ACUTE TOXICITY TO FISH (FATHEAD MINNOW)

TEST SUBSTANCE

Identity: A mixture containing perfluorooctanesulfonate, which may also be referred to as PFOS, FC-95, or as a component of FC-603EF. (1-Octanesulfonic acid) (CAS # 2795-39-3).

Remarks: The 3M production lot number is not noted. The test sample is FC-603EF. Current information indicates it is a mixture of 1.30% PFOS, 10% diethylene glycol butyl ether, 54.15% water, 22.5% ethylene glycol, 2.6% propanesultone foamer, 5% Alkylether sulfate, 0.1% N-(3-Chloroallyl)hexaminium chloride, 0.9% Xanthan gum, 0.9% starch, 1.0% triethanolamine, and 0.05% tolyl triazole.

The following summary applies a mixture with incompletely characterized concentrations of impurities. Data may not accurately reflect toxicity of the fluorochemical component of the test sample.

METHOD:

Method: OECD Guideline 203.
Type: Acute static
GLP: No
Year completed: 1997
Species: *Pimephales promelas*
Supplier: In-house cultures maintained at AScI-DETD, Duluth, MN.
Analytical monitoring: Temperature, pH, conductivity, and dissolved oxygen.
Exposure period: 96-hours
Statistical methods: LC_{50} values were calculated by the Trimmed Spearman-Karber method. The NOEC was calculated using Fisher's Exact Test (Toxstat software, Version 3.5).
Test fish age: Juveniles
Length and weight: Average length = 12.7 mm
Average weight = 0.0165 g (wet)
Loading: 0.066 g/L
Pretreatment: None.
Test conditions:
Dilution water: Dechlorinated City of Duluth, MN, tap water (post carbon filter) that was aerated for 24 hours prior to use.
Dilution water chemistry:
Hardness: 48 mg/L as $CaCO_3$
Alkalinity: 46 mg/L as $CaCO_3$
pH: 7.80

Lighting: Cool-white fluorescent lights. A daily photoperiod of 16 hours light and 8 hours dark was maintained throughout the testing period.
Stock and test solution preparation: The test material was added directly to the dilution water on a weight/volume basis and mixed by mechanical stirring.
Concentrations dosing rate: Once.
Stability of the test chemical solutions: Not noted.
Exposure vessels: 4 L glass jars containing 2.5 L test solution.
Number of replicates: 2.
Number of fish per replicate: 10.
Number of concentrations: five plus a blank control
Water chemistry during the study:

pH range (0-96 hours):
7.73 – 7.83 (control exposure)
7.31 – 7.85 (1000 mg/L exposure) -

Temperature range (0-96 hours): 21.1 – 21.3 °C

Conductivity range (0-96 hours):
133 – 149 µmhos/cm (control exposure)
150 – 159 µmhos/cm (1000 mg/L exposure)

Dissolved oxygen range (0-96 hours):
7.1 – 9.2 mg/L (control exposure)
6.7 – 9.4 mg/L (1000 mg/L exposure)

Remarks: Solutions were aerated continuously during the exposures using single bubble aeration to minimize foaming.

RESULTS

Nominal concentrations: Blank control, 1, 130, 216, 360, 600, and 1000 mg/L

Element values: 96-hour LC_{50} = >1000 mg/L
96-hour NOEC = 1000 mg/L

Element values based on nominal concentrations

Remarks: Testing was conducted on the mixture as described in the Test Substance Remarks field. The values reported apply to that mixture and not the fluorochemical proportion alone.

CONCLUSIONS

The test sample 96-hour LC_{50} for fathead minnow was determined to be >1000 mg/L, the highest concentration tested. Mortality observed at this concentration was 15%.

The 96-hour No Observed Effect Concentration (NOEC) was determined to be 1000 mg/L.

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

DATA QUALITY

Reliability: Klimisch ranking = 2. This study meets the criteria for quality testing. However, sample purity was not properly characterized and the study lacks analytical confirmation of the amount of fluorochemical proportion in solution.

REFERENCES

This study was conducted by AScI Corporation, AScI-Duluth Environmental Testing Division, Duluth, MN at the request of the 3M Company, Lab Request number P2581, 1997.

OTHER

Last changed: 6/26/00

STUDY TITLE

ACUTE TOXICITY OF FC-603EF, LIGHT WATER(TM) ATC 3%, AR-AFFF TO FATHEAD MINNOW (*PIMEPHALES PROMELAS*)

DATA STANDARD

OECD GUIDELINE 203

STUDY COMPLETED

January 28, 1997

TESTING FACILITY

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

Tel. No. (218) 722-4040

STUDY IDENTIFICATION NUMBERS

ASci Study ID# 5010-068

3M Company Lab Request# P2581

1.0 GENERAL INFORMATION

Study Title	Acute Toxicity of FC-603EF Light Water(TM) ATC 3%, AR-AFFF 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.	
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#	P2581.	
Testing Facility	ASci Corporation/ASci-Duluth Environmental Testing Division, 4444 Airpark Boulevard, Duluth, MN 55811; Tel. No. (218) 722-4040.	
Principal Investigator	Todd McGuire.	
Project Director	Joe Amato.	
Testing Facility Director	Donald Mount.	
Definitive Test Dates	January 21-25, 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-h LC_{50} and 96-h NOEC (no observed effect concentration) of the test substance, FC-603EF, 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*: FC603EF, amber-colored liquid, specific gravity ca. 1.06 (water = 1), vapor pressure ca. 15.8 mmHg, stable, and completely soluble in water. *Test concentrations*: 0 (dilution water control), 130, 216, 360, 600, and 1000 mg/L. Each test substance concentration obtained by mass additions to dilution water followed by mechanical stirring. These solutions were split into two aliquots to accommodate test replicates.
Test Organisms	*Source*: Healthy, juvenile fathead minnows (*Pimephales promelas*) obtained from ASci-DETD in-house culture. The batch of fish used in the study was hatched between December 18 and 20, 1996. *Holding/acclimation*: Held and acclimated from hatching until test initiation (app. 32 days) in the test dilution water. For the 48 hours preceding the test, 0% mortality occurred. At test termination, the fish were 12.7 $\pm$ 2.4 mm in mean length and 16.5 $\pm$ 8.9 mg in mean weight. *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 7.80. Aerated for 24 h prior to use in the test.

Exposure Chambers	*Type*: 4-L glass jars. *Use in test*: 2.5 L of dilution water or test solution/chamber. Solutions were aerated continuously during the exposures using single bubble aeration to minimize foaming.
Incubation	*Duration*: 96 h. *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_{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 Tables 1 and 2)	*24-h* LC_{50}: >1000 mg/L (Not Calculable). *48-h* LC_{50}: >1000 mg/L (Not Calculable). *72-h* LC_{50}: >1000 mg/L (Not Calculable). *96-h* LC_{50}: >1000 mg/L (Not Calculable). *96-h NOEC*: 1000 mg/L.

Ranges of Water Chemistry Parameters	*Total hardness (as $CaCO_3$ mg/L)*: 48. *Alkalinity (as $CaCO_3$ mg/L)*: 46-52. *Temperature (°C)*: 21.1-21.3. *DO (mg/L)*: 6.7-9.4. *pH*: 7.31-7.85. *Specific conductivity (µmhos/cm)*: 133-159.

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 75% of the air saturation value at 21.0°C. The test solution dissolved oxygen concentrations were maintained via single bubble aeration for the entire duration of the test.
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).

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

TABLE 1. ACUTE TOXICITY OF FC-603EF LIGHT WATER(TM) ATC 3% AR-AFFF TO FATHEAD MINNOWS (*P. promelas*)

Test Substance Nominal 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)
130	0 (0)	0 (0)	1 (5)	4 (20)
216	0 (0)	0 (0)	0 (0)	1 (5)
360	0 (0)	0 (0)	0 (0)	2 (10)
600	0 (0)	0 (0)	0 (0)	0 (0)
1000	0 (0)	0 (0)	1 (5)	3 (15)

[a] Percentages are in parenthesis.

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

LC_{50} and NOEC	Data (mg/L)[b]	95% Confidence Interval
24-h LC_{50}	>1000	Not calculable
48-h LC_{50}	>1000	Not calculable
72-h LC_{50}	>1000	Not calculable
96-h LC_{50}	>1000	Not calculable
96-h NOEC	1000	Not applicable

[b] LC_{50} values were calculated based on test substance nominal concentrations.