

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, or FC-109-X. (Glycine, N-ethyl-N-[(heptadecafluorooctyl)sulfonyl]-, potassium salt, CAS # 2991-51-7)

Remarks: The test sample is FC-109-X referred to by the test laboratory as E2566-2. Current information indicates it is a mixture of 19-23% test substance, 61% water, 12% 2-methoxymethylethoxypropanol, 2% ethanol, and 2-6% C₄-C₇ fluoroalkyl carboxylate compounds. Test sample was from 3M production lot 2.

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: Yes

Year Completed: 1988

Species: *Daphnia magna*

Analytical monitoring: Temperature, pH, and DO

RESULTS

Nominal concentrations: Bk control, 1.0, 1.8, 3.2, 5.6, 10, 18, 32 mg/L

Element values: 24-hour LC₅₀ = >32 mg/L (estimated)

24-hour NOEL = 32 mg/L

48-hour LC₅₀ = 1.5 (1.0-1.8) mg/L

48-hour NOEL = 1.0 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-109-X 48-hour LC₅₀ for *Daphnia magna* was determined to be 1.5 mg/L with a 95% confidence interval of 1.0-1.8 mg/L. The 48-hour no observed effect level was 1.0 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 Analytical Bio-Chemistry Laboratories, Inc. of Columbia, Missouri at the request of the 3M Company.

OTHER

Last changed: 5/18/00



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STUDY TITLE

Acute Toxicity of E2566-2 to
Daphnia magna

DATA REQUIREMENT

OECD Guideline 202

AUTHOR

Scott Frazier
Biologist II

STUDY COMPLETED ON

January 9, 1988

PERFORMING LABORATORY

Analytical Bio-Chemistry Laboratories, Inc.
Aquatic Toxicology Division
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P.O. Box 1097
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ABC LABORATORY PROJECT ID

#36616

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PROJECT PERSONNEL

The Study Director of the project for Analytical Bio-Chemistry Laboratories, Inc. was Scott Frazier, Biologist II. Supervising the conduct of the study for 3M Company was Mr. Mohamed Elnabarawy. The following ABC personnel assisted with various phases of the study.

<u>Name</u>	<u>Title</u>	<u>Initials</u>
Michael Ruddy	Biological Technician II	<u>MR</u>
Scott Frazier	Biologist II	<u>SF</u>
Jeffrey Latham	Biological Technician I	<u>JL</u>

INVERTEBRATE ACUTE TOXICITY COMPENDIUM

Subject: "Acute Toxicity of E2566-2 to Daphnia magna,"
ABC Study Number 36616.

Sponsor: 3M Company

Testing Facility: Analytical Bio-Chemistry Laboratories, Inc.
Aquatic Toxicology Division
P.O. Box 1097
Columbia, Missouri 65205 (314-474-8579)

Location of Original Raw Data and Final Report:

3M Company
EE & PC Environmental Labs
Building 21-2W-05, 900 Bush Avenue
St. Paul, Minnesota

Test Material: E2566-2

Test Concentrations: Control, 1.0, 1.8, 3.2, 5.6, 10, 18 and 32 mg/l.

Dilution Water: 246 mg/l (hardness), 178 mg/l (alkalinity),
8.0 (pH).

Test Dates: Initiation - January 7, 1988

Termination - January 9, 1988

Length of Study: 48 hours

Results: 24-hour LC₅₀ = >32 mg/l (estimated)

24-hour NOEL = 32 mg/l

48-hour LC₅₀ = 1.5 mg/l (95% C.I. - 1.0-1.8 mg/l)

48-hour NOEL = 1.0 mg/l

Test Species: Daphnia magna

Source of Organisms: ABC Laboratories in-house culture.

Age of Organisms at Study Initiation: First-instar (<24 hours old)

SUMMARY

The acute toxicity of E2566-2 to Daphnia magna was assessed using the methods outlined by the Committee on Methods for Toxicity Tests with Aquatic Organisms. Water quality parameters of temperature, dissolved oxygen and pH were measured at the termination of the test and were within acceptable limits.

The results of the 48-hour static Daphnia magna toxicity study are summarized below. All reported values were based upon nominal concentrations.

<u>Compound</u>	<u>24-Hour LC₅₀</u>	<u>48-Hour LC₅₀</u> <u>(95% C.I.)</u>
E2566-2	>32 mg/l (estimated)	1.5 mg/l (1.0-1.8 mg/l)

The 24 and 48 hour no-effect levels of E2566-2 based on the absence of mortality and abnormal effects were 32 and 1.0 mg/l, respectively.

INTRODUCTION

This definitive static bioassay was performed by the Aquatic Toxicology Division of Analytical Bio-Chemistry Laboratories, Inc., Columbia, Missouri, for 3M Company, from January 7, 1988 to January 9, 1988. The purpose of this test was to determine the 24- and 48-hour LC_{50} levels for E2566-2 to Daphnia magna. The study was performed using ABC Protocol #7806 as approved by Mr. Mohamed Elnabarawy, 3M Company, on January 7, 1988.

METHODS AND MATERIALS

The procedures for static bioassay, as described in Methods of Acute Toxicity Tests with Fish, Macroinvertebrates and Amphibians (1) and Standard Methods for Examination of Water and Wastewater (2), were used in this experiment.

I. Test Organisms

The Daphnia magna used in the test were cultured at the ABC facilities in well water (Table 1). The adult daphnids were fed algae (Selenastrum capricornutum) at least every three days prior to testing and supplemented with a suspension of Tetramin[®], cereal leaves, vitamin solution and yeast. The daphnids were identified to species using the taxonomic key presented by Pennak (3).

II. Test Procedure

The static Daphnia bioassay was conducted in 250-ml glass beakers containing 200 ml of daphnid culture/test water. All test vessels were covered with loose-fitting petri dish covers to minimize evaporation and protect against contamination during the study. These vessels were kept at 20 (± 2.0) °C in a temperature controlled area. The lighting was maintained at 50-70 footcandles on a 16-hour daylight photoperiod with 30-minute simulated dawn and dusk periods.

An initial range-finding experiment was conducted using ten Daphnia each in exposure concentrations of 1.0, 10, and 100 mg/l. From these results, seven treatment concentrations in duplicate with ten Daphnia (first instar less than 24 hours old) per beaker were selected for the definitive bioassay. These concentrations were a logarithmic series ranging from 1.0 to 32 mg/l and included a control. All concentrations were observed once every 24 hours for mortality and abnormal effects such as surfacing, clumping of the daphnids together and daphnids tending to the bottom of test chambers. Water chemistry parameters of temperature, dissolved oxygen and pH were measured in the control, low, middle and high concentrations at 0 and 48 hours.

III. Test Compound

The E2566-2 sample was received on December 29, 1987 as a foamy, yellow liquid and was stored at room temperature in darkness. Sample purity was unspecified. Test concentrations were prepared based on total product. All standard weights and dilutions can be found in the Appendix. Daphnia culture/test water was used in the preparation of all working stock solutions.

IV. Statistics

Statistical analysis of the concentration vs. effect data (generally mortality) was obtained by employing a computerized LC_{50} program developed by Stephan et al. (4). This program calculated the LC_{50} statistic and its 95 percent confidence limits using the binomial, the moving average and the probit tests. Three different methods of analyzing the data were used since no one method of analysis is appropriate for all possible sets of data that may be obtained. The method of calculation selected for presentation in this report was that which gave the narrowest confidence limits for the LC_{50} (4, 5) although all three models are valid. However, if no mortality occurred or if a dose response could not be demonstrated over a reasonable range (<37 to >63%) an LC_{50} and/or its 95 percent confidence limits could not be calculated.

RESULTS

Table 2 presents the estimated and calculated LC_{50} values and 95-percent confidence intervals for E2566-2. The 24- and 48-hour LC_{50} values were >32 (estimated) and 1.5 mg/l, respectively. All results were based on the nominal concentrations of 1.0, 1.8, 3.2, 5.6, 10, 18 and 32 mg/l. The no-effect concentration based on the absence of mortality and abnormal effects was 32 mg/l at 24 hours. At 48 hours the no-effect level was 1.0 mg/l. The abnormal effect of mortality was observed in the 1.8, 3.2, 5.6, 10, 18 and 32 mg/l test concentrations. The 10, 18 and 32 mg/l test concentrations were observed at 24 and 48 hours to have a surface film and precipitate present.

Table 3 presents the percent mortality and water quality parameters measured during the test. The dissolved oxygen concentrations ranged between 8.7 and 9.5 mg/l. These values represented 100 and 107-percent saturation at 20 and 19 °C (corrected for local altitudinal pressure), respectively, and were considered adequate for testing (1). The pH values of the test solutions ranged from 8.2 to 8.4 and were consistent with the control.

The study was conducted following the intent of the Good Laboratory Practice Regulations (6) and the final report was reviewed by Analytical Bio-Chemistry Laboratories' Quality Assurance Unit. All original raw data were provided to 3M Company, with a copy retained at Analytical Bio-Chemistry Laboratories.

TABLE 1
Chemical Characteristics of Well Water Used by
ABC's Aquatic Toxicology Division.

Parameter	Concentration
Temperature	17 - 22°C ^b
Dissolved Oxygen ^a	7.4 - 9.1 ppm ^b
pH	7.6 - 8.4 ^b
Hardness (CaCO ₃)	206 - 275 ppm ^b
Alkalinity (CaCO ₃)	224 - 336 ppm ^b
Conductivity	500 - 650 umhos/cm ^b
Total Organic Carbon	1.4 ppm ^c (0.9 - 2.8 ppm) ^d
Suspended Solids	0 - 1.3 ppm ^b
Aluminum	<10 ppb
Arsenic	<1 ppb
Boron	2.25 ppm
Cadmium	<3 ppb
Chromium	<7 ppb
Cobalt	<3 ppb
Copper	<3 ppb
Iron	14 ppb
Lead	<30 ppb
Mercury	0.43 ppb
Nickel	<8 ppb
Silver	<4 ppb
Zinc	2 ppb
Measured organophosphorus pesticides	<0.14 ppb
Measured organochlorine pesticides plus PCB's	<1.0 ppb

^aAfter aeration

^bRepresents seasonal variation, difference from one month to the next, will not exceed 10%

^cAverage of all well water values from January-September, 1987

^dTotal monthly range of organic carbon

Sample dates: Trace elements = June 3, 1987, organophosphate and organochlorine analyses = May 28, 1987.

TABLE 2
Acute Toxicity of E2566-2
to Daphnia magna^a

Compound	LC ₅₀ (mg/l)	
	24 hours	48 hours
E2566-2	>32 (estimated) b	1.5 (1) (1.0-1.8) ^c

^a Bioassay conducted at 20°C (±2.0).

^b 95% confidence limits could not be established.

^c 95% confidence limits.

The 24- and 48-hour no-effect concentrations were 32 and 1.0 mg/l, respectively, based on the absence of mortality and abnormal effects.

All values were rounded to two significant figures following the technique provided in ABC S.O.P. #8.7.

LC₅₀ calculated using:

(1) Binomial Method

TABLE 3

Percent Mortality and Water Quality Measurements During the
Acute Toxicity Test of E2566-2 to Daphnia magna.

Nominal Concentration (mg/l)	Percent Mortality		Water Quality					
			0 hours			48 hours		
	24 hr	48 hr	Temp. ^a °C	D.O. ^b mg/l	pH ^c	Temp. °C	D.O. mg/l	pH
Control	0	0	19	9.3	8.2	20	8.7	8.3
1.0	0	0	19	9.4	8.2	20	8.7	8.3
1.6	0	80						
3.2	0	100						
5.6	0	100	19	9.4	8.2	20	8.7	8.3
10	0	100						
18	0	100						
32	0	100	19	9.5	8.2	20	8.7	8.4

^aTemperature - mercury thermometer.

^bDissolved oxygen concentrations - YSI 54 ARC dissolved oxygen meter.

^cpH - Corning 140 pH/mV meter with Beckman 39831 electrode.

NOTE: Dissolved oxygen saturations at the test temperatures of 19 and 20°C are 8.9 and 8.7 mg/l, respectively (corrected for altitudinal pressure at ABC Labs).

LITERATURE CITED

- (1) Committee on Methods for Toxicity Tests with Aquatic Organisms. Methods of Acute Toxicity Tests with Fish, Macroinvertebrates and Amphibians. 1975. U.S. EPA, Ecol. Res. Ser. 660/3-75-009.
- (2) American Public Health Association. 1980. Standard Methods for the Examination of Water and Wastewater. 15th ed. Washington, DC. 1134 p.
- (3) Pennak, R. W. 1978. Freshwater Invertebrates of the United States. John Wiley & Sons, New York, 2nd ed. 803 p.
- (4) Stephan, C. E., K. A. Busch, R. Smith, J. Burke and R. W. Andrews. 1978. A Computer Program for Calculating an LC_{50} . U. S. Environmental Protection Agency, Duluth, Minnesota, pre-publication manuscript, August, 1978.
- (5) Stephan, C. 1977. Methods for Calculating an LC_{50} . p. 65-84. In F. L. Mayer and J. L. Hamelink (eds.). Aquatic Toxicology and Hazard Evaluation. ASTM Special Technical Publication 634. ASTM. Philadelphia.
- (6) Organization for Economic Cooperation and Development. 1981. OECD Guidelines for Testing of Chemicals, Principles of Good Laboratory Practice Annex 2, C(81) 30(Final):7-28.

QUALITY ASSURANCE STATEMENT

Quality Assurance Statement for report #36616 entitled, "Acute Toxicity of E2566-2 to Daphnia magna," for Mr. Mohamed Elnabarawy, 3M Company, St. Paul, Minnesota.

In accordance with ABC Laboratories' intent that all studies conducted at our facilities are designed and function in conformance with good laboratory practice regulations and the protocols for individual laboratory studies, an inspection of the final report for E2566-2 was conducted and found to be in an acceptable form by a member of our Quality Assurance Unit. A procedure audit was conducted on December 14, 1987. The study director was notified of the results on the same day. The results of this audit were reported to ABC Laboratories' management on December 30, 1987. A final inspection of all data and records on January 14, 1988, indicated that the report submitted to you is an accurate reflection of the study as it was conducted by ABC Laboratories.

Should you have any questions relating to the information provided in this statement or the function of our Quality Assurance Unit, please contact the Quality Assurance Unit at your convenience.


Kathy Landholt
Quality Assurance Officer

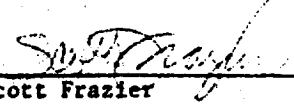
1/18/88
Date

STUDY COMPLIANCE STATEMENT

Study Compliance Statement for ABC report #36616 entitled, "Acute Toxicity of E2566-2 to Daphnia magna," for Mr. Mohamed Elnabarawy, 3M Company, St. Paul, Minnesota.

In accordance with ABC Laboratories' intent that all aquatic toxicity tests conducted by our facility follow good laboratory practices, ABC's study director for the above test herein confirms that the study was conducted in compliance with the OECD Guidelines for Testing of Chemicals, Principles of Good Laboratory Practice Annex 2, C(81) 30(Final):7-28.

All original raw data were provided to 3M Company, and a copy retained at Analytical Bio-Chemistry Laboratories.



Scott Frazier
ABC Study Director

11/11/81

Date

APPENDIX - RAW DATA

NOTE: Some of the records which appear in this raw data appendix have been provided as photocopies of original records on file at ABC. This has been done by necessity for certain data which are used commonly in several studies at ABC. Such records may include analytical standard solution preparation sheets, organism culture logs and well water chemical screen results.