

ACUTE TOXICITY OF CADMIUM TO BLUEGILL  
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BIONOMICS



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BY

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## INTRODUCTION

The current concern regarding the protection of aquatic life in surface waters has prompted the evaluation of the effects of exposure to chemicals on aquatic organisms.

The primary objective of these studies was to provide the Environmental Protection Agency with information to evaluate the relative susceptibility of aquatic organisms to acute exposure to cadmium. The acute toxicity of cadmium to bluegill and rainbow trout in both a soft and a hard water, and to pink shrimp in sea water was estimated during static bioassays.

The bioassays with fishes were conducted at the Aquatic Toxicology Laboratory of E G & G, Bionomics, Wareham, Massachusetts. The shrimp bioassay was conducted at the Marine Research Laboratory of E G & G, Bionomics, Pensacola, Florida.

MATERIALS AND METHODS

The methodology for acute toxicity testing of fishes and shrimp closely followed the recommended bioassay procedures except for certain conditions described below.

The chemical evaluated in these bioassays was cadmium, as cadmium chloride ( $\text{CdCl}_2$ , 61.25% cadmium), a white anhydrous powder manufactured by Matheson, Coleman & Bell (lot #Z273). Results for all tests were expressed as the median lethal concentration (LC50), the nominal concentration of the test compound in water causing 50 percent mortality of test animals. The LC50 value and its 95% confidence interval were calculated by converting the test concentrations and the corresponding observed percent response to logs and probits, respectively. These values were then utilized in a least squares regression analysis, and the LC50 value and its confidence interval were estimated from the calculated regression equation.

The animals used in these tests were bluegill (Lepomis macrochirus), rainbow trout (Salmo gairdneri) and pink shrimp (Penaeus duorarum). The bluegill were acquired from

a commercial fish hatchery in Nebraska, and had a mean wet weight of 1.1 g and a mean standard length of 37 mm. The rainbow trout were obtained from a commercial fish farmer in Washington, and had a mean wet weight of 1.0 g and a mean standard length of 32 mm. The shrimp were collected by laboratory personnel from Big Lagoon in Pensacola, Florida and had rostrum-telson lengths of 35-50 mm.

The bluegill and rainbow trout were held in 1700-l concrete raceways which are coated with an epoxy resin paint to prevent leaching of materials into the water. Flow of well water (having a temperature of  $21 \pm 1.0^{\circ}\text{C}$  for the bluegill, and  $12 \pm 1.0^{\circ}\text{C}$  for the rainbow trout) into these raceways was at a minimum flow of 4 l/minute, providing an adequate rate of turnover for holding these species. This water had a hardness of 35 mg/l as  $\text{CaCO}_3$ , a pH of 7.1 and a dissolved oxygen concentration of at least 6.0 mg/l (60% of saturation). These species were maintained in the laboratory hatchery facilities for at least 30 days prior to testing. During the 30 day period, mortality was <2%; no mortality was observed during the 48 hours immediately prior to testing, and these fish were judged to be in excellent condition. The shrimp were held in 1100-l fiberglass tanks in constantly flowing filtered (10 micrometers)

natural sea water. The salinity of this water was 25 parts per thousand (o/oo) and the temperature was  $20 \pm 1.0^{\circ}\text{C}$ .

The static bioassays were conducted in 19.6-l wide-mouth soft-glass bottles containing 15 liters of test solution. Exposure mixtures for the bluegill bioassays were maintained in water baths at  $21 \pm 1.0^{\circ}\text{C}$  by immersion coil heaters and mercury column thermoregulators. Test solutions for the rainbow trout and shrimp were maintained in water baths at  $12 \pm 1.0^{\circ}\text{C}$  and  $20 \pm 1.0^{\circ}\text{C}$ , respectively, by use of commercial refrigeration units. Each species was from the same year class, and the standard length of the longest fish or shrimp was no more than two times that of the shortest fish or shrimp. The bluegill and rainbow trout were acclimated to test conditions of temperature and water quality over a 96-hour period prior to testing. These species were not fed during the 48 hours immediately prior to testing or during the tests. The shrimp were acclimated to test conditions of water quality and temperature for at least seven days prior to testing. Water in the test vessels was not aerated. The test compound dissolved in distilled water was added to each jar in the bluegill and rainbow trout bioassays. In the shrimp bioassays, the test material was introduced into each jar directly. Animals were introduced into the test vessel within 30 minutes

after the compound was added. Ten bluegill or rainbow trout were randomly assigned to each test vessel. Ten shrimp (2 replicates, 5 animals/vessel) were exposed to each concentration.

The dilution water used in the fish bioassays was the same as previously described for holding these fish. The hard water for these bioassays was prepared by adding 192 mg of  $\text{NaHCO}_3$ , 120 mg of  $\text{CaSO}_4$ , 120 mg of  $\text{MgSO}_4$ , and 8 mg of KCl per liter of deionized water. The resulting water had a pH of 7.6 and a total hardness of 200 mg/l as  $\text{CaCO}_3$ . The dilution water for the shrimp bioassays consisted of filtered (10 micrometers) natural sea water with a salinity of 25 o/oo and a pH of  $8.0 \pm 0.5$ . Concentrations of dissolved oxygen were measured with a combination temperature-oxygen probe and meter in selected concentrations at 24, 48, and 96 hours of exposure.

Two series of concentrations were established within a bioassay, a series of range-finding (preliminary) concentrations and a series of definitive concentrations. The preliminary test was conducted to determine the approximate range of concentrations for evaluating the dose-response relationship. The definitive test, consisting of at least

five concentrations, evaluated the dose-response relationship to a degree allowing the LC50 to be calculated from the data with optimum accuracy. A control, which consisted of the same dilution water, conditions, procedures, and organisms, was maintained for each species tested.

### RESULTS AND DISCUSSION

The estimated LC50 values (95% confidence intervals) for cadmium and the species tested are presented in Table 1 along with the highest nominal concentration tested at which there were no discernible effects on test animals due to exposure to cadmium. A summary of observed mortality for each individual test concentration at 24, 48 and 96 hours of exposure to cadmium is also presented (Table 2). The mortality syndrome among fish from those concentrations where mortality was observed was similar. Fish generally became dark and lethargic, lost equilibrium, and expired. Affected shrimp generally lost equilibrium, swam erratically, and died.

The concentrations of dissolved oxygen, measured at 0, 24, 48 and 96 hours, are presented in Table 3. Final pH was  $7.0 \pm 0.5$  for all test concentrations and controls where



bluegill and rainbow trout were exposed in soft water. Comparable pH's for the test concentrations where bluegill and rainbow trout were exposed in hard water were  $7.5 \pm 0.5$ . Final pH was  $8.0 \pm 0.5$  for all test concentrations and the control for the shrimp bioassay.

Cadmium appeared to be more toxic in soft water than in hard water, indicating the effect of water quality was on the toxicant and not on the test species. The bluegill exhibited a 96-hour LC50 value of 6.62 mg/l in soft water, and 48.2 mg/l in hard water. A similar situation was observed for rainbow trout tested in soft and hard water. The 96-hour LC50 value was 0.007 mg/l in soft water, while that for the hard water bioassay was 0.016 mg/l. The 96-hour LC50 value for pink shrimp was 4.9 mg/l.

LITERATURE CITED

A.P.H.A. 1971. Standard Methods for the Examination  
of Water and Wastewater. 13th Edition, 874 pp.

Table 1 -- Acute toxicity of cadmium<sup>a</sup> to bluegill<sup>b</sup> (Lepomis macrochirus), rainbow trout<sup>c</sup> (Salmo gairdneri), and pink shrimp<sup>d</sup> (Penaeus duorarum). These data are based on the results of bioassays conducted at the Aquatic Toxicology Laboratory of E G & G, Bionomics, Wareham, Massachusetts and Pensacola, Florida.

Species/ diluent	LC50 (mg active ingredient/l)			No discernible effect level (mg/l)
	24 hour	48 hour	96 hour	
bluegill/ soft water	11.20 (6.98-17.80) <sup>e</sup>	7.41 (5.43-10.10)	6.62 (5.45-8.05)	4.20
bluegill/ hard water	76.3 (61.1-95.4)	48.2 (39.6-58.6)	48.2 (39.6-58.6)	32.0
rainbow trout/ soft water	0.94 (0.32-2.73)	0.030 (0.016-0.056)	0.007 (0.005-0.011)	0.0018
rainbow trout/ hard water	>7.50	0.043 (0.025-0.073)	0.016 (0.011-0.023)	0.012
pink shrimp/ sea water	17 (13-23)	8.6 (5.6-12)	4.9 (2.9-8.6)	<2.0

a  
Cadmium chloride (CdCl<sub>2</sub>), 61.25% cadmium.

b  
Bioassays conducted at 21 ± 1.0°C, mean wet weight of bluegill, 1.1 g.

c  
Bioassays conducted at 12 ± 1.0°C, mean wet weight of rainbow trout, 1.0 g.

d  
Bioassays conducted at 20 ± 1.0°C, rostrum-telson lengths of pink shrimp, 35-50 mm.

e  
95% confidence interval.

Table 2 -- Concentrations tested and corresponding observed percentage mortalities at 24, 48 and 96 hours for bluegill (Lepomis macrochirus), rainbow trout (Salmo gairdneri) and pink shrimp (Penaeus duorarum).

Species/ diluent	Nominal concentration (mg/l)	% mortality observed		
		24 hour	48 hour	96 hour
bluegill/ soft water	18.0	100	100	100
	10.0	20	100	100
	7.5	0	40	50
	5.6	0	0	10
	4.2	0	0	0
	control	0	0	0
bluegill/ hard water	140.0	100	100	100
	100.0	60	100	100
	75.0	40	100	100
	56.0	0	60	60
	42.0	10	30	30
	32.0	0	0	0
	control	0	0	0

Table 2 -- Continued.

Species/ diluent	Nominal concentration (mg/l)	% mortality observed		
		24 hour	48 hour	96 hour
rainbow trout/ soft water	3.2000	90	100	100
	1.8000	60	100	100
	1.0000	40	100	100
	0.5600	20	100	100
	0.3200	40	100	100
	0.1000	10	80	100
	0.0320	0	50	100
	0.0180	0	40	80
	0.0100	0	40	70
	0.0075	0	0	30
	0.0056	0	0	20
	0.0042	0	0	20
	0.0024	0	0	10
	0.0018	0	0	0
control		0	0	0
rainbow trout/ hard water	7.500	0	100	100
	3.200	0	100	100
	1.000	0	100	100
	0.320	0	100	100
	0.075	0	50	100
	0.042	0	30	100
	0.024	0	40	100
	0.016	0	40	80
	0.012	0	0	0
	control		0	0

Table 2 -- Continued.

Species/ diluent	Nominal concentration (mg/l)	% mortality observed		
		24 hour	48 hour	96 hour
pink shrimp	34.0	60	100	100
	20.0	20	100	100
	11.0	10	10	80
	6.1	0	20	40
	2.0	0	0	10
	control	0	0	0

Table 3 -- Measured concentrations of dissolved oxygen during 96-hour exposures of bluegill (Lepomis macrochirus), rainbow trout (Salmo gairdneri) and pink shrimp (Penaeus duorarum) to cadmium.

Species/ diluent	Nominal concentration (mg/l)	Dissolved oxygen (mg/l and % of saturation)			
		0 hour	24 hour	48 hour	96 hour
bluegill/ soft water	18.0	8.5 (96)	- <sup>a</sup>	-	-
	7.5	8.2 (92)	6.2 (67)	5.8 (63)	4.5 (49)
	4.2	8.1 (91)	5.8 (63)	4.7 (52)	2.7 (31)
	control	8.4 (95)	5.9 (64)	4.7 (52)	4.2 (46)
bluegill/ hard water	140.0	8.4 (95)	-	-	-
	56.0	8.2 (92)	6.9 (76)	6.4 (71)	6.6 (73)
	32.0	8.0 (90)	6.5 (72)	5.5 (61)	5.5 (61)
	control	8.4 (95)	5.8 (63)	5.2 (56)	4.1 (45)
rainbow trout/ soft water	3.2000	9.2 (85)	7.7 (70)	-	-
	0.0320	9.1 (83)	9.1 (83)	6.9 (63)	-
	0.0056	8.8 (81)	8.7 (80)	8.0 (73)	6.0 (55)
	control	8.8 (81)	8.8 (81)	7.8 (71)	5.8 (53)
rainbow trout/ hard water	7.500	9.5 (87)	9.1 (83)	-	-
	0.320	9.2 (84)	9.2 (84)	-	-
	0.012	9.0 (83)	6.2 (57)	5.3 (49)	4.6 (42)
	control	9.1 (84)	6.4 (58)	5.7 (52)	5.0 (46)

Table 3 -- Continued.

Species/ diluent	Nominal concentration (mg/l)	Dissolved oxygen (mg/l and % of saturation)			
		0 hour	24 hour	48 hour	96 hour
pink shrimp/ sea water	34.0	6.8 (89)	- <sup>a</sup>	-	-
	11.0	6.7 (88)	6.3 (83)	5.7 (75)	5.7 (75)
	2.0	6.8 (89)	6.6 (87)	6.1 (80)	4.4 (58)
	control	6.8 (89)	6.8 (89)	6.9 (79)	3.5 (46)

<sup>a</sup>

Dissolved oxygen not measured due to 100% mortality.