ACUTE TOXICITY TO FISH (FATHEAD MINNOW)

TEST SUBSTANCE

Identity: Perfluorooctanesulfonylamido(ethyl)acetate; may also be referred to as PFOSAA, FC-128 or as the major component of FC-127, FC-129, FC-109, FC-109-X or R1904. (Glycine, N-ethyl-N-[(heptadecafluorooctyl)sulfonyl]-, potassium salt, CAS # 2991-51-7)

Remarks: The test sample is FC-129 referred to by the test laboratory as R1904. Current information indicates it is a mixture of 42% test substance, 32% water, 14% 2-butoxyethanol, 4% ethanol, and 8% C_4-C_7 fluoroalkyl carboxylate compounds. Test material was from 3M production lot 930014, mfg. date 3/97.

The following summary is abbreviated because the test sample is a mixture of the test substance in a water/organic solvent solution. Data may not accurately relate toxicity of the test sample with that of the test substance.

METHOD:

Methods: OECD 203
Test type: Static acute
GLP: No
Year Completed: 1997
Species: *Pimephales promelas*
Analytical monitoring: Temperature, pH, DO, and conductivity

RESULTS

Nominal concentrations: Bk control, 19, 32, 54, 90, 150, 250 mg/L (in duplicate)

Element values:
24-hour LC_{50} = >250 mg/L (C.I. not calculable)
48-hour LC_{50} = 211 mg/L (C.I. not calculable)
72-hour LC_{50} = 146 (128-167) mg/L
96-hour LC_{10} = 45 (22-80) mg/L
96-hour LC_{50} = 97 (81-116) mg/L
96-hour NOEC = 54 mg/L

All element values based on nominal concentrations.

Remarks: Testing was conducted on the mixture of the test substance as described in the test substance remarks field. The value reported applies to that mixture and not the test substance. No attempt was made to

determine the impact of the presence of the organic solvents or what portion of the toxicity can be contributed to the Perfluorooctanesulfonylamido(ethyl)acetate.

CONCLUSIONS

The FC-129 96-hour LC_{50} for *Pimephales promelas* was determined to be 97 mg/L with a 95% confidence interval of 81-116 mg/L. The 96-hour no observed effect concentration was 54 mg/L.

Submitter: 3M Company, Environmental Laboratory, P.O. Box 33331, St. Paul, Minnesota, 55133

DATA QUALITY

Reliability: Klimisch ranking 2. Testing meets all criteria for quality testing, but lacks analytical confirmation of test substance concentrations. There is a lack of characterization of the test sample.

REFERENCES

This study was conducted at AScI Corporation of Duluth, Minnesota at the request of the 3M Company.

OTHER

Last changed: 5/17/00

STUDY TITLE

ACUTE TOXICITY OF R1904 TO
FATHEAD MINNOW (*Pimephales promelas*)

DATA STANDARD

OECD GUIDELINE 203

STUDY COMPLETED

June 27, 1997

TESTING FACILITY

AScI Corporation/AScI-Duluth
Environmental Testing Division
4444 Airpark Boulevard
Duluth, MN 55811

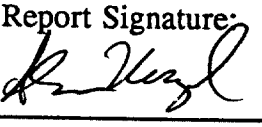
Tel. No. (218) 722-4040
Fax No. (218) 722-2592

STUDY IDENTIFICATION NUMBERS

AScI Study ID# 5010-068

3M Company Lab Request# R1904

1.0 GENERAL INFORMATION

Study Title	Acute Toxicity of R1904 to Fathead Minnow (*Pimephales promelas*).	
Data Standard	OECD Guideline 203.	
Sponsor	3M Environmental Technology and Safety Services, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55133; Tel No. (612) 778-7452.	
Sponsor's Representative	Susan Beach, 3M Environmental Technology and Safety Services, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55133; Tel No. (612) 778-7452.	
Sponsor's Lab Request#	R1904.	
Testing Facility	AScI Corporation/AScI-Duluth Environmental Testing Division, 4444 Airpark Boulevard, Duluth, MN 55811; Tel. No. (218) 722-4040.	
Principal Investigator	Joe Dierkes	
Project Director	Joe Amato	
Testing Facility Director	Donald Mount	
Definitive Test Dates	June 17-21, 1997.	
Data Validation and Report Review	Alan Mozol	Report Signature: 
Retention of Raw Data and Final Report	Two years from the date study is completed.	
Location of Raw Data and Final Report	AScI Corporation/AScI-Duluth Environmental Testing Division, 4444 Airpark Boulevard, Duluth, MN 55811; Tel. No. (218) 722-4040.	

2.0 OBJECTIVE(S)

To determine the 96-hour 10% and 50% effect concentrations (LC_{10} and LC_{50}), and the 96-hour no observable effect concentration (NOEC) of the test substance, R1904, for fathead minnow (*Pimephales promelas*) under static test conditions.

This study was conducted according to the AScI study plan retained by the Sponsor and OECD Guideline 203 (OECD 1993).

3.0 TEST METHODS

Test Substance	*Properties*: R1904, bright-yellow liquid, specific gravity = 1.3 (water=1.0), stable, and completely soluble in water. *Test concentrations*: 0 (dilution water control), 19, 32, 54, 90, 150, and 250 mg/L. Test solutions prepared by mass addition of test substance to dilution water followed by vigorous stirring.
Test Organisms	*Source*: Healthy, juvenile fathead minnows (*Pimephales promelas*) obtained from AScI-DETD in-house culture. *Holding/acclimation*: The fish used in the tests were obtained from AScI's in-house culture. The fish used in the study were hatched on May 24, 1997. For the 48 hours preceding test initiation mortality was 0%, and the fish were not fed for 24 hours before test initiation. The fish mean length was 17.0 ± 2.6 mm, and mean wet weight was 38 ± 23 mg. Loading rate was calculated to be 127 mg of fish per L test solution. *Use in test*: Used 10 organisms/replicate and 2 replicates/test concentration.
Dilution Water	*Source*: Dechlorinated City of Duluth, MN, tap water. The most recent chemical analysis is in Raw Data Package. *Characteristics*: At test time, the water had a hardness of 48 mg/L (as $CaCO_3$) and a pH of 8.03. Aerated for 24 h prior to use in the test.

Exposure Chambers	*Type*: 4-L glass jars. *Use in test*: 3.0 L of dilution water or test solution/chamber. During the test, kept covered, except when experimental observations were made. Solutions were not aerated during the exposures.
Incubation	*Duration*: 96 hours. *Daily photoperiod*: 16 h light and 8 h dark, maintained using cool white fluorescent lamps. *Temperature*: 21.0° ± 1°C.
Observations	*Biological*: Daily- Test organisms with abnormal behavior and signs of stress, and dead test organisms. *Water chemistry*: (1) At test initiation- total hardness and alkalinity, (2) daily- temperature, DO, conductivity, and pH. All determinations were made according to APHA methods (1995).
Endpoint Calculations	LC_{10}: Inhibition Concentration (ICp) Method. LC_{50}'s: Trimmed Spearman-Karber Method. *NOEC*: Fisher's Exact Test (TOXSTAT statistical software, Version 3.5, West Inc., and University of Wyoming).

3.0 RESULTS

Test Substance Toxicity (Pertinent data are in Table 1)	*24-h* LC_{50}: >250 mg/L (Not Calculable). *48-h* LC_{50}: 211 mg/L (Not Calculable). *72-h* LC_{50}: 146 mg/L (128-167). *96-h* LC_{10}: 45 mg/L (22-80). *96-h* LC_{50}: 97 mg/L (81-116). *96-h NOEC*: 54 mg/L.
Ranges of Water Chemistry Parameters	*Total hardness (as* $CaCO_3$ *mg/L)*: 46-48. *Alkalinity (as* $CaCO_3$ *mg/L)*: 44-46. *Temperature (°C)*: 20.8 (No Range). *DO (mg/L)*: 5.4-8.8. *pH*: 7.60-8.05. *Specific conductivity (µmhos/cm)*: 116-133.

4.0 TEST QA/QC

QA/QC Criteria	Data
Mortality in control exposure must not be more than 10%.	0% mortality.
The lowest DO concentration at test termination must be at least 60% of the air saturation value at test temperature.	The lowest DO concentration measured was 61% of the air saturation value at 21.0°C.
Test duration must be 96 h.	The test duration was 96 h.
Reference Substance Toxicity Test (this test is optional).	Control chart for sodium chloride reference toxicity testing is in Raw Data Package.

5.0 REFERENCES

American Public Health Association (APHA). 1995. Standard Methods for the Examination of Water and Wastewater. APHA, Washington, D.C.

Organization for Economic Cooperation and Development (OECD). 1981/1993. OECD Guidelines for Testing of Chemicals. OECD Publication Information Center, Washington, D.C.

Hamilton, M.A., R.C. Russo, and R.V. Thurston. 1977. Trimmed Spearman-Karber Method for Estimating Median Lethal Concentrations in Toxicity Bioassays. Environ. Sci. Technol. 11(7): 714-719; Correction 12 (4): 417 (1978).

USEPA. 1993. A Linear Interpolation Method for Sublethal Toxicity: The Inhibition Concentration (ICp) Approach. Version 2.0.

West Inc., and University of Wyoming. Toxstat Software Package. Version 3.5. Cheyenne, Wyoming.

TABLE 1. ACUTE TOXICITY OF R1904 TO FATHEAD MINNOWS (*P. promelas*)

Test Substance Concentration (mg/L)	Cumulative Number of Dead Organisms and Percent Mortality[a]			
	24 h	48 h	72 h	96 h
0 (control)	0 (0)	0 (0)	0 (0)	0 (0)
19	0 (0)	0 (0)	0 (0)	1 (5)
32	0 (0)	0 (0)	0 (0)	0 (0)
54	0 (0)	0 (0)	0 (0)	3 (15)
90	0 (0)	0 (0)	2 (10)	8 (40)
150	0 (0)	0 (0)	9 (45)	16 (80)
250	0 (0)	15 (75)	20 (100)	20 (100)

[a] Percentages are in parenthesis.

LETHAL AND NO-EFFECT CONCENTRATIONS WITH CORRESPONDING 95% CONFIDENCE INTERVALS

LC's and NOEC	Data (mg/L)[a]	95% Confidence Interval
24-h LC_{50}	>250	Not calculable
48-h LC_{50}	211	Not reliable
72-h LC_{50}	146	128-167
96-H LC_{10}	45	22-80
96-h LC_{50}	97	81-116
96-h NOEC	54	Not applicable

[a] Values based on nominal test concentrations.

ACUTE TOXICITY TO AQUATIC INVERTABRATES (DAPHNIA MAGNA)

TEST SUBSTANCE

Identity: Perfluorooctanesulfonylamido(ethyl)acetate; may also be referred to as PFOSAA, FC-128 or as the major component of FC-127, FC-129, FC-109, FC-109-X or R1904. (Glycine, N-ethyl-N-[(heptadecafluorooctyl)sulfonyl]-, potassium salt, CAS # 2991-51-7)

Remarks: The test sample is FC-129 referred to by the test laboratory as R1904. Current information indicates it is a mixture of 42% test substance, 32% water, 14% 2-butoxyethanol, 4% ethanol, and 8% C_4-C_7 fluoroalkyl carboxylate compounds. Test material was from 3M production lot 930014, mfg. Date 3/97.

The following summary is abbreviated because the test sample is a mixture of the test substance in a water/organic solvent solution. Data may not accurately relate toxicity of the test sample with that of the test substance.

METHOD:

Methods: OECD 202
Test type: Static acute
GLP: No
Year Completed: 1997
Species: *Daphnia magna*
Analytical monitoring: Temperature, pH, DO, and conductivity

RESULTS

Nominal concentrations: Bk control, 1.4, 2.3, 3.9, 6.5, 10.8, 18, 30, 50 mg/L (in duplicate)

Element values:
24-hour EC_{50} = >50 mg/L (C.I. not calculable)
48-hour EC_{10} = 4.4 (4.3 – 4.6) mg/L
48-hour EC_{50} = 9.1 (7.4 - 11) mg/L
48-hour NOEC = 3.9 mg/L

All element values based on nominal concentrations.

Remarks: Testing was conducted on the mixture of the test substance as described in the test substance remarks field. The value reported applies to that mixture and not the test substance. No attempt was made to determine the impact of the presence of the organic solvents or what

portion of the toxicity can be contributed to the Perfluorooctanesulfonylamido(ethyl)acetate.

CONCLUSIONS

The FC-129 48-hour EC_{50} for *Daphnia magna* was determined to be 9.1 mg/L with a 95% confidence interval of 7.4 - 11 mg/L. The 48-hour no observed effect concentration was 3.9 mg/L.

Submitter: 3M Company, Environmental Laboratory, P.O. Box 33331, St. Paul, Minnesota, 55133

DATA QUALITY

Reliability: Klimisch ranking 2. Testing meets all criteria for quality testing, but lacks analytical confirmation of test substance concentrations. There is a lack of characterization of the test sample.

REFERENCES

This study was conducted at AScI Corporation of Duluth, Minnesota at the request of the 3M Company.

OTHER

Last changed: 5/17/00

STUDY TITLE

ACUTE TOXICITY OF R1904 TO *DAPHNIA MAGNA*

DATA STANDARD

OECD GUIDELINE 202

STUDY COMPLETED

June 17, 1997

TESTING FACILITY

AScI Corporation/AScI-Duluth
Environmental Testing Division
4444 Airpark Boulevard
Duluth, MN 55811

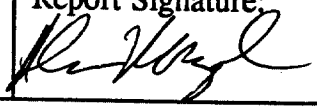
Tel. No. (218) 722-4040
Fax No. (218) 722-2592

STUDY IDENTIFICATION NUMBERS

AScI Study ID# 5010-068

3M Company Lab Request# R1904

1.0 GENERAL INFORMATION

Study Title	Acute Toxicity of R1904 to *Daphnia magna*.	
Data Standard	OECD Guideline 202.	
Sponsor	3M Environmental Technology and Safety Services, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55133.	
Sponsor's Representative	Susan A. Beach, 3M Environmental Technology and Safety Services, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55133. Tel No. (612) 778-7452.	
Sponsor's Lab Request#	R1904.	
Testing Facility	AScI Corporation/AScI-Duluth Environmental Testing Division, 4444 Airpark Boulevard, Duluth, MN 55811; Tel. No. (218) 722-4040.	
Principal Investigator	Holly Kitterman.	
Project Director	Joe Amato.	
Testing Facility Director	Donald Mount.	
Definitive Test Dates	June 10-12, 1997.	
Data Validation and Report Review	Alan Mozol.	Report Signature: 
Retention of Raw Data and Final Report	Two years from the date study is completed.	
Location of Raw Data and Final Report	AScI Corporation/AScI-Duluth Environmental Testing Division, 4444 Airpark Boulevard, Duluth, MN 55811; Tel. No. (218) 722-4040.	

2.0 OBJECTIVE(S)

To determine the 48-h EC_{10} and EC_{50}, and 48-h NOEC (no observed effect concentration) of the test substance, R1904, for *Daphnia magna*.

This study was conducted according to the ASci study plan retained by the Sponsor and OECD Guideline 202 (OECD 1993).

3.0 TEST METHODS

Test Substance	*Properties*: R1904, yellow-colored liquid, specific gravity Ca. 1.3 (water = 1), stable, and completely soluble in water. *Test concentrations*: 0 (dilution water control), 1.4, 2.3, 3.9, 6.5, 10.8, 18, 30, and 50 mg/L. Stock solution prepared by diluting 200 mg of test substance in 1 L of dilution water. Test substance concentrations were prepared by direct addition of stock solution into dilution water. A 0.6 dilution series was utilized.
Test Organisms	*Source*: Healthy, less than 24-h old *Daphnia magna* neonates obtained from ASci stock cultures maintained in water of test dilution water quality. *Use in test*: Gravid adults isolated in dilution water 24 h before the test initiation. Used 5 organisms/replicate and 4 replicates/test concentration. Handled with a smooth bore pipet with an inside diameter of 6 mm. Did not feed during the test.
Dilution Water	*Source*: Dechlorinated City of Duluth, MN, tap water. The most recent chemical analysis is in Raw Data Package. *Characteristics*: At test time, the water had a hardness of 46 mg/L (as $CaCO_3$) and a pH of 7.93. Aerated for 24 h prior to using in the test.
Exposure Chambers	*Type*: 250-ml borosilicate glass beakers. *Use in test*: 200 ml of dilution water or test solution/chamber. During the test, kept covered, except when experimental observations were made.

Incubation	*Duration*: 48 h. *Daily photoperiod*: 16 h light and 8 h dark, maintained using cool-white fluorescent bulbs. *Temperature*: 21°C.
Observations	*Biological*: Daily- gently agitated the exposure chambers and then counted immobilized and dead test organisms (immobilized organisms were those that did not swim within 15 seconds after exposure chambers were gently agitated). Did not remove any daphnids from the exposure chambers at 24 h. *Water chemistry*: (1) At test initiation- total hardness and alkalinity, (2) daily- temperature, pH, DO, and conductivity. All determinations were made according to APHA methods (1995).
Endpoint Calculations	EC_{50}: Trimmed Spearman-Karber method. EC_{10}: Inhibition Concentration (10%) USEPA software package for Determining ICp concentrations. *NOEC*: Fisher's Test (TOXSTAT statistical software, Version 3.5, West, Inc., and University of Wyoming).

4.0 RESULTS

Test Substance Toxicity (Pertinent data are in Table 1)	*24-h* EC_{50}: >50 mg/L. *48-h* EC_{50}: 9.1 mg/L (7.4-11.0). *48-h* EC_{10}: 4.4 mg/L (4.3-4.6). *48-h NOEC*: 3.9 mg/L.

Ranges of Water Chemistry Parameters	*Total hardness (as $CaCO_3$ mg/L)*: 44-46. *Alkalinity (as $CaCO_3$ mg/L)*: 42-44. *Temperature (°C)*: 20.5-20.8°C. *DO (mg/L)*: 8.1-8.5. *pH*: 7.81-8.04. *Specific conductivity (µmhos/cm)*: 118-126.

5.0 TEST QA/QC

QA/QC Criteria	Data
Test organism dead, immobilized or trapped in control exposure must not be more than 10%.	0% dead, immobilized or trapped.
The lowest DO concentration at test termination must be at least 60% of the air saturation value at test temperature.	The lowest DO concentration measured was 91% of the air saturation value at 21.0°C.
Test duration must be 48 h.	The test duration was 48 h.
Reference substance toxicity test (this test is optional).	Current control chart for *D. magna* acute testing is included in the raw data package.

5.0 REFERENCES

American Public Health Association (APHA). 1995. Standard Methods for the Examination of Water and Wastewater. APHA, Washington, D.C.

Hamilton, M.A., R.C. Russo, and R.V. Thurston. 1977. Trimmed Spearman-Karber Method for Estimating Median Lethal Concentrations in Toxicity Bioassays. Environ. Sci. Technol. 11(7):714-719; Correction 12(4):417 (1978).

Organization for Economic Cooperation and Development (OECD). 1993. OECD Guidelines for Testing of Chemicals. OECD Publication Information Center, Washington, D.C.

West Inc., & University of Wyoming. TOXSTAT Software Package Version 3.5.

US EPA. 1993. A Linear Interpolation Method for Sublethal Toxicity: The Inhibition Concentration (ICp) Approach (Version 2.0). Environmental Research Laboratory, Duluth, Minnesota.

TABLE 1. ACUTE TOXICITY OF R1904 TO *DAPHNIA MAGNA*

Test Substance Concentration (mg/L)	Cumulative Number and Percent of Dead and Immobilized Organisms[a]	
	24 h	48 h
0	0 (0)	0 (0)
1.4	0 (0)	0 (0)
2.3	0 (0)	0 (0)
3.9	0 (0)	0 (0)
6.5	0 (0)	10 (50)
10.8	1 (5)	17 (85)
18	1 (5)	14 (70)
30	0 (0)	16 (80)
50	4 (20)	20 (100)

[a] Percentages are in parenthesis.

EFFECT AND NO-EFFECT CONCENTRATIONS WITH CORRESPONDING 95% CONFIDENCE INTERVALS

EC_{50} and NOEC	Data (mg/L)	95% Confidence Interval
24-h EC_{50}	>50	Not Calculable
48-h EC_{50}	9.1	7.4-11.0
48-h EC_{10}	4.4	4.3-4.6
48-h NOEC	3.9	Not Applicable