

Tech Data Book  
Chemical Review

EC-200

10/19/72

LIOASSAY REPORT

Acute toxicity of Light Water<sup>®</sup> to  
bluegill (Lepomis macrochirus).

Bionomics, Inc.  
790 Main Street  
Wareham, Massachusetts  
September, 1972

**Exhibit  
1095**

State of Minnesota v. 3M Co.,  
Court File No. 27-CV-10-28862

3M\_MN01998404

1095.0001

## 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<sup>(R)</sup> aqueous film forming foam (tested as 100% active) under dynamic conditions was reported as the incipient median tolerance limit (TL<sub>50</sub>), 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 TL<sub>50</sub> 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)<sup>1</sup>.

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<sup>1</sup>Mount, 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<sub>3</sub>, and a constant temperature of 18C ( $\pm$  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<sub>50</sub> 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.

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

Compound	TL <sub>50</sub> mg active ingredient/liter			No Effect Level (mg/l)
	24 hour	96 hour	Incipient <sup>b</sup>	
Light Water <sup>®</sup>	>33.4	20.4(16.4-25.3) <sup>c</sup>	15.9(12.5-20.2)	5.9
DDT		0.008(0.004-0.012)		

<sup>a</sup> Assay conducted at 18C ( $\pm$  1.0) mean weight of bluegill 2.4 g.

<sup>b</sup> Incipient TL<sub>50</sub> estimated over 264 hours.

<sup>c</sup> 95% confidence intervals.

Table 2 -- Concentrations tested and corresponding observed percent mortalities for bluegill (Lepomis macrochirus) exposed to Light Water<sup>®</sup> 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

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

Concentration (mg/l)	pH		
	24 hour	96 hour	Incipient
			<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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Toxic Rate Bank - Clam P. cc JTL  
Chickens →

BIOASSAY REPORT

Acute toxicity of Light Water<sup>®</sup> to grass shrimp (Palaemonetes vulgaris), fiddler crab (Uca pugilator) and mummichog (Fundulus heteroclitus).

Bionomics, Inc.  
790 Main Street  
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September, 1972

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## METHODS AND MATERIALS

The investigations were performed at the aquatic toxicology laboratory of Bionomics, Inc. in Wareham, Massachusetts. The susceptibility of grass shrimp (Palaemonetes vulgaris), fiddler crab (Uca pugilator) and mummichog (Fundulus heteroclitus) to Light Water<sup>®</sup> was measured in terms of the median tolerance limit (TL<sub>50</sub>), the concentration of the chemical in water which causes 50 percent response under the test conditions. The criteria utilized in these studies was death. The prediction of a TL<sub>50</sub> value, and its 95% confidence interval, was based on conversion of the concentrations tested and the corresponding observed percent mortalities to logs and probits respectively, and the subsequent mathematical calculation of a linear regression equation. The sample tested was a clear amber liquid identified as Light Water<sup>®</sup> aqueous film forming foam and was tested as 100% active.

The test procedures used in this evaluation are in accordance with the Fish Bioassay Procedures described in the 1970 edition of Standard Methods (APHA). The fiddler crab, and mummichog assays were conducted in 5-gallon glass jars containing 15 liters of diluent. The grass shrimp were assayed in 1-gallon jars containing 3 liters of diluent. The grass shrimp, fiddler crab, and mummichog used in these investigations were collected locally by laboratory personnel.

The test species were observed in the laboratory hatchery facility for at least 10 days prior to testing. During that period, mortality in the test populations was less than 3% and these animals were judged to be in excellent physical condition. The mean length of the grass shrimp was 18 mm. The mean carapace width of the fiddler crabs was 15 mm. The mummichog had a mean weight of 0.75 g and a mean length of 22 mm. The bioassays were conducted at 18 C ( $\pm$  1.0) for 96 hours with a single introduction of toxicant and without aeration. The test diluent consisted of synthetic sea water<sup>1</sup>. Dissolved oxygen levels for the tests ranged from 5.0 - 8.4 mg/l. Ten specimens were assayed at each concentration.

## RESULTS

The TL<sub>50</sub>'s and 95% confidence intervals for the species tested are presented in Table 1. The concentrations of Light Water<sup>Ⓟ</sup> tested and the corresponding percent mortalities are shown in Table 2.

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<sup>1</sup>Laroche, G. R. Eisler, and C. R. Tarzwell, 1970. Bioassay procedures for oil and oil dispersant toxicity evaluations. J. W. Poll. Cont. Fed. 42 (11): 1982-1989.

Moribund grass shrimp and fiddler crabs became lethargic before expiring. Moribund mummichog darkened in color, displayed a general loss of equilibrium and fell to the bottom of the test vessel where they expired. Table 3 presents the pH of selected concentrations from each bioassay at 24 and 96 hours.

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Table 1 - The acute toxicity of Light Water<sup>®</sup> to some marine organisms at 24 and 96 hours. The data are based on results of acute bioassays<sup>a</sup> conducted at the aquatic toxicology laboratory of Bionomics, Inc. in Wareham, Massachusetts.

Species	TL <sub>50</sub> -mg active ingredient/liter		No Effect Level (mg/l)
	24 hour	96 hour	
grass shrimp <sup>b</sup> ( <u>Palaemonetes vulgaris</u> )	> 750.0	93.0(53.3-162.0) <sup>c</sup>	56.0
fiddler crab <sup>d</sup> ( <u>Uca pugilator</u> )	> 10,000.0	> 10,000.0	10-1-73 show > 10,000.0 water
mummichog <sup>e</sup> ( <u>Fundulus heteroclitus</u> )	111.0(75.4-164.0)	36.1(28.8-45.3)	18.0

<sup>a</sup>Bioassays conducted in synthetic sea water at 18 C ( $\pm$  0.5).

<sup>b</sup>Mean length of shrimp 18 mm.

<sup>c</sup>95% confidence intervals.

<sup>d</sup>Mean carapace width of crabs 15 mm.

<sup>e</sup>Mean weight of mummichog 0.75 g.

Table 2 - Concentrations of Light Water<sup>(R)</sup> tested and corresponding observed percent mortality of some marine organisms at 24 and 96 hours.

Species	Concentration (mg/l)	% Mortality	
		24 hour	96 hour
grass shrimp ( <u>Palaemonetes vulgaris</u> )	750.0	0	100
	490.0	0	100
	370.0	0	100
	280.0	0	100
	160.0	0	100
	87.0	0	20
	56.0	0	0
	Control	0	0
fiddler crab ( <u>Uca pugilator</u> )	10,000.0	0	0
	5,000.0	0	0
	1,000.0	0	0
	500.0	0	0
	250.0	0	0
	100.0	0	0
	Control	0	0

Table 2 - Continued

Species	Concentration (mg/l)	% Mortality	
		24 hour	96 hour
mummichog ( <u>Fundulus heteroclitus</u> )	240.0	100	100
	180.0	70	100
	75.0	30	100
	56.0	0	70
	42.0	0	60
	32.0	0	30
	24.0	0	30
	18.0	0	0
Control	0	0	0



Table 3 - The pH of selected concentrations from bioassays with some marine organisms exposed to Light Water<sup>®</sup> for 24 and 96 hours.

Species	Concentration (mg/l)	pH	
		24 hour	96 hour
grass shrimp ( <u>Palaemonetes vulgaris</u> )	750.0	7.2	7.1
	370.0	7.2	7.0
	160.0	7.2	7.0
	Control	8.0	8.0
fiddler crab ( <u>Uca pugilator</u> )	10,000.0	6.8	7.1
	1,000.0	7.0	7.1
	100.0	7.1	7.3
	Control	8.1	8.0
mummichog ( <u>Fundulus heteroclitus</u> )	240.0	7.0	7.2
	75.0	7.0	7.2
	24.0	6.9	7.1
	Control	8.0	8.0