

ACUTE TOXICITY TO FISH (BLUEGILL SUNFISH)

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 not noted. 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: Fish bioassay procedures, Standard Methods, 1970 edition.

Type: Flow-Through Acute

GLP: No

Year completed: 1972

Species: *Lepomis macrochirus*

Supplier: Commercial hatchery in Nebraska.

Analytical monitoring: Temperature, pH, conductivity, and DO

Exposure period: 24, 96, and 264 hours.

Statistical methods: TL₅₀ (incipient median tolerance limit) values calculated using a linear regression equation.

Test fish age: Juveniles

Length and weight: Average length = 56 mm
Average weight = 2.4 g (wet)

Loading: Not given

Pretreatment: None

Test conditions:

Dilution water: Aerated well water

Dilution water chemistry:

Hardness: 38 mg/L as CaCO₃

Lighting: Not given.

Stock and test solution preparation: Sufficient amounts of stock solution containing the compound initially added to each test vessel. Proportional diluter used to maintain desired concentrations.

Flow-through rate: Continuous at 5 L every hour.

Stability of the test chemical solutions: Not noted

Exposure vessels: 30 L

Number of replicates: 1

Number of fish per replicate: 30
Number of concentrations: seven plus a blank control
Water chemistry during the study:
 pH range (0-96 hours):
 7.1 – 7.1 (control exposure)
 6.8 – 6.9 (33.4 mg/L exposure)
 Temperature range (0-96 hours): 18 ± 1 °C
 Dissolved oxygen range (0-96 hours): 9.1-9.3 mg/L

RESULTS

Nominal concentrations: Blank control, 5.9, 7.9, 10.5, 14.1, 18.8, 25.0, and 33.4 mg/L
Element values: 96-hour TL50 = 20.4 (16.4 – 25.3) mg/L
 264-hour TL50 = 15.9 (12.5 – 20.2) mg/L
 264-hour NOEC = 5.9 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 FC-203 96-hour TL₅₀ for fathead minnow was determined to be 20.4 mg/L with a 95% confidence interval of 16.4 to 25.3 mg/L.

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

DATA QUALITY

Reliability: Klimisch ranking = 3. Testing was done using laboratory procedures not consistent with current regulatory approaches. 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 Bionomics, Inc., of Wareham, MA at the request of the 3M Company, St. Paul, MN, 1972.

OTHER

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METHODS AND MATERIALS

The investigation was performed at the aquatic toxicology laboratory of Bionomics, Inc., Wareham, Massachusetts. The susceptibility of bluegill (Lepomis macrochirus) to Light Water^(R) aqueous film forming foam (tested as 100% active) under dynamic conditions was reported as the incipient median tolerance limit (TL₅₀), the concentration of the test compound in water causing 50 percent mortality with no additional significant response (>10%) during the final 48 hours of exposure. The predicted TL₅₀ value and its 95% confidence intervals were arrived at by converting the concentrations tested and the corresponding observed percent mortalities to logs and probits, respectively. These values were then used to calculate a linear regression equation.

Test procedures for the dynamic bioassay are those described for fish Bioassay Procedures in the 1970 edition of Standard Methods (APHA). The bluegill were obtained from a commercial fish hatchery in Nebraska and had a mean weight of 2.4 g and a mean length of 56 mm. The dynamic bioassay was conducted using a continuous-flow proportional dilution apparatus (Mount and Brungs, 1967)¹.

¹Mount, D. I. and W. A. Brungs. 1967. A simplified dosing apparatus for fish toxicology studies. Water Research. 1:21.

The apparatus provides for intermittent introduction of seven concentrations of the test compound into test vessels and diluent water to a vessel serving as a control unit. Flow rate to each of the 30-liter test vessels was 5 l/hour throughout the test period.

The test diluent consisted of aerated well water of pH 7.1, total hardness 38 mg/l as CaCO_3 , and a constant temperature of 18C (± 1.0). Dissolved oxygen levels for the test ranged from 9.1 to 9.3 mg/l. Thirty specimens were introduced 48 hours prior to the start of the assay into each test unit. The desired concentrations of the test compound were established after the 48 hour acclimation period in the test vessels by adding sufficient amounts of stock solution containing the compound to each test vessel. The proportional dilution apparatus was then used to maintain the desired concentration of the compound in each test vessel.

RESULTS

The predicted TL_{50} values and 95% confidence intervals are presented in Table 1. The data for p,p' - DDT, determined at Bionomics by a static bioassay to serve as a "standard" indicate that the population of test animals is representative of an "average population". Table 2 presents a summary of observed mortality for bluegill after 24 and 96 hours

of exposure and end of test. Moribund fish generally became dark and lethargic, lost equilibrium, and expired. Table 3 presents the pH of selected concentrations from the test at 24 and 96 hours and end of test.

SUBMITTED BY:

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September, 1972

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Table 1 -- Acute toxicity of Light Water[®] to bluegill^a (Lepomis macrochirus). The data are based on dynamic bioassays conducted at the Fish Toxicology Laboratory of Bionomics, Inc. in Wareham, Massachusetts.

Compound	TL ₅₀ mg active ingredient/liter			No Ef Level (mg/l)
	24 hour	96 hour	Incipient ^b	
Light Water [®]	>33.4	20.4(16.4-25.3) ^c	15.9(12.5-20.2)	5.
DDT		0.008(0.004-0.012)		

^a Assay conducted at 18C ($\pm$ 1.0) mean weight of bluegill 2.4 g.

^b Incipient TL₅₀ estimated over 264 hours.

^c 95% confidence intervals.

Table 2 -- Concentrations tested and corresponding observed percent mortalities for bluegill (Lepomis macrochirus) exposed to Light Water[®] after 24 and 96 hours and end of test.

Concentration (mg/l)	% mortality observed		
	24 hour	96 hour	Incipient
			<u>264 hours</u>
33.4	0	100	100
25.0	0	30	64
18.8	0	17	50
14.1	0	10	20
10.5	0	0	17
7.9	0	0	3
5.9	0	0	0
Control	0	0	0

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Table 3 -- pH of selected concentrations from a dynamic bioassay conducted with bluegill (Lepomis macrochirus) exposed to Light Water^(R) after 24 and 96 hours and end of test.

Concentration (mg/l)	pH		Incipient
	24 hour	96 hour	
			<u>264 hours</u>
33.4	6.9	6.8	6.9
14.1	7.1	7.1	7.1
5.9	7.4	7.1	7.1
Control	7.1	7.1	7.1

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