ACUTE TOXICITY TO FISH (MUMMICHOG)

TEST SUBSTANCE

Identity: A mixture containing perfluorooctanesulfonate, which may also be referred to as PFOS, FC-95, or as a component of FC-203. (1-Octanesulfonic acid) (CAS # 2795-39-3).

Remarks: The 3M production lot number was 1. The test sample is FC-203. Current information indicates it is a mixture of 1.34% PFOS, 35% diethylene glycol butyl ether, 37.85% water, 20% ethylene glycol, 2.66 % Sultone foamer, 3% sodium octyl sulfate, 0.1% sodium lauryl sulfate, and 0.05% tolyltriazole.

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

METHOD:

Method: DTNSRDC Standard Static Marine Bioassay Procedure for Shipboard Chemicals (Liberatore).
Type: Acute static
GLP: No
Year completed: 1979
Species: *Fundulus heteroclitus*
Supplier: Commercial supplier in Massachusetts.
Analytical monitoring: Temperature, pH, salinity, and DO
Exposure period: 96-hours
Statistical methods: LC_{50} values calculated using the Stephan computer program, 1978.
Test fish age: Juveniles
Length and weight: Average length = 43 mm
Average weight = 0.91 g (wet)
Loading: Not given.
Pretreatment: None
Test conditions:
Dilution water: Instant Ocean® dissolving in deionized water
Dilution water chemistry:

pH	8.2
Salinity	15 ppt
Alkalinity:	52 mg/L as $CaCO_3$
Conductivity:	23,000 µmhos/cm

Lighting: A daily photoperiod of 16 hours light and 8 hours dark was maintained throughout the testing period.

Stock and test solution preparation: The test solution was added directly to the dilution water and mixed by stirring with a glass rod.

Concentrations dosing rate: Once
Stability of the test chemical solutions: Not noted
Exposure vessels: 19.6 L glass jars containing 15 L of test solution.
Number of replicates: 2
Number of fish per replicate: 5
Number of concentrations: five plus a blank control
Water chemistry during the study:

Salinity range: (0-96 hours)

15 - 17 ppt (control exposure)
16 - 17 ppt (3600 mg/L exposure)*

pH range (0-96 hours):

7.2 – 8.1 (control exposure)
6.7 – 8.1 (3600 mg/L exposure)

Temperature range (0-96 hours): 19.5 °C

Dissolved oxygen range (0-96 hours):

6.2 – 8.3 mg/L (control exposure)
3.0 – 8.2 mg/L (3600 mg/L exposure)

RESULTS

Nominal concentrations: Bk control, 600, 1000, 1700, 2400, and 3600 mg/L
Element values: 96-hour LC50 = 2500 (1700 – 3600) mg/L
96-hour NOEC = <600 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 mummichog was determined to be 2500 mg/L with a 95% confidence interval of 1700 to 3600 mg/L.

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

DATA QUALITY

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

REFERENCES

Test was conducted by EG&G Bionomics, of Wareham, MA at the request of the 3M Company, St. Paul, MN, Lab Request number 4971S, 1979.

OTHER

Last changed: 6/27/00

ACUTE TOXICITY OF 7902 TO THE COMMON MUMMICHOG (Fundulus heteroclitus).

FC-203 lot 1
LR 4971 S
EAI ~~4988-9~~ 79245

TOXICITY TEST REPORT

SUBMITTED TO

THE 3M COMPANY

ST. PAUL, MINNESOTA

REPORT #BW-79-8-526

E G & G, Bionomics
Aquatic Toxicology Laboratory
790 Main Street
Wareham, Massachusetts
August, 1979

FC-203

INTRODUCTION

The purpose of this study was to estimate the acute toxicity of 7902 to the common mummichog (Fundulus heteroclitus) under static conditions. A 96-hour definitive test was conducted from 27-31 July 1979 at the Aquatic Toxicology Laboratory of E G & G, Bionomics, Wareham, Massachusetts. All raw data generated are stored at the above location.

MATERIALS AND METHODS

Unless otherwise stated, procedures used in this acute toxicity test followed those described in "DTNSRDC Standard Static Marine Bioassay Procedure for Shipboard Chemicals" (Liberatore). All values are reported to various levels of significance depending on the accuracy of the measuring devices employed during any one process.

The 7902, an amber liquid, received from the 3M Company, St. Paul, Minnesota on 6 July 1979, was tested on a formulation basis. Nominal test concentrations are reported as milligrams of 7902 per liter of test solution (mg/ℓ).

The common mummichog (lot #79A7) used in this study were obtained from a commercial fish supplier in Massachusetts. Upon receipt at Bionomics, the fish were held in a 600-ℓ fiberglass

tank containing a solution of Instant Ocean^{R} which was recirculated through dolomite in a biologically active filter bed to preserve water quality during holding and acclimation. This water was characterized as having a pH range of 7.0-7.2, a dissolved oxygen (DO) range of 89-90% of saturation (Weekly Record of Fish Holding Water Characteristics, Vol. I), a specific conductance range of 18,000-23,000 micromhos per centimeter (µmhos/cm), a salinity of 15-19 parts per thousand (o/oo) and a temperature range of 19.0-20.5°C (Daily Record of Fish Holding Conditions, July 1979). Experimental animals were maintained under these conditions for a minimum of 14 days. All fish were fed a dry pelleted food, ad libitum, daily except during the 48-hours prior to testing. There was no mortality during this 2 day period. The pH was measured with a Model #175 Instrumentation Laboratory pH meter and combination electrode, the temperature with a dial thermometer, the DO with a Model #57 YSI meter and probe, the specific conductance with a Model #33 YSI conductivity meter, and the salinity with an American Optical refractometer.

The definitive test was conducted in 19.6-ℓ glass jars which contained 15 ℓ of test solution. The dilution water used in this test was prepared by adding Instant Ocean^{R} to deionized water and mixing until dissolved. This water had a pH of 8.2, a total alkalinity as calcium carbonate of 52 mg/ℓ (APHA et al., 1975), a specific conductivity of 23,000 µmhos/cm and a salinity

of 15 o/oo (Reconstituted Water, Water Quality Analysis, Vol. II). The 7902 was added directly to the dilution water and mixed by stirring with a glass rod.

Control jars (A and B replicates) containing the same dilution water and maintained under the same conditions as test concentrations, but containing no 7902, were established. All test solution temperatures were controlled by a system designed to maintain temperatures at 19 $\pm$ 1°C. All test solutions were gently aerated during the exposure period. The photoperiod during acclimation and testing was 16 hours light and 8 hours darkness.

Ten common mummichog (five per replicate) with a mean (range, N=30) wet weight and total length of 0.91 (0.28-2.79) grams and 43 (32-60) millimeters (Fish Weights and Lengths Log, Vol. III) were randomly distributed to each test concentration within 15 minutes after the solutions were prepared.

Mortalities were recorded and removed from each test jar every 24 hours during testing. Biological observations of the fish and observations of the physical characteristics of the test solutions were made and recorded at 0, 24, 48, 72 and 96 hours. The pH, DO concentration and salinity of test solutions were measured at 0, 24, 48, 72 and 96 hours in both replicates of the control, high, middle and low test concentrations. Temperature

was also measured in each control jar replicate at the above mentioned time intervals.

When results permit, the concentrations tested and the corresponding mortality data derived from the toxicity test are used to estimate 24-, 48-, 72- and 96-hour median lethal concentrations (LC50) and 95% confidence intervals. The LC50 is defined as the concentration (nominal or measured) of the test compound in dilution water which caused 50% mortality in the test animal population at the stated exposure interval.

The computer program utilized (Stephan, 1978, personal communication) estimates LC50 values using one of three statistical methods in the following order of preference: moving average angle analysis, probit analysis, binomial probability. The method selected is determined by the characteristics of the data base (i.e. presence or absence of 100% mortality, number of partial mortalities, etc.). The computer program scans the data base, identifies the most preferred statistical method and performs the analysis.

RESULTS

The 96-hour LC50 for the common mummichog exposed to 7902, estimated by the binomial probability method, was 2,500 mg/l. Table 1

summarizes the 24-, 48-, 72- and 96-hour LC50's and 95% confidence intervals and states the no discernible effect concentration through 96 hours. The no effect concentration is the highest concentration tested at which there were no mortalities or observed behavioral and physical abnormalities, e.g. loss of equilibrium, at surface, darkened pigmentation.

The pH, DO and salinity measured during the test are presented in Table 2. The temperature of the control solution was 19.5°C during exposure.

Table 3 presents the nominal test concentrations, the corresponding cumulative mortalities and the observations made.

Deviations from the Protocol Cited

- The common mummichog were ~4.3 centimeters long.
- Nineteen point six liter widemouth glass bottles were used as test containers.
- Instant Ocean® solution was used for acclimation and testing.
- The fish were acclimated in recirculating water.
- The salinity for acclimation and testing was 15 o/oo.
- Fish were acclimated in water with a pH range of 7.0-7.2 and tested in water having a pH of 8.2.

- The test jars contained 15 ℓ of solution and 5 test fish.
- The test concentrations were replicated.
- LC50's were calculated by Stephan, 1978, personal communication.

It is our judgment that none of these deviations have affected the integrity of the test results.

LITERATURE CITED

APHA, AWWA, WPCF. 1975. Standard methods for the examination of water and wastewater. 14th Edition, Washington, D.C. 1193 pp.

Liberatore, Giacomo. David W. Taylor Navel Ship Research and Development Center. Standard Static Marine Bioassay Procedure for Shipboard Chemicals. Report TM-28-76-29.

Stephan, Charles. 1978. U.S. EPA, Environmental Research Laboratory, Duluth, Minnesota. Personal communication.

SUBMITTED BY:

E G & G, Bionomics
Aquatic Toxicology Laboratory
790 Main Street
Wareham, Massachusetts
August, 1979

PREPARED BY: Charlotte Stiefel

Charlotte Stiefel RPH

Principal Investigator

REVIEWED BY: Robert J. Buccafusco

Robert J. Buccafusco

Study Director

APPROVED BY: George A. Cary

George A. Cary

Director, Aquatic Biology

DATA AUDITED BY: Ronni Krasny

Ronni Krasny

Director, Quality Assurance Unit

Table 1 -- The 24-, 48-, 72- and 96-hour LC50 values and 95% confidence intervals for the common mummichog (Fundulus heteroclitus) exposed to 7902.

LC50 (mg/ℓ)				No discernible effect concentration through 96 hours (mg/ℓ)
24 hour[a]	48 hour[b]	72 hour[b]	96 hour[b]	
>3600	2900 (2400-3600)[c]	2900 (2400-3600)	2500 (1700-3600)	<600[d]

a Empirally estimated.
b Estimated by the binomial probability method.
c 95% confidence intervals.
d Lowest concentration tested.

3M_MN01656897

Table 2 -- The pH, DO and salinity measurements made during the 96-hour toxicity test with 7902 and the common mummichog (Fundulus heteroclitus). Ranges of measurements made in replicate concentrations are reported.

	Nominal concentration (mg/ℓ)	0 hour	24 hour	48 hour	72-hour	96 hour
pH	3,600	8.1	8.0-8.1	6.7	6.7	6.7
	1,700	8.0	8.0	6.8	6.7-6.8	6.7-6.8
	600	8.0	7.9-8.1	7.0-7.1	7.1-7.2	6.9-7.2
	control	8.0-8.1	7.9-8.0	7.2-7.5	7.2-7.6	7.3-7.6
DO (mg/ℓ)	3,600	8.2(88)[a]	7.8-7.9(84-85)	3.1-3.7(33-40)	3.1-3.8(33-41)	3.0-4.6(32-48)
	1,700	8.0-8.4(86-88)	7.7-7.9(83-85)	4.5-5.2(48-56)	1.6-4.7(17-51)	1.5-4.6(16-49)
	600	8.2(88)	7.7(83)	6.0-7.3(65-78)	5.3-6.9(57-74)	4.9-7.1(53-76)
	control	8.0-8.3(86-89)	7.4-7.5(80-81)	7.1-8.1(76-87)	6.2-8.1(67-87)	6.8-8.0(73-86)
salinity (o/oo)	3,600	16	16	17	17	17
	1,700	15-16	15-16	16	17	16
	600	15	15	16	16-17	16
	control	15	15	16	17-17	16

[a] % of saturation at 19°C.

3M_MN01656898

Table 3 -- Concentrations tested and corresponding cumulative mortalities of the common mummichog (Fundulus heteroclitus) exposed to 7902 for 24, 48, 72 and 96 hours. Ten fish were exposed to each concentration, five fish per replicate.

Nominal[a] concentration (mg/ℓ)	Cumulative mortalities (A and B replicates) 24 hour	48 hour	72 hour	96 hour
3,600	0[b,c,d,e]	10	10	10
2,400	0[b,d]	1[b,d]	1[b]	2[b,d]
1,700	0[b]	0[b]	0	0[b,d]
1,000	0	0	0[b]	0[b]
600	0	0[b]	0[b]	0[b]
control	0	0	0	0

[a] Foam present on surface of all solutions, except the controls, throughout testing.
[b] Some fish were at the surface.
[c] Some fish had partially lost their equilibrium.
[d] Some fish were lethargic.
[e] Some fish were respiring rapidly.