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# Toxicity of Pulsed Monochloramines to Goldfish (*Carassius auratus*)

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Intermittent chlorination employed as a biocide in water treatment and antifouling chemical by industry (particularly electric generating stations) has the potential for large scale negative impact on biota of receiving waters. In addition to free chlorine which is seldom detected in most treated wastewater and treatment plant effluents (BRUNGS 1973) chlorine reacts with water to form hypochlorus and hydrochloric acids. In the presence of ammonia varying amounts of inorganic chloramines are created including mono-, di- and trichloramines. The relative amounts of these three chloro-derivatives are mainly dependent upon pH and initial ammonia concentration (MERKINS 1958). In alkaline waters and when the initial molar ratio of $Cl_2$:$NH_3$<1.0, a mass conversion to $NH_2Cl$ (monochloramine) occurs. Although the effects of monochloramine exposure are not fully known, it is a potential hazard to non-target organisms at all trophic levels of aquatic environments (MERKINS 1958, ARTHUR and EATON 1971, ZILLICH 1972, TSAI 1973, JOHNSON et al. 1977, LARSEN et al. 1977 and SEEGERT et al. 1979). The biota of most receiving waters experience pulses of chlorine and chloro-derivatives rather than uniform concentrations; however, most toxicity studies present relatively constant, short term (i.e. 24 h ) exposures. Therefore, these data are not directly applicable to existent field conditions. Our research was designed to determine the toxicity of pulsed monochloramine concentrations to a hardy species, the goldfish, Carassius auratus.

## MATERIALS AND METHODS

Goldfish (2.6 to 27.4 g) were obtained during spring from a state hatchery in Fort Worth, Texas. Prior to tests fish were held in the laboratory in 200-l tanks containing dechlorinated, continuously filtered tapwater. Holding temperatures equalled 22°C ( = hatchery water temperature) except for one group which was slowly acclimated to 32°C. Hatchery and holding waters were analyzed for monochloramines to insure their absence. Amperometric titration (STANDARD METHODS 1971) was employed to quantify all monochloramine concentrations. Monochloramine concentrations were prepared by mixing a predetermined amount of ammonium hydroxide and calcium hypochlorite in 20 l of water. At 22°C, goldfish were exposed to the following nominal concentrations (mg/l): 10, 8, 4, 2, 1.75, 1.5, 1.0, 0.5, 0.1, 0.01, 0.001 and control (0.000).

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Chemicals were added to 22-l test tanks, mixed by gentle aeration and $NH_2Cl$ concentrations were determined. After one hour, five or six fish were randomly distributed into each tank. Temperatures equalled 22 or 32°C and were controlled ( ± 0.1 C) by circulating thermoregulators mounted in an outer water bath.

Checks of chloramine concentrations and fish were made at 1, 2, 4 and 8 h and at 6 h intervals thereafter for a total of 96 h . Fish behavior and physical changes were also observed during these times. When the monochloramine levels fell below nominal concentration, a "pulse" of monochlorine was added to return concentration as near as possible to the nominal concentration. In a few cases, the pulse increased concentrations above nominal levels, but these were reduced by increasing air flow into the tank.

## RESULTS AND DISCUSSION

Since monochloramine concentrations varied, exposure concentrations were quantified as both 2 h mean values and maximum concentration during each 96 h exposure. A total of 26 trials (135 fish) were conducted at 22°C. Chloramine concentrations of ≤0.5 mg/l resulted in no mortalities. Several fish exhibited stress including air gulping, excessive mucus production and rapid operculation at concentrations approaching 1.0 mg/l; however, in 5 trials of 26 fish only 2 died. A "lethality threshold" appeared at approximately 1.25 ± 0.10 mg/l. In two of the trials at this concentration, no mortalities occured in 96 h ; however, 100% lethality resulted in two other trials, even though mean, minimum and maximum chloramine concentrations were not appreciably different among the four trials. Median lethal tolerance times (TLm) equalled 37 and 62 h in these latter trials. Both mean and maximum chloramine concentrations were slightly higher in the 37 h TLm group than the 62 h TLm group. These findings support those of LARSEN et al. (1977) for coho salmon, Oncorhynchus nerka. They reported that lethality rose rapidly from 0 to 100% with only minor increases in chloramine concentration. In all 9 trials at mean chloramine concentrations ≥ 1.5 mg/l, 100% mortality occurred before 96 h had elapsed.

Over a range of mean chloramine concentrations of 1.13 to 9.20 mg/l, logarithmically transformed TLm times and chloramine concentrations (Figure 1) were significantly inversely correlated ($r = -0.943$; ANOVA $F = 71.7$, $p << 0.001$). The best fit, least squares regression equation equalled: Log TLm (h ) = 1.80 - 1.94 Log concentration (mg/l). Standard error for the slope was ± 0.23.

This model predicts 24, 48, 72 and 96 h TLms of 1.64, 1.15, 0.93 and 0.80 mg/l. These are similar to 96 h LC-50 values reported for other nonsalmonid, freshwater fish species (see references in introduction). A major problem in directly comparing results of earlier studies relates to differences in physical-chemical parameters (eg. temperature, pH and alkalinity), experimental design particularly in the presentation of chloramine (continuous flow, static, frequency of pulses), test duration (24 h to 21 weeks), life history stage and species. Our results for goldfish are similar to those reported by SEEGERT et al. (1979) for carp, Cyprinus carpio, also a member of the family Cyprinidae. They reported LC-50 values of 1.82 mg/l at 20°C and 1.50 at 30°C for

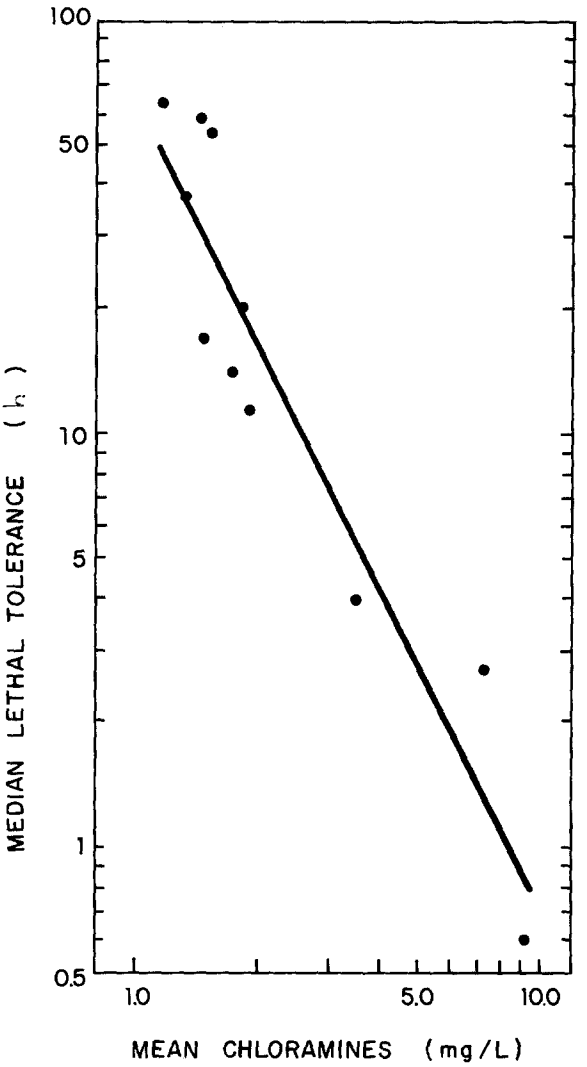


FIG. 1. Relationship between median lethal tolerance times and mean chloramine concentrations

carp subjected to four 4 h exposures of chloramine at 5 h intervals over 24 h . At these temperatures, carp were the most resistant to chloramines of 10 riverine species examined.

In 1975, CAIRNS et al. concluded from available data that the relationship between temperature and chlorine toxicity could not be predicted. We examined the influence of temperature on chloramine toxicity by exposing 32°C acclimated goldfish to a concentration (approximately 1 mg/l) that was sublethal to 22°C acclimated fish. In 5 trials with mean concentrations ranging from 0.71 to 0.99 mg/l, only 2 of 26 fish died at 22°C. Conversely, in two 96 h exposures at 32°C (mean concentrations of 0.91 and 0.93 mg/l), 9 of 11

goldfish died. The TLm for these trials equalled 41 and 68 h . GROTHE and EATON (1975) reported that the mode of chloramine lethality is tissue anoxia produced by a great increase in the proportion of blood methemoglobin and possible inhibition of methemoglobin reductase system. Given this mode of action, it is not surprising that chloramine toxicity is enhanced by increased temperature. At higher temperatures, ambient oxygen tension is less and tissue oxygen demand is increased. Since KLICKA (1965) found that metabolic rates of goldfish approach the van't Hoff rule (i.e. $Q_{10} \simeq 2.0$), tissue demands for oxygen at 32°C would be approximately twice those at 22°C. Higher temperatures would also augment the rate of chloramine uptake via gill surface. Our results for goldfish corroborate those of SEEGERT et al. (1979) for 10 riverine fish species.

Our results suggest that chloramine LC-50 values for goldfish, similar to the carp, are approximately an order of magnitude higher than those for salmonid species, and that resistance times for goldfish are inversely related to exposure temperatures.

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