Bull Environ Contam Toxicol (1984) 32:565–569

Environmental Contamination and Toxicology

Antimony and Thallium Toxicity to Embryos and Larvae of Fathead Minnows (*Pimephales promelas*)

Gerald A. LeBlanc[1] and Jerry W. Dean

EG & G Bionomics, 790 Main Street, Wareham, MA 02571

The establishment of water quality criteria by the U.S. Environmental Protection Agency for a potential aquatic pollutant requires the development of data regarding the chronic toxicity of the material (U.S. EPA 1976). Full life cycle toxicity tests with many species of fish require exposures lasting nine months or longer (Macek et al. 1976). The embryos and early larval stages of fish have been identified as the stages most susceptible to the toxic effects of many toxicants (McKim 1977; Macek and Sleight 1977). Tests involving the exposure of these stages of fish development have been accepted as a means of predicting chronic toxicity (U.S. EPA 1982).

Antimony, as antimony trioxide, is a commercially important metal used as a flame retarding agent. Thallium has industrial application as a catalyst for many organic reactions and in the production of alloys and electronic devices. Accordingly, both metals are potential pollutants through anthropogenic sources. The purpose of this study was to assess the effects of these metals on the embryos and larvae of fathead minnows (Pimephales promelas).

MATERIALS AND METHODS

These tests were performed according to procedures described by U.S. EPA (1972). Antimony solutions were prepared using antimony trioxide. Thallium solutions were prepared from thallium sulfate. Dilution water was well water which was extensively aerated prior to use. The water had a pH range of 6.7-7.1, a total hardness of 28-40 mg/L as $CaCO_3$ and a specific conductance of 140-170 umhos/cm. Proportional diluters (Mount and Brungs 1967) were used to deliver the solutions of

[1]Present address: Biology Dept., University of South Florida, Tampa, FL, 33620, USA, Correspondence and reprint requests.

test metal and dilution water to the aquaria. Aquaria were constructed of glass and measured 40X20X25 cm. The aquaria rested in a water bath containing circulating water heated by immersion coil heaters and regulated by a mercury thermoregulator. Illumination was provided with Cool White fluorescent lights centrally located above the aquaria. Light intensity was 3 to 14 hectolux at the water surface and was maintained on a 12-hour photoperiod. Solutions were delivered to the aquaria at a minimum rate of 6 aquarium volume replacements per day. Mariotte bottle-dipping bird delivery devices (Lemke et al. 1978) were used to deliver the metals to the mixing chamber of the diluters. Antimony was dissolved in distilled water containing 1% HCl to facilitate solubilization. Thallium was dissolved in distilled water.

Exposure concentrations were based on preliminary acute toxicity tests where no fathead minnows died after 96-hours exposure to a measured antimony concentration of approximately 4 ug/L. The 96-hour LC50 value for thallium was 860 ug/L. The tests were initiated with eggs obtained within 48 hours after fertilization. Fifty-five eggs (for antimony) or fifty eggs (for thallium) were impartially distributed to each of 12 embryo incubation chambers for each test. The embryo incubation chambers were glass cups (5 cm O.D., 8 cm high) with 40 mesh Nitex screened bottoms. The embryos were incubated in the test solutions according to Mount (1968). Each concentration was replicated 1X.

To initiate the 30-day post-hatch larvae exposure, 40 larvae were impartially selected from each incubation chamber and transferred to the respective aquarium upon completion of hatching. Larvae were fed live brine shrimp (Artemia salina) nauplii three times daily. At 30-days post-hatch, the larvae were anesthetized with MS-222 (tricaine methane-sulfonate) and percentage survival, mean total length, and average wet weight were determined. The larvae were measured individually to calculate the mean total length while the larvae from each aquarium were group weighted to calculate average wet weight. The biological parameters measured were analyzed using ANOVA and Dunnett's procedure (Steel and Torrie 1960) to determine significant ($p=0.05$) differences from the controls.

Dissolved oxygen concentrations, pH values, and temperatures of the test solutions were measured daily. Water samples (100 ml) were taken from each aquarium on test day 0, day of hatch (3-5), and weekly thereafter. These samples were analyzed for antimony or thallium concentrations using atomic absorption spectrophotometry

(U.S. EPA 1976).

RESULTS AND DISCUSSION

The temperature of the test solutions was maintained at 25±1 C during these tests. The mean (and standard deviation) dissolved oxygen concentrations and pH range of test solutions during the exposure to antimony was 8.7 (0.4) mg/L and 6.2-7.3, respectively. The mean (and standard deviation) dissolved oxygen concentrations and pH range of test solutions during the exposure to thallium was 8.4 (0.4) mg/L and 6.7-7.2, respectively. These values remained within acceptable ranges for the normal survival and growth of fathead minnows.

Fathead minnow embryos hatched normally when exposed to antimony concentrations as high as 7.5 ug/L (Table 1). Survival and growth of larvae was also unaffected from exposure to antimony concentrations as high as 7.5 ug/L. These data indicate that solutions saturated with antimony, as antimony trioxide, are not toxic to fathead minnows.

No fathead minnow embryos survived exposure to 720 ug/L thallium (Table 1). Significantly fewer embryos exposed to 350 ug/L successfully hatched. Embryos were not affected from exposure to thallium concentrations as high as 200 ug/L. No fathead minnow larvae survived exposure to 350 ug/L thallium. Larvae survival was significantly reduced from exposure to thallium concentrations as low as 40 ug/L. Larvae growth was significantly reduced from exposure to thallium concentrations of 200 and 120 ug/L. Based on the reduced survival of fathead minnow larvae exposed to thallium concentrations as low as 40 ug/L, the Maximum Acceptable Toxicant Concentration (MATC) for thallium , as thallium sulfate, was estimated to be less than 40 ug/L.

Toxicity and water solubility of metals vary depending upon the metal species (LeBlanc et al. 1983; Windholz 1976). LeBlanc et al. (1983) demonstrated that the toxicity of silver varied tremendously depending upon the ligand to which it was bound. The effects of antimony, as antimony trioxide, on fathead minnows were assessed during this study at the highest concentrations which could be consistently maintained in solution. The data suggests that antimony in this form poses no threat to the survival of fathead minnows.

Thallium, however, exibited considerable chronic toxicity. Chronic toxicity can be quantitated by calculating the Acute-Chronic Ratio (ACR). The ACR is

calculated by dividing the 96-hour LC50 of the test material by the geometric mean of the MATC. The ACR for thallium, as thallium sulfate, was greater than 22. An absolute value cannot be applied to the ACR since a no-effect concentration was not determined during the test. Xenobiotic metals tend to exibit considerable chronic toxicity (lead ACR= 49, Holcombe et al. 1976; nickel ACR= 50, Pickering 1974; cadmium ACR= 130, Pickering and Gast 1972). The chronic toxicity of thallium appears to be within the range of ACR's for these metals.

Table 1. The effects of antimony and thallium on the embryos and larvae of fathead minnows (Pimephales promelas)

Measured Concentration[a] (ug/L)	Hatch[a] (%)	30 Days Post Hatch Survival[a] (%)	Length[a] (mm)	Weight[a] (mg)
ANTIMONY				
7.5(0.2)	92(9)	84(16)	22(3)	80(14)
3.2(0.8)	84(5)	92(4)	22(2)	71(4)
1.4(0.4)	92(5)	94(6)	22(2)	71(4)
0.74(0.2)	96(0)	74(37)	23(3)	100(36)
0.62(0.4)	87(3)	95(7)	22(2)	76(5)
HCl control	94(4)	89(6)	22(2)	82(0)
Control	92(4)	94(8)	22(2)	72(2)
THALLIUM				
720(20)	0(0)[b]	-	-	-
350(50)	22(17)[b]	0(0)[b]	-	-
200(40)	75(1)	26(9)[b]	12(2)[b]	28(1)[b]
120(10)	79(1)	74(5)[b]	16(3)[b]	66(25)
40(10)	77(1)	60(0)[b]	25(2)	130(16)
Control	75(7)	87(0)	24(1)	104(1)

[a]Mean (and standard deviation).
[b]Significantly different from the control.

Acknowledgement. This study was performed under U.S. EPA contract number 78-01-4646.

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Received September 10, 1983; Accepted October 5, 1983