0.09	Boron	0.001
Erbium		Palladium		Manganese	0.006	Beryllium	
Holmium		Rhodium		Chromium	0.006	Lithium	0.002
Dysprosium						Hydrogen	NR

STD – Internal Standard
NR – Not Reported
All elements not detected < 0.004 µg/ml
MC – Major Component >10 µg/ml
INT – Interference

* Heterogeneous

Approved: M Spencer

COMMERCIAL TESTING & ENGINEERING CO.
GENERAL OFFICES: 330 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9321

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 25, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: A-45 Blank SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS IAD No.: 97-E697-116-28

FILTER BLANK CONCENTRATION IN $\mu g/cm^2$

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium	≤0.008	Terbium		Ruthenium		Vanadium	0.02
Thorium	0.01	Gadolinium		Molybdenum	0.005	Titanium	0.9
Bismuth		Europium		Niobium	0.01	Scandium	0.002
Lead	0.007	Samarium		Zirconium	0.05	Calcium	MC
Thallium		Neodymium	0.004	Yttrium	0.003	Potassium	0.5
Mercury	NR	Praseodymium	0.003	Strontium	0.04	Chlorine	0.4
Gold	<0.001	Cerium	0.01	Rubidium	<0.001	Sulfur	0.8
Platinum		Lanthanum	0.02	Bromine	0.03	Phosphorus	0.07
Iridium		Barium	0.2	Selenium	0.005	Silicon	MC
Osmium		Cesium		Arsenic	0.001	Aluminum	MC
Rhenium		Iodine	0.001	Germanium		Magnesium	1
Tungsten	0.01	Tellurium		Gallium	0.008	Sodium	>2
Tantalum		Antimony		Zinc	0.01	Fluorine	≈0.005
Hafnium		Tin		Copper	0.01	Oxygen	NR
Lutetium		Indium	STD	Nickel	0.01	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	<0.001	Carbon	NR
Thulium		Silver	0.001	Iron	0.2	Boron	0.9
Erbium		Palladium		Manganese	0.003	Beryllium	
Holmium		Rhodium		Chromium	0.02	Lithium	<0.001
Dysprosium						Hydrogen	NR

STD — Internal Standard
NR — Not Reported
All elements not detected <0.001 $\mu g/cm^2$
MC — Major Component > 10 $\mu g/cm^2$
INT — Interference

Approved: M Speeks

COMMERCIAL TESTING & ENGINEERING CO.
GENERAL OFFICES: 220 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-279-9321

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 24, 1980

Analyst: L. Jacobs

P. O. No.:

Sample No.: 80-1312
TAP WATER BLANK

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN µg/ml

IAD No.: 97-E697-116-28

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	<0.001
Thorium		Gadolinium		Molybdenum	0.1	Titanium	0.1
Bismuth		Europium		Niobium	0.005	Scandium	
Lead	0.01	Samarium		Zirconium	0.02	Calcium	10
Thallium		Neodymium		Yttrium		Potassium	0.05
Mercury		Praseodymium		Strontium	0.02	Chlorine	MC
Gold		Cerium		Rubidium	≤0.002	Sulfur	0.4
Platinum		Lanthanum		Bromine	0.1	Phosphorus	0.9
Iridium		Barium	0.02	Selenium		Silicon	0.2
Osmium		Cesium		Arsenic	0.004	Aluminum	0.004
Rhenium		Iodine		Germanium		Magnesium	MC
Tungsten		Tellurium		Gallium	0.009	Sodium	0.2
Tantalum		Antimony		Zinc	0.04	Fluorine	≈0.004
Hafnium		Tin		Copper	0.002	Oxygen	NR
Lutetium		Indium	STD	Nickel	0.003	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	*≤0.002	Carbon	NR
Thulium		Silver		Iron	0.2	Boron	<0.001
Erbium		Palladium		Manganese	0.006	Beryllium	
Holmium		Rhodium		Chromium	0.07	Lithium	INT
Dysprosium						Hydrogen	NR

* Heterogeneous

STD – Internal Standard
NR – Not Reported
All elements not detected < 0.002 µg/ml
MC – Major Component > 10 µg/ml
INT – Interference

Approved: M Jacobs

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 230 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14235 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 24, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 80-1313
WATER TANK BLANK

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN µg/ml

IAD No.: 97-E697-116-28

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.003
Thorium		Gadolinium		Molybdenum	*0.1	Titanium	0.1
Bismuth		Europium		Niobium		Scandium	<0.001
Lead	0.03	Samarium		Zirconium	0.02	Calcium	10
Thallium		Neodymium		Yttrium		Potassium	2
Mercury	NR	Praseodymium		Strontium	0.03	Chlorine	8
Gold		Cerium		Rubidium	0.006	Sulfur	2
Platinum		Lanthanum		Bromine	0.3	Phosphorus	0.4
Iridium		Barium	0.2	Selenium		Silicon	2
Osmium		Cesium		Arsenic	0.002	Aluminum	0.2
Rhenium		Iodine	0.01	Germanium		Magnesium	0.5
Tungsten		Tellurium		Gallium	≤0.007	Sodium	2
Tantalum		Antimony		Zinc	*2	Fluorine	≈0.1
Hafnium		Tin		Copper	4	Oxygen	NR
Lutetium		Indium	STD	Nickel	0.04	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	0.005	Carbon	NR
Thulium		Silver		Iron	0.3	Boron	0.002
Erbium		Palladium		Manganese	0.01	Beryllium	
Holmium		Rhodium		Chromium	0.008	Lithium	0.004
Dysprosium						Hydrogen	NR

* Heterogeneous

STD – Internal Standard
NR – Not Reported
All elements not detected <0.003 µg/ml
MC – Major Component > 10 µg/ml
INT – Interference

Approved: M J Jacobs

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 228 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 24, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 80-1368
TANK SAMPLE PRIOR TO TEST

IAD No.: 97-E697-116-28

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN µg/ml

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.007
Thorium		Gadolinium		Molybdenum	*0.4	Titanium	0.3
Bismuth		Europium		Niobium		Scandium	0.004
Lead	0.1	Samarium		Zirconium	0.05	Calcium	MC
Thallium		Neodymium		Yttrium	0.05	Potassium	0.5
Mercury	NR	Praseodymium		Strontium	0.05	Chlorine	0.4
Gold		Cerium	0.008	Rubidium	<0.001	Sulfur	MC
Platinum		Lanthanum	0.01	Bromine	0.4	Phosphorus	0.4
Iridium		Barium	0.07	Selenium	0.1	Silicon	1
Osmium		Cesium		Arsenic	INT	Aluminum	0.1
Rhenium		Iodine		Germanium	0.002	Magnesium	0.6
Tungsten	0.02	Tellurium	0.02	Gallium	0.004	Sodium	0.8
Tantalum		Antimony		Zinc	MC	Fluorine	*=0.2
Hafnium		Tin		Copper	MC	Oxygen	NR
Lutetium		Indium	STD	Nickel	1	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	0.02	Carbon	NR
Thulium		Silver	0.03	Iron	MC	Boron	0.002
Erbium		Palladium		Manganese	0.2	Beryllium	
Holmium		Rhodium		Chromium	0.7	Lithium	0.002
Dysprosium						Hydrogen	NR

* Heterogeneous

STD — Internal Standard
NR — Not Reported
All elements not detected < 0.002 µg/ml
MC — Major Component > 10 µg/ml
INT — Interference

Approved: M Y Jacobs

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 230 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Avenue
Mountain View, California 94042

Date: September 26, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 1471
MAN TEST
FILTER

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN $\mu g/cm^2$

IAD No.: 97-E697-116-28

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.2
Thorium		Gadolinium		Molybdenum	0.05	Titanium	1
Bismuth		Europium		Niobium	0.005	Scandium	$\leq$0.001
Lead	0.2	Samarium		Zirconium	0.006	Calcium	MC
Thallium		Neodymium	0.005	Yttrium	0.008	Potassium	0.5
Mercury	NR	Praseodymium	0.003	Strontium	0.08	Chlorine	0.3
Gold		Cerium	0.03	Rubidium	<0.001	Sulfur	8
Platinum		Lanthanum	0.04	Bromine	0.04	Phosphorus	0.5
Iridium		Barium	* 0.9	Selenium		Silicon	MC
Osmium		Cesium		Arsenic	$\leq$0.001	Aluminum	MC
Rhenium		Iodine		Germanium	$\leq$0.006	Magnesium	4
Tungsten		Tellurium		Gallium	0.009	Sodium	MC
Tantalum		Antimony		Zinc	0.6	Fluorine	$\approx$1
Hafnium		Tin	0.02	Copper	0.3	Oxygen	NR
Lutetium		Indium	STD	Nickel	0.04	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	0.02	Carbon	NR
Thulium		Silver	0.002	Iron	0.7	Boron	MC
Erbium		Palladium		Manganese	0.02	Beryllium	
Holmium		Rhodium		Chromium	0.1	Lithium	0.02
Dysprosium						Hydrogen	NR

* Heterogeneous

STD – Internal Standard
NR – Not Reported
All elements not detected $< 0.001\ \mu g/cm^2$
MC – Major Component $> 10 \mu g/cm^2$
INT – Interference

Approved: M Speeth

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 228 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8484
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent HIgginbotham
Acurex Corporation
485 Clyde Avenue
Mountain View, California 94042

Date: September 26, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 1476
MAN TEST
1st IMPINGER + OMC

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN µg/ml

IAD No.: 97-E697-116-28

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.001
Thorium		Gadolinium		Molybdenum	0.008	Titanium	0.3
Bismuth		Europium		Niobium		Scandium	<0.001
Lead	0.04	Samarium		Zirconium	0.002	Calcium	1
Thallium		Neodymium		Yttrium		Potassium	3
Mercury	NR	Praseodymium		Strontium	0.002	Chlorine	0.04
Gold		Cerium	<0.001	Rubidium	<0.001	Sulfur	MC
Platinum		Lanthanum		Bromine	0.004	Phosphorus	3
Iridium		Barium	0.05	Selenium	0.006	Silicon	1
Osmium		Cesium	<0.001	Arsenic		Aluminum	>0.7
Rhenium		Iodine		Germanium		Magnesium	0.6
Tungsten		Tellurium	0.01	Gallium	0.002	Sodium	MC
Tantalum	≤0.002	Antimony		Zinc	0.2	Fluorine	≈0.3
Hafnium		Tin	0.05	Copper	0.2	Oxygen	NR
Lutetium		Indium	STD	Nickel	0.2	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	0.004	Carbon	NR
Thulium		Silver	0.03	Iron	0.4	Boron	0.003
Erbium		Palladium		Manganese	0.04	Beryllium	
Holmium		Rhodium		Chromium	0.08	Lithium	0.005
Dysprosium						Hydrogen	NR

STD – Internal Standard
NR – Not Reported
All elements not detected < 0.001 µg/ml
MC – Major Component >10 µg/ml
INT – Interference

Approved: M. J. Jacobs

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 228 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Avenue
Mountain View, California 94042

Date: September 26, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 1369 MAN TEST TANK WATER SAMPLE AFT. TEST

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS

IAD No.: 97-E697-116-28

CONCENTRATION IN µg/ml

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.002
Thorium		Gadolinium		Molybdenum	0.2	Titanium	0.2
Bismuth		Europium		Niobium		Scandium	0.01
Lead	0.09	Samarium		Zirconium	0.03	Calcium	MC
Thallium		Neodymium		Yttrium	≤0.2	Potassium	3
Mercury	NR	Praseodymium		Strontium	0.009	Chlorine	* MC
Gold		Cerium		Rubidium		Sulfur	MC
Platinum		Lanthanum		Bromine	0.1	Phosphorus	0.8
Iridium		Barium	0.02	Selenium	0.1	Silicon	6
Osmium		Cesium		Arsenic	≤0.009	Aluminum	0.7
Rhenium		Iodine	0.004	Germanium		Magnesium	2
Tungsten		Tellurium	0.006	Gallium	0.002	Sodium	2
Tantalum	≤0.04	Antimony	0.02	Zinc	MC	Fluorine	≈0.2
Hafnium		Tin	≤0.006	Copper	MC	Oxygen	NR
Lutetium		Indium	STD	Nickel	* 0.7	Nitrogen	NR
Ytterbium		Cadmium	0.006	Cobalt	* 0.07	Carbon	NR
Thulium		Silver		Iron	7	Boron	* 0.03
Erbium		Palladium		Manganese	0.1	Beryllium	
Holmium		Rhodium		Chromium	0.5	Lithium	0.003
Dysprosium						Hydrogen	NR
						* Heterogeneous	

STD – Internal Standard
NR – Not Reported
All elements not detected < 0.002 µg/ml
MC – Major Component >10 µg/ml
INT – Interference

Approved: M Spooks

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 230 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-8434
Reply to · INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 23, 1980

Analyst: T. Bouts

P. O. No.:

Sample No.: 80-1473
XAD - MAN TEST

IAD No.: 97-E697-116-28

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN PPM WEIGHT

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	<0.1
Thorium		Gadolinium		Molybdenum	1	Titanium	*2
Bismuth		Europium		Niobium		Scandium	*<0.1
Lead	0.3	Samarium		Zirconium	<0.1	Calcium	62
Thallium		Neodymium		Yttrium		Potassium	22
Mercury	NR	Praseodymium		Strontium	0.2	Chlorine	1
Gold		Cerium	0.3	Rubidium		Sulfur	14
Platinum		Lanthanum	0.4	Bromine	0.8	Phosphorus	15
Iridium		Barium	0.9	Selenium	<0.1	Silicon	*500
Osmium		Cesium		Arsenic	<0.1	Aluminum	5
Rhenium		Iodine		Germanium		Magnesium	8
Tungsten		Tellurium		Gallium	*<0.1	Sodium	22
Tantalum	0.7	Antimony		Zinc	6	Fluorine	*=0.3
Hafnium		Tin		Copper	6	Oxygen	NR
Lutetium		Indium	STD	Nickel	180	Nitrogen	NR
Ytterbium		Cadmium		Cobalt	0.7	Carbon	NR
Thulium		Silver		Iron	26	Boron	<0.1
Erbium		Palladium		Manganese	1	Beryllium	
Holmium		Rhodium		Chromium	11	Lithium	<0.1
Dysprosium						Hydrogen	NR

* Heterogeneous

STD – Internal Standard
NR – Not Reported
All elements not detected < 0.1 ppm
MC – Major Component
INT – Interference

Approved: M Spacks

COMMERCIAL TESTING & ENGINEERING CO.

GENERAL OFFICES: 330 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-9494
Reply to INSTRUMENTAL ANALYSIS DIVISION, 14335 WEST 44TH AVENUE, GOLDEN, COLORADO 80401, PHONE: 303-278-9521

To: Mr. Brent Higginbotham
Acurex Corporation
485 Clyde Ave.
Mountain View, CA 94042

Date: September 22, 1980

Analyst: L. Jacobs

P. O. No.:

Sample No.: 80-1562

IAD No.: 97-E697-116-28

DIESEL OIL - MAN TEST

SPARK SOURCE MASS SPECTROGRAPHIC ANALYSIS
CONCENTRATION IN PPM WEIGHT

ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.	ELEMENT	CONC.
Uranium		Terbium		Ruthenium		Vanadium	0.02
Thorium		Gadolinium		Molybdenum	0.1	Titanium	5
Bismuth		Europium		Niobium		Scandium	<0.01
Lead	0.2	Samarium		Zirconium	0.04	Calcium	MC
Thallium		Neodymium		Yttrium	0.02	Potassium	7
Mercury		Praseodymium		Strontium		Chlorine	*70
Gold		Cerium		Rubidium	<0.01	Sulfur	0.2
Platinum		Lanthanum		Bromine	0.3	Phosphorus	1
Iridium		Barium	0.3	Selenium		Silicon	25
Osmium		Cesium		Arsenic	0.02	Aluminum	MC
Rhenium		Iodine		Germanium		Magnesium	2
Tungsten		Tellurium	0.2	Gallium	0.02	Sodium	4
Tantalium		Antimony		Zinc	0.7	Fluorine	=1
Hafnium		Tin	0.03	Copper	0.5	Oxygen	NR
Lutetium		Indium	STD	Nickel	1	Nitrogen	NR
Ytterbium		Cadmium	0.02	Cobalt	0.04	Carbon	NR
Thulium		Silver		Iron	10	Boron	0.04
Erbium		Palladium		Manganese	0.2	Beryllium	
Holmium		Rhodium		Chromium	0.4	Lithium	0.02
Dysprosium						Hydrogen	NR

* Heterogeneous

STD – Internal Standard
NR – Not Reported
All elements not detected < 0.01 ppm
MC – Major Component > 100 ppm
INT – Interference

Approved: M. Jacobs

ACUREX Corporation
ANALYSIS LABORATORIES

DATA REPORTING FORM

CUSTOMER CMEA DATE July 10, 1980

CUSTOMER CONTRACT NO. 307605.22 ACUREX CONTRACT NO. _____

RESULTS REPORT TO _____ TELEPHONE _____

ADDRESS _____

Mountain View May 21, 1980 MAN FURNACE SYSTEM

SAMPLE ID (CUSTOMER)	Fuel Hg	XAD Hg	1 Imp Hg	2&3 Hg	2&3 As	2&3 Sb	Water Hg			
SAMPLE ID (LAB)										
PARAMETER										UNITS
Blank	Less 1	Less 1	Less 1	Less 1	Less 10	Less 3	Less 1*			ug/L
Sample Aliquot	Less 1	Less 1	Less 1	Less 1	Less 10	Less 3	Less 1			ug/L
Total Sample			Less 1	Less 1	Less 10	Less 3	Less 1			ug/L
Total Sample	Less 0.1	Less 0.1								ug/g
Total Sample		Less 10	Less 1	Less 1	Less 10	Less 3				ug

*All water tank samples were below 1 ug/L

ANALYST G. Nicoll

REVIEWER Greg Nicoll

Form EED-057 4/80

COMMERCIAL TESTING & ENGINEERING CO.
GENERAL OFFICES: 220 NORTH LA SALLE STREET, CHICAGO, ILLINOIS 60601 · AREA CODE 312 726-6434

Reply to
Instrumental Analysis Division
490 Orchard Street
Golden, CO 80401

Phone: 303-278-9521

November 18, 1980

Mr. Brent Higginbotham
Acurex Aerotherm Corp.
485 Clyde Avenue
Mountain View, CA 94042

RE: IAD #97-E697-116-28

Analytical Report

We were asked as per phone conversation with Carlo Castaldini, on October 20, 1980, to check by atomic absorption, several elemental values on samples that were analyzed by spark source mass spectrometry (SSMS) and reported September 24, 1980.

Nickel was checked on the XAD Resins Acurex ID Nos. 80-1473 & 1533-37. Nickel contamination was due to combustion in the Parr combustion apparatus as they were prepared for SSMS. The samples were reprepped by ashing at approximately 600° C, dissolving the ash in aqua regia, and diluting with deionized water. Nickel was determined from this solution as well by flame atomic absorption.

Copper was found to be a major component on samples 80-1368 and 80-1369 by SSMS. This was quantitated by flame atomic absorption on a diluted portion of each of the samples.

The results of these analyses are presented in Table No. 1 and are reported in the appropriate units on the samples "as received".

Table No. 1

Sample #	Nickel (µg/g)	Copper (mg/l)
80-1473 MAN KAD	2.4	---
80-1368 Tank water prior to test---		505
80-1369 Tank water after test ---		480

If there are any questions concerning these results, please call.

Bruce A. Hale
Section Supervisor

M. L. Jacobs, Ph.D., Mngr.
Instrumental Analysis Div.

as

IC ANALYSIS SHEET

F, Cl, Br, NO_2, NO_3

Contractor: Acurex

Sample Site: Mountain View — Sample Acquisition Date: May 21, 1980

Type of Source:

Test Number: — Sample ID Number:

Sample Description: Water Tank

Analyst Responsible: E. Hagmann — Date Analyzed: July 21, 1980 — Time: 8pm - 2am

Calculations and Report Reviewed By: G. Nicoll — Report Date: July 23, 1980

Instrument: Micromeritics - Wescan Conductivity

Eluent: 0.0025M Sodium Benzoate @ pH 6.0

Column Flow Rate: 1.50 ml/min — Pressure: 1.08 KPSI — Recorder Speed: 0.5 cm/min

Sample Size: 100 ul — Attenuator Setting: 2

Original Sample Volume or Mass: — Multiple Standard Addition: Yes ____ No X

Observations: 30 min run, good separation

IC ANALYSIS SHEET

SO_3, SO_4

Contractor: Acurex

Sample Site: Mountain View — Sample Acquisition Date: May 21, 1980

Type of Source:

Test Number: — Sample ID Number:

Sample Description: Water Tank

Analyst Responsible: E. Hagmann — Date Analyzed: July 11, 1980 — Time: 8am - 8pm

Calculations and Report Reviewed By: G. Nicoll — Report Date: July 23, 1980

Instrument: Micromeritics - Wescan Conductivity

Eluent: 0.0047M KHP @ pH 4.5

Column Flow Rate: 2.00 ml/min — Pressure: 0.79 KPSI — Recorder Speed: 1 cm/min

Sample Size: 10 ul — Attenuator Setting: 4

Original Sample Volume or Mass: — Multiple Standard Addition: Yes ____ No X

Observations: Spike and recovery: average recovery at 88%

IC ANALYSIS SHEET
PO_4

Contractor: Acurex

Sample Site: Mountain View — Sample Acquisition Date: May 21, 1980

Type of Source:

Test Number: — Sample ID Number:

Sample Description: Water Tank

Analyst Responsible: E. Hagmann — Date Analyzed: July 22, 1980 — Time: 8am - 3pm

Calculations and Report Reviewed By: G. Nicoll — Report Date: July 23, 1980

Instrument: Micromeritics - Wescan Conductivity

Eluent: 5×10^{-3}M Phthalic Acid

Column Flow Rate: 2.00 ml/min — Pressure: 1.43 KPSI — Recorder Speed: 0.5 cm/min

Sample Size: 100 ul — Attenuator Setting: 2

Original Sample Volume or Mass: — Multiple Standard Addition: Yes ____ No X

Observations: Spike and recovery: 106% recovery

Water Tank Blank- Tap Water

Ion	Uncorrected Sample Value	Blank Value	Corrected Sample Value	High/Low Calibration Standards or Concentration Added	Dilution Factor	Assigned Concentration*	Detection Limit*
F^-	ND	ND		5, 50		ND	1
Cl^-	2	ND	2	5, 50		2	1
Br^-	ND	ND		5, 50		ND	5
NO_2^-	ND	ND		5, 50		ND	2
NO_3^-	ND	ND		5, 50		ND	5
$SO_3^=$	ND	ND		50		ND	10
$SO_4^=$	ND	ND		50, 500, 1000		ND	10
$PO_4^{\equiv}$	ND	ND		10, 50		ND	3

*Results: Mg/L values (in original sample or I – Interference; MC – major constituent, not quantified; NC – not computed; NG – sample value below blank; ND – not detectable (< 2 σ blank or baseline).

Water Tank- Sample Blank

Ion	Uncorrected Sample Value	Blank Value	Corrected Sample Value	High/Low Calibration Standards or Concentration Added	Dilution Factor	Assigned Concentration*	Detection Limit*
F^-	ND	ND		5, 50		ND	1
Cl^-	2	ND	2	5, 50		2	1
Br^-	ND	ND		5, 50		ND	5
NO_2^-	ND	ND		5, 50		ND	2
NO_3^-	ND	ND		5, 50		ND	5
$SO_3^=$	ND	ND		50		ND	10
$SO_4^=$	ND	ND		50, 500, 1000		ND	10
$PO_4^{\equiv}$	ND	ND		10, 50		ND	3

*Results: mg/L values (in original sample or I – Interference; MC – major constituent, not quantified; NC – not computed; NG – sample value below blank; ND – not detectable (< 2 σ blank or baseline).

Water Tank - Prior to Test

Ion	Uncorrected Sample Value	Blank Value	Corrected Sample Value	High/Low Calibration Standards or Concentration Added	Dilution Factor	Assigned Concentration*	Detection Limit*
F^-	ND	ND		5, 50		ND	3
Cl^-	3	ND	3	5, 50		3	2
Br^-	ND	ND		5, 50		ND	10
NO_2^-	ND	ND		5, 50		ND	10
NO_3^-	7	ND	7	5, 50		7	5
$SO_3^=$	ND	ND		50		ND	10
$SO_4^=$	990	ND	990	50, 500, 1000		990	10
$PO_4^{\equiv}$	ND	ND		10, 50		ND	20

*Results: mg/L values (in original sample or I – Interference; MC – major constituent, not quantified; NC – not computed; NG – sample value below blank; ND – not detectable (< 2 σ blank or baseline).

Water Tank - Sample End of test

Ion	Uncorrected Sample Value	Blank Value	Corrected Sample Value	High/Low Calibration Standards or Concentration Added	Dilution Factor	Assigned Concentration*	Detection Limit*
F^-	ND	ND		5, 50		ND	3
Cl^-	3	ND	3	5, 50		ND	2
Br^-	ND	ND		5, 50		ND	10
NO_2^-	ND	ND		5, 50		ND	10
NO_3^-	7	ND	7	5, 50		7	5
$SO_3^=$	ND	ND		50		ND	10
$SO_4^=$	1000	ND	1000 gn	50, 500, 1000		1000	10
$PO_4^{\equiv}$	ND	ND		10, 50		ND	20

*Results: mg/L values (in original sample or I – Interference; MC – major constituent, not quantified; NC – not computed; NG – sample value below blank; ND – not detectable (< 2 σ blank or baseline).

5.5 ORGANIC ANALYSIS REPORT

ORGANIC COMPOUNDS (bp < 100° C)

Contractor: Acurex

Sample Site: Mountain View — Sample Acquisition Date: May 21, 1980 Time: 14:05

Type of Source: Residential Hot Water Heating System

Test Number: No 1 — Sample ID Number: End of burn cycle

Sample Description: Exhaust gas

Analyst Responsible: M. Gardner — Date Analyzed: May 23, 1980 Time: _____

Calculations and Report Reviewed By: G. Nicoll — Report Date: May 23, 1980

Workup

1. Column Flow Rate (mL/min): _____
2. Recorder Speed: 1 cm/min
3. Full Scale (mV): 1.0
4. Column Pressure (psi): _____
5. Electrometer Set (A/mV): 10^{-12}
6. Calibration Date: May 21, 1980
7. Sample Size (mL): 1.0
8. Oven Temperature (°C): 150
9. Flame Flow Rates (mL/min): H_2 _____ Air _____
10. Attenuation: 8
11. Range: _____
12. Observations: Integrator at 12,000 cft/min

Results: PPM value (in original sample) or I – interference; NC – not computed; NG – sample value below blank; ND – not detectable (< 2 σ blank or baseline).

	Uncorrected Sum of Peak Areas	Blank Value	Retention Time	Corrected Sum of Peak Areas	Conc. (%)	ppm/cft³ Sensitivity	High/Low Calibration Standards	Conc. (ppm)
E_1	1.0	ND	0.7 min	1.0		0.087	9.9 ppm	< 0.2
E_2	ND	ND	1.0 min	ND		0.11	10.5 ppm	< 0.2
E_3	ND	ND	1.7 min	ND		0.17	11.2 ppm	< 0.5
E_4	ND	ND	3.9 min	ND		0.28	11.3 ppm	< 0.5
E_5	ND	ND	9.4 min	ND		0.37	12 ppm	< 1.0
E_6	ND	ND	23.2 min	ND		0.53	11.1 ppm	< 1.0
E_7	NC							

ORGANIC COMPOUNDS (bp < 100° C)

Contractor: HCUREX

Sample Site: MOUNTAIN VIEW Sample Acquisition Date: May 21, 1980 Time: 12:45

Type of Source: Residential Hot Water Heating System

Test Number: No. 1 Sample ID Number: 9 min into burn cycle

Sample Description: exhaust gas

Analyst Responsible: M. Gardner Date Analyzed: May 23, 1980 Time: _____

Calculations and Report Reviewed By: G. Nicoll Report Date: May 23, 1980

Workup

1. Column Flow Rate (mL/min): _____
2. Recorder Speed: 1 cm/min
3. Full Scale (mV): 1.0
4. Column Pressure (psi): _____
5. Electrometer Set (A/mV): 10^{-12}
6. Calibration Date: May 21, 1980
7. Sample Size (mL): 1.0
8. Oven Temperature (°C): 150
9. Flame Flow Rates (mL/min): H_2 _____ Air _____
10. Attenuation: 8
11. Range: _____
12. Observations: Integrator at 12,000 ft³/min

Results: PPM value (in original sample) or I – interference; NC – not computed; NG – sample value below blank; ND – not detectable (<2 σ blank or baseline).

#	Uncorrected Sum of Peak Areas	Blank Value	Retention Time	Corrected Sum of Peak Areas	Conc. (%)	ppm/acf Sensitivity	High/Low Calibration Standards	Conc. (ppm)
K_1	1.	ND	0.7 min	1.0		0.087	9.9 ppm	<0.2
K_2	ND	ND	1.0 min	ND		0.11	10.5 ppm	<0.2
K_3	ND	ND	1.7 min	ND		0.17	11.2 ppm	<0.5
K_4	ND	ND	3.3 min	ND		0.28	11.3 ppm	<0.5
K_5	ND	ND	9.4 min	ND		0.37	12 ppm	<1.
K_6	ND	ND	23.8 min	ND		0.53	11.1 ppm	<1.
K_7	NC							

ORGANIC COMPOUNDS (bp < 100° C)

Contractor Acurex

Sample Site Mountain View Sample Acquisition Date May 21, 1980 Time 15:10

Type of Source Residential Hot Water Heating System

Test Number No. 1 Sample ID Number Beginning of burn cycle

Sample Description Exhaust gas

Analyst Responsible M. Gardner Date Analyzed May 23, 1980 Time

Calculations and Report Reviewed By G. Nicoll Report Date May 23 1980

Workup

1. Column Flow Rate (mL/min) 2. Recorder Speed 1 cm/min
3. Full Scale (mV) 1.0 4. Column Pressure (psi)
5. Electrometer Set (A/mV) 10^{-12} 6. Calibration Date May 21, 1980
7. Sample Size (mL) 1.0 8. Oven Temperature (°C) 150
9. Flame Flow Rates (mL/min): H_2 Air
10. Attenuation 8 11. Range
12. Observations Integrator at 12,000 ft³/min

Results: PPM value (in original sample) or I – interference; NC – not computed; NG – sample value below blank; ND – not detectable (< 2 σ blank or baseline).

#	Uncorrected Sum of Peak Areas	Blank Value	Retention Time	Corrected Sum of Peak Areas	Conc. (%)	ppm/pct Sensitivity	High/Low Calibration Standards	Conc. (ppm)
K_1	5	ND	0.7 min	5		0.087	9.7 ppm	0.4
K_2	ND	ND	1.0 min	ND		0.11	10.5 ppm	< 0.2
K_3	ND	ND	1.7 min	ND		0.17	11.2 ppm	< 0.5
K_4	ND	ND	3.9 min	ND		0.28	11.3 ppm	< 0.5
K_5	ND	ND	9.4 min	ND		0.37	12.0 ppm	< 1.0
K_6	ND	ND	23.8 min	ND		0.53	11.1 ppm	< 1.0
K_7	ND							

ACUREX Corporation
ANALYSIS LABORATORIES

DATA REPORTING FORM

CUSTOMER CMEA DATE July 15, 1980

CUSTOMER CONTRACT NO. 307605.22 ACUREX CONTRACT NO. _____

RESULTS REPORT TO _____ TELEPHONE _____

ADDRESS _____

Mountain View May 21, 1980 Residential Wall Furnace

SAMPLE ID (CUSTOMER)	Filter	XAD	OMC	Tank Blank	Tap Water	Prior to Test	Tank Sample			
SAMPLE ID (LAB)										
PARAMETER										UNITS
TCO Blank		0.07	Less 0.02	Less 0.02	Less 0.02	Less 0.02	Less 0.02			mg
TCO Sample		74	Less 0.02	Less 0.02	Less 0.02	Less 0.02	0.50			mg
TCO, Corrected		74	Less 0.02	Less 0.02	Less 0.02	Less 0.02	0.50			mg
GRAV Blank	Less 2	4	Less 2	Less 2	Less 2	Less 2	Less 2			mg
GRAV Sample	3	22	5	6	Less 2	Less 2	Less 2			mg
GRAV, Corrected	3	18	5	6	Less 2	Less 2	Less 2			mg
TCO & GRAV	3	92	5	6	Less 2	Less 2	Less 2			mg
TCO & GRAV	0.1	3.2	0.2	0.2	Less 0.1	Less 0.1	Less 0.1			mg/dscm
Volume Analyzed				3665	3720	1300	10,000			ml

ANALYST M. Gardner, G. Nicoll

REVIEWER _____

Form EED-057 4 80

IR REPORT

SAMPLE: Filter Mountain View 5/21/80

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: Filter Blank

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: XAD Mountain View 5/21/80

Wave Number (cm^{-1})	Intensity	Assignment	Comments
3575-3070	W	O-H	
2905	S	C-H Alkane	
2845	S	C-H Alkane	
2750-2450	W	O-H	
1725	M	C=O	
1445.	W	C-H Alkane	
1265	W	C-O	
1095	W	C-O	
805	W	Not Assigned	

IR REPORT

SAMPLE: XAD Blank

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: OMC Mountain View 5/21/80

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: Tap Water Mountain View 5/21/80

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: Tank Blank Mountain View 4/21/80

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: Tank Sample Mountain View 5/21/80

Wave Number - (cm^{-1})	Intensity	Assignment	Comments
2905	S	C-H Alkane	
2845	S	C-H Alkane	
1190	S	Sulfonamide	Found in GC/MS PNA

COMPOUNDS SOUGHT
Detection Limit in nanograms

8	* 4-bromophenyl phenyl ether
1	* bis (2-chloroisopropyl) ether
2	* bis (2-chloroethoxy) methane
8	* hexachlorobutadiene
40	* hexachlorocyclopentadiene
1	* isophorone
1	* naphthalene
8	* nitrobenzene
4	* N-nitrosodiphenylamine
40	* N-nitrosodi-n-propylamine
3	* bis (2-ethylhexyl) phthalate
3	* butyl benzyl phthalate
1	* di-n-butyl phthalate
2	* di-n-octyl phthalate
2	* diethyl phthalate
2	* dimethyl phthalate
5	* benzo(a)anthracene
7	* benzo(a)pyrene
8	* 3,4-benzofluoranthene
8	* benzo(k)fluoranthene
5	* chrysene
1	* acenaphthylene
1	* anthracene
40	* benzo(ghi)perylene
2	* fluorene
1	* phenanthrene
40	* dibenzo(a,h)anthracene
40	* indeno(1,2,3-cd)pyrene
2	* pyrene
20	2,3,7,8-tetrachlorodibenzo-p-dioxin
2	* acenaphthene
100	* benzidine
8	* 1,2,4-trichlorobenzene
8	* hexachlorobenzene
8	* hexachloroethane
3	* bis(2-chloroethyl)ether
2	* 2-chloronaphthalene
4	* 1,2-dichlorobenzene
8	* 1,3-dichlorobenzene
4	* 1,4-dichlorobenzene
20	* 3,3-dichlorobenzindine
10	* 2,4-dinitrotoluene
10	* 2,6-dinitrotoluene
1	* 1,2-diphenylhydrazine (as azobenzene)
2	* fluoranthene
4	* 4-chlorophenyl phenyl ether
40	anthanthrene
40	benzo(e)pyrene
@	dibenzo(a,H)pyrene
@	dibenzo(a,i)pyrene
40	dibenzo(c,g)carbozole
40	7,12 dimethyl benz(a)anthracene
40	3-methyl cholanthrene
40	perylene
40	Benzo(c)phenanthrene

*Authenic standard ran

@Molecular weight too high for direct analysis by Base/Neutral run

ACUREX Corporation
ANALYSIS LABORATORIES

DATA REPORTING FORM

CUSTOMER CMEA DATE July 21, 1980

CUSTOMER CONTRACT NO. 307605.22 ACUREX CONTRACT NO. _____

RESULTS REPORT TO _____ TELEPHONE _____

ADDRESS _____

Mountain View 5/21/80 28.5 dscm

SAMPLE ID (CUSTOMER)	Filter Bk	Filter	XAD Blank	Xad	OMC	@ Water Blk	Tank water			
SAMPLE ID (LAB)										
PARAMETER										UNITS
Amount injected	2	2	2	1	2	3	3			uL
Naphthalene*	Less 0.5	Less 0.5	Less 0.5	94	Less 0.5	Less 0.3	9			mg/L
Phenanthrene/										
Anthracene*	Less 0.5	Less 0.5	Less 0.5	4	Less 0.5	Less 0.3	2			mg/L
Naphthalene		Less 0.02		36	Less 0.02		0.4			ug/dscm
Phenanthrene/										
Anthracene		Less 0.02		2	Less 0.02		0.08			ug/dscm
Volume analyzed							10,000			mL

*Sample Aliquot @All water tank blanks are clean

ANALYST _____

REVIEWER Greg Nicol

Form EED-057 4/80

SAMPLE: Man XAD #1473

	TCO mg	GRAV mg	TCO + GRAV Total mg	Concentration mg/dscm
Total Sample[1]	74	18	92	3.2
Taken for LC[2]	36	9	45	1.6
Recovered[3]	26	14	40	1.4

Fraction	TCO in mg				GRAV in mg				TCO + GRAV Total mg	Concentration mg/dscm
	Found in Fraction	Blank	Corrected	Total[4]	Found in Fraction	Blank	Corrected	Total[4]		
1	25	<0.02	25	52	11	<1	11	23	75	2.6
2	0.09	<0.01	0.09	0.2	1.0	<0.8	1	2	2	0.07
3	0.41	<0.01	0.41	0.85	<0.8	<0.8	< 0.8	< 2	< 3	0.1
4	0.06	<0.01	0.06	0.1	1.2	1.2	< 0.8	< 2	< 2	< 0.07
5	<0.01	<0.01	<0.01	<0.02	<0.8	<0.8	< 0.8	< 2	< 2	< 0.07
6	<0.01	<0.01	<0.01	<0.02	1.0	<0.8	1.0	2	2	0.07
7	<0.01	<0.01	<0.01	<0.02	2.0	<0.8	2.0	4	4	0.1
Sum	26	< 0.08	26	54	16	< 6	15	31	85	3.0

1. Quantity in entire sample, determined before LC
2. Portion of whole sample used for LC, actual mg
3. Quantity recovered from LC column, actual mg
4. Total mg computed back to total sample

IR REPORT

SAMPLE: MAN XAD 1473 F1

Wave Number (cm^{-1})	Intensity	Assignment	Comments
2920	s	CH Aliphatic hydrocarbons	
2840	s	CH Aliphatic hydrocarbons	

IR REPORT

SAMPLE: MAN XAD 1473 F2

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: MAN XAD 1473 F3

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: MAN XAD 1473 F4

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: MAN XAD 1473 F5

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: MAN XAD 1473 F6

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

IR REPORT

SAMPLE: MAN XAD 1473 F7

Wave Number (cm^{-1})	Intensity	Assignment	Comments
		No Peaks	

LRMS REPORT

SAMPLE: 1473 F1 Mountain View XAD

Major Categories

Intensity	Category	MW Range
1	Aliphatic hydrocarbons	

Sub-Categories, Specific Compounds

Intensity	Category	m/e	Composition

Other

5.6 BIOASSAY ANALYSES REPORT

GENETICS ASSAY NO. 5397

LBI SAFETY NO. 6391

CYTOTOXIC EVALUATION OF

SAMPLE #1473
(CMEA MOUNTAIN VIEW)

IN THE
RODENT CELL (CHO)
CLONAL TOXICITY ASSAY

FINAL REPORT

SUBMITTED TO:

ACUREX CORPORATION
485 CLYDE AVENUE
MOUNTAIN VIEW, CALIFORNIA 94042

SUBMITTED BY:

LITTON BIONETICS, INC.
5516 NICHOLSON LANE
KENSINGTON, MARYLAND 20795

LBI PROJECT NO. 20933

REPORT DATE: DECEMBER, 1980

PREFACE

This report contains a summary of the data compiled during the evaluation of the test compound. The report is organized to present the results in a concise and easily interpretable manner. The first part contains items I - IX. Items I - IV provide sponsor and compound identification information, type of assay, and the assay design reference number. All assay design references indicate a standard procedure described in the Litton Bionetics, Inc. "Screening Program for the Identification of Potential Mutagens and Carcinogens." Item V provides the initiation and completion dates for the study, and Item VI provides identification of supervisory personnel. Item VII identifies the tables and figures containing the data used by the study director in interpreting the test results. The interpretation itself is in Item VIII. Item IX provides the conclusion and evaluation.

The second part of the report, entitled Assay Design, describes the materials and procedures employed in conducting the assay. This part of the report also contains any appendices, as well as evaluation criteria used by the study director. The evaluation criteria are included to acquaint the sponsor with the methods used to develop and analyze the test results.

All test and control results presented in this report are supported by fully documented raw data which are permanently maintained in the files of the Department of Genetics and Cell Biology or in the archives of Litton Bionetics, Inc., 5516 Nicholson Lane, Kensington, Maryland, 20795.

Copies of raw data will be supplied to the sponsor upon request.

I. SPONSOR: Acurex Corporation

II. MATERIAL (TEST COMPOUND): GENETICS ASSAY NUMBER: 5397

A. Identification: Sample #1473 (CMEA Mountain View)

B. Date Received: October 9, 1980

C. Physical Description: Brown, opaque liquid

III. TYPE OF ASSAY: Rodent Cell (CHO) Clonal Toxicity Assay

IV. ASSAY DESIGN NUMBER: 442

V. STUDY DATES:

A. Initiation: October 20, 1980

B. Completion: November 24, 1980

VI. SUPERVISORY PERSONNEL:

A. Study Director: Brian C. Myhr, Ph.D.

B. Laboratory Supervisor: Robert Young, M.S.

VII. RESULTS:

The data are presented in Table 1 on page 4 and in Figure 1 on page 5.

VIII. INTERPRETATION OF RESULTS:

The test material, Sample No. 1473, was supplied as a SASS train organic extract in 10 ml of methylene chloride. The total organic content was given as 26 mg. A portion of the sample (8.20 ml) was solvent exchanged into an equal volume of dimethylsulfoxide (DMSO). The concentration of organics was therefore unchanged, and the value of 2.6 mg/ml (or 2.6 µg/µl) was used to convert the doses obtained on a volume basis into the equivalent amount of organics per milliliter of culture medium.

The test material remained completely soluble after exchange into DMSO. Diluted stocks were prepared with DMSO just prior to testing, and the treatments were initiated with culture media containing 1:100 dilutions of the stocks. The highest test concentration was achieved with a 1:50 dilution of the solvent-exchanged sample, which introduced 2% DMSO into the medium. Vehicle control cultures were therefore exposed to

VIII. INTERPRETATION OF RESULTS: (continued)

culture media containing 1% and 2% DMSO in order to provide the reference points for determining the survival of the cells to treatments with the test material. No precipitation of test material in the culture medium was observed at any of the tested concentrations.

Seven test concentrations from 0.05 µl/ml to 20 µl/ml were evaluated for their effects on colony survival. As shown in Table 1 and Figure 1, none of the assayed concentrations caused significant changes in the numbers of colonies. This lack of observable toxicity was not conclusive, however, because the highest concentration corresponded to only 52 µg of organics/ml, which was essentially the midpoint of the concentration range where an EC50 would yield a moderate toxicity classification (see Evaluation Criteria). A sharp increase in toxicity could occur with a small increase in concentration, but it is reasonable to assume that 50% survival would not have been observed had the high dose been doubled. This concentration would have fallen just within the low toxicity region. Thus, the test material was evaluated as possibly having low toxicity or no detectable toxicity to CHO cells. The position of the EC50 could be further investigated by concentrating the extracted organic material.

IX. CONCLUSIONS:

The test material, Sample No. 1473, was evaluated as having low or nondetectable toxicity in the CHO Clonal Toxicity Assay. No toxicity was observed for concentrations up to the highest testable level of 52 µg of organics/ml.

Submitted by:

Study Director

Brian C. Myhr 12/1/80

Brian C. Myhr, Ph.D. Date
Section Chief
Mammalian Genetics
Department of Genetics
and Cell Biology

Reviewed by:

David J. Brusick, Ph.D. 12/1/80 Date
Director
Department of Genetics
and Cell Biology

TABLE 1

RODENT CELL (CHO) CLONAL TOXICITY ASSAY

Sample Identity: Sample #1473, CMEA MOUNTAIN VIEW

Description of Test Sample: Clear, pale yellow solution in DMSO.

LBI Assay No.: 5397

Date Received: October 9, 1980

Test Date: November 17, 1980

Vehicle: DMSO

Cell Type: CHO-K1

Cells Seeded per Dish: 200

EC50 Value: >20 µl/ml (>52 µg/ml)

Toxicity Classification: Probably Low or Non-detectable

pH Alterations: NONE

Comments on Treatment:

CLONAL TOXICITY DATA

Sample	Applied Concentration µl/ml	Dish #1	Dish #2	Dish #3	Average Count	Relative Survival* %	Cloning Efficiency %
NC	---	207	185	192	194.7	---	97.4
VC, 1%	10.0	180	177	172	176.3	100.0	88.2
VC, 2%	20.0	173	162	166	167.0	100.0	83.5
Test	0.05	183	192	189	188.0	106.6	
Test	0.2	C	192	183	187.5	106.4	
Test	0.5	185	189	179	184.3	104.5	
Test	2.0	161	187	183	177.0	100.4	
Test	5.0	161	190	164	171.7	97.4	
Test	10.0	166	178	180	174.7	99.1	
Test	20.0	170	171	154	165.0	98.8	

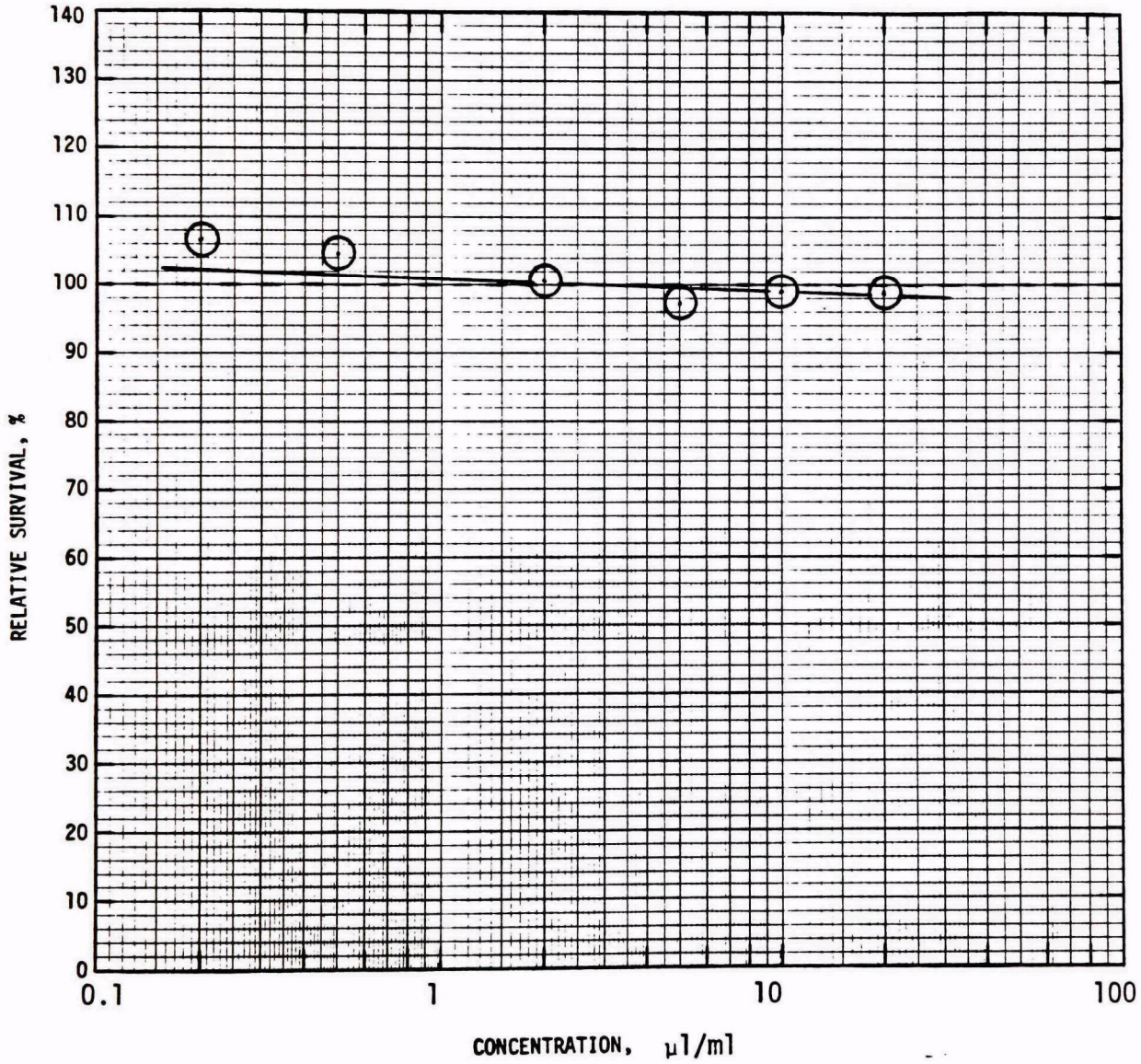
NC = Negative Control, F12 medium.
VC = Vehicle Control, percent DMSO in F12 as noted.
* Relative to 2% VC for 20 µl/ml and to 1% VC for other treatments.
C = Contaminated.

FIGURE 1

RODENT CELL (CHO) CLONAL TOXICITY ASSAY

EC_{50} DETERMINATION

SAMPLE #1473, CMEA MOUNTAIN VIEW



ASSAY DESIGN NO. 442

RODENT CELL CLONAL (CHO) TOXICITY ASSAY

This assay conforms to one of the bioassay tests in the US Environmental Protection Agency (EPA) IERL - RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests (September, 1980). Level 1 bioassays obtain preliminary information on the harmful biological effects of chemicals found in industrial feed and waste streams by using mammalian cells in culture to measure metabolic impairment and cellular death.

1. OBJECTIVE

The objective of this assay is to determine the concentration of a test article that will reduce the colony forming ability of Chinese hamster cells by 50% after a 24-hour exposure. This concentration is referred to as the EC_{50} value.

2. MATERIALS

A. Indicator Cells

The indicator cells used in this study are Chinese hamster CHO-K1 cells (American Type Culture Collection No. CCL 61). This cell type was derived from ovarian tissue and has spontaneously transformed to a stable, hypodiploid line of rounded, fibroblastic cells with unlimited growth potential. Monolayer cultures have a fast doubling time of 11 to 14 hours, and untreated cells can normally be cloned with an efficiency of 80% or greater. Permanent stocks are maintained in liquid nitrogen and laboratory cultures are maintained by serial subculturing. Laboratory cultures are periodically checked by culturing methods for the absence of mycoplasma contamination. This test system is specified by the IERL- RTP Procedures Manual.

B. Medium

The CHO-K1 cell line has an absolute requirement for proline and therefore must be maintained in a culture medium containing sufficient amounts of this amino acid. Ham's F12 medium, which contains 3×10^{-4}M L-proline, is used, supplemented with 10% fetal bovine serum, 100 units per milliliter of penicillin, 100 µg streptomycin per milliliter, and 0.5 µg amphotericin B (Fungizone) per milliliter.

C. Controls

Untreated cells are cloned to establish the control cloning efficiency. If the test article is dissolved in an organic solvent (usually dimethylsulfoxide), cells exposed to solvent in the medium

2. MATERIALS (Continued)

are cloned to provide the reference cloning efficiency for the effect of the test article. The final concentration of solvent in the growth medium is generally 1% or less. All controls are performed in triplicate.

D. Sample Forms

Solid samples are tested as a solution or are ground to fine particles (less than 5 μm) and tested as a suspension in growth medium. Dry particulate articles, aqueous liquids, suspensions and slurries are added directly to the growth medium and tested as a suspension or solution. Liquids containing less than 0.2% organic solvent are generally tested directly; samples dissolved in organic solvents are solvent exchanged in dimethylsulfoxide (DMSO) before testing. Original sample volumes may be reduced a maximum of 10-fold during solvent exchange, and the concentration factor is used to convert assayed volumes into equivalent original sample volumes. All sample manipulations are performed as described in the IERL-RTP Level 1 Procedures Manual.

3. EXPERIMENTAL DESIGN

A. Dose Selection

Unless the approximate toxicity is already known or the sample size is limiting, the following dose ranges are tested for different sample forms. Dry particulate articles are dissolved or suspended in growth medium and tested at five dose levels from 1000 μg/ml to 10 μg/ml. Aqueous samples, suspensions or slurries are tested from 600 μliters/ml to 6 μliters/ml in five dose steps. Samples that are solvent exchanged into DMSO are tested from 20 μliters/ml to 0.2 μliters/ml, also in five dose steps. Solvent concentrations are 1% for all dose levels except the maximum applicable dose (MAD) of 20 μl/ml, which contains 2% solvent. A second dose study is performed with an appropriate dose range if the EC_{50} has not been properly located in the initial test; EC_{50} values greater than 1,000 μg/ml, 600 μliters of aqueous sample/ml or 20 μliters nonaqueous sample/ml are not determined.

B. Clonal Toxicity Assay

Cells from monolayer stock cultures in logarithmic growth phase are trypsinized, counted by hemacytometer and reseeded into a series of 60- or 100-mm culture dishes at 200 cells per dish. The cultures are incubated for 6 to 16 hours at 37°C to allow attachment of the cells and recovery of growth rate.

Test article is then applied (three dishes per dose), and the cultures returned to the incubator. If the test article causes a color change, an additional dish is treated with the high dose and the pH of the

3. EXPERIMENTAL DESIGN (Continued)

medium is determined. The pH is also recorded for the highest dose which results in a slight color change. After a 24-hour exposure period the medium is aspirated and the cells washed with Dulbecco's phosphate buffered saline (PBS; prewarmed to 37°C). The pH of the discarded medium for which initial pH measurements were made is again recorded. Fresh medium is placed on each culture and incubation continued for an additional 6 days to allow colony development. Medium is drained from the cultures and the surviving colonies are washed with PBS, fixed in methanol, and stained with Giemsa. Colony counting is performed by eye; colonies smaller than 50 to 100 cells are not counted.

4. ASSAY ACCEPTANCE CRITERIA

The assay is considered acceptable for evaluation of the test results if the following criteria are met:

- The average cloning efficiency of the CHO-K1 cells in the negative controls is 70% or greater, but not exceeding 115%.
- The distribution of colonies in the treated cultures is generally uniform over the surface of the culture dish.
- The data points for each test concentration critical to the location of the EC_{50} are the averages of at least two treated cultures.
- A sufficient number of test concentrations are available to clearly locate the EC_{50} within a toxicity region as defined under Assay Evaluation Criteria.
- If the EC_{50} value is greater than 1000 µg/ml, 600 µliters of aqueous sample/ml, or 20 µliters of nonaqueous sample/ml, the plotted curve does not exceed 110% of the negative control.

5. ASSAY EVALUATION CRITERIA

The screened doses, pH values (if appropriate), colony counts, percent survivals (colony counts relative to control colony counts) and EC_{50} values are provided. The percent survival is plotted as a function of applied concentration and the EC_{50} value determined graphically by fitting a curve by eye through the data points. The EC_{50} is used to rank the test material using the standard evaluation criteria defined in the table below.

Sorbent extracts of known organic content are evaluated as nonaqueous liquids and in terms of their solid contents (µg organics/ml), and are ranked using the more sensitive parameter. Where data on the original liquid or gas sample volumes is provided by the Sponsor, the EC_{50} value is calculated in terms of these equivalent volumes per milliliter of culture medium; however, no evaluations are made as yet on this basis.

5. ASSAY EVALUATION CRITERIA (Continued)

Toxicity[a]	Solids (EC_{50} in µg/ml)	Aqueous Liquids (EC_{50} in µl/ml)	Nonaqueous Liquids[b] (EC_{50} in µl/ml)
High	<10	<6	<.2
Moderate	10 to 100	6 to 60	.2-2
Low	100 to 1000	60 to 600	2-20
Not Detectable	>1000	>600	>20

[a]Evaluation criteria formulated by Litton Bionetics, Inc. for IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests.

[b]Criteria for nonaqueous liquids are tentative and under evaluation.

6. RECORDS TO BE MAINTAINED

All raw data, protocols, protocol modifications, test article weight and dispensation records and correspondence between LBI and the Sponsor are being maintained in a central file within the Department of Genetics and Cell Biology. These records will be filed under departmental assay number and held up to 2 years following submission of the final report to the Sponsor. After 2 years they will be transferred to the LBI archives for permanent storage.

Q.A. Inspection Statement
(reference 21 CFR 58.35(b)(7))

PROJECT 10993 LBI Assay No. 5397

TYPE of STUDY Rodent Cell (CHO) Clonal Toxicity Assay

This final study report was reviewed by the LBI Quality Assurance Unit on December 1, 1980 A report of findings was submitted to the Study Director and to Management on December 1, 1980

The short-term nature of this study precluded inspection while it was in process. The Quality Assurance Unit inspects an in-process study of this type approximately once per month to assure that no significant problems exist that are likely to affect the integrity of this type of study.

Patrice M. Cuccaro
Auditor, Quality Assurance Unit

GENETICS ASSAY NO. 5397

LBI SAFETY NO. 6391

MUTAGENICITY EVALUATION OF SAMPLE #1473

IN THE AMES SALMONELLA/MICROSOME PLATE TEST

FINAL REPORT

SUBMITTED TO:

ACUREX CORPORATION
485 CLYDE AVENUE
MOUNTAIN VIEW, CALIFORNIA 94042

SUBMITTED BY:

LITTON BIONETICS, INC.
5516 NICHOLSON LANE
KENSINGTON, MARYLAND 20795

LBI PROJECT NO. 20988

REPORT DATE: DECEMBER, 1980

PREFACE

This report contains a summary of the data compiled during the evaluation of the test compound. The report is organized to present the results in a concise and easily interpretable manner. The first part contains items I-IX. Items I-IV provide sponsor and compound identification information, type of assay, and the protocol reference number. All protocol references indicate a standard procedure described in the Litton Bionetics, Inc. "Screening Program for the Identification of Potential Mutagens and Carcinogens." Item V provides the initiation and completion dates for the study, and Item VI provides identification of supervisory personnel. Item VII identifies the tables and/or figures containing the data used by the study director in interpreting the test results. The interpretation itself is in Item VIII. Item IX provides the conclusion and evaluation.

The second part of the report describes the materials and procedures employed in conducting the assay. This part of the report also contains evaluation criteria used by the study director, and any appendices. The evaluation criteria are included to acquaint the sponsor with the methods used to develop and analyze the test results.

All test and control results presented in this report are supported by fully documented raw data which are permanently maintained in the files of the Department of Genetics and Cell Biology or in the archives of Litton Bionetics, Inc., 5516 Nicholson Lane, Kensington, Maryland 20795.

I. SPONSOR: Acurex Corporation

II. MATERIAL (TEST COMPOUND): GENETICS ASSAY NUMBER: 5397

A. Identification: Sample #1473

B. Date Received: October 9, 1980

C. Physical Description: Pale yellow liquid

III. TYPE OF ASSAY: Ames Salmonella/microsome Mutagenesis Assay

IV. PROTOCOL NUMBER: 401 (EPA-Level I)

V. STUDY DATES:

A. Initiation: November 7, 1980

B. Completion: November 13, 1980

VI. STUDY DIRECTOR: D.R. Jagannath, Ph.D.

VII. RESULTS:

The results of this assay are presented in Table 1.

VIII. INTERPRETATION OF RESULTS:

The test material, Sample No. 1473, was supplied as a SASS train organic extract in the 10 ml of methylene chloride. The total organic content was given as 26.0 mg. A portion of the sample (8.20 ml) was solvent exchanged into dimethylsulfoxide (DMSO) to a final volume of 8.20 ml. This solvent exchanged sample (test material) was examined for mutagenic activity in the Ames/Salmonella assay in the presence and absence of liver microsomal enzyme preparations from Aroclor-induced rats.

A negative control consisting of the solvent DMSO and specific positive compounds were also assayed concurrently with the test material.

DOSE RANGE:

The dose range employed for the evaluation of this test material was from 10.0 µl to 200.0 µl per plate. Based on the organic content of the sample the doses employed would equal to 26.0 µg to 520.0 µg per plate. The tests were conducted using two plates per dose level.

VIII. INTERPRETATION OF RESULTS: (continued)

TOXICITY

The test material did not exhibit toxicity with any of the indicator strains used in this assay.

The results of the tests conducted on the test material in the absence of an activation system was positive with the strain TA-98. The lowest dose at which the response was observed was at 100 µl or 260.0 µg organics per plate.

The results of the test conducted on the test material in the presence of rat liver activation system were negative.

IX. CONCLUSIONS:

(1) The SASS train organic extract sample #1473 exhibited genetic activity with the strain TA-98 in the nonactivation assays conducted in this evaluation and is considered as mutagenic under these test conditions. (2) Based on the EPA's 'Definition of Toxicity Categories for Health Effects Tests' the mutagenic activity of the sample is classified as 'Moderate' (see Table A). The specific Activity at the Minimum Effective Concentration (which is the number of revertants minus the background revertants divided by µg organics x 1000) of the sample (260.0 µg) based on the results from TA-98 (nonactivation) was 92.31 revertants per mg of organics.

The classification of the mutagenic activity (item 2 above) and the Specific Activity of the sample (item 3 above) are given here to compare the potency of various samples belonging to the same class.

Submitted by:

Study Director

D.R. Jagannath 12/3/80

D.R. Jagannath, Ph.D. Date
Section Chief
Submammalian Genetics
Department of Genetics
and Cell Biology

Reviewed by:

for: Brian C. Myhr 12/3/80

David J. Brusick, Ph.D. Date
Director
Department of Genetics
and Cell Biology

TABLE A

DEFINITION OF TOXICITY CATEGORIES FOR HEALTH EFFECTS ASSAYS*

Assay[a]	Activity Measured[b]	Sample Type[c]	MAD[d]	Units	Range of Concentration or Dosage			
					High	Moderate	Low	Not Detectable (ND)
Ames	MEC (mutagenesis)	S	5	mg/plate	<0.05	0.05-0.5	0.5-5	ND at >5
		AL,NAL	200	µl/plate	<2	2-20	20-200	ND at >200
RAM	EC_{50} (lethality)	S	1	mg/ml	<0.01	0.01-0.1	0.1-1	ND at >1
		AL	600	µl/ml	<6	6-60	60-600	ND at >600
		NAL	20	µl/ml	<0.2	0.2-2	2-20	ND at >20
CHO	EC_{50} (lethality)	S	1	mg/ml	<0.01	0.01-0.1	0.1-1	ND at >1
		AL	600	µl/ml	<6	6-60	60-600	ND at <600
		NAL	20	µl/ml	<0.2	0.2-2	2-20	ND at >20
WAT	LD_{50} (lethality and toxic signs)	S	5	gm/kg	<0.05	0.05-0.5	0.5-5	ND at <5
		AL,NAL	5	ml/kg	<0.05	0.5-0.5	0.5-5	ND at <5

[a]Standard test abbreviations are as follows:
Ames: Ames Salmonella/microsome mutagenesis assay
RAM: Rabbit alveolar macrophage cytotoxicity assay
CHO: Rodent cell clonal toxicity assay
WAT: Acute in vivo test in rodents (whole animal test)

[b]Standard abbreviations for measured endpoints are as follows:
MEC: Minimum effective concentration
EC_{50}: Calculated concentration expected to produce effect in 50 percent of population
LD_{50}: Calculated dose expected to kill 50 percent of population

[c]S = Solid, AL = Aqueous liquid, NAL = Nonaqueous liquid

[d]MAD = Maximum applicable dose

*Evaluation criteria formulated by Litton Bionetics, Inc. for IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests.

RESULTS

TABLE 1

A. NAME OR CODE DESIGNATION OF THE TEST COMPOUND: SAMPLE #1473
B. SOLVENT: DMSO
C. TEST INITIATION DATES: 11/07/80
D. TEST COMPLETION DATE: 11/13/80
E. S-9 LOT#: 00009
NOTE: CONCENTRATIONS ARE GIVEN IN MICROLITERS PER PLATE

TEST	SPECIES	TISSUE	REVERTANTS PER PLATE TA-1535			TA-1537			TA-98			TA-100		
			1	2	3	1	2	3	1	2	3	1	2	3
NONACTIVATION														
SOLVENT CONTROL	---	---	12	11		6	9		22	18		168	193	
POSITIVE CONTROL**	---	---	990	1050		191	225		861	727		1383	1274	
TEST COMPOUND														
10.000000 UL	---	---	22	21		10	10		28	21		155	141	
25.000000 UL	---	---	34	25		9	6		30	21		169	155	
50.000000 UL	---	---	17	21		10	3		30	44		173	152	
100.000000 UL	---	---	21	23		10	8		58	30		176	140	
200.000000 UL	---	---	24	28		14	9		63	33		153	146	
ACTIVATION														
SOLVENT CONTROL	RAT	LIVER	9	9		9	9		14	15		190	198	
POSITIVE CONTROL***	RAT	LIVER	118	150		179	161		980	1066		1942	1746	
TEST COMPOUND														
10.000000 UL	RAT	LIVER	9	10		16	15		32	21		151	118	
25.000000 UL	RAT	LIVER	8	10		6	9		37	29		152	135	
50.000000 UL	RAT	LIVER	11	10		8	5		33	18		145	152	
100.000000 UL	RAT	LIVER	6	11		3	3		23	29		193	181	
200.000000 UL	RAT	LIVER	19	14		1	5		29	24		223	228	

**

TA-1535	SODIUM AZIDE	10 UG/PLATE
TA-1537	9-AMINOACRIDINE	50 UG/PLATE
TA-98	2-NITROFLUORENE	10 UG/PLATE
TA-100	SODIUM AZIDE	10 UG/PLATE

SOLVENT 200 UL/PLATE

***

TA-1535	2-ANTHRAMINE	2.5 UG/PLATE
TA-1537	2-ANTHRAMINE	2.5 UG/PLATE
TA-98	2-ANTHRAMINE	2.5 UG/PLATE
TA-100	2-ANTHRAMINE	2.5 UG/PLATE

AMES SALMONELLA/MICROSOME PLATE ASSAY

1. OBJECTIVE

The objective of this study was to evaluate a test material for mutagenic activity in a bacterial assay with and without a mammalian S9 activation system.

2. RATIONALE

The Salmonella typhimurium strains used at LBI are all histidine auxotrophs by virtue of mutations in the histidine operon. When these histidine-dependent cells are grown in a minimal media petri plate containing a trace of histidine, only those cells that revert to histidine independence (his+) are able to form colonies. The trace amount of histidine allows all the plated bacteria to undergo a few divisions; this growth is essential for mutagenesis to occur. The his+ revertants are easily scored as colonies against the slight background growth. The spontaneous mutation frequency of each strain is relatively constant, but when a mutagen is added to the agar the mutation frequency is increased 2- to 100-fold. Cells which grow to form colonies on the minimal media petri plates are therefore assumed to have reverted, either spontaneously or by the action of a test substance to his+ genotype.

3. MATERIALS

A. Indicator Microorganisms

The Salmonella typhimurium strains used in this assay were obtained from Dr. Bruce Ames, University of California at Berkeley.[1-5] The following 4 strains were used:

Strain Designation	Gene Affected	Additional Mutations			Mutation Type Detected
		Repair	LPS	R Factor	
TA-1535	his G	Δ uvr B	rfa	-	Base-pair substitution
TA-1537	his C	Δ uvr B	rfa	-	Frameshift
TA-98	his D	Δ uvr B	rfa	pKM101	Frameshift
TA-100	his G	Δ uvr B	rfa	pKM101	Base pair substitution

3. MATERIALS (Continued)

The aforementioned strains have, in addition to the mutation in the histidine operon, a mutation (rfa-) that leads to defective lipopolysaccharide coat, a deletion that covers genes involved in the synthesis of vitamin biotin (bio-) and in the repair of ultraviolet (uv) - induced DNA damage (uvrB-). The rfa- mutation makes the strains more permeable to many large molecules. The uvrB- mutation decreases repair of some types of chemically or physically damaged DNA and thereby enhances the strain's sensitivity to some mutagenic agents. The resistant transfer factor plasmid (R factor) pKM101 in TA-98 and TA-100 is believed to cause an increase in error-prone DNA repair that leads to many more mutations for a given dose of most mutagens[5]. In addition, plasmid pKM101 confers resistance to the antibiotic ampicillin, which is a convenient marker to detect the presence of plasmid in the cells.

All indicator strains are kept at 4°C on minimal medium plates supplemented with a trace of biotin and an excess of histidine. The plates with plasmid-carrying strains contain in addition ampicillin (25 µg/ml) to ensure stable maintenance of plasmid pKM101. New stock culture plates are made as often as necessary from frozen master cultures or from single colony reisolates that were checked for their genotypic characteristics (his, rfa, uvrB, bio) and for the presence of plasmid. For each experiment, an inoculum from the stock culture plates is grown overnight at 37°C in nutrient broth (Oxoid CM67).

B. Media

The bacterial strains were cultured in Oxoid Media #2 (nutrient Broth). The selective medium was Vogel Bonner Medium E with 2% glucose[7]. The overlay agar consisted of 0.6% purified agar with 0.5 mM histidine, 0.05 mM biotin and 0.1 M NaCl according to the methods of Ames et. al.[6]

C. Activation System

(1) S9 Homogenate

A 9,000 x g supernatant prepared from Sprague-Dawley adult male rat liver induced by Aroclor 1254 (described by Ames et. al.[6]) was purchased from Bionetics Laboratory Products, Litton Bionetics, Inc. and used in this assay.

(2) S9 Mix

Components	Concentration per Milliliter S9 Mix
NADP (sodium salt)	4 µmoles
D-glucose-6-phosphate	5 µmoles
$MgCl_2$	8 µmoles
KCl	33 µmoles
Sodium phosphate buffer pH 7.4	100 µmoles
Organ homogenate from rat liver (S9 fraction)	100 µliters

4. EXPERIMENTAL DESIGN

A. Dosage Selection

All tests are run at a minimum of four concentrations. In the Standard EPA Level I Ames assays, five dose levels of the test material, dissolved in a suitable solvent, are added to the test system. The standard test doses for the extracted material are 10, 25, 50, 100 and 200 µliters per plate. The solids are tested up to 5 mg per plate and at lower concentrations of 2.5, 1, 0.5, 0.1 and 0.05 mg per plate. The samples are retested over a narrower range of concentrations with strains showing positive results if there is enough sample.

B. Mutagenicity Testing

The procedure used is based on the paper published by Ames et. al.[6] and is performed as follows:

(1) Nonactivation Assay

To a sterile 13 x 100 mm test tube placed in a 43°C water bath the following is added in order:

(a) 2.00 ml of 0.6% agar containing 0.05 mM histidine and 0.05 mM biotin.

(b) 0.05 ml of a solution of the test chemical to give the appropriate dose.

(c) 0.1 ml - 0.2 ml of indicator organism(s).

(d) 0.50 ml of 0.2M phosphate buffer, pH 7.4.

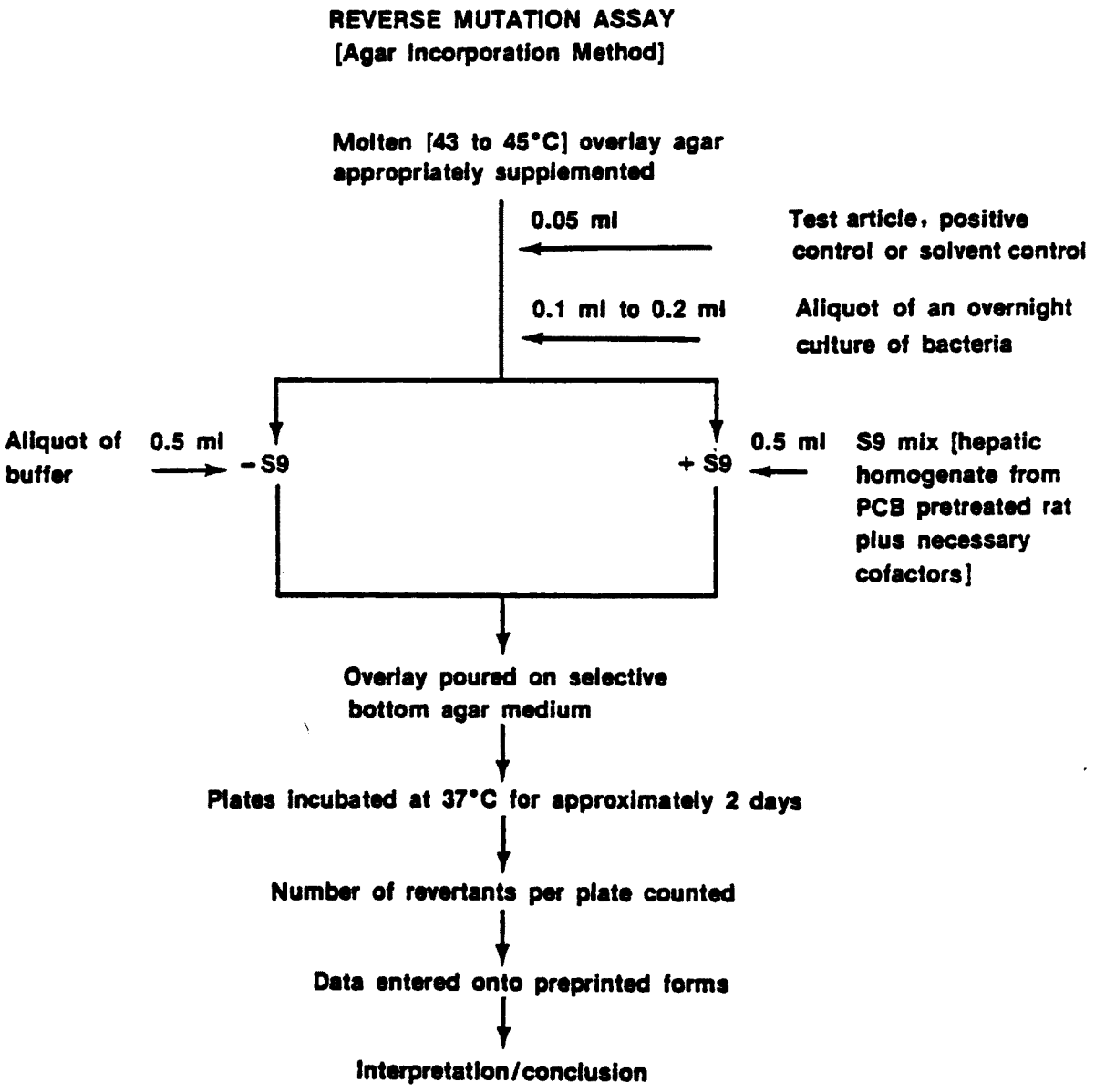
This mixture is swirled gently and then poured onto minimal agar plates (see 3B, Media). After the top agar has set, the plates are incubated at 37°C for approximately 2 days. The number of his+ revertant colonies growing on the plates is counted and recorded.

(2) Activation Assay

The activation assay is run concurrently with the nonactivation assay. The only difference is the addition of 0.5 ml of S9 mix (see 3C:2, Activation System) to the tubes in place of 0.5 ml of phosphate buffer which is added in nonactivation assays. All other details are similar to the procedure for nonactivation assays.

A detailed flow diagram for the plate incorporation assay is provided in Figure 1.

FIGURE 1


REVERSE MUTATION ASSAY
[Agar Incorporation Method]
Molten [43 to 45°C] overlay agar appropriately supplemented
0.05 ml
Test article, positive control or solvent control
0.1 ml to 0.2 ml
Aliquot of an overnight culture of bacteria
Aliquot of buffer
0.5 ml
- S9
+ S9
0.5 ml
S9 mix [hepatic homogenate from PCB pretreated rat plus necessary cofactors]
Overlay poured on selective bottom agar medium
Plates incubated at 37°C for approximately 2 days
Number of revertants per plate counted
Data entered onto preprinted forms
Interpretation/conclusion

4. EXPERIMENTAL DESIGN (Continued)

C. Control Compounds

A negative control consisting of the solvent used for the test material is performed in all cases. For negative controls, step 'b' of Nonactivation Assays is replaced by 0.05 ml of the solvent. The negative controls are employed for each indicator strain and are performed in the absence and presence of S9 mix. The solvent used to prepare the stock solution of the test material is given in the Results section of this report. All dilutions of the test material are made using this solvent. The amount of solvent used is equal to the maximum volume used to give the appropriate test dose.

Specific positive control compounds known to revert each strain are also used in the assays. The concentrations and specificities of these compounds to specific strains are given in the following table:

Assay	Chemical	Solvent	Concentration per Plate (µg)	Salmonella Strains
Nonactivation	Sodium azide	Water	1	TA-1535, TA-100
	2-Nitrofluorene (NF)	Dimethyl-sulfoxide	10	TA-98
	9-aminoacridine (9AA)	Ethanol	50	TA-1537
Activation	2-anthramine (ANTH)	Dimethyl-sulfoxide	2.5	For all strains

5. EVALUATION CRITERIA

Statistical methods are not currently used and evaluation is based on the criteria included in this protocol.

Plate test data consist of direct revertant colony counts obtained from a set of selective agar plates seeded with populations of mutant cells suspended in a semisolid overlay. Because the test material and the cells are incubated in the overlay for approximately 2 days and a few cell divisions occur during the incubation period, the test is semiquantitative in nature. Although these features reduce the quantitation of result, they provide certain advantages not contained in a quantitative suspension test:

- The small number of cell divisions permits potential mutagens to act on replicating DNA, which is often more sensitive than nonreplicating DNA.
- The combined incubation of the test article and the cells in the overlay permits constant exposure of the indicator cells for approximately 2 days.

A. Surviving Populations

Plate test procedures do not permit exact quantitation of the number of cells surviving chemical treatment. At low concentrations of the test material, the surviving population on the treatment plates is essentially the same as that on the negative control plate. At high concentrations, the surviving population is usually reduced by some fraction. Our protocol will normally employ several doses ranging over two or three log concentrations. This does not apply to spot tests and tests performed on fabrics and like materials which are tested at a single concentration.

B. Dose-Response Phenomena

The demonstration of dose-related increases in mutant counts is an important criterion in establishing mutagenicity. A factor that might modify dose-response results for a mutagen would be the selection of doses that are too low (usually mutagenicity and toxicity are related). If the highest dose is far lower than a toxic concentration, no increases may be observed over the dose range selected. Conversely, if the lowest dose employed is highly cytotoxic, the test material may kill any mutants that are induced, and the test material will not appear to be mutagenic.

5. EVALUATION CRITERIA (Continued)

C. Control Tests

Positive and negative control assays are conducted with each experiment and consist of direct-acting mutagens for nonactivation assays and mutagens requiring metabolic biotransformation in activation assays. Negative controls consist of the test material solvent in the overlay agar together with the other essential components. The negative control plate for each strain gives a reference point to which the test data is compared. The positive control assay is conducted to demonstrate that the test systems are functional with known mutagens.

D. Evaluation Criteria for Ames Assay

Because the procedures used to evaluate the mutagenicity of the test material are semiquantitative, the criteria used to determine positive effects are inherently subjective and are based primarily on a historical data base. Most data sets are evaluated using the following criteria:

(1) Strains TA-1535, TA-1537

If the solvent control value is within the normal range, a test material producing a positive response equal to three times the solvent control value is considered mutagenic.

(2) Strains TA-98 and TA-100

If the solvent control value is within the normal range, a test material producing a positive response equal to twice the solvent control value for TA-98 and TA-100 is considered mutagenic.

(3) Pattern

Because TA-1535 and TA-100 are both derived from the same parental strain (G-46) and because TA-1538 and TA-98 are both derived from the same parental strain (D3052), to some extent there is a built-in redundancy in the microbial assay. In general, the two strains of a set respond to the same mutagen and such a pattern is sought. Generally, if a strain responds to a mutagen in nonactivation tests, it will do so in activation tests.

(4) Reproducibility

If a test material produces a response in a single test which cannot be reproduced in additional runs, the initial positive test data lose significance.

5. EVALUATION CRITERIA (Continued)

E. Evaluation Criteria for Toxicity

(1) Complete toxicity

When there are no revertants observed on the plate(s) treated with the test compound, the test compound is defined as toxic to all or any of the indicator strains at that (those) particular dose(s).

(2) Slight toxicity

When there are fifty or less percent revertants on the plate(s) treated with the test compound as compared to the solvent control plate(s), the test compound is defined as slightly toxic to all or any of the indicator strains at that (those) particular dose(s).

F. Relation Between Mutagenicity and Carcinogenicity

It must be emphasized that the Ames Salmonella/Microsome Plate Assay is not a definitive test for chemical carcinogens. It is recognized, howerver, that correlative and functional relations have been demonstrated between these two endpoints. The results of comparative tests on 300 chemicals by McCann et. al.[1] show an extremely good correlation between results of microbial mutagenesis tests and in vivo rodent carcinogenesis assays.

All evaluations and interpretation of the data to be presented in the final report will be based only on the demonstration, or lack, of mutagenic activity.

REFERENCES

1. McCann,J., Choi, E., Yamasaki, E. and Ames, B.N.: Detection of carcinogens as mutagens in the Salmonella/microsome test: Assay of 300 chemicals. Proc. Nat. Acad. Sci. USA, 72:5135-5139, 1975.

2. Ames, B.N., Gurney, E.G., Miller, J.A. and Bartsch, H.: Carcinogens as frameshift mutagens: Metabolites and derivatives of 2-acetylaminofluorene and other aromatic amine carcinogens. Proc. Nat. Acad. Sci. USA, 69:3128-3132, 1972.

3. Ames, B.N, Lee, F.D. and Durston, W.E.: An improved bacterial test system for the detection and classification of mutagens and carcinogens. Proc. Nat. Acad. Sci. USA, 70:782-786, 1973.

4. Ames, B.N, Durston, W.E., Yamasaki, E. and Lee, F.D.: Carcinogens are mutagens: A simple test system combining liver homogenates for activation and bacteria for detection. Proc. Nat. Acad. Sci. USA, 70:2281-2285, 1973.

5. McCann, J., Springarn, N.E., Kobori, J. and Ames, B.N.: Detection of carcinogens as mutagens: Bacterial tester strains with R factor plasmids. Proc. Nat. Acad. Sci. USA, 72:979-983, 1975.

6. Ames, B.N., McCann, J. and Yamasaki, E.: Methods for detecting carcinogens and mutagens with the Salmonella/mammalian-microsome mutagenicity test. Mutation Res., 31:347-364, 1975.

7. Vogel, H.J. and Bonner, D.M.: Acetylornithinase of E. coli; Partial purification and some properties. J. Biol. Chem., 218:97-106, 1956.

Q.A. Inspection Statement
(reference 21 CFR 58.35(b)(7))

PROJECT 20988 LBI Assay No. 5397

TYPE of STUDY AMES PLATE TEST

This final study report was reviewed by the LBI Quality Assurance Unit on 12/3/80. A report of findings was submitted to the Study Director and to Management on 12/3/80.

The short-term nature of this study precluded inspection while it was in process. The Quality Assurance Unit inspects an in-process study of this type approximately once per month to assure that no significant problems exist that are likely to affect the integrity of this type of study.

Mitchell S. Ehrlich
Auditor, Quality Assurance Unit

GENETICS ASSAY NO. 5394

LBI SAFETY NO. 6378

CYTOTOXIC EVALUATION OF

SAMPLE # 1452
(WATER TANK DRAIN, END OF TEST)

IN THE
RODENT CELL (CHO)
CLONAL TOXICITY ASSAY

FINAL REPORT

SUBMITTED TO:

ACUREX CORPORATION
485 CLYDE AVENUE
MOUNTAIN VIEW, CA 94042

SUBMITTED BY:

LITTON BIONETICS, INC.
5516 NICHOLSON LANE
KENSINGTON, MARYLAND 20795

LBI PROJECT NO. 20933

REPORT DATE: DECEMBER 1980

PREFACE

This report contains a summary of the data compiled during the evaluation of the test compound. The report is organized to present the results in a concise and easily interpretable manner. The first part contains items I - IX. Items I - IV provide sponsor and compound identification information, type of assay, and the assay design reference number. All assay design references indicate a standard procedure described in the Litton Bionetics, Inc. "Screening Program for the Identification of Potential Mutagens and Carcinogens." Item V provides the initiation and completion dates for the study, and Item VI provides identification of supervisory personnel. Item VII identifies the tables and figures containing the data used by the study director in interpreting the test results. The interpretation itself is in Item VIII. Item IX provides the conclusion and evaluation.

The second part of the report, entitled Assay Design, describes the materials and procedures employed in conducting the assay. This part of the report also contains any appendices, as well as evaluation criteria used by the study director. The evaluation criteria are included to acquaint the sponsor with the methods used to develop and analyze the test results.

All test and control results presented in this report are supported by fully documented raw data which are permanently maintained in the files of the Department of Genetics and Cell Biology or in the archives of Litton Bionetics, Inc., 5516 Nicholson Lane, Kensington, Maryland, 20795.

Copies of raw data will be supplied to the sponsor upon request.

I. SPONSOR: Acurex Corporation

II. MATERIAL (TEST COMPOUND): GENETICS ASSAY NUMBER: 5394

A. Identification: Sample #1452 (Man Test 5/21/80, #5 Gal, Water Tank Drain, End of Test)

B. Date Received: October 6, 1980

C. Physical Description: Clear, colorless liquid

III. TYPE OF ASSAY: Rodent Cell (CHO) Clonal Toxicity Assay

IV. ASSAY DESIGN NUMBER: 442

V. STUDY DATES:

A. Initiation: November 19, 1980

B. Completion: December 1, 1980

VI. SUPERVISORY PERSONNEL:

A. Study Director: Brian C. Myhr, Ph.D.

B. Laboratory Supervisor: Robert Young, M.S.

VII. RESULTS:

The data are presented in Table 1 on page 3 and in Figure 1 on page 4.

VIII. INTERPRETATION OF RESULTS:

The test material, Sample No. 1452, was supplied as a colorless, aqueous solution that did not contain any obvious suspended material. A portion of the sample (18 ml) was combined with 10X F10 medium and supplements to yield 30 ml of culture medium containing the test material at a concentration of 600 µl/ml. This was the highest concentration applied to the CHO cells. Lower concentrations were achieved by serial dilution with F10 culture medium. The treatments were initiated by replacing the F12 medium in the cell cultures with F10 medium containing the test material. After the 24 hour exposure period, the cultures were returned to F12 medium and incubated for colony development. The test material did not cause any visible precipitation of culture medium components, but a shift to a slightly acidic pH of 6.26 was observed for the 600 µl/ml concentration. The pH remained normal (above 7.0) for the other test concentrations.

VIII. INTERPRETATION OF RESULTS: (continued)

Two trials of the assay were performed. The first trial was considered insufficient for evaluation because the average cloning efficiency of the negative controls was 67.4%, which was slightly below the 70% criterion used for an acceptable assay. The results of the second trial are presented in Table 1 and Figure 1.

Seven test concentrations from 6 µl/ml to 600 µl/ml were evaluated for their effects on colony survival. As shown in Table 1, complete lethality was obtained for treatments in the 60 to 600 µl/ml concentration range. In contrast, the treatments with 6 to 30 µl/ml were essentially nontoxic to the cells. Thus, an extremely sharp survival curve was obtained, as shown in Figure 1, with an EC50 apparently located at 45 µl/ml. [In the first trial (results not shown), an EC50 value of 52 µl/ml was obtained for a somewhat broader survival curve]. The EC50 was therefore located at the upper end of the 6-60 µl/ml concentration range, which is the range defined for moderate toxicity for aqueous liquids (see Evaluation Criteria). Since complete lethality was obtained at the boundary between moderate and low toxicity, an evaluation of moderate toxicity to CHO cells appeared appropriate.

IX. CONCLUSIONS:

The test material, Sample No. 1452, was evaluated as having moderate toxicity in the CHO Clonal Toxicity Assay. The EC50 value was 45 µl/ml.

Submitted by:

Study Director:

Brian C. Myhr 12/3/80

Brian C. Myhr, Ph.D. Date
Section Chief
Mammalian Genetics
Department of Genetics
and Cell Biology

Reviewed by:

D.J. Jaymuth 12/3/80

For David J. Brusick, Ph.D. Date
Director
Department of Genetics
and Cell Biology

TABLE 1

RODENT CELL (CHO) CLONAL TOXICITY ASSAY

Sample Identity: Sample #1452, (Water Tank Drain, End of Test)

Description of Sample: Clear, colorless aqueous liquid

LBI Assay No.: 5394

Date Received: October 6, 1980

Test Date: November 25, 1980 (Trial 2)

Vehicle: None

Cell Type: CHO-K1

Cells Seeded per Dish: 200

EC50 Value: 45 µl/ml

Toxicity Classification: Moderate

pH Alterations: pH 6.26 at 600 µl/ml

Comments on Treatment: Normal pH for treatments with 6 to 300 µl/ml

CLONAL TOXICITY DATA

Sample	Applied Concentration µl/ml	Dish #1	Dish #2	Dish #3	Average Count	Relative Survival %	Cloning Efficiency %
NC	---	187	193	187	189.0	100.0	94.5
Test	6.0	190	219	183	197.3	104.4	
Test	10.0	214	210	181	201.7	106.7	
Test	30.0	167	190	178	178.3	94.3	
Test	60.0	0	0	0	0	0	
Test	100.0	0	0	0	0	0	
Test	300.0	0	0	0	0	0	
Test	600.0	0	0	0	0	0	

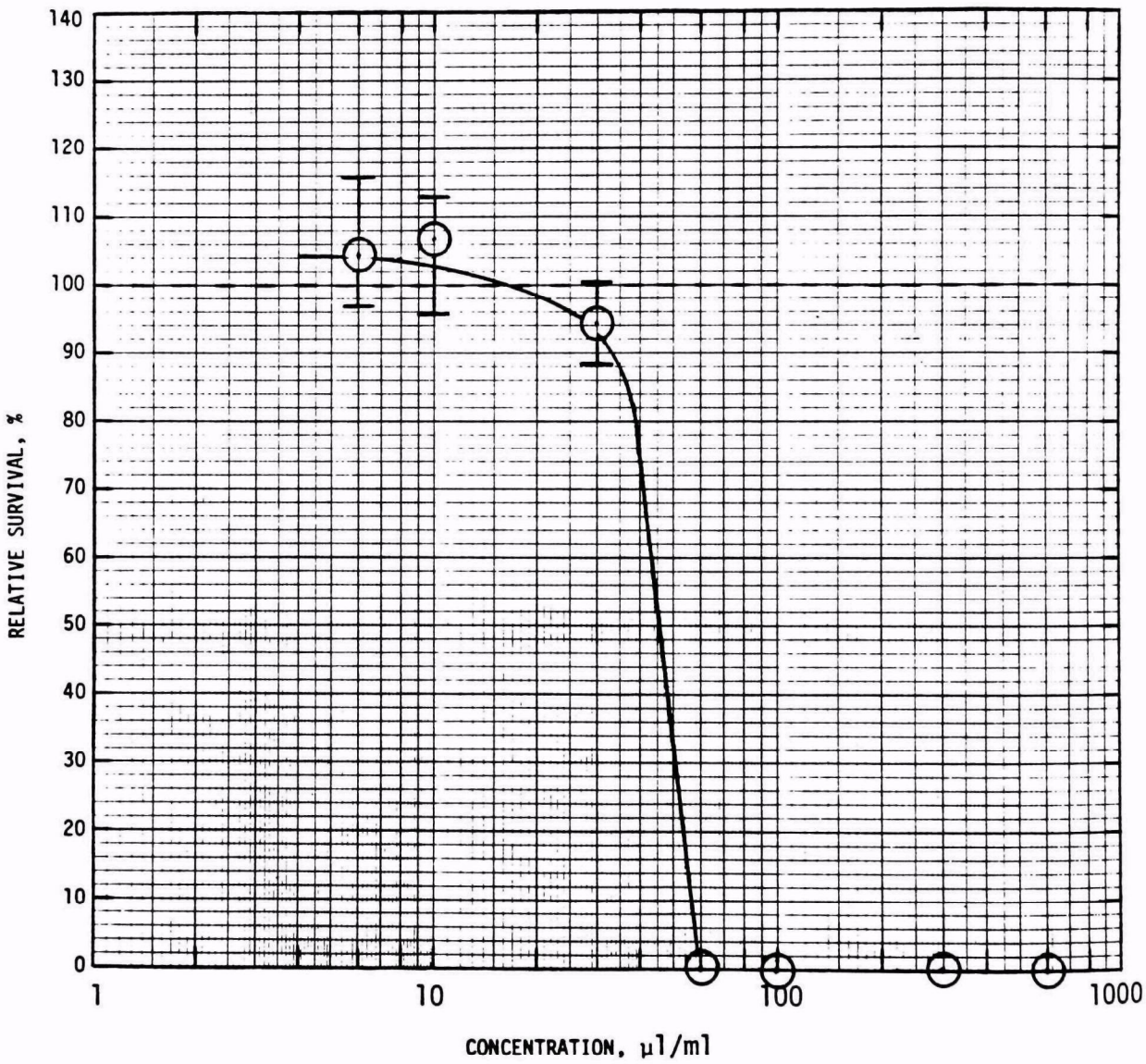
NC = Negative Control, F12 medium.

FIGURE 1

RODENT CELL (CHO) CLONAL TOXICITY ASSAY

EC_{50} DETERMINATION

SAMPLE #1452 (WATER TANK DRAIN, END OF TEST)



ASSAY DESIGN NO. 442

RODENT CELL CLONAL (CHO) TOXICITY ASSAY

This assay conforms to one of the bioassay tests in the US Environmental Protection Agency (EPA) IERL - RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests (September, 1980). Level 1 bioassays obtain preliminary information on the harmful biological effects of chemicals found in industrial feed and waste streams by using mammalian cells in culture to measure metabolic impairment and cellular death.

1. OBJECTIVE

The objective of this assay is to determine the concentration of a test article that will reduce the colony forming ability of Chinese hamster cells by 50% after a 24-hour exposure. This concentration is referred to as the EC_{50} value.

2. MATERIALS

A. Indicator Cells

The indicator cells used in this study are Chinese hamster CHO-K1 cells (American Type Culture Collection No. CCL 61). This cell type was derived from ovarian tissue and has spontaneously transformed to a stable, hypodiploid line of rounded, fibroblastic cells with unlimited growth potential. Monolayer cultures have a fast doubling time of 11 to 14 hours, and untreated cells can normally be cloned with an efficiency of 80% or greater. Permanent stocks are maintained in liquid nitrogen and laboratory cultures are maintained by serial subculturing. Laboratory cultures are periodically checked by culturing methods for the absence of mycoplasma contamination. This test system is specified by the IERL- RTP Procedures Manual.

B. Medium

The CHO-K1 cell line has an absolute requirement for proline and therefore must be maintained in a culture medium containing sufficient amounts of this amino acid. Ham's F12 medium, which contains 3×10^{-4}M L-proline, is used, supplemented with 10% fetal bovine serum, 100 units per milliliter of penicillin, 100 µg streptomycin per milliliter, and 0.5 µg amphotericin B (Fungizone) per milliliter.

C. Controls

Untreated cells are cloned to establish the control cloning efficiency. If the test article is dissolved in an organic solvent (usually dimethylsulfoxide), cells exposed to solvent in the medium

2. MATERIALS (Continued)

are cloned to provide the reference cloning efficiency for the effect of the test article. The final concentration of solvent in the growth medium is generally 1% or less. All controls are performed in triplicate.

D. Sample Forms

Solid samples are tested as a solution or are ground to fine particles (less than 5 μm) and tested as a suspension in growth medium. Dry particulate articles, aqueous liquids, suspensions and slurries are added directly to the growth medium and tested as a suspension or solution. Liquids containing less than 0.2% organic solvent are generally tested directly; samples dissolved in organic solvents are solvent exchanged in dimethylsulfoxide (DMSO) before testing. Original sample volumes may be reduced a maximum of 10-fold during solvent exchange, and the concentration factor is used to convert assayed volumes into equivalent original sample volumes. All sample manipulations are performed as described in the IERL-RTP Level 1 Procedures Manual.

3. EXPERIMENTAL DESIGN

A. Dose Selection

Unless the approximate toxicity is already known or the sample size is limiting, the following dose ranges are tested for different sample forms. Dry particulate articles are dissolved or suspended in growth medium and tested at five dose levels from 1000 μg/ml to 10 μg/ml. Aqueous samples, suspensions or slurries are tested from 600 μliters/ml to 6 μliters/ml in five dose steps. Samples that are solvent exchanged into DMSO are tested from 20 μliters/ml to 0.2 μliters/ml, also in five dose steps. Solvent concentrations are 1% for all dose levels except the maximum applicable dose (MAD) of 20 μl/ml, which contains 2% solvent. A second dose study is performed with an appropriate dose range if the EC_{50} has not been properly located in the initial test; EC_{50} values greater than 1,000 μg/ml, 600 μliters of aqueous sample/ml or 20 μliters nonaqueous sample/ml are not determined.

B. Clonal Toxicity Assay

Cells from monolayer stock cultures in logarithmic growth phase are trypsinized, counted by hemacytometer and reseeded into a series of 60- or 100-mm culture dishes at 200 cells per dish. The cultures are incubated for 6 to 16 hours at 37°C to allow attachment of the cells and recovery of growth rate.

Test article is then applied (three dishes per dose), and the cultures returned to the incubator. If the test article causes a color change, an additional dish is treated with the high dose and the pH of the

3. EXPERIMENTAL DESIGN (Continued)

medium is determined. The pH is also recorded for the highest dose which results in a slight color change. After a 24-hour exposure period the medium is aspirated and the cells washed with Dulbecco's phosphate buffered saline (PBS; prewarmed to 37°C). The pH of the discarded medium for which initial pH measurements were made is again recorded. Fresh medium is placed on each culture and incubation continued for an additional 6 days to allow colony development. Medium is drained from the cultures and the surviving colonies are washed with PBS, fixed in methanol, and stained with Giemsa. Colony counting is performed by eye; colonies smaller than 50 to 100 cells are not counted.

4. ASSAY ACCEPTANCE CRITERIA

The assay is considered acceptable for evaluation of the test results if the following criteria are met:

- The average cloning efficiency of the CHO-K1 cells in the negative controls is 70% or greater, but not exceeding 115%.
- The distribution of colonies in the treated cultures is generally uniform over the surface of the culture dish.
- The data points for each test concentration critical to the location of the EC_{50} are the averages of at least two treated cultures.
- A sufficient number of test concentrations are available to clearly locate the EC_{50} within a toxicity region as defined under Assay Evaluation Criteria.
- If the EC_{50} value is greater than 1000 µg/ml, 600 µliters of aqueous sample/ml, or 20 µliters of nonaqueous sample/ml, the plotted curve does not exceed 110% of the negative control.

5. ASSAY EVALUATION CRITERIA

The screened doses, pH values (if appropriate), colony counts, percent survivals (colony counts relative to control colony counts) and EC_{50} values are provided. The percent survival is plotted as a function of applied concentration and the EC_{50} value determined graphically by fitting a curve by eye through the data points. The EC_{50} is used to rank the test material using the standard evaluation criteria defined in the table below.

Sorbent extracts of known organic content are evaluated as nonaqueous liquids and in terms of their solid contents (µg organics/ml), and are ranked using the more sensitive parameter. Where data on the original liquid or gas sample volumes is provided by the Sponsor, the EC_{50} value is calculated in terms of these equivalent volumes per milliliter of culture medium; however, no evaluations are made as yet on this basis.

5. ASSAY EVALUATION CRITERIA (Continued)

Toxicity[a]	Solids (EC_{50} in µg/ml)	Aqueous Liquids (EC_{50} in µl/ml)	Nonaqueous Liquids[b] (EC_{50} in µl/ml)
High	<10	<6	<.2
Moderate	10 to 100	6 to 60	.2-2
Low	100 to 1000	60 to 600	2-20
Not Detectable	>1000	>600	>20

[a]Evaluation criteria formulated by Litton Bionetics, Inc. for IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests.

[b]Criteria for nonaqueous liquids are tentative and under evaluation.

6. RECORDS TO BE MAINTAINED

All raw data, protocols, protocol modifications, test article weight and dispensation records and correspondence between LBI and the Sponsor are being maintained in a central file within the Department of Genetics and Cell Biology. These records will be filed under departmental assay number and held up to 2 years following submission of the final report to the Sponsor. After 2 years they will be transferred to the LBI archives for permanent storage.

Q.A. Inspection Statement
(reference 21 CFR 58.35(b)(7))

PROJECT 20993 LBI Assay No. 5394

TYPE of STUDY Rodent Cell (CHO) Clonal Toxicity Assay

This final study report was reviewed by the LBI Quality Assurance Unit on December 3, 1982 A report of findings was submitted to the Study Director and to Management on December 3, 1982

The short-term nature of this study precluded inspection while it was in process. The Quality Assurance Unit inspects an in-process study of this type approximately once per month to assure that no significant problems exist that are likely to affect the integrity of this type of study.

Patrice M. Ciccaro
Auditor, Quality Assurance Unit

GENETICS ASSAY NO. 5394

LBI SAFETY NO. 6378

MUTAGENICITY EVALUATION OF SAMPLE #1452

IN THE AMES SALMONELLA/MICROSOME PLATE TEST

FINAL REPORT

SUBMITTED TO:

ACUREX CORPORATION
485 CLYDE AVENUE
MOUNTAIN VIEW, CA 94042

SUBMITTED BY:

LITTON BIONETICS, INC.
5516 NICHOLSON LANE
KENSINGTON, MARYLAND 20795

LBI PROJECT NO. 20988

REPORT DATE: DECEMBER, 1980

PREFACE

This report contains a summary of the data compiled during the evaluation of the test compound. The report is organized to present the results in a concise and easily interpretable manner. The first part contains items I-IX. Items I-IV provide sponsor and compound identification information, type of assay, and the protocol reference number. All protocol references indicate a standard procedure described in the Litton Bionetics, Inc. "Screening Program for the Identification of Potential Mutagens and Carcinogens." Item V provides the initiation and completion dates for the study, and Item VI provides identification of supervisory personnel. Item VII identifies the tables and/or figures containing the data used by the study director in interpreting the test results. The interpretation itself is in Item VIII. Item IX provides the conclusion and evaluation.

The second part of the report describes the materials and procedures employed in conducting the assay. This part of the report also contains evaluation criteria used by the study director, and any appendices. The evaluation criteria are included to acquaint the sponsor with the methods used to develop and analyze the test results.

All test and control results presented in this report are supported by fully documented raw data which are permanently maintained in the files of the Department of Genetics and Cell Biology or in the archives of Litton Bionetics, Inc., 5516 Nicholson Lane, Kensington, Maryland 20795.

I. SPONSOR : Acurex Corporation

II. MATERIAL (TEST COMPOUND): GENETIC ASSAY NUMBER: 5394

A. Identification: Sample Number 1452 and its Extract

B. Date Received : October 6, 1980

C. Physical Description: Neat Sample: Colorless Liquid
Extract : Green Liquid

III. TYPE OF ASSAY : Ames Salmonella/microsome Mutagenesis Assay

IV. PROTOCOL NUMBER: 401 (EPA-LEVEL I)

V. STUDY DATES:

A. Initiation: November 21, 1980

B. Completion: November 25, 1980

VI. SUPERVISORY PERSONNEL:

A. Study Director: D.R. Jagannath, Ph.D.

VII. RESULTS:

The results of this are presented in Tables 1 and 2.

VIII. INTERPRETATION OF RESULTS:

The test material, Sample No. 1452 was received as a clear colorless liquid (1 gallon) with no visible suspended particulates. An aliquot of the neat sample (3250 ml) was concentrated 500 fold by passing through the XAD-2 resin column at a flow rate of 3 ml per minute. The resin was then extracted with methylene chloride in a Soxhlet apparatus for 24 h. The extract (250 ml) was concentrated to approximately 5 ml using a Kuderna Concentrator. The concentrated extract was then solvent exchanged by sequential addition of Dimethylsulfoxide (DMSO) and evaporation of residual methylene chloride in a warm water bath under a stream of nitrogen. Final sample size in DMSO was 6.5 ml which represents a 500 fold concentration factor. The concentrate was a clear solution with a pale blue/green color.

The test materials, neat sample #1452 and its extract were examined for mutagenic activity in the Ames Salmonella assay in the presence and absence of liver microsomal enzymes from Aroclor induced rats.

Solvent controls, distilled water for the neat sample and DMSO for the extract of the sample, and specific positive controls were also assayed concurrently with the test materials.

DOSE RANGE: The dose range employed for the neat sample was from 10.0 ul to 200.0 ul per plate and for the extract it was from 5.0 ul to 100.0 ul per plate (due to the limited amount of the extract). The organic content of the sample was not supplied by the sponsor.

TOXICITY: The test materials, neat sample and its extract were not toxic to any of the strains used in this assays.

RESULTS:

The results of the tests conducted on the test material, neat sample, in the absence of an activation system was negative. However, the extract of the sample exhibited genetic activity with the strain TA-98 in the absence of an activation system. The response was slightly above two times the background level and was observed at the highest concentration of 100 ul per plate.

The results of the tests conducted on the test materials neat sample and its extract were all negative in the presence of rat liver activation system.

Dose-related increases in the number of revertants were also observed with the base-pair substitution strain TA-100 in the non-activation and activation assays performed on the extract. However these increases did not meet the criteria for a positive response.

IX. CONCLUSION:

The test material, neat sample #1452 and its extract were tested in the Ames/Salmonella assay and were compared for their mutagenic activities. The neat sample did not exhibit genetic activity in any of the assays conducted in this evaluation and was considered not mutagenic under these conditions. However, the extract of the sample #1452 exhibited genetic activity with the strain TA-98 in the nonactivation assays conducted in this evaluation and is considered as mutagenic. Based on the EPA's "Definition of Toxicity Catagories for Health Effects Tests" the mutagenic activity of the neat sample #1452 is classified as 'ND' and the mutagenic activity of the extract of the sample #1452 is classified as 'low'. Since organic content of the sample was not available the specific activity has not been calculated.

Submitted by:

Study Director

Reviewed by:

for, Bruce C. Myhr 12/3/80
David J. Brusick, Ph.D. Date
Director
Department of Genetics
and Cell Biology

D.R. Jagannath 12/3/80
D.R. Jagannath, Ph.D. Date
Section Chief
Submammalian Genetics
Department of Genetics
and Cell Biology

RESULTS — TABLE 1

A. NAME OR CODE DESIGNATION OF THE TEST COMPOUND: SAMPLE #1452 (EXTRACT)
B. SOLVENT: DMSO
C. TEST INITIATION DATES: 11/21/80
D. TEST COMPLETION DATE: 11/25/80
E. S-9 LOT#: DR009
NOTE: CONCENTRATIONS ARE GIVEN IN MICROLITERS PER PLATE

TEST	SPECIES	TISSUE	REVERTANTS PER PLATE: TA-1535			TA-1537			TA-98			TA-100		
			1	2	3	1	2	3	1	2	3	1	2	3
NONACTIVATION														
SOLVENT CONTROL	---	---	28	39		13	11		25	25		126	176	
POSITIVE CONTROL**	---	---	620	662		797	848		1013	990		701	654	
TEST COMPOUND														
10.000000 UL	---	---	31	28		8	11		20	22		142	139	
25.000000 UL	---	---	31	27		9	9		38	28		146	142	
50.000000 UL	---	---	32	31		16	9		22	22		115	112	
100.000000 UL	---	---	29	30		8	8		29	33		128	150	
200.000000 UL	---	---	22	19		9	12		33	28		139	128	
ACTIVATION														
SOLVENT CONTROL	RAT	LIVER	27	31		10	16		46	50		145	135	
POSITIVE CONTROL***	RAT	LIVER	163	164		262	210		1302	1263		1585	1521	
TEST COMPOUND														
10.000000 UL	RAT	LIVER	27	11		10	7		36	28		100	78	
25.000000 UL	RAT	LIVER	27	19		8	9		29	33		77	95	
50.000000 UL	RAT	LIVER	42	15		9	10		26	17		87	87	
100.000000 UL	RAT	LIVER	28	16		8	9		28	26		92	76	
200.000000 UL	RAT	LIVER	21	14		7	6		16	29		106	99	

** | | | |
|---|---|---|
| TA-1535 | SODIUM AZIDE | 10 UG/PLATE |
| TA-1537 | 9-AMINOACRIDINE | 50 UG/PLATE |
| TA-98 | 2-NITROFLUORENE | 10 UG/PLATE |
| TA-100 | SODIUM AZIDE | 10 UG/PLATE |
| SOLVENT | 200 UL/PLATE | |

*** | | | |
|---|---|---|
| TA-1535 | 2-ANTHRAMINE | 2.5 UG/PLATE |
| TA-1537 | 2-ANTHRAMINE | 2.5 UG/PLATE |
| TA-98 | 2-ANTHRAMINE | 2.5 UG/PLATE |
| TA-100 | 2-ANTHRAMINE | 2.5 UG/PLATE |

RESULTS

TABLE 2

A. NAME OR CODE DESIGNATION OF THE TEST COMPOUND: SAMPLE #1452 (EXTRACT)
B. SOLVENT: DMSO
C. TEST INITIATION DATES: 11/21/90
D. TEST COMPLETION DATE: 11/25/90
E. S-9 LOT#: D8009
NOTE: CONCENTRATIONS ARE GIVEN IN MICROLITERS PER PLATE

TEST	SPECIES	TISSUE	REVERTANTS PER PLATE TA-1535			TA-1537			TA-98			TA-100		
			1	2	3	1	2	3	1	2	3	1	2	3
NONACTIVATION														
SOLVENT CONTROL	---	---	32	35		7	7		31	25		144	141	
POSITIVE CONTROL**	---	---	620	662		797	848		1013	890		701	654	
TEST COMPOUND														
5.000000 UL	---	---	11	19		10	4		30	33		164	162	
10.000000 UL	---	---	17	22		11	13		28	31		174	173	
25.000000 UL	---	---	20	32		17	16		38	32		170	164	
50.000000 UL	---	---	23	23		16	14		44	46		213	199	
100.000000 UL	---	---	27	15		19	20		69	56		233	228	
ACTIVATION														
SOLVENT CONTROL	RAT	LIVER	34	27		18	9		39	37		153	143	
POSITIVE CONTROL***	RAT	LIVER	163	164		262	210		1302	1263		1585	1523	
TEST COMPOUND														
5.000000 UL	RAT	LIVER	42	42		7	8		33	37		152	119	
10.000000 UL	RAT	LIVER	53	40		9	13		41	43		121	154	
25.000000 UL	RAT	LIVER	26	34		13	19		39	37		172	142	
50.000000 UL	RAT	LIVER	25	45		18	23		45	49		208	193	
100.000000 UL	RAT	LIVER	32	34		20	29		57	53		239	182	

** TA-1535 SODIUM AZIDE 10 UG/PLATE
TA-1537 9-AMINOACRIDINE 50 UG/PLATE
TA-98 2-NITROFLUORENE 10 UG/PLATE
TA-100 SODIUM AZIDE 10 UG/PLATE
SOLVENT 100 UL/PLATE

*** TA-1535 2-ANTHRAMINE 2.5 UG/PLATE
TA-1537 2-ANTHRAMINE 2.5 UG/PLATE
TA-98 2-ANTHRAMINE 2.5 UG/PLATE
TA-100 2-ANTHRAMINE 2.5 UG/PLATE

TABLE A

DEFINITION OF TOXICITY CATEGORIES FOR HEALTH EFFECTS ASSAYS*

Assay[a]	Activity Measured[b]	Sample Type[c]	MAD[d]	Units	Range of Concentration or Dosage			
					High	Moderate	Low	Not Detectable (ND)
Ames	MEC (mutagenesis)	S	5	mg/plate	<0.05	0.05-0.5	0.5-5	ND at >5
		AL,NAL	200	µl/plate	<2	2-20	20-200	ND at >200
RAM	EC_{50} (lethality)	S	1	mg/ml	<0.01	0.01-0.1	0.1-1	ND at >1
		AL	600	µl/ml	<6	6-60	60-600	ND at >600
		NAL	20	µl/ml	<0.2	0.2-2	2-20	ND at >20
CHO	EC_{50} (lethality)	S	1	mg/ml	<0.01	0.01-0.1	0.1-1	ND at >1
		AL	600	µl/ml	<6	6-60	60-600	ND at <600
		NAL	20	µl/ml	<0.2	0.2-2	2-20	ND at >20
WAT	LD_{50} (lethality and toxic signs)	S	5	gm/kg	<0.05	0.05-0.5	0.5-5	ND at <5
		AL,NAL	5	ml/kg	<0.05	0.5-0.5	0.5-5	ND at <5

[a]Standard test abbreviations are as follows:
Ames: Ames Salmonella/microsome mutagenesis assay
RAM: Rabbit alveolar macrophage cytotoxicity assay
CHO: Rodent cell clonal toxicity assay
WAT: Acute in vivo test in rodents (whole animal test)

[b]Standard abbreviations for measured endpoints are as follows:
MEC: Minimum effective concentration
EC_{50}: Calculated concentration expected to produce effect in 50 percent of population
LD_{50}: Calculated dose expected to kill 50 percent of population

[c]S = Solid, AL = Aqueous liquid, NAL = Nonaqueous liquid

[d]MAD = Maximum applicable dose

*Evaluation criteria formulated by Litton Bionetics, Inc. for IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests.

AMES SALMONELLA/MICROSOME PLATE ASSAY

1. OBJECTIVE

The objective of this study was to evaluate a test material for mutagenic activity in a bacterial assay with and without a mammalian S9 activation system.

2. RATIONALE

The Salmonella typhimurium strains used at LBI are all histidine auxotrophs by virtue of mutations in the histidine operon. When these histidine-dependent cells are grown in a minimal media petri plate containing a trace of histidine, only those cells that revert to histidine independence (his+) are able to form colonies. The trace amount of histidine allows all the plated bacteria to undergo a few divisions; this growth is essential for mutagenesis to occur. The his+ revertants are easily scored as colonies against the slight background growth. The spontaneous mutation frequency of each strain is relatively constant, but when a mutagen is added to the agar the mutation frequency is increased 2- to 100-fold. Cells which grow to form colonies on the minimal media petri plates are therefore assumed to have reverted, either spontaneously or by the action of a test substance to his+ genotype.

3. MATERIALS

A. Indicator Microorganisms

The Salmonella typhimurium strains used in this assay were obtained from Dr. Bruce Ames, University of California at Berkeley.[1-5] The following 4 strains were used:

Strain Designation	Gene Affected	Additional Mutations			Mutation Type Detected
		Repair	LPS	R Factor	
TA-1535	his G	Δ uvr B	rfa	-	Base-pair substitution
TA-1537	his C	Δ uvr B	rfa	-	Frameshift
TA-98	his D	Δ uvr B	rfa	pKM101	Frameshift
TA-100	his G	Δ uvr B	rfa	pKM101	Base pair substitution

3. MATERIALS (Continued)

The aforementioned strains have, in addition to the mutation in the histidine operon, a mutation (rfa-) that leads to defective lipopolysaccharide coat, a deletion that covers genes involved in the synthesis of vitamin biotin (bio-) and in the repair of ultraviolet (uv) - induced DNA damage (uvrB-). The rfa- mutation makes the strains more permeable to many large molecules. The uvrB- mutation decreases repair of some types of chemically or physically damaged DNA and thereby enhances the strain's sensitivity to some mutagenic agents. The resistant transfer factor plasmid (R factor) pKM101 in TA-98 and TA-100 is believed to cause an increase in error-prone DNA repair that leads to many more mutations for a given dose of most mutagens[5]. In addition, plasmid pKM101 confers resistance to the antibiotic ampicillin, which is a convenient marker to detect the presence of plasmid in the cells.

All indicator strains are kept at 4°C on minimal medium plates supplemented with a trace of biotin and an excess of histidine. The plates with plasmid-carrying strains contain in addition ampicillin (25 µg/ml) to ensure stable maintenance of plasmid pKM101. New stock culture plates are made as often as necessary from frozen master cultures or from single colony reisolates that were checked for their genotypic characteristics (his, rfa, uvrB, bio) and for the presence of plasmid. For each experiment, an inoculum from the stock culture plates is grown overnight at 37°C in nutrient broth (Oxoid CM67).

B. Media

The bacterial strains were cultured in Oxoid Media #2 (nutrient Broth). The selective medium was Vogel Bonner Medium E with 2% glucose[7]. The overlay agar consisted of 0.6% purified agar with 0.5 mM histidine, 0.05 mM biotin and 0.1 M NaCl according to the methods of Ames et. al.[6]

C. Activation System

(1) S9 Homogenate

A 9,000 x g supernatant prepared from Sprague-Dawley adult male rat liver induced by Aroclor 1254 (described by Ames et. al.[6]) was purchased from Bionetics Laboratory Products, Litton Bionetics, Inc. and used in this assay.

(2) S9 Mix

Components	Concentration per Milliliter S9 Mix
NADP (sodium salt)	4 µmoles
D-glucose-6-phosphate	5 µmoles
$MgCl_2$	8 µmoles
KCl	33 µmoles
Sodium phosphate buffer pH 7.4	100 µmoles
Organ homogenate from rat liver (S9 fraction)	100 µliters

4. EXPERIMENTAL DESIGN

A. Dosage Selection

All tests are run at a minimum of four concentrations. In the Standard EPA Level I Ames assays, five dose levels of the test material, dissolved in a suitable solvent, are added to the test system. The standard test doses for the extracted material are 10, 25, 50, 100 and 200 µliters per plate. The solids are tested up to 5 mg per plate and at lower concentrations of 2.5, 1, 0.5, 0.1 and 0.05 mg per plate. The samples are retested over a narrower range of concentrations with strains showing positive results if there is enough sample.

B. Mutagenicity Testing

The procedure used is based on the paper published by Ames et. al.[6] and is performed as follows:

(1) Nonactivation Assay

To a sterile 13 x 100 mm test tube placed in a 43°C water bath the following is added in order:

(a) 2.00 ml of 0.6% agar containing 0.05 mM histidine and 0.05 mM biotin.

(b) 0.05 ml of a solution of the test chemical to give the appropriate dose.

(c) 0.1 ml - 0.2 ml of indicator organism(s).

(d) 0.50 ml of 0.2M phosphate buffer, pH 7.4.

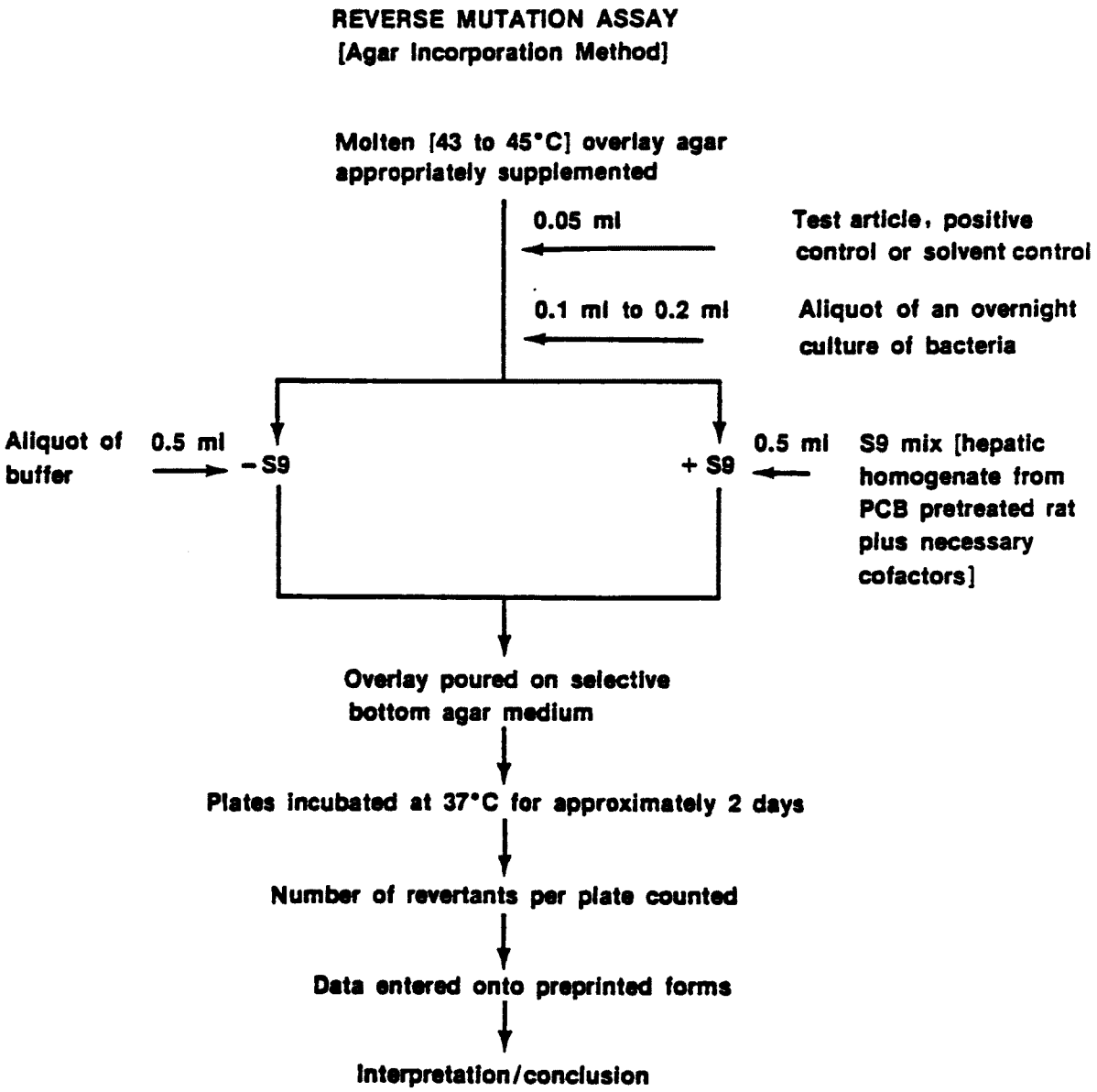
This mixture is swirled gently and then poured onto minimal agar plates (see 3B, Media). After the top agar has set, the plates are incubated at 37°C for approximately 2 days. The number of his+ revertant colonies growing on the plates is counted and recorded.

(2) Activation Assay

The activation assay is run concurrently with the nonactivation assay. The only difference is the addition of 0.5 ml of S9 mix (see 3C:2, Activation System) to the tubes in place of 0.5 ml of phosphate buffer which is added in nonactivation assays. All other details are similar to the procedure for nonactivation assays.

A detailed flow diagram for the plate incorporation assay is provided in Figure 1.

FIGURE 1


REVERSE MUTATION ASSAY
[Agar Incorporation Method]
Molten [43 to 45°C] overlay agar appropriately supplemented
0.05 ml
Test article, positive control or solvent control
0.1 ml to 0.2 ml
Aliquot of an overnight culture of bacteria
Aliquot of buffer
0.5 ml
- S9
+ S9
0.5 ml
S9 mix [hepatic homogenate from PCB pretreated rat plus necessary cofactors]
Overlay poured on selective bottom agar medium
Plates incubated at 37°C for approximately 2 days
Number of revertants per plate counted
Data entered onto preprinted forms
Interpretation/conclusion

4. EXPERIMENTAL DESIGN (Continued)

C. Control Compounds

A negative control consisting of the solvent used for the test material is performed in all cases. For negative controls, step 'b' of Nonactivation Assays is replaced by 0.05 ml of the solvent. The negative controls are employed for each indicator strain and are performed in the absence and presence of S9 mix. The solvent used to prepare the stock solution of the test material is given in the Results section of this report. All dilutions of the test material are made using this solvent. The amount of solvent used is equal to the maximum volume used to give the appropriate test dose.

Specific positive control compounds known to revert each strain are also used in the assays. The concentrations and specificities of these compounds to specific strains are given in the following table:

Assay	Chemical	Solvent	Concentration per Plate (µg)	Salmonella Strains
Nonactivation	Sodium azide	Water	1	TA-1535, TA-100
	2-Nitrofluorene (NF)	Dimethyl-sulfoxide	10	TA-98
	9-aminoacridine (9AA)	Ethanol	50	TA-1537
Activation	2-anthramine (ANTH)	Dimethyl-sulfoxide	2.5	For all strains

5. EVALUATION CRITERIA

Statistical methods are not currently used and evaluation is based on the criteria included in this protocol.

Plate test data consist of direct revertant colony counts obtained from a set of selective agar plates seeded with populations of mutant cells suspended in a semisolid overlay. Because the test material and the cells are incubated in the overlay for approximately 2 days and a few cell divisions occur during the incubation period, the test is semiquantitative in nature. Although these features reduce the quantitation of result, they provide certain advantages not contained in a quantitative suspension test:

- The small number of cell divisions permits potential mutagens to act on replicating DNA, which is often more sensitive than nonreplicating DNA.
- The combined incubation of the test article and the cells in the overlay permits constant exposure of the indicator cells for approximately 2 days.

A. Surviving Populations

Plate test procedures do not permit exact quantitation of the number of cells surviving chemical treatment. At low concentrations of the test material, the surviving population on the treatment plates is essentially the same as that on the negative control plate. At high concentrations, the surviving population is usually reduced by some fraction. Our protocol will normally employ several doses ranging over two or three log concentrations. This does not apply to spot tests and tests performed on fabrics and like materials which are tested at a single concentration.

B. Dose-Response Phenomena

The demonstration of dose-related increases in mutant counts is an important criterion in establishing mutagenicity. A factor that might modify dose-response results for a mutagen would be the selection of doses that are too low (usually mutagenicity and toxicity are related). If the highest dose is far lower than a toxic concentration, no increases may be observed over the dose range selected. Conversely, if the lowest dose employed is highly cytotoxic, the test material may kill any mutants that are induced, and the test material will not appear to be mutagenic.

5. EVALUATION CRITERIA (Continued)

C. Control Tests

Positive and negative control assays are conducted with each experiment and consist of direct-acting mutagens for nonactivation assays and mutagens requiring metabolic biotransformation in activation assays. Negative controls consist of the test material solvent in the overlay agar together with the other essential components. The negative control plate for each strain gives a reference point to which the test data is compared. The positive control assay is conducted to demonstrate that the test systems are functional with known mutagens.

D. Evaluation Criteria for Ames Assay

Because the procedures used to evaluate the mutagenicity of the test material are semiquantitative, the criteria used to determine positive effects are inherently subjective and are based primarily on a historical data base. Most data sets are evaluated using the following criteria:

(1) Strains TA-1535, TA-1537

If the solvent control value is within the normal range, a test material producing a positive response equal to three times the solvent control value is considered mutagenic.

(2) Strains TA-98 and TA-100

If the solvent control value is within the normal range, a test material producing a positive response equal to twice the solvent control value for TA-98 and TA-100 is considered mutagenic.

(3) Pattern

Because TA-1535 and TA-100 are both derived from the same parental strain (G-46) and because TA-1538 and TA-98 are both derived from the same parental strain (D3052), to some extent there is a built-in redundancy in the microbial assay. In general, the two strains of a set respond to the same mutagen and such a pattern is sought. Generally, if a strain responds to a mutagen in nonactivation tests, it will do so in activation tests.

(4) Reproducibility

If a test material produces a response in a single test which cannot be reproduced in additional runs, the initial positive test data lose significance.

5. EVALUATION CRITERIA (Continued)

E: Evaluation Criteria for Toxicity

(1) Complete toxicity

When there are no revertants observed on the plate(s) treated with the test compound, the test compound is defined as toxic to all or any of the indicator strains at that (those) particular dose(s).

(2) Slight toxicity

When there are fifty or less percent revertants on the plate(s) treated with the test compound as compared to the solvent control plate(s), the test compound is defined as slightly toxic to all or any of the indicator strains at that (those) particular dose(s).

F. Relation Between Mutagenicity and Carcinogenicity

It must be emphasized that the Ames Salmonella/Microsome Plate Assay is not a definitive test for chemical carcinogens. It is recognized, howerver, that correlative and functional relations have been demonstrated between these two endpoints. The results of comparative tests on 300 chemicals by McCann et. al.[1] show an extremely good correlation between results of microbial mutagenesis tests and in vivo rodent carcinogenesis assays.

All evaluations and interpretation of the data to be presented in the final report will be based only on the demonstration, or lack, of mutagenic activity.

REFERENCES

1. McCann,J., Choi, E., Yamasaki, E. and Ames, B.N.: Detection of carcinogens as mutagens in the Salmonella/microsome test: Assay of 300 chemicals. Proc. Nat. Acad. Sci. USA, 72:5135-5139, 1975.

2. Ames, B.N., Gurney, E.G., Miller, J.A. and Bartsch, H.: Carcinogens as frameshift mutagens: Metabolites and derivatives of 2-acetylaminofluorene and other aromatic amine carcinogens. Proc. Nat. Acad. Sci. USA, 69:3128-3132, 1972.

3. Ames, B.N, Lee, F.D. and Durston, W.E.: An improved bacterial test system for the detection and classification of mutagens and carcinogens. Proc. Nat. Acad. Sci. USA, 70:782-786, 1973.

4. Ames, B.N, Durston, W.E., Yamasaki, E. and Lee, F.D.: Carcinogens are mutagens: A simple test system combining liver homogenates for activation and bacteria for detection. Proc. Nat. Acad. Sci. USA, 70:2281-2285, 1973.

5. McCann, J., Springarn, N.E., Kobori, J. and Ames, B.N.: Detection of carcinogens as mutagens: Bacterial tester strains with R factor plasmids. Proc. Nat. Acad. Sci. USA, 72:979-983, 1975.

6. Ames, B.N., McCann, J. and Yamasaki, E.: Methods for detecting carcinogens and mutagens with the Salmonella/mammalian-microsome mutagenicity test. Mutation Res., 31:347-364, 1975.

7. Vogel, H.J. and Bonner, D.M.: Acetylornithinase of E. coli; Partial purification and some properties. J. Biol. Chem., 218:97-106, 1956.

Q.A. Inspection Statement
(reference 21 CFR 58.35(b)(7))

PROJECT 20988 LBI Assay No. 5394

TYPE of STUDY AMES PLATE TEST

This final study report was reviewed by the LBI Quality Assurance Unit on 12/5/80. A report of findings was submitted to the Study Director and to Management on 12/5/80.

The short-term nature of this study precluded inspection while it was in process. The Quality Assurance Unit inspects an in-process study of this type approximately once per month to assure that no significant problems exist that are likely to affect the integrity of this type of study.

Mitchell S. Ehrl

Auditor, Quality Assurance Unit

GENETICS ASSAY NO. 5394
LBI SAFETY NO. 6378

TOXIC EVALUATION OF SAMPLE #1452 (WATER TANK DRAIN, END OF TEST) IN THE RODENT QUANTAL TOXICITY ASSAY

FINAL REPORT

SUBMITTED TO:

ACUREX CORPORATION
485 CLYDE AVENUE
MOUNTAIN VIEW, CA 94042

SUBMITTED BY:

LITTON BIONETICS, INC.
5516 NICHOLSON LANE
KENSINGTON, MARYLAND 20795

LBI PROJECT NO. 22064

REPORT DATE: DECEMBER, 1980

PREFACE

This report contains a summary of the data compiled during the evaluation of the test compound. The report is organized to present the results in a concise and easily interpretable manner. The first part contains items I - VIII. Items I - III provide sponsor and compound identification information and identify the type of assay. The assay was conducted and evaluated according to procedures recommended in IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests (Litton Bionetics, Inc., Kensington, MD, September, 1980, in press). Item IV provides the initiation and completion dates for the study, and Item V provides identification of supervisory personnel. Item VI identifies the tables and figures containing the data used by the study director in interpreting the test results. The interpretation itself is in Item VII. Item VIII provides the conclusion and evaluation.

The second part of the report, entitled Assay Design, describes the materials and procedures employed in conducting the assay. This part of the report also contains evaluation criteria used by the study director, and any appendices. The evaluation criteria are included to acquaint the sponsor with the methods used to develop and analyze the test results.

All test and control results presented in this report are supported by fully documented raw data which are permanently maintained in the files of the Department of Genetics and Cell Biology or in the archives of Litton Bionetics, Inc., 5516 Nicholson Lane, Kensington Maryland, 20795.

Copies of raw data will be supplied to the sponsor upon request.

I. SPONSOR: Acurex Corporation

II. MATERIAL (TEST COMPOUND): GENETICS ASSAY NO.: 5394

A. Identification: Sample #1452 (Man Test 5/21/80, #5 Gal, water tank drain, end of test)

B. Date Received: October 6, 1980

C. Physical Description: Clear, colorless liquid

III. TYPE OF ASSAY: Rodent Quantal Toxicity Assay

IV. STUDY DATES:

A. Initiation: October 22, 1980

B. Completion: November 24, 1980

V. SUPERVISORY PERSONNEL:

A. Study Director: David J. Brusick, Ph.D.

B. Laboratory Supervisor: Joan McGowan

VI. RESULTS:

The data are presented in Table 1 on pages 3 and 4.

VII. INTERPRETATION OF RESULTS:

The test material, Sample No. 1452, was concentrated 10-fold (as described under Assay Design) prior to administration by oral gavage to 10 male and 10 female weanling CD-1 mice. A single average dose of 5 ml/kg was given, which corresponded to 0.08 ml for the males and 0.07 ml for the females. The animals were observed for 14 days for toxic signs or lethality. Survivors were weighed and necropsied on Day 14 after dosing.

All twenty animals survived the exposure with no evidence of any compound-related toxic signs. Both the male and female mice showed good weight gains, and no observations were obtained at necropsy that indicated compound-related lesions (Table 1). The test material was therefore considered to have no detectable toxicity in this assay, and according to the Evaluation Criteria, an LD50 determination was unnecessary.

VIII. CONCLUSIONS:

The test material, Sample No. 1452, concentrated 10-fold, had no detectable toxicity to weanling CD-1 mice in the Rodent Quantal Toxicity Assay. The applied dose was 5 ml/kg.

Submitted by:

Study Director

for: Brian C. Myhr 12/3/80

David J. Brusick, Ph.D. Date
Director
Department of Genetics
and Cell Biology

TABLE 1

QUANTAL TOXICITY DATA WITH WEANLING MICE

Quantal Toxicity: Weanling mice
Sponsor: Acurex Corporation
Test Article: Sample No. 1452
Vehicle: Not Applicable
Study Dates: 11/10/80 to 11/24/80
Animal Room No.: 4S
Animals: Charles River CD-1 mice, P.O. 100760
Dose: 5 ml/kg administered P.O.

Animal No.	Initial Weight gm	Final Weight gm	Visible Toxic Signs[a]	Gross Necropsy Findings
Males				
1101	16.1	24.2	NTS[b]	Small mucoid lump in bladder
1102	16.7	25.5	NTS	Both eyes opaque
1103	14.2	22.1	NTS	Small mucoid lump in bladder
1104	14.4	21.7	NTS	NSL[c]
1105	15.6	23.2	NTS	NSL
1106	13.5	22.2	NTS	Both eyes opaque
1107	16.2	23.7	NTS	NSL
1108	13.0	23.7	NTS	NSL
1109	14.8	23.8	NTS	·Both eyes opaque ·Mesenteric lymph nodes slightly enlarged ·Left kidney slightly pale
1110	16.6	25.2	NTS	Both eyes opaque

Mean Body Weight:

Initial 15.1 ± 1.3 gm (Standard Deviation)
Final 23.5 ± 1.3 gm (Standard Deviation)

[a] Animals observed for 14 days.

[b] NTS = No Toxic Signs.

[c] NSL = No Significant Lesions.

TABLE 1 (continued)

QUANTAL TOXICITY DATA WITH WEANLING MICE

Animal No.	Initial Weight gm	Final Weight gm	Visible Toxic Signs[a]	Gross Necropsy Findings
Females				
1111	14.1	19.8	NTS[b]	NSL[c]
1112	14.4	20.0	NTS	NSL
1113	14.3	20.4	Possible nasal irritation	Uterine horns enlarged/ fluid
1114	14.5	20.2	NTS	NSL
1115	12.5	19.0	NTS	NSL
1116	14.0	21.2	NTS	Uterine horns enlarged/ fluid
1117	13.3	18.7	NTS	NSL
1118	12.1	19.9	NTS	NSL
1119	13.5	19.7	NTS	Left kidney slightly smaller than right
1120	11.9	16.7	NTS	NSL

Mean Body Weight:

Initial 13.5 ± 1.0 gm (Standard Deviation)
Final 19.6 ± 1.2 gm (Standard Deviation)

[a]Animals observed for 14 days.

[b]NTS = No Toxic Signs.

[c]NSL = No Significant Lesions.

ASSAY DESIGN

RODENT QUANTAL TOXICITY ASSAY

This assay conforms to one of the bioassay tests in the US Environmental Protection Agency (EPA) IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests (September, 1980). Level 1 bioassays obtain preliminary information on the harmful biological effects of chemicals found in industrial feed and waste streams.

1. OBJECTIVE

The objective of this assay was to evaluate the acute toxicity of a test material when administered by oral gavage to male and female weanling mice.

The assay consisted of recording any lethality and toxic signs that occurred over a 14-day period following a single treatment and then collecting necropsy information on animals that died or were killed at the end of the observation period.

2. TEST MATERIAL

A test material described as Sample #1452 (Man Test 5/21/80, #5 Gal, Water Tank Drain, End of Test) was received as a clear, aqueous liquid containing no obvious particulate material. The sample was stored at 4°C in its original one-gallon amber glass container.

An aliquot of 600 ml of test material was concentrated 10-fold to 60 ml by lyophilization prior to testing. A total volume of 1.5 ml of the concentrate was then utilized for oral dosing of a group of 20 weanling mice. The concentrate was stored at 4°C in an amber glass bottle.

3. EXPERIMENTAL DESIGN

Nine nursing female Charles River CD-1 mice with six pups each (three male and three female) were obtained from Charles River Breeding Laboratories, Inc., Wilmington, Massachusetts on November 5, 1980. The birth date of the pups was October 19, 1980. The animals were quarantined for 5 days upon receipt. The litters were individually housed on Absorb-Dri bedding in polycarbonate cages and were cared for according to Litton Bionetics, Inc., Department of Genetics and LAMS Standard Operating Procedures. Purina certified laboratory chow and water (pH 2.5) were provided ad libitum. The pups were

3. EXPERIMENTAL DESIGN (continued)

maintained with mothers until weaned. The animals were identified by eartags and cage cards and were released for study on November 10, 1980.

Prior to initiation of dosing, ten male and ten female weanling mice were individually weighed; the volume of the test material to be administered was based on the mean weight values for each sex. The test material (10-fold concentrate of Acurex Sample #1452) was administered once at 5 ml/kg on November 10, 1980. The weanling mice were 23 days old. The animals were observed at the time of dosing, 2 hours later, and daily thereafter. On day 14, the surviving animals were fasted overnight. They were subjected to gross necropsy the next day.

4. ASSAY EVALUATION CRITERIA

Immediately following administration of the test material and at frequent intervals during the first day, observations of the frequency and severity of all toxic signs or pharmacological effects (as listed in Table A) will be recorded. Particular attention will be paid to the time of onset and disappearance of the signs. Daily observations will be made and recorded on all animals through a 14-day period. At termination of the observation period, all surviving animals will be weighed, killed, and then gross necropsies performed. Necropsies will also be performed on all animals that die during the course of this study.

If no mortality occurs in the quantal study, no further studies will be performed with the test substance and the LD_{50} should be reported as greater than 5 ml/kg or 5 g/kg. The test material is ranked as having nondetectable toxicity (ND) at the maximum applicable dose (MAD). Effluent samples which produce harmful effects *in vivo* and do not result in deaths will be noted in the results summary. Such observations are difficult to quantitate but provide insight into the sublethal effects of a sample on rodents. Further investigations may be recommended from observations of nonlethal toxic effects.

If a single animal in the quantal study dies in the 14-day observation period, a quantitative study will be performed. An LD_{50} will be calculated by the method of Litchfield and Wilcoxin. If the data are not suitable for calculation of a precise LD_{50}, i.e., total mortality occurs for the lowest dose, an estimate of the LD_{50} could be made or the LD_{50} could be expressed as 0.05 ml/kg or 0.05 g/kg or less. Occasionally, it may be necessary to use a different series of dosages in a repeat study to accurately locate the LD_{50}. The calculated LD_{50} value is used to rank the toxicity of the test material according to the dose ranges presented in Table B.

TABLE A. DEFINITION OF PHARMACOLOGICAL TOXIC SIGNS

Organ System	Observation and Examination	Common Signs of Toxicity
CNS and somatomotor	Behavior	Change in attitude to observer, unusual vocalization, restlessness, sedation
	Movements	Twitch, tremor, ataxia, catatonia, paralysis, convulsion, forced movements
	Reactivity to various stimuli	Irritability, passivity, anaesthesis, hyperaesthesis
	Cerebral and spinal reflexes	Sluggishness, absence
	Muscle tone	Rigidity, flaccidity
Autonomic nervous system	Pupil size	Myosis, mydriasis
	Secretion	Salivation, lacrimation
Respiratory	Nostrils	Discharge
	Character and rate of breathing	Bradypnoea, dyspnoea, Cheyne-Stokes breathing, Kussmaul breathing
Cardiovascular	Palpation of cardiac region	Thrill, bradycardia, arrhythmia, stronger or weaker beat
Gastrointestinal	Events	Diarrhea, constipation,
	abdominal shape	Flatulence, contraction
	feces consistency and color	Unformed, black or clay colored
	Vulva, mammary glands	Swelling
	Penis	Prolapse
	Perianal region	Soiled
Skin and fur	Color, turgor, integrity	Reddening, flaccid skinfold, eruptions, piloerection
Mucous membranes	Conjunctiva, mouth	Discharge, congestion, hemorrhage cyanosis, jaundice
Eye	Eyeball	Exophthalmus, nystagmus
	Transparency	Opacities
Others	Rectal or paw skin	Subnormal, increased temperature
	Injection site	Swelling
	General Condition	Abnormal posture, emaciation

4. ASSAY EVALUATION CRITERIA (continued)

Observations are also made and recorded daily on all animals through the 14-day period. As in the quantal phase, no attempt is made to quantitate or rank the observations. The average animal body weight of each group is determined initially and at the termination of the experiment. The average weights and the weights as fractions of the control are reported for each dose level. Necropsy observations are recorded and reported.

TABLE B. ACUTE IN VIVO RODENT ASSAY EVALUATION CRITERIA

Toxicity[a]	Solids (LD_{50} in g/kg)	Liquids (LD_{50} in ml/kg)
High	<0.05	<0.05
Moderate	0.05 to 0.5	0.05 to 0.5
Low	0.5 to 5	0.5 to 5
Not Detectable	>5	>5

[a]Evaluation criteria formulated by Litton Bionetics, Inc. for IERL-RTP Procedures Manual: Level 1 Environmental Assessment Biological Tests.

5. RECORDS TO BE MAINTAINED

All raw data, protocols, protocol modifications, test article weight and dispensation records and correspondence between LBI and the Sponsor are being maintained in a central file within the Department of Genetics and Cell Biology. These records will be filed under departmental assay number and held up to 2 years following submission of the final report to the Sponsor. After 2 years they will be transferred to the LBI archives for permanent storage.

Q.A. Inspection Statement
(reference 21 CFR 58.35(b)(7))

PROJECT 22064 LBI Assay No. 5394

TYPE of STUDY Rodent Quantal Toxicity Assay

This final study report was reviewed by the LBI Quality Assurance Unit on 12/10/80. A report of findings was submitted to the Study Director and to Management on 12/10/80.

The short-term nature of this study precluded inspection while it was in process. The Quality Assurance Unit inspects an in-process study of this type approximately once per month to assure that no significant problems exist that are likely to affect the integrity of this type of study.

Mitchell L Elvid

Auditor, Quality Assurance Unit

SECTION 6

SAM IA WORK SHEETS

1. SOURCE/CONTROL OPTION: RESIDENTIAL FURNACE M.A.N / HADWICK

LEVEL 1 X
LEVEL 2 _____

2. EFFLUENT STREAM
CODE NO. _____ NAME GAS STREAM

3. TOTAL MASS RATE OF DISCHARGE
Q = 98 g/s

4. COMPLETE THE FOLLOWING TABLE FOR THE EFFLUENT STREAM OF LINE 2.

A	B	C	D	E	F	G	H	I	J	K	L	M
SPECIES OR MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (C/D)	DISCHARGE SEVERITY — ECOLOGICAL (C/F)	✓ IF HEALTH DMEG EXCEEDED	✓ IF ECOL DMEG EXCEEDED	WEIGHTED DISCHARGE SEVERITY (HEALTH BASED) (H x LINE 2)	WEIGHTED DISCHARGE SEVERITY (ECOLOGICAL BASED) (I x LINE 2)
UNITS	—	$\mu g/m^3$	$\mu g/m^3$	—		—	—	—	—	—	g/s	g/s
ALIPHATIC HYDROCARBONS	01	2.6×10^3	3.5×10^5	01A200			7.4×10^{-3}				7.25×10^{-1}	
METHANE	01A020	3.9×10^3	3.3×10^6	01A020			1.2×10^{-3}				1.2×10^{-1}	
ALDEHYDES	07A	1.0×10^2	2.5×10^2	07A060			4.0×10^{-1}				3.92×10^1	
CARBOXYLIC ACIDS	08A,B	2.0×10^2	1.0×10^3	08A120			2.0×10^{-1}				1.96×10^1	
NAPHTHALENE	021A020	3.6×10^1	2.2×10^5	021A020			1.6×10^{-4}				1.6×10^{-2}	
PHENANTHRENE	21A180	2.0	1.6×10^3	21A180			1.3×10^{-3}				1.27×10^{-1}	
LITHIUM	27	2.4×10^{-1}	2.2×10^1	27A000			1.1×10^{-2}				1.1	
(IF MORE SPACE IS NEEDED USE A CONTINUATION SHEET)												

5 TOTAL DISCHARGE SEVERITY
HEALTH BASED (Σ col H) 5a 2.72×10^1
ECOLOGICAL BASED (Σ col I) 5b _____
(ENTER HERE AND AT LINE 8 OF THE SUMMARY SHEET)

6 TOTAL WEIGHTED DISCHARGE SEVERITY
HEALTH BASED (Σ col L) 6a 2.69×10^3 g/s
ECOLOGICAL BASED (Σ col M) 6b _____ g/s
(ENTER HERE AND AT LINE 8 OF THE SUMMARY SHEET)

CONTINUATION SHEET FOR WORKSHEET Page 2 of 5

1. SOURCE/CONTROL OPTION RESIDENTIAL FURNACE M.A.N/HADWICK LEVEL 1 X LEVEL 2 ____

2. EFFLUENT STREAM CODE NO. ____ NAME GAS STREAM

A	B	C	D	E	F	G	H	I	J	K	L	M
SPECIES OR MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (C÷D)	DISCHARGE SEVERITY — ECOLOGICAL (C÷F)	√ IF HEALTH DMEG EXCEEDED	√ IF ECOL DMEG EXCEEDED	WEIGHTED DISCHARGE SEVERITY: HEALTH BASED (% × LINE 3)	WEIGHTED DISCHARGE SEVERITY: ECOLOGICAL BASED (% × LINE 3)
UNITS	—	$\mu g/m^3$	$\mu g/m^3$	—		—	—	—	—	—	%	%
SODIUM	28	$>2.2\times10^{2}$	2.0×10^{3}	298100			$>1.1\times10^{-1}$				$>1.1\times10^{1}$	
POTASSIUM	29	4.2×10^{1}	2.0×10^{3}	298100			2.1×10^{-2}				2.1	
RUBIDIUM	30	4.2×10^{-2}	1.2×10^{5}	309200			3.5×10^{-7}				3.4×10^{-5}	
CESIUM	31	4.2×10^{-2}	8.2×10^{4}	312300			5.1×10^{-7}				5.0×10^{-5}	
MAGNESIUM	33	3.8×10^{1}	6.0×10^{3}	334000			6.3×10^{-3}				6.2×10^{-1}	
CALCIUM	34	$>5.4\times10^{1}$	1.6×10^{4}	344200			3.4×10^{-3}				3.3×10^{-1}	
STRONTIUM	35	2.2×10^{-1}	3.1×10^{3}	354000			7.1×10^{-5}				7.0×10^{-3}	
BARIUM	36	3.9	5.0×10^{2}	364000			7.8×10^{-3}				7.7×10^{-1}	
BORON	37	$>5.4\times10^{1}$	3.1×10^{3}	374000			$>1.7\times10^{-2}$				>1.7	
ALUMINUM	38	$>7.1\times10^{1}$	5.2×10^{3}	384000			$>1.4\times10^{-2}$				>1.4	
GALLIUM	39	8.9×10^{-2}	5.0×10^{3}	394200			1.8×10^{-5}				1.8×10^{-3}	

CONTINUATION SHEET FOR WORKSHEET

Page 3 of 5

LEVEL 1 7

LEVEL 2 ____

1. SOURCE/CONTROL OPTION RESIDENTIAL FURNACE M.A.N / HADWICK

2. EFFLUENT STREAM GAS STREAM

CODE NO. ____ NAME

A	B	C	D	E	F	G	H	I	J	K	L	M
SPECIES OR MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (C÷D)	DISCHARGE SEVERITY — ECOLOGICAL (C÷F)	√ IF HEALTH DMEG EXCEEDED	√ IF ECOL DMEG EXCEEDED	WEIGHTED DISCHARGE SEVERITY	
											HEALTH BASED, IF > LINE 2,	ECOLOGICAL BASED, IF > LINE 3,
UNITS	—	$\mu g/m^3$	$\mu g/m^3$	—		—	—	—	—	—	—	—
CO	428100	3.1×10^{4}	4.0×10^{4}	428100			7.7×10^{-1}				7.6×10^{1}	
SILICON	43	$>7.9\times10^{1}$	1.0×10^{4}	438000			$>7.9\times10^{-3}$				$>7.74\times10^{-1}$	
GERMANIUM	44	$<3.3\times10^{-2}$	5.6×10^{2}	440000			$<5.9\times10^{-5}$				$<5.8\times10^{-3}$	
TIN	46	1.1×10^{-1}	1.0×10^{4}	458100			1.1×10^{-5}				1.1×10^{-3}	
LEAD	46	2.8	1.5×10^{2}	464000			1.9×10^{-2}				1.9	
NO.	478100	9.7×10^{4}	9.0×10^{3}	478100			1.1×10^{1}		✓		1.1×10^{3}	
PHOSPHOROUS	48	2.2	1.0×10^{2}	489200			2.2×10^{-2}				2.2	
ARSENIC	49	1.7×10^{-1}	2.0	490000			8.5×10^{-2}				8.4	
ANTIMONY	50	5.1×10^{-2}	5.0×10^{1}	500000			1.0×10^{-4}				9.8×10^{-3}	
SULFUR	53	4.8×10^{2}	1.0×10^{3}	538140			4.8×10^{-1}				4.7×10^{1}	
SO2	538100	1.3×10^{5}	1.3×10^{4}	538100			10×10^{1}		✓		9.8×10^{2}	

CONTINUATION SHEET FOR WORKSHEET

1. SOURCE/CONTROL OPTION: RESIDENTIAL FURNACE M.A.N/HADWICK — LEVEL 1: X — LEVEL 2: ____

2. EFFLUENT STREAM — CODE NO. ____ — NAME: GAS STREAM

A	B	C	D	E	F	G	H	I	J	K	L	M
SPECIES OR WEG CATEGORY NAME	WEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH BASED WEG CONCENTRATION	WEG ASSUMED — HEALTH	ECOLOGICAL BASED WEG CONCENTRATION	WEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (C÷D)	DISCHARGE SEVERITY — ECOLOGICAL (C÷F)	√ IF HEALTH BASED WEG EXCEEDED	√ IF ECOL BASED WEG EXCEEDED	WEIGHTED DISCHARGE SEVERITY: HEALTH BASED (H × LINE 2)	WEIGHTED DISCHARGE SEVERITY: ECOLOGICAL BASED (I × LINE 2)
UNITS	—	$\mu g/m^3$	$\mu g/m^3$	—		—	—	—	—	—	—	—
SELENIUM	54	2.5×10^{-1}	2.0×10^{2}	54A000			1.3×10^{-3}				1.3×10^{-1}	
TELLURIUM	55	4.2×10^{-1}	1.0×10^{2}	55A000			4.2×10^{-3}				4.1×10^{-1}	
FLUORINE	56	1.4×10^{1}	2.0×10^{3}	56B100			7.0×10^{-3}				6.9×10^{-1}	
BROMINE	58	1.7	1.0×10^{4}	58B100			1.7×10^{-4}				1.7×10^{-2}	
YTTRIUM	61	2.8×10^{-1}	1.0×10^{3}	61A200			2.8×10^{-4}				2.7×10^{-2}	
TITANIUM	62	1.3×10^{1}	6.0×10^{3}	62A000			2.2×10^{-3}				2.2×10^{-1}	
VANADIUM	65	1.1	5.0×10^{2}	65A000			2.2×10^{-3}				2.2×10^{-1}	
TANTALUM	67	5.1×10^{-1}	5.0×10^{3}	67A200			1.0×10^{-4}				9.8×10^{-3}	
CHROMIUM	68	3.4	1.0	68A000			3.4		✓		3.3×10^{2}	
MOLYBDENUM	69	2.8	5.0×10^{3}	69A000			5.6×10^{-4}				5.5×10^{-2}	
MANGANESE	71	1.4	5.0×10^{3}	71A000			2.8×10^{-4}				2.8×10^{-2}	

CONTINUATION SHEET FOR WORKSHEET

1. SOURCE/CONTROL OPTION *Residential Furnace M.A.N / Hadwick* LEVEL 1 ✓ LEVEL 2 ___

2. EFFLUENT STREAM *Gas Stream*

CODE NO. NAME

A	B	C	D	E	F	G	H	I	J	K	L	M
Species or MEG category name	MEG ID number or category	Species or category concentration	Health DMEG concentration	MEG assumed — health	Ecological DMEG concentration	MEG assumed — ecology	Discharge severity — health (C/D)	Discharge severity — ecological (C/F)	# health DMEG exceeded	# ecol DMEG exceeded	Weighted discharge severity: Health based (B × line 3)	Weighted discharge severity: Ecological based (B × line 3)
UNITS	—			—		—	—	—	—	—	g/s	g/s
Iron	72	1.5×10^{1}	1.0×10^{3}	72A200			1.5×10^{-2}				1.47	
Cobalt	74	1.1×10^{-1}	5.0×10^{1}	74A000			2.2×10^{-3}				2.2×10^{-1}	
Nickel	76	8.6	1.5×10^{1}	76A000			5.7×10^{-1}				5.6×10^{1}	
Copper	78	5.9	2.0×10^{2}	78A000			3.0×10^{-2}				3.0	
Silver	79	5.4×10^{-3}	1.0×10^{1}	79A000			5.4×10^{-4}				5.3×10^{-2}	
Zinc	81	7.5	4.0×10^{3}	81A000			1.9×10^{-3}				1.9×10^{-1}	
Mercury	83	4.8×10^{-1}	5.0×10^{1}	83A000			9.6×10^{-3}				9.4×10^{-1}	
Lanthanum	84	1.1×10^{-1}	1.1×10^{5}	84A010			1.0×10^{-6}				9.8×10^{-5}	
Cerium	84	1.5×10^{-1}	3.7×10^{4}	84A020			4.1×10^{-6}				4.0×10^{-4}	
Neodymium	84	5.4×10^{-3}		84A040			--				--	

1. SOURCE/CONTROL OPTION: RESIDENTIAL FURNACE M.A.N. HADWICK

LEVEL 1 X
LEVEL 2 ____

2. EFFLUENT STREAM
CODE NO. ____ NAME: WASTE WATER (LIQUID STREAM)

3. TOTAL MASS RATE OF DISCHARGE
Q = 4.7×10^{-2} g/s

4. COMPLETE THE FOLLOWING TABLE FOR THE EFFLUENT STREAM OF LINE 2

A	B	C	D	E	F	G	H	I	J	K	L	M
SPECIES OR MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	*HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (CD)	DISCHARGE SEVERITY — ECOLOGICAL (CF)	% HEALTH DMEG EXCEEDED	% ECOL DMEG EXCEEDED	WEIGHTED DISCHARGE SEVERITY: HEALTH BASED (= LINE 2k)	WEIGHTED DISCHARGE SEVERITY: ECOLOGICAL BASED (= LINE 2l)
UNITS	—	ug/l	ug/l	—		—	—	—	—	—	g/s	g/s
NAPHTHALENE	21A020	4.0×10^{1}	7.5×10^{5}	21A020	1.0×10^{2}	21A020	5.3×10^{-7}	4.0×10^{-3}			2.49×10^{-8}	1.88×10^{-4}
PHENANTHRENE	21A180	8.0×10^{-2}	2.4×10^{4}	21A180	--	--	3.3×10^{-6}	--			1.55×10^{-7}	--
MAGNESIUM	33	1.0×10^{2}	9.0×10^{4}	33A000	8.7×10^{4}	33A000	1.1×10^{-3}	1.2×10^{-3}			5.2×10^{-6}	5.6×10^{-5}
CALCIUM	34	$>1.0 \times 10^{4}$	2.4×10^{5}	34A000	1.6×10^{4}	34A000	4.2×10^{-2}	6.2×10^{-1}			$>2.0 \times 10^{-3}$	$>2.9 \times 10^{-2}$
STRONTIUM	35	2.0×10^{1}	4.6×10^{4}	35A000	--	--	4.4×10^{-4}	--			2.1×10^{-5}	--
GERMANIUM	44	2.0	8.4×10^{3}	44A000	--	--	2.4×10^{-4}	--			1.1×10^{-5}	--
LEAD	46	7.0×10^{1}	2.5×10^{2}	46A000	5.0×10^{1}	46A000	2.8×10^{-1}	1.4		V	1.3×10^{-2}	6.6×10^{-2}
(IF MORE SPACE IS NEEDED USE A CONTINUATION SHEET)												

5. TOTAL DISCHARGE SEVERITY
HEALTH BASED (Σ col H) 5a = 1.86×10^{2}
ECOLOGICAL BASED (Σ col I) 5b = 1.25×10^{4}
(ENTER HERE AND AT LINE 8 OF THE SUMMARY SHEET)

6. TOTAL WEIGHTED DISCHARGE SEVERITY
HEALTH BASED (Σ col L) 6a = 8.7 g/s
ECOLOGICAL BASED (Σ col M) 6b = 5.8×10^{2} g/s
(ENTER HERE AND AT LINE 8 OF THE SUMMARY SHEET)

CONTINUATION SHEET FOR WORKSHEET

LEVEL 1 X
LEVEL 2 ____

1. SOURCE/CONTROL OPTION *Residential Furnace M.A.N Hadwick*

2. EFFLUENT STREAM

CODE NO. ____ *Waste Water (Liquid Stream)* NAME

A	B	C	D	E	F	G	H	I	J	K	L	M
											WEIGHTED DISCHARGE SEVERITY	
SPECIES OF MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (DS)	DISCHARGE SEVERITY — ECOLOGICAL (EDS)	√ IF HEALTH DMEG EXCEEDED	√ IF ECOL. DMEG EXCEEDED	HEALTH BASED (H × LINE 3)	ECOLOGICAL BASED (I × LINE 3)
UNITS	—	ug/l	ug/l	—		—	—	—	—	—	g/s	g/s
Nitrate	47A300	7.0×10^{3}	7.5×10^{4}	47B180	4.5×10^{2}	47B180	9.3×10^{-2}	1.6×10^{1}		✓	4.4×10^{-3}	7.5×10^{-1}
Sulfate	53A300	1.0×10^{6}	1.5×10^{4}	53B160	4.5×10^{2}	53B160	6.7×10^{1}	2.2×10^{3}	✓	✓	3.2	1.0×10^{2}
Selenium	54	1.0×10^{2}	5.0×10^{1}	54A000	2.5×10^{1}	54A000	2.0	4.0	✓	✓	9.4×10^{-2}	1.9×10^{-1}
Tellurium	55	2.0×10^{1}	1.5×10^{3}	55A000	--	--	1.3×10^{-2}	--			6.1×10^{-4}	--
Fluorine	56	1.0×10^{2}	3.0×10^{4}	56B100	--	--	3.3×10^{-3}	--			1.6×10^{-4}	--
Chloride	57A200	1.0×10^{3}	1.1×10^{5}	57B100	--	--	9.1×10^{-3}	--			4.28×10^{-4}	--
Bromine	58	1.0×10^{2}	1.5×10^{5}	58B100	--	--	6.7×10^{-4}	--			3.1×10^{-5}	--
Scandium	60	4.0	8.0×10^{5}	60A000	--	--	5.0×10^{-6}	--			2.4×10^{-7}	--
Yttrium	61	5.0×10^{1}	1.5×10^{4}	61A200	--	--	3.3×10^{-3}	--			1.6×10^{-4}	--
Titanium	62	2.0×10^{2}	9.0×10^{4}	62A000	8.2×10^{2}	62A000	2.2×10^{-3}	2.4×10^{-1}	—	—	1.0×10^{-4}	1.1×10^{-2}
Zirconium	63	3.0×10^{1}	7.5×10^{4}	63A000	--	--	4.0×10^{-4}	--			1.9×10^{-5}	--

CONTINUATION SHEET FOR WORKSHEET

LEVEL 1 X
LEVEL 2 ____

1. SOURCE/CONTROL OPTION: RESIDENTIAL FURNACE M·A·N HADWICK

2. EFFLUENT STREAM: WASTE WATER (LIQUID STREAM)

CODE NO. ____ NAME ____

A	B	C	D	E	F	G	H	I	J	K	L	M
											WEIGHTED DISCHARGE SEVERITY	
SPECIES OR MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (DS)	DISCHARGE SEVERITY — ECOLOGICAL (EDS)	√ IF HEALTH DMEG EXCEEDED	√ IF ECOL. DMEG EXCEEDED	HEALTH BASED (H × LINE 2)	ECOLOGICAL BASED (I × LINE 2)
UNITS	—	μg/l	μg/l	—		—	—	—	—	—	[illegible]	[illegible]
VANADIUM.	65	4.0	2.5×10^{3}	65A000	1.5×10^{2}	65A000	1.6×10^{-3}	2.7×10^{-2}			7.5×10^{-5}	1.3×10^{-3}
CHROMIUM	68	7.0×10^{2}	2.5×10^{2}	68A000	2.5×10^{2}	68A000	2.8	2.8	✓	✓	1.3×10^{-1}	1.3×10^{-1}
MOLYBDENUM	69	3.0×10^{2}	7.5×10^{4}	69A000	7.0×10^{3}	69A000	4.0×10^{-3}	4.3×10^{-2}			1.9×10^{-4}	2.0×10^{-3}
TUNGSTEN	70	2.0×10^{1}	1.5×10^{4}	70A000	--	--	1.3×10^{-3}	--			6.1×10^{-5}	--
MANGANESE	71	1.9×10^{2}	2.5×10^{2}	71A000	1.0×10^{2}	71A000	7.6×10^{-1}	1.9		✓	3.6×10^{-2}	8.9×10^{-2}
IRON	72	$>1.0\times10^{4}$	1.5×10^{3}	72A200	2.5×10^{2}	72A200	6.7	4.0×10^{1}	✓	✓	3.2×10^{-1}	1.9
COBALT	74	2.0×10^{1}	7.5×10^{2}	74A000	2.5×10^{2}	74A000	2.7×10^{-2}	8.0×10^{-2}			1.3×10^{-3}	3.8×10^{-3}
NICKEL	76	1.0×10^{3}	2.3×10^{2}	76A000	1.0×10^{1}	76A000	4.4	1.0×10^{2}	✓	✓	2.1×10^{-1}	4.7
COPPER	78	5.0×10^{5}	5.0×10^{3}	78A000	5.0×10^{1}	78A000	100.	1.0×10^{4}	✓	✓	4.7	4.7×10^{2}
SILVER	79	3.0×10^{1}	2.5×10^{2}	79A000	5.0	79A000	1.2×10^{-1}	6.0		✓	5.6×10^{-3}	2.8×10^{-1}
ZINC	81	$>1.0\times10^{4}$	2.5×10^{4}	81A000	1.0×10^{2}	81A000	$>4.0\times10^{-1}$	$>1.0\times10^{2}$		✓	$>1.9\times10^{-2}$	>4.7

CONTINUATION SHEET FOR WORKSHEET

LEVEL 1 X
LEVEL 2 ____

1. SOURCE/CONTROL OPTION RESIDENTIAL FURNACE M.A.N HADWICK

2. EFFLUENT STREAM

CODE NO. ____ NAME WASTE WATER (LIQUID STREAM)

A	B	C	D	E	F	G	H	I	J	K	L	M
SPECIES OR MEG CATEGORY NAME	MEG ID NUMBER OR CATEGORY	SPECIES OR CATEGORY CONCENTRATION	HEALTH DMEG CONCENTRATION	MEG ASSUMED — HEALTH	ECOLOGICAL DMEG CONCENTRATION	MEG ASSUMED — ECOLOGY	DISCHARGE SEVERITY — HEALTH (C÷D)	DISCHARGE SEVERITY — ECOLOGICAL (C÷F)	J IF HEALTH DMEG EXCEEDED	K IF ECOL DMEG EXCEEDED	WEIGHTED DISCHARGE SEVERITY	
											HEALTH BASED (H × LINE 2)	ECOLOGICAL BASED (I × LINE 2)
UNITS	—	ug/l	ug/l	—		—	—	—	—	—		
LANTHANUM	84	1.0×10^{1}	1.7×10^{6}	8HA010	--	--	5.9×10^{-6}	--			2.8×10^{-7}	--
CERIUM	84	8.0	5.5×10^{5}	8HA020	--	--	1.4×10^{-5}	--			6.6×10^{-7}	--

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

1. REPORT NO.	2.	3. RECIPIENT'S ACCESSION NO.
EPA-600/7-82-038b		

4. TITLE AND SUBTITLE	5. REPORT DATE
Environmental Assessment of a Low-emission oil-fired Residential Hot Water Condensing Heating System; Volume II. Data Supplement	May 1982
	6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S)	8. PERFORMING ORGANIZATION REPORT NO.
C. Castaldini	
9. PERFORMING ORGANIZATION NAME AND ADDRESS	10. PROGRAM ELEMENT NO.
Acurex Corporation 485 Clyde Avenue Mountain View, California 94042	11. CONTRACT/GRANT NO. 68-02-3188
12. SPONSORING AGENCY NAME AND ADDRESS	13. TYPE OF REPORT AND PERIOD COVERED
EPA, Office of Research and Development Industrial Environmental Research Laboratory Research Triangle Park, NC 27711	Final; 7/80-2/81 14. SPONSORING AGENCY CODE EPA/600/13

15. SUPPLEMENTARY NOTES IERL-RTP project officer is Robert E. Hall, Mail Drop 65, 919/541-2477. Volume I. gives technical results.

16. ABSTRACT The report gives results of a test program measuring air and water emissions from a high-efficiency hot-water residential heating system of European design, utilizing a condensing flue gas system and a low emission burner. Criteria and noncriteria emissions, including trace elements and organic species in both flue gas and condensate waste water streams, were measured. NO (as NO2), CO, total UHC (as propane), and total particulate emissions measured about 37, 12, 1.5, and 2.7 ng/J heat input, respectively. Absorption of sulfates and nitrates in the waste water resulted in a constant pH of 3.0. Total organic emissions in the flue gas measured 3.5 mg/dscm; they were below the detectable limit in the waste water. Several inorganic trace elements, including chromium, copper, iron, and nickel, in the waste water were attributed to leaching of heat transfer metal surfaces by the warm acidic water. Bioassays were also performed to evaluate the potential health hazard of the streams. Results indicate nondetectable to moderate toxicity and mutagenicity.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Pollution Assessments Hot Water Heating Residential Buildings Fuel Oil Flue Gases Waste Water Condensates Measurement	Pollution Control Stationary Sources Environmental Assessment	13B 14B 07D 13A 13M 21D 21B

18. DISTRIBUTION STATEMENT	19. SECURITY CLASS (This Report)	21. NO. OF PAGES
Release to Public	Unclassified	193
	20. SECURITY CLASS (This page) Unclassified	22. PRICE

EPA Form 2220-1 (9-73)