

# Enviro-Kleen®

## ENVIRONMENTAL DATA

VOC, SEMI-VOLATILES, METALS, TCLP, PAH TESTS

**PERFORMED BY:** TSL, Tri-State Laboratories

**REPORT DATA:** July 15, 2002

**SUMMARY:** As part of the US EPA Environmental Technology Verification (ETV) Program Enviro-Kleen® was tested to determine major, minor and trace constituents using various EPA test methods.

Bulk analysis techniques were used to quantitatively determine the presence of Title 22 metals, Volatile Organic Compounds (VOC), Semi-volatiles, pesticides herbicides and Polynuclear Aromatic Hydrocarbons (PAH) in Enviro-Kleen®. Bulk analysis is performed on the sample in the "as received" form and does not consider application rates, dilution ratios or environmental conditions. The vast majority of the analytes were found to be Below Detection Limits (BDL). Ever evolving sophistication of analytical methods and techniques have made detection limits well below regulatory levels. Some metals were detected at low levels, primarily iron and that can be attributed to the use of carbon steel tanks in transportation and storage.

Toxicity Characteristic Leaching Procedure (TCLP) is a sample preparation and battery of tests that can determine the presence of various elements and chemical compounds in a landfill type situation. In this test Enviro-Kleen® was subjected to chemical extractions to "leach" the analytes from the product. This includes metals, volatiles and semivolatiles analysis. All analytes were determined to be "below detection limits".

**RESULTS:** Results indicate that Enviro-Kleen® contains no bulk analysis or TCLP elements or compounds above regulatory levels. Most materials were not detected in Enviro-Kleen®. Please see attached for results.

# TSL Tri-State Laboratories, Inc.

2870 Salt Springs Road • Youngstown, Ohio 44509

Ph: (330) 797-8844 • Fax: (330) 797-3264 • 1-800-523-0347

E-mail: trislabs@aol.com

## Laboratory Analysis Report

<b>Client:</b> RTI	<b>Lab Number:</b> 22061406
Attn: DEBBIE FRANKE	<b>Sample ID:</b> B-MIDWEST KANSAS CITY
PO BOX 12194	
RESEARCH TRIANGLE PARK, NC 27709	
<b>Date Sampled:</b>	<b>Sample Description:</b>
<b>Time Sampled:</b>	<b>Sampler Name:</b>
<b>Date Received:</b> 6/14/2002	<b>Sample Matrix:</b> Aqueous
<b>Report Date:</b> 7/15/2002	<b>PO#:</b> 19820
<b>Comments:</b>	

Analyte	Result	Unit	Detection Limit	Method	Analysis Date	Analyst
Aluminum	1.0	mg/kg	0.47	200.7	6/19/2002	SCB
Antimony	BDL	mg/kg	0.047	200.7	6/19/2002	SCB
Arsenic	BDL	mg/kg	0.14	200.7	6/19/2002	SCB
Arsenic-TCLP	BDL	mg/L	0.10	6010B	6/19/2002	SCB
BariumTCLP	BDL	mg/L	0.040	6010B	6/19/2002	SCB
Barium	BDL	mg/kg	0.047	200.7	6/19/2002	SCB
Beryllium	BDL	mg/kg	0.0074	200.7	6/19/2002	SCB
Cadmium	BDL	mg/L	0.020	6010B	6/19/2002	SCB
Cadmium	BDL	mg/kg	0.023	200.7	6/19/2002	SCB
Chromium	BDL	mg/kg	0.023	200.7	6/19/2002	SCB
Chromium-TCLP	BDL	mg/L	0.020	6010B	6/19/2002	SCB
Copper	BDL	mg/kg	0.023	200.7	6/19/2002	SCB
Iron	25.0	mg/kg	0.50	200.7	6/19/2002	SCB
Lead-TCLP	BDL	mg/L	0.10	6010B	6/19/2002	SCB
Lead	BDL	mg/kg	0.12	200.7	6/19/2002	SCB
Manganese	0.12	mg/kg	0.047	200.7	6/19/2002	SCB
Mercury-TCLP	BDL	mg/L	0.001	7472	6/21/2002	SCB
Mercury	BDL	mg/kg	0.0012	245.2	6/21/2002	SCB
Nickel	BDL	mg/kg	0.047	200.7	6/19/2002	SCB
Selenium-TCLP	BDL	mg/L	0.16	6010B	6/19/2002	SCB
Selenium	BDL	mg/kg	0.19	200.7	6/19/2002	SCB
Silver	BDL	mg/kg	0.023	200.7	6/19/2002	SCB
Silver-TCLP	BDL	mg/L	0.020	6010B	6/19/2002	SCB
Thallium	BDL	mg/kg	0.12	200.7	6/19/2002	SCB
Zinc	0.137	mg/kg	0.047	200.7	6/19/2002	SCB
Herbicides	SEE ATTACHED			8270	6/19/2002	JP
Pesticides	SEE ATTACHED			8270	6/19/2002	JP
Polynuclear Aromatic Hydrocarbons	SEE ATTACHED			8270/610	6/19/2002	JP
Semi-Volatile Organic Compounds	SEE ATTACHED			8270A/625	6/19/2002	JP
TCLP-Semi-Volatiles	SEE ATTACHED			1311/8270	6/19/2002	JP
TCLP-Volatiles (VOC)	SEE ATTACHED			1311/8260	6/17/2002	JP
Volatile Organic Compounds (VOC)	SEE ATTACHED			8260/624	6/17/2002	JP

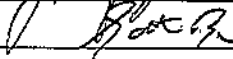
BDL = Below Detection Limit

Results approved by:

John Pflugh, Lab Manager

  
\_\_\_\_\_

Scott Bolam, QA/QC Officer

  
\_\_\_\_\_

**TRI-STATE LABORATORIES**

2870 Salt Springs Road  
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Fax: (330) 797-3264

Client: RTI

Date Received: 06.14.02

Sample: 22061406

Date Analyzed: 06.19.02

Sample Description: B

Date Reported: 07.15.02

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**HERBICIDES**

Method #: 8270

COMPOUND	CONCENTRATION (mg/L)	MDL (mg/L)
2,4-D	BDL	0.145
Silvex	BDL	0.145
<b>Surrogates</b>	<b>Recovery</b>	<b>Accept. Limits</b>
DCAA	86	35-114

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**PESTICIDES**

Method #: 8270

COMPOUND	CONCENTRATION (mg/L)	MDL (mg/L)
TECHNICAL CHLORDANE	BDL	0.008
ENDRIN	BDL	0.003
HEPTACHLOR	BDL	0.003
LINDANE	BDL	0.003
METHOXYCHLOR	BDL	0.033
TOXAPHENE	BDL	0.073
<b>Surrogates</b>	<b>Recovery</b>	<b>Accept. Limits</b>
TCMX	80	35-114
DBCP	92	43-116

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Client: RTI

Date Received: 06/14/02

Sample: 22061406

Date Analyzed: 06/19/02

Sample Description: B

Date Reported: 06/28/02

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**POLYNUCLEAR AROMATIC HYDROCARBONS**

Method #: 8270

COMPOUND	CONCENTRATION (mg/kg)	MDL(mg/kg)
Acenaphthene	BDL	50
Acenaphthylene	BDL	50
Anthracene	BDL	50
Benzo [a] anthracene	BDL	50
Benzo [a] pyrene	BDL	50
Benzo [b] fluoranthene	BDL	50
Benzo [k] fluoranthene	BDL	50
Benzo [g,h,l] perylene	BDL	50
Chrysene	BDL	50
Dibenzo [a,h] anthracene	BDL	50
Fluoranthene	BDL	50
Fluorene	BDL	50
Indeno (1,2,3-cd) pyrene	BDL	50
Naphthalene	BDL	50
Phenanthrene	BDL	50
Pyrene	BDL	50

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Surrogates	Recovery	Accept.Limits
Nitrobenzene-d5	67	23-123
2-Fluorobiphenyl	66	30-107
p-Terphenyl	87	18-129

BDL = below detection limit  
MDL = method detection limit

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Date Received: 06/14/02

Sample: 22061406

Date Analyzed: 06/19/02

Sample Description: B

Date Reported: 06/28/02

## BASE/NEUTRAL & ACID COMPOUNDS: PRIORITY POLLUTANTS

Method #: EPA 8270

COMPOUND	CONCENTRATION (mg/kg)	MDL (mg/kg)
Acenaphthene	BDL	50
Acenaphthylene	BDL	50
Anthracene	BDL	50
Benzidine	BDL	500
Benzo [a] anthracene	BDL	50
Benzo [a] pyrene	BDL	50
3,4-Benzofluoranthene	BDL	50
Benzo (g,h,i) perylene	BDL	50
Benzo (b) fluoranthene	BDL	50
Benzo (k) fluoranthene	BDL	50
Bis (2-chloroethoxy) methane	BDL	50
Bis (2-chloroethyl) ether	BDL	50
Bis (2-chloroisopropyl) ether	BDL	50
Bis (2-ethylhexyl) phthalate	BDL	50
4-Bromophenyl phenyl ether	BDL	50
Butyl benzyl phthalate	BDL	50
Carbazole	BDL	50
2-Chloronaphthalene	BDL	50
4-Chlorophenyl phenyl ether	BDL	50
Chrysene	BDL	50
Dibenzo [a,h] anthracene	BDL	50
1,2-Dichlorobenzene	BDL	50
1,3-Dichlorobenzene	BDL	50
1,4-Dichlorobenzene	BDL	50
3,3'-Dichlorobenzidine	BDL	500
Diethyl phthalate	BDL	50
Dimethyl phthalate	BDL	50
Di-n-octyl phthalate	BDL	50
2,4-Dinitrotoluene	BDL	50
2,6-Dinitrotoluene	BDL	50
Di-n-octyl phthalate	BDL	50
1,2-Diphenylhydrazine (as azobenzene)	BDL	50

BDL = below detection limit

MDL = method detection limit

Client: RTI

Sample: 22061406

COMPOUND	CONCENTRATION (mg/kg)	MDL (mg/kg)
Fluoranthene	BDL	50
Fluorene	BDL	50
Hexachlorobenzene	BDL	50
Hexachlorobutadiene	BDL	50
Hexachlorocyclopentadiene	BDL	50
Hexachloroethane	BDL	50
Indeno (1,2,3-cd) pyrene	BDL	50
Isophorone	BDL	50
Naphthalene	BDL	50
Nitrobenzene	BDL	50
N-Nitrosodimethylamine (as diphenylamine)	BDL	50
N-Nitrosodi-n-propylamine	BDL	50
N-Nitrosodiphenylamine	BDL	50
Phenanthrene	BDL	50
Pyrene	BDL	50
1,2,4-Trichlorobenzene	BDL	50
2-Chlorophenol	BDL	100
2,4-Dichlorophenol	BDL	100
2,4-Dimethylphenol	BDL	100
4,6-Dinitro-o-cresol	BDL	100
2,4-Dinitrophenol	BDL	100
2-Methyl phenol	BDL	100
3&4-Methyl phenol	BDL	100
2-Nitrophenol	BDL	100
4-Nitrophenol	BDL	100
Pentachlorophenol	BDL	100
Phenol	BDL	100
2,4,5-Trichlorophenol	BDL	100
2,4,6-Trichlorophenol	BDL	100
4-Chloro-3-Methyl Phenol	BDL	100
Benzoic Acid	BDL	100
2,3,7,8-tetrachloro-dibenzo-p-dioxin	ABSENT	

Surrogates	Recovery	Accept.Limits
Nitrobenzen-d5	67	35-114
2-Fluorobiphenyl	66	43-116
p-Terphenyl	87	33-141
Phenol-d6	92	11-94
2-Fluorophenol	80	25-100
2,4,6- Tribromophenol	58	16-123

BDL = below detection limit

MDL = method detection limit



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Client: RTI

Date Received: 06.14.02

Sample: 22061406

Date Analyzed: 06.19.02

Sample Description: B

Date Reported: 06.28.02

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**TCLP SEMI-VOLATILES - GC/MS**

Method #: 1311/8270

COMPOUND	CONCENTRATION (mg/L)	MDL (mg/L)
Cresols	BDL	0.575
1,4-Dichlorobenzene	BDL	0.112
2,4-Dinitrotoluene	BDL	0.112
Hexachlorobenzene	BDL	0.112
Hexachloro-1,3-butadiene	BDL	0.112
Hexachloroethane	BDL	0.112
Nitrobenzene	BDL	0.112
Pentachlorophenol	BDL	0.575
Pyridine	BDL	0.288
2,4,5-Trichlorophenol	BDL	0.575
2,4,6-Trichlorophenol	BDL	0.575

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Surrogates	Recovery	Accept.Limits
Nitrobenzene-d5	78	35-114
2-Fluorobiphenyl	77	43-116
p-Terphenyl	100	33-141
Phenol-d6	93	25-100
2-Fluorophenol	79	11-94
2,4,6-Tribromophenol	68	16-123

BDL = below detection limits  
MDL = method detection limit  
GC/MS = gas chromatography/mass  
spectrometry

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Client: RTI

Date Received: 06.14.02

Sample: 22061406

Date Analyzed: 06.17.02

Sample Description: B

Date Reported: 06.28.02

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**TCLP VOLATILES - GC/MS**

Method #: 1311/8260

COMPOUND	CONCENTRATION (mg/L)	MDL (mg/L)
Benzene	BDL	2.14
Carbon Tetrachloride	BDL	2.14
Chlorobenzene	BDL	2.14
Chloroform	BDL	2.14
1,2-Dichloroethane	BDL	2.14
1,1-Dichloroethene	BDL	2.14
Methyl ethyl ketone (2-Butanone)	BDL	2.14
Tetrachloroethene	BDL	2.14
Trichloroethene	BDL	2.14
Vinyl Chloride	BDL	4.28

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Surrogates	Recovery	Accept. Limits
Dibromofluorobenzene	114	86-118
Toluene-d8	94	88-110
Bromofluorobenzene	99	86-115

BDL = below detection limit

MDL = method detection limit

GC/MS = gas chromatography/mass spectrometry

# TRI-STATE LABORATORIES

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Youngstown, OH 44509  
Phone: (330) 797-8844/1-800-523-0347  
Fax: (330) 797-3264

Client: RTI

Date Received: 06.14.02

Sample: 22061406

Date Analyzed: 06.14.02

Sample Description: B

Date Reported: 06.22.02

## WATER Method #: 8260

COMPOUND	CONCENTRATION	MDL (mg/kg)
Acetone	BDL	25
Benzene	BDL	2.5
Bromobenzene	BDL	2.5
Bromochloromethane	BDL	2.5
Bromodichloromethane	BDL	2.5
Bromoform	BDL	2.5
Bromomethane	BDL	2.5
2-Butanone	BDL	5
n-Butylbenzene	BDL	25
sec-Butylbenzene	BDL	2.5
tert-Butylbenzene	BDL	2.5
Carbon Tetrachloride	BDL	2.5
Chlorobenzene	BDL	2.5
Chloroethane	BDL	2.5
Chloroform	BDL	5
Chloromethane	BDL	2.5
2-Chlorotoluene	BDL	5
4-Chlorotoluene	BDL	2.5
1,2-Dibromo-3-chloropropane	BDL	2.5
Dibromochloromethane	BDL	2.5
1,2-Dibromoethane	BDL	2.5
Dibromomethane	BDL	2.5
1,2-Dichlorobenzene	BDL	2.5
1,3-Dichlorobenzene	BDL	2.5
1,4-Dichlorobenzene	BDL	2.5
Dichlorodifluoromethane	BDL	2.5
1,1-Dichloroethane	BDL	5
1,2-Dichloroethane	BDL	2.5
1,1,-Dichloroethene	BDL	2.5
cis-1,2-Dichloroethene	BDL	2.5
trans-1,2-Dichloroethene	BDL	2.5
1,2-Dichloropropane	BDL	2.5
1,3-Dichloropropane	BDL	2.5
2,2-Dichloropropane	BDL	2.5
1,1-Dichloropropene	BDL	2.5
Ethyl Benzene	BDL	2.5
Hexachlorobutadiene	BDL	2.5

BDL = below detection limit  
MDL = method detection limit

COMPOUND	CONCENTRATION	MDL (mg/kg)
2-Hexanone	BDL	25
Isopropylbenzene	BDL	2.5
p-Isopropyltoluene	BDL	2.5
Methylene Chloride	BDL	2.5
Methyl Isobutyl Ketone	BDL	25
Naphthalene	BDL	2.5
n-Propylbenzene	BDL	2.5
Styrene	BDL	2.5
1,1,1,2-Tetrachloroethane	BDL	2.5
1,1,2,2-Tetrachloroethane	BDL	2.5
Tetrachloroethene	BDL	2.5
Toluene	BDL	2.5
1,2,3-Trichlorobenzene	BDL	2.5
1,2,4-Trichlorobenzene	BDL	2.5
1,1,1-Trichloroethane	BDL	2.5
1,1,2-Trichloroethane	BDL	2.5
Trichloroethene	BDL	2.5
Trichlorofluoromethane	BDL	5
1,2,3-Trichloropropane	BDL	2.5
1,2,4-Trimethylbenzene	BDL	2.5
1,3,5-Trimethylbenzene	BDL	2.5
Vinyl Chloride	BDL	5
m,p-Xylene	BDL	2.5
o-Xylene	BDL	2.5
<b>Surrogates</b>	<b>Recovery</b>	<b>Accept.Limits</b>
Dibromofluorobenzene	114	86-118
Toluene-d8	94	88-110
Bromofluorobenzene	99	86-115

BDL = below detection limit  
MDL = method detection limit

# EnviroKleen®

## ENVIRONMENTAL DATA

### Acute and Chronic Aquatic Toxicity

**PERFORMED BY:** ABC Laboratories, Inc.

**REPORT DATA:** September 16, 2002

**SUMMARY:** As part of the US EPA Environmental Technology Verification (ETV) Program EnviroKleen® (Liquid Sample B) was tested to determine chronic and acute toxicity to aquatic species: Ceriodaphnia dubia, Fathead minnow and Americamysis bahia (Mysid shrimp).

Concentrated EnviroKleen® was evaluated per the following EPA test methods:

\*Methods for Measuring the Acute Toxicity of Effluents and Receiving Water to Freshwater and Marine Organisms, EPA/600/4-90/027F.

\*Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, EPA/600/4-91/002.

\*Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Water to Marine and Estuarine Organisms, EPA/600/4-91/003.

**RESULTS:** The results indicate that EnviroKleen® has very low aquatic toxicity levels and is not considered an aquatic pollutant.

Ceriodaphnia dubia Acute, EC <sub>50</sub> :	>1000mg/L
Fathead minnow Acute, LC <sub>50</sub> :	>1000mg/L
Americamysis bahia Acute, EC <sub>50</sub> :	>1000mg/L
Ceriodaphnia dubia Chronic, EC <sub>50</sub> :	>1000mg/L
Fathead minnow Chronic, LC <sub>50</sub> :	>1000mg/L
Americamysis bahia Chronic, EC <sub>50</sub> :	>1000mg/L

## EPA Method 24 Analysis

ID#	Sample ID	ASTM D1475	ASTM D2369	ASTM D3792
		Density (g/mL)	Total Volatiles (Wt%)	Water (Wt%)
2	EnviroKleen	0.8473	10.57%	0.00%

NOTE: Each value is the average of two measurements.

\*Duplicate measurements did not meet criterion (analysis repeated four times).



Laboratories

7200 E. ABC Lane, Columbia, MO 65202 • Tel: 573/474-8579 Fax: 573/443-9033

September 17, 2002

RTI  
Attn: Debbie Frankie  
P.O. Box 12194  
Research Triangle Park, NC 27709

Dear Ms. Frankie:

Enclosed please find ABC Laboratories' summary report for aquatic toxicity testing on five dust suppression products (ABC study numbers 47551, 47552, 47553, 47554, 47555, and 47556).

If you have any other questions or need additional information pertaining to the enclosed materials, please call me direct at 573-876-8187, by fax at 573-443-9089, or by E-mail at [warbritonr@abclabs.com](mailto:warbritonr@abclabs.com). For additional information regarding ABC Laboratories, please visit our web site at [www.abclabs.com](http://www.abclabs.com). We appreciate your consideration of ABC for this testing and look forward to a continued association with you in the future.

Sincerely,

A handwritten signature in black ink, appearing to read 'Ryan Warbriton', written over a horizontal line.

Ryan Warbriton  
Scientist/Manager, Environmental Toxicology  
Chemical Development Group

Enclosure

**STUDY TITLE**

Acute and Chronic Toxicity of Dust Suppression Products A, B, E, Perma-Zyme 11X,  
and Soil Sement Engineered Formula to *Ceriodaphnia dubia*, Fathead Minnow  
(*Pimephales promelas*), and *Americamysis bahia*

**SPONSOR**

RTI International  
3040 Cornwallis Road  
Research Triangle Park, NC 27709-2194

**AUTHORS**

Ryan Warbritton  
Scientist

**REPORT COMPLETED ON**

September 16, 2002

**PERFORMING LABORATORY**

ABC Laboratories, Inc.  
Chemical Development Group  
7200 E. ABC Lane  
Columbia, MO 65202

**PROJECT ID**


47551 *Ceriodaphnia dubia* Acute  
47552 Fathead Minnow Acute  
47553 *Americamysis bahia* Acute  
47554 *Ceriodaphnia dubia* Chronic  
47555 Fathead Minnow Chronic  
47556 *Americamysis bahia* Chronic



**SIGNATURE PAGE**

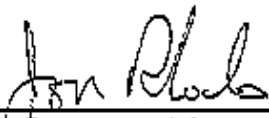
Submitted by: ABC Laboratories, Inc.  
Chemical Development Group  
7200 E. ABC Lane  
Columbia, MO 65202

Prepared by:

  
\_\_\_\_\_  
Ryan Warbritton  
Scientist/Manager, Environmental Toxicology  
Chemical Development Group

*Sept 16, 02*  
\_\_\_\_\_  
Date

Reviewed by:

  
\_\_\_\_\_  
Jon Rhodes, M.S.  
Senior Research Scientist/Biological Services Director  
Chemical Development Group

*16 Sep 02*  
\_\_\_\_\_  
Date

**ACUTE TOXICITY COMPENDIUM**

**Subject:** Acute and Chronic Toxicity of EnviroKleen Formula to *Ceriodaphnia dubia*, Fathead Minnow (*Pimephales promelas*), and *Americamysis bahia*

**Sponsor:** RTI International  
3040 Cornwallis Road  
Research Triangle Park, NC 27709-2194

**Test Substances:** EnviroKleen

**Definitive Test Concentrations:**

**47551 *Ceriodaphnia dubia* Acute:**

EnviroKleen                      Control and 1,000 mg/L

**47552 Fathead Minnow Acute:**

EnviroKleen                      Control, 62.5, 125, 250, 500, and 1,000 mg/L

**47553 *Americamysis bahia* Acute:**

EnviroKleen                      Control and 1,000 mg/L

**47554 *Ceriodaphnia dubia* Chronic:**

EnviroKleen                      Control, 250, 500, and 1,000 mg/L

47555 Fathead Minnow Chronic

EnviroKleen                      Control, 62.5, 125, 250, 500, and 1,000 mg/L

47556 Americamysis bahia Chronic:

EnviroKleen                      Control, 250, 500, and 1,000 mg/L

**Solution Preparation:**

EnviroKleen                      WAF (Water Accommodated Fraction) Stirred overnight

**Definitive Test Dates:**

47551 Ceriodaphnia dubia Acute:

EnviroKleen                      August 20 to 22, 2002

47552 Fathead Minnow Acute:

EnviroKleen                      August 14 to 21, 2002

47553 Americamysis bahia Acute:

EnviroKleen                      August 22 to 26, 2002

47554 *Ceriodaphnia dubia* Chronic:

EnviroKleen                      August 21 to 28, 2002

47555 Fathead Minnow Chronic:

EnviroKleen                      August 14 to 21, 2002

47556 *Americamysis bahia* Chronic:

EnviroKleen                      August 29 to September 05, 2002

**Duration of Test:**

47551 *Ceriodaphnia dubia* Acute:      48 hours  
47552 Fathead Minnow Acute:          96 hours  
47553 *Americamysis bahia* Acute:      96 hours  
47554 *Ceriodaphnia dubia* Chronic:    7 days  
47555 Fathead Minnow Chronic:        7 days  
47556 *Americamysis bahia* Chronic:    7 days

**Organism Source:**

47551 *Ceriodaphnia dubia* Acute:      In-house cultures  
47552 Fathead Minnow Acute:          In-house cultures  
47553 *Americamysis bahia* Acute:      In-house cultures  
47554 *Ceriodaphnia dubia* Chronic:    In-house cultures  
47555 Fathead Minnow Chronic:        In-house cultures  
47556 *Americamysis bahia* Chronic:    In-house cultures

**Age at Initiation:**

47551 *Ceriodaphnia dubia* Acute:      <24 hours for all samples  
47552 Fathead Minnow Acute:          7 days post hatch for all samples  
47553 *Americamysis bahia* Acute:      <24 hours for all samples  
47554 *Ceriodaphnia dubia* Chronic:    <24 hours for all samples  
47555 Fathead Minnow Chronic:        <24 hours post hatch for all samples  
47556 *Americamysis bahia* Chronic:    7 days old for all samples

## Test Procedures and Conditions:

### 47551 *Ceriodaphnia dubia* Acute:

Duration:	48 hours
Temperature:	25 ± 1°C
Lighting:	Ambient laboratory lighting, 16:8-hr light:dark
Observations:	24 and 48 hours
Test chambers:	50-mL glass containers
Volume per chamber:	25 mL
Replicates per treatment:	4
Organisms per chamber:	5
Organisms per treatment:	20
Dilution water:	Age blended freshwater
Solution renewal:	None
Aeration:	None

### 47552 Fathead minnow Acute:

Duration:	96 hours
Temperature:	25 ± 1°C
Lighting:	Ambient laboratory lighting, 16:8-hr light:dark
Observations:	24, 48, 72, and 96 hours
Test chambers:	900-mL glass containers
Volume per chamber:	400 mL
Replicates per treatment:	
EnviroKleen	2
Organisms per chamber:	10
Organisms per treatment:	
EnviroKleen	20
Dilution water:	Blended freshwater
Solution renewal:	None
Aeration:	None

### 47553 *Americamysis bahia* Acute:

Duration:	96 hours
Temperature:	26 ± 1°C
Lighting:	Ambient laboratory lighting, 16:8-hr light:dark
Observations:	24, 48, 72, and 96 hours
Test chambers:	900-mL glass containers

Volume per chamber: 400 L  
Replicates per treatment:

EnviroKleen 3

Organisms per chamber: 10  
Organisms per treatment:

EnviroKleen 30

Dilution water: Saltwater  
Solution renewal: None  
Aeration: None

47554 *Ceriodaphnia dubia* Chronic:

Duration: 7 Days  
Temperature: 25 ± 1°C  
Lighting: Ambient laboratory lighting, 16:8-hr light:dark  
Observations: Daily  
Test chambers: 50-mL glass containers  
Volume per chamber: 30 mL  
Replicates per treatment: 10  
Organisms per chamber: 1  
Organisms per treatment: 10  
Dilution water: Age blended freshwater  
Solution renewal: Daily  
Aeration: None

47555 Fathead minnow Chronic:

Duration: 7 Days  
Temperature: 25 ± 1°C  
Lighting: Ambient laboratory lighting, 16:8-hr light:dark  
Observations: Daily  
Test chambers: 900-mL glass containers  
Volume per chamber: 400 L  
Replicates per treatment: 3  
Organisms per chamber: 10  
Organisms per treatment: 30  
Dilution water: Aged blended freshwater  
Solution renewal: Daily  
Aeration: None

47556 *Americamysis bahia* Chronic:

Duration:	7 Days
Temperature:	27 ± 1°C
Lighting:	Ambient laboratory lighting, 16:8-hr light:dark
Observations:	Daily
Test chambers:	1,000-mL glass beakers
Volume per chamber:	200 mL
Replicates per treatment:	8
Organisms per chamber:	5
Organisms per treatment:	40
Dilution water:	Saltwater
Solution renewal:	Daily
Aeration:	None

**Methods:**

Weber, C.I. 1993. Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms, 4th ed. U.S. Environmental Protection Agency, EPA/600/4-90/027F, 293 p.

U.S. EPA. 1994. Short-Term Methods For Estimating The Chronic Toxicity Of Effluents And Receiving Water To Freshwater Organisms, Third Edition. EPA/600/4-91/002. 341 p.

U.S. EPA. 1994. Short-Term Methods For Estimating The Chronic Toxicity Of Effluents And Receiving Water To Marine and Estuarine Organisms, Second Edition. EPA/600/4-91/003. 483 p.

Toxcalc™ Version 5.0, Release 6.12. Copyright 1994 by Tidepool Scientific Software: Michael A. Ives.

Results:

I. Acute Test Mortality

47551 – <i>Ceriodaphnia dubia</i> Acute Test -- Percent Mortality					
Concentration		EnviroKleen			
Control		0			
L1		0			
L2		–			
L3		–			
L4		–			
L5		–			
L6		–			

Note:  
EnviroKleen concentrations were control, and 1,000 mg/L.



47552 – Fathead Minnow Acute Test –Percent Mortality

Concentration		EnviroKleen			
Control		0			
L1		0			
L2		0			
L3		0			
L4		0			
L5		0			
L6		0			

Note:

EnviroKleen concentrations were control, 62.5, 125, 250, 500, and 1,000 mg/L.

47553 – *Americamysis bahia* Acute Test –Percent Mortality

Concentration	EnviroKleen			
Control	0			
L1	0			
L2	-			
L3	-			
L4	--			
L5	--			

Note:

EnviroKleen concentrations were control and 1,000 mg/L.

47554 -- *Ceriodaphnia dubia* Chronic Test -- Mortality and Reproduction  
 EnviroKleen

Concentration (mg/L)	Percent Mortality at 7 Days	Total Reproduction	Mean Reproduction per Adult
Control	0	706	47.1
250	0	440	44.0
500	10	436	43.6
1,000	10	465	46.5

<sup>A</sup> Control replicated 15 times all other concentrations replicated 10 times.

<sup>B</sup> Based upon original number of 15 for control and 10 for other concentrations.

47555 -- Fathead minnow Chronic Test -- Mortality and Growth  
EnviroKleen

Concentration (mg/L)	Percent Mortality at 7 Days	Mean Weight per Fish (mg)
Control	3	1.062
62.5	10	0.9556
125	10	1.085
250	3	1.020
500	3	0.9448
1,000	3	0.9207

47556 – *Americamysis bahia* Chronic Test – Mortality and Reproduction  
 EnviroKleen

Concentration (mg/L)	Percent Mortality at 7 Days	No. Females at 7-days	Percent Females with Eggs	Mean Dry Weight per Adult (mg)
Control	15	6	100	0.562
250	15	4	100	0.587
500	8	12	100	0.512
1000	5	7	100	0.485

III. Acute Test Water Quality Ranges

47551 <i>Ceriodaphnia dubia</i> Acute Test – Water Quality Ranges			
Sample ID	Temperature (°C)	Dissolved Oxygen (mg/L)	pH
Control	24.0 – 25.0	7.1 – 8.5	8.19 – 8.66
EnviroKleen	24.8 – 25.0	8.2 – 8.5	8.39 – 8.67

47552 Fathead Minnow Acute Test – Water Quality Ranges			
Sample ID	Temperature (°C)	Dissolved Oxygen (mg/L)	pH
Control	24.0 – 25.0	6.5 – 8.6	7.96 – 8.60
EnviroKleen	24.5 – 25.0	7.3 – 8.6	8.43 – 8.62

47551 *Americamysis bahia* Acute Test- Water Quality Ranges

Sample ID	Temperature (°C)	Dissolved Oxygen (mg/L)	pH
Control	24.0 – 24.8	5.8 – 8.0	7.91 – 8.25
EnviroKleen	24.6 – 24.9	6.4 – 7.9	8.05 – 8.26

IV. Chronic Test Water Quality Ranges

47554 <i>Ceriodaphnia dubia</i> Chronic Test Old Solution Water Quality Ranges			
Sample ID	Temperature (°C)	Dissolved Oxygen (mg/L)	pH
Control	24.3 – 25.3	7.0 – 8.3	8.30 – 8.69
EnviroKleen	24.8 – 25.7	7.5 – 8.2	8.37 – 8.59



47555 Fathead Minnow Chronic Test Old Solution Water Quality Ranges			
Sample ID	Temperature (°C)	Dissolved Oxygen (mg/L)	pH
Control	24.3 – 24.9	5.5 – 6.73	8.02 – 8.55
EnviroKleen	24.6 – 25.0	4.4 – 7.06	7.90 – 8.54

47556 <i>Americamysis bahia</i> Chronic Test Old Solution Water Quality Ranges				
Sample ID	Temperature (°C)	Dissolved Oxygen (mg/L)	pH	Salinity
Control	23.7 – 26.0	4.6 – 7.5	7.68 – 8.23	19.1 – 21.8
EnviroKleen	24.7 – 25.6	4.5 – 6.3	7.62 – 8.22	19.1 – 22.1

**Statistical Analysis:**

I. Acute Tests

47551 <i>Ceriodaphnia dubia</i> Acute Test			
Statistical Analysis For Survival (mg/L)			
Sample ID	48 Hour EC <sub>50</sub> (95% CI)	NOEC	LOEC
EnviroKleen	>1,000	1,000	>1,000

47552 Fathead Minnow Acute Test			
Statistical Analysis For Survival (mg/L)			
Sample ID	96 Hour LC <sub>50</sub> (95% CI)	NOEC	LOEC
EnviroKleen	>1,000	1,000	>1,000

47553 *Americamysis bahia* Acute Test  
 Statistical Analysis For Survival (mg/L)

Sample ID	48 Hour EC <sub>50</sub> (95% CI)	NOEC	LOEC
EnviroKleen	>1,000	1,000	>1,000

II. Chronic Tests

47554 <i>Ceriodaphnia dubia</i> Chronic Test Statistical Analysis (mg/L)						
Sample ID	7-Day Survival			7-Day Reproduction		
	EC <sub>50</sub> (95% CI)	NOEC	LOEC	EC <sub>50</sub> (95% CI)	NOEC	LOEC
Enviro-Kleen	>1,000	1,000	>1,000	>1,000	1,000	>1,000

47555 Fathead Minnow Chronic Test – Statistical Analysis (mg/L)

Sample ID	7-Day Survival			7-Day Growth		
	EC <sub>50</sub> (95% CI)	NOEC	LOEC	EC <sub>50</sub> (95% CI)	NOEC	LOEC
Enviro-Kleen	>1,000	1,000	>1,000	>1,000	1,000	>1,000

**47556 *Americamysis bahia* Chronic Test  
Statistical Analysis (mg/L)**

Sample ID	7-Day Survival			7-Day Dry Weight			7-Day Fecundity		
	EC <sub>50</sub> (95% CI)	NOEC	LOEC	EC <sub>50</sub> (95% CI)	NOEC	LOEC	EC <sub>50</sub> (95% CI)	NOEC	LOEC
EnviroKleen	>1,000	1,000	>1,000	>1,000	1,000	>1,000	>1,000	1,000	>1,000

## Discussion:

### Solution Preparation:

Solutions were prepared on a weight to volume basis for all compounds. Liquid samples A and B were not water-soluble and were conducted as the water accommodated fraction (WAF). Liquid sample A was weighed out on large glass microscope slides and suspended in a beakers of water containing a Teflon stir bar. The beakers were placed on a stir plate and stirred overnight. Solutions were drawn off by siphoning the solutions into another glass container leaving a small amount of solution in the beaker. The remaining solution contained undissolved test compound that was floating on the surface of the water in the beaker. This undissolved test compound was not included in solutions for two reasons. One, so that it would not cause a decrease in dissolved oxygen transfer by covering the surface of the test vessels, and two, so that it would not cause secondary toxicity from impairment of the animals respiratory system in the case of the fathead minnows gills or cause any impairment in the appendages of the *Ceriodaphnia dubia* or *Americamysis bahia*. The Liquid B sample was weighed out into 20mL glass vials and mixed directly into beakers to stir overnight. Liquid sample E, Perma zyme 11X, and Soil Sement samples were prepared as required and not stirred overnight.

### Test Design:

Where preliminary testing indicated no mortality at concentrations of 1,000 mg/L, abbreviated or limit studies were performed. Acute studies run as limit tests were conducted with a control and a single concentration at 1,000 mg/L. Chronic studies were conducted with a control and three levels of 250; 500, and 1,000 mg/L. All other studies were conducted with five or six test levels and a control.

### Statistical Analysis:

Statistical analysis of the concentration versus effect data was performed using a custom computer program, ToxCalc. This program is designed to calculate the LC<sub>50</sub>/EC<sub>50</sub> statistic and its 95% confidence interval (CI), where possible, using the appropriate EPA recommended analysis. Statistical significance of comparison of means for *Ceriodaphnia dubia*, fathead minnow, and *Americamysis bahia* survival and reproduction, growth, and/or fecundity was determined by hypothesis testing using EPA recommended methods, typically either Fisher's Exact test or Dunnett's test. Point estimates testing to calculate the LC<sub>50</sub> or EC<sub>50</sub> was determined with the Trimmed Spearman-Kärber method where possible.

Generally, the statistical approach was as follows. Analysis of each endpoint between samples was evaluated by first analyzing the data for normality and homogeneity of variance with Shapiro-Wilk's Test and Kolmogorov D's Test before comparison of means. If the data were normally distributed and the variances were homogeneous, then analysis of variance (ANOVA) was utilized for the weight data along with Fisher's Exact Test or Dunnett's procedure for comparing the means. Survival data were analyzed using Fisher's Exact test and growth or reproduction were analyzed using Dunnett's. If the assumptions of

normality or homogeneity of variance were not met, transformations of the survival data were employed to allow the use of parametric procedures. If transformations (e.g., arcsine-square root transformation) of the survival data still did not meet assumptions of normality and homogeneity, then the non-parametric test, Steel's Many-One Rank Test, was used to analyze these data.

47551 *Ceriodaphnia dubia* Acute Tests:

EnviroKleen

This test was conducted as a limit test with levels of control and 1,000 mg/L. Mortality was 0% in both the control and the 1,000-mg/L concentrations. The 48 hour EC<sub>50</sub> for survival was >1,000 mg/L. The no observed effect concentration, or NOEC, was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L.

47552 Fathead Minnow Acute Tests:

EnviroKleen

This test was conducted as a multi-concentration test with levels of control, 62.5, 125, 250, 500, and 1,000 mg/L. Mortality was 0% in the control. Mortality was also 0% in all test levels. The 96-hour LC<sub>50</sub> for survival was >1,000 mg/L. The no observed effect concentration, or NOEC was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L.

47553 *Americamysis bahia* Acute Tests:

EnviroKleen

This test was conducted as a limit test with levels of control and 1,000 mg/L. Mortality was 5% in the control and 0% in the 1,000 mg/L concentration. The 96 hour EC<sub>50</sub> for survival was >1,000 mg/L. The no observed effect concentration, or NOEC, was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L.

47554 *Ceriodaphnia dubia* Chronic Tests:

EnviroKleen

This test was conducted as a limit test with levels of control, 250, 500, and 1,000 mg/L. Mortality was 0% in the control and all test levels. The 7-day EC<sub>50</sub> for survival was >1,000 mg/L. For survival the no observed effect concentration, or NOEC, was 1,00 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L. The 7-day EC<sub>50</sub> for reproduction was >1,000 mg/L. For reproduction the no observed effect concentration, or NOEC, was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L.



#### 47552 Fathead Minnow Chronic Tests:

##### Liquid Sample B:

This test was conducted as a multi-concentration test with levels of control, 62.5, 125, 250, 500, and 1,000 mg/L. Mortality was 3% in the control. Mortality was 13, 10, 3, 3, and 3% in the 62.5, 125, 250, 500, and 1,000 mg/L test levels respectively. The 7-day LC<sub>50</sub> for survival was >1,000 mg/L. For survival the no observed effect concentration, or NOEC was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L. The 7-day LC<sub>50</sub> for growth was >1,000 mg/L. For growth the no observed effect concentration, or NOEC, was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L.

#### 47553 *Americamysis bahia* Chronic Tests:

##### Liquid Sample B:

This test was conducted as a limit test with levels of control, 250, 500, and 1,000 mg/L. Mortality was 15% in the control. Mortality was 15, 7, and 5% in the 250, 500, and 1,000 mg/L test levels respectively. The 7-day LC<sub>50</sub> for survival was >1,000 mg/L. For survival the no observed effect concentration, or NOEC was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L. The 7-day LC<sub>50</sub> for growth was >1,000 mg/L. For growth the no observed effect concentration, or NOEC, was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L. The 7-day LC<sub>50</sub> for fecundity was >1,000 mg/L. For fecundity the no observed effect concentration, or NOEC, was 1,000 mg/L and the lowest observed effective concentration, or LOEC, was >1,000 mg/L.

## RAW DATA APPENDICES

- I. STUDY 47551 *Ceriodaphnia dubia* ACUTE TEST DATA
- II. STUDY 47552 FATHEAD MINNOW ACUTE TEST DATA
- III. STUDY 47553 *Americamysis bahia* ACUTE TEST DATA
- IV. STUDY 47554 *Ceriodaphnia dubia* CHRONIC TEST DATA
- V. STUDY 47555 FATHEAD MINNOW CHRONIC TEST DATA
- VI. STUDY 47556 *Americamysis bahia* CHRONIC TEST DATA

NOTE: Some of the records that appear in the raw data appendix have been provided as photocopies of original records on file at ABC. This has been done by necessity for certain data commonly used in several studies at ABC. Such records include organism culture logs and blended water chemical screen results.

The following symbols may appear on the raw data pages included in this report:

C - Calculation error

S - Spelling error

D - Date error

T - Transcription error

F - Form change

W - Write over

R - Recording error



United States Department of Agriculture  
Animal and Plant Health Inspection Service

**INSPECTION REPORT**

JANESMANN  
1596 cust\_id  
000000 site\_id  
1000 site\_id

**A B C LABORATORIES INC.**

Customer ID: 1596

Certificate: 43-R-0108

Site: 001

ABC LABORATORIES

7200 E ABC LN  
COLUMBIA, MO 65202

Inspection

Type: ROUTINE INSPECTION

Date: SEP-17-2002

This inspection was conducted by Noel Premkumar, director, Jessica Ambroz, IACUC chair, and Dr. Jami Niemann, VMO.

NO NON-COMPLIANT ITEMS IDENTIFIED THIS INSPECTION.

**COPY**

Prepared By:

Jamilon Niemann, DVM  
JAMILON NIEMANN, USDA, APHIS, Animal Care

Title:

VETERINARY MEDICAL OFFICER, Inspector ID: 4054

Date:

SEP-17-2002

Received By:

Noel Premkumar  
NOEL PREMUMAR

Date:

SEP-17-2002

Title: DIRECTOR



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY  
 WASHINGTON, D.C. 20460

MAR 5 2001

OFFICE OF  
 ENFORCEMENT AND  
 COMPLIANCE ASSURANCE

Mr. Eric Lorenz  
 Manager, Program Development  
 ABC Laboratories, Inc.  
 7200 East ABC Lane  
 Columbia, MO 65202

**COPY**

Dear Mr. Lorenz:

This is to inform you that a Good Laboratory Practice (GLP) inspection and data audit was conducted by the United States Environmental Protection Agency on February 27 - March 2, 2001; April 9 - 13, 1996; May 12 - 16, 1992; September 18 - 22, 1990 and February 14 - 18, 1989 at your facility named below:

ABC Laboratories, Inc.  
 (Ecotoxicity, Residue Chemistry, Environmental Fate)  
 7200 East ABC Lane  
 Columbia, MO 65202

Based on information presently available to the Agency, the GLP inspection and data audit conducted at that time did not reveal anything, such as significant departures from the GLP regulations, that would prevent EPA at this time from considering as reliable for the purposes of supporting a pesticide application for a research or marketing permit data developed at the above-named facility.

Sincerely,

Rick Colbert, Director  
 Agriculture Division

cc: