same time, thus removing infected oysters that might carry the disease to future planting.

**LITERATURE CITED**


**Acute Toxicity of Zinc to the Killfish, Fundulus heteroclitus**

**ABSTRACT:** Groups of adult mummichogs, *Fundulus heteroclitus*, were subjected to different concentrations of zinc as zinc chloride at 24% salinity, pH 8.0 and 20°C. At 192 hours, all mummichogs exposed to 43 ppm or less of zinc were alive and outwardly indistinguishable from the controls. Among these survivors there were no appreciable differences at 192 hours in zinc content of various tissues, as determined by atomic absorption spectroscopy, regardless of the initial amount of zinc in solution. All mummichogs subjected to 157 ppm or 180 ppm of zinc died between 24 and 48 hours.

Dead mummichogs contained about 7 and 8 times more zinc in whole fish and in gill arch, respectively, than controls and this affords an index of zinc-caused mortality for the species.

**Introduction**

During preliminary experiments on effects of sublethal concentrations of insecticides to marine organisms, I observed that zinc and other metals accumulate in selected tissues of several species. Literature on the toxicity of zinc to aquatic organisms, as reviewed by Skidmore (1964), notes that levels of zinc in solution lethal to 50% of various species of freshwater molluscs and fishes in 96 hours (LC-50, 96 h) range from 0.3 to 12.3 ppm. However, quantitative data on the toxicity of zinc to marine fishes are lacking. This account reports on effects of different levels of zinc in seawater on the mummichog, *Fundulus heteroclitus*, an euryhaline cyprinodontiform fish, during a period of 192 hours, including effects on mortality, uptake of zinc from solution, and accumulation of zinc by selected tissues. I am indebted to George R. Gardner and Philip H. Edmunds for technical assistance throughout the study.

**Methods**

The experiment was conducted in a windowless room at 20°C under subdued artificial light. The test medium was seawater of 24% salinity and pH 8.0 pumped from an underground well (Clark and Eisler, 1964). Aquaria were 20-liter glass jars filled with 19 liters of test medium. Each jar was covered with a glass disc and aerated via 6 mm glass tubing inserted through a hole in the disc; dissolved oxygen levels ranged from 7.2 to 7.4 ppm.

Test fish were adult mummichogs, mean weight 4.5 g, from a single seine collection in Sandy Hook Bay, N. J. During a two week acclimatization period, mummichogs were held in large holding aquaria filled with the test medium and fed adult brine shrimp.

A saturated solution of zinc chloride was prepared by dissolving metallic zinc in concentrated hydrochloric acid. Using serial dilution, 2 ml of ZnCl2 were added to each jar to produce zinc concentrations calculated to range from 0.8 to 180 ppm. After 30 minutes, 70 mummichogs were distributed, in groups of five, among 14 jars (7 concentrations, including controls, in replicate). Food was withheld for the balance of the study. Immediately before the fish were added, aliquots from each jar were analyzed for zinc by a Perkin-Elmer Model 303 atomic absorption spectrophotometer according to methods outlined in the manual accompanying the instrument. Water from each jar was analyzed for zinc every 24 hours during the 192-hour study.

Dead animals were removed daily, blotted dry, and separated into liver, gill arch, and remainder (primarily carcass). For each jar, like tissues were pooled, placed in preweighed porcelain crucibles, charred and then ashed in a muffle furnace at 600°C for 24 hours. The weighed ash was dissolved in 0.1 ml of concentrated HCl, and filtered through Whatman No. 42 paper. Each crucible was rinsed with two 3 ml aliquots of 6N HCl and two 3 ml aliquots of distilled water. Each wash was filtered and added to the original filtrate. Samples were diluted to 25 ml with distilled water and analyzed for zinc by atomic absorption. Animals surviving the 192-hour exposure were killed, and their tissues analyzed for zinc in the manner described.

**Results**

Levels of zinc, in ppm, in test jars immediately before fish were added were 0.78, 3.5, 9.3, 43.0,


Adult mummichogs can tolerate seawater solutions containing up to 43 ppm of zinc for 192 hours without apparent ill effects, and this indicates that mummichogs are more resistant to zinc than various species of freshwater fishes for which data are available. For example, the LC-50 (96-h) values for zinc to bluegills range from 2.9 to 12.3 ppm (Skidmore, 1964). Moreover, some deaths occur in 96 hours among 5 species of freshwater fishes, primarily centrachids, at zinc concentrations between 5 and 35 ppm (Mount, 1964). A strict comparison between susceptibility to zinc of marine and of freshwater fishes is not feasible, primarily because of differences in the salinity of the test medium; viz, zinc is demonstrably less toxic to freshwater teleosts in hard water than in soft water (Cairns and Scheier, 1957; Lloyd, 1960; Herbert and Wakeford, 1964).

Figure 1 suggests that mummichogs lose zinc to media which contains less than 3.5 ppm of zinc and remove zinc from more concentrated media. The reasons for this are unknown, but may be associated with a short (two weeks) acclimatization period to media containing comparatively little (0.075 ppm) zinc. Saiki and Mori (1955) observed a similar phenomenon in clams. They found that clams that have taken up zinc—65 from seawater lost 40% within two days after return to zinc-free seawater.

Mummichogs start to accumulate zinc from the medium when the concentration approaches 6.5 ppm (Fig. 1) or about 1/10 of the LC-50 (96-h) value. Although accumulation of zinc by fishes from both saline and freshwater media is well-documented (Skidmore, 1964), Herbert and Wakeford (1964) report that toxicity of zinc to salmonids in seawater is at a minimum in a solution approximately isotonic with the fish's blood or about 1/10 of a lethal dose.

The high concentration of zinc in gill arch of mummichogs not surviving exposure of 157 or 180 ppm of zinc (Fig. 2) was expected. Mount (1964) states that bony parts of freshwater fish accumulate zinc at the expense of other tissues. Joyner and Eisler (1961), in work with rapidly-growing salmon fingerlings, aver that the bone surface acts as an ion exchange bed that readily absorbs zinc and that overgrowth of normal bone tissue seals in and isolates zinc.

Data from the present study on zinc content in gill arch and in total animal can be used to determine zinc-caused mortality among mummichogs in marine and estuarine environments. This procedure can probably be extended to establish a quantitative method of determining zinc-caused mortality among other species of marine fishes and invertebrates.