# Toxicity of Methylmercury and Copper, Applied Singly and Jointly, to the Blue Gourami, *Trichogaster trichopterus**

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## Introduction

In recent years, much work has concerned itself with the determination of the various toxicants in the environment, and with the establishment of the median tolerance limit ($Tl_m$) for many contaminants. Mercury and copper are among the most toxic of the heavy metals to fishes.

Much work has been reported on the toxicity of various mercury compounds to fishes (JONES 1939a; GREESON 1970; REHWOLDT et al. 1972), and the toxic effects of copper on fishes has also been noted (JONES 1938a; CALVENTI and NIGRELLI 1961; SPRAGUE 1964; CAIRNS and SCHEIER 1968; MOUNT 1968; REHWOLDT et al. 1971). However, most often, contaminated waters contain combinations of pollutants, and therefore, several studies have been concerned with the interaction of various contaminants (JONES 1938b, 1939b; LLOYD 1961; SPRAGUE 1964; HERBERT and VANDYKE 1964; SPRAGUE and RAMSAY 1965; SPRAGUE et al. 1965; CAIRNS and SCHEIER 1968; WILSON 1972; ROALES and PERLMUTTER 1974).

The purpose of this study was to determine the toxicity of an organic mercury compound (methylmercuric chloride) and copper (cupric chloride), singly, and to determine the type of toxic interaction between the two contaminants on adult blue gouramis (Trichogaster trichopterus).

Since most of the mercury isolated from fishes is in the form of methylmercury (ACKEFORS et al. 1970), and since it is very stable within an organism (BACKSTROM 1969), and is the most readily absorbed form (HANSON 1968), methylmercury was chosen as the mercurial compound to be tested along with copper in this study.

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## Materials and Methods

Adult blue gouramis were obtained commercially, and allowed to acclimate in modified New York City water at 26 - 28 C for two weeks prior to experimentation. Modification of the water was as follows: New York City tap water was passed through a mixed-bed ion-exchange resin column at the rate of one liter/min. To 45 gallons of this de-ionized water, five gallons of untreated tap water were added, and the pH was adjusted to 7.4 by the addition of sodium bicarbonate.

The toxicity of methylmercury and copper was obtained by determining the 24, 48 and 96 hour $Tl_m$ for each toxicant. Two fish weighing 1.5 - 2.0 g each were placed in a polyethylene bag-lined aquarium containing four liters of test solution. During the test periods the solutions were aerated constantly, and the fish were not fed so as not to foul the water. A total of ten fish were used at each dose tested, and ten fish were used as controls at a chloride ion concentration equal to the chloride ion concentration of the highest dosage of toxicant tested. The $Tl_m$ was determined using probit analysis (BLISS 1937, 1952).

The type of interaction between methylmercury and copper was determined using the method of ROALES and PERLMUTTER (1974). Twenty-five fish (2 fish/4 liters of test solution) were exposed for 96 hours to combinations of varying percentages of the respective 96 hour $Tl_m$ dosages for methylmercury and copper, and the mortality was recorded. Thirty fish were used as controls at a chloride ion concentration equal to the chloride ion concentration of the highest dosage of toxicant tested.

In an effort to remove as many variables as possible, e.g. water hardness, calcium and magnesium hardness, modified New York City water was used as the dilution water in all experiments. The dissolved $O_2$ levels were maintained at 10 ppm, and dissolved $CO_2$ did not exceed 2.0 ppm. The biomass per test solution volume did not exceed 1 g/liter.

## Results and Discussion

The results of the 24, 48 and 96 hour $Tl_m$ experiments are shown in Table I. Both compounds appear to be highly toxic to the fish. However, methylmercury appears to be slightly more toxic, especially at 24 and 48 hours.

The results of the methylmercury/copper interaction experiments are shown in Table II, and are represented graphically in Figure 1. According to this method, a horizontal line indicates an additive effect; a bell-shaped curve shows synergism, while an inverted-bell curve indicates antag-

TABLE I

$Tl_m$ data for blue gourami exposed to methylmercury and copper

| Toxicant | $Tl_m$ (ppb ± S. E.) | | |
|---|---|---|---|
| | 24 hrs. | 48 hrs. | 96 hrs. |
| Methylmercury | 123 ± 7.38 | 94.2 ± 8.70 | 89.5 ± 4.12 |
| Copper | 209 ± 49.0 | 123.0 ± 49.0 | 91.2 ± 10.90 |

onism. The shape of the curve (Figure 1) seems to indicate antagonism, especially at low to moderately-high doses of copper.

TABLE II

Percent kill of various methylmercury/copper combinations during 96 hours

Methylmercury dose: 90 ppb
Copper dose: 90 ppb

| Test solution composition | | % Kill ± S. E. |
|---|---|---|
| % Methylmercury | % Copper | |
| 100 | 0 | 56 ± 8.7 |
| 80 | 20 | 0 ± 0.0 |
| 60 | 40 | 8 ± 8.3 |
| 40 | 60 | 0 ± 0.0 |
| 20 | 80 | 32 ± 7.9 |
| 0 | 100 | 44 ± 7.8 |
| Control | | 0 ± 0.0 |

A t test of significance indicated that 80% methylmercury 20% copper, 60% methylmercury/ 40% copper and 40% methylmercury 60% copper were statistically different from all other dosages tested (Table III). It was possible, however, that the results were due to dilution of one component of the mixture by the other and vice versa. Since the mortality for 100% methylmercury/0% copper and 0% methylmercury/100% copper in the interaction tests were both within 6% of the 50% mortality expected for the $Tl_m$ dose, the expected mortality due to dilution was computed from the 96 hour $Tl_m$ data (Table IV).

The data in Table IV seems to indicate that although the 60% methylmercury/40% copper dose could be due to dilution,

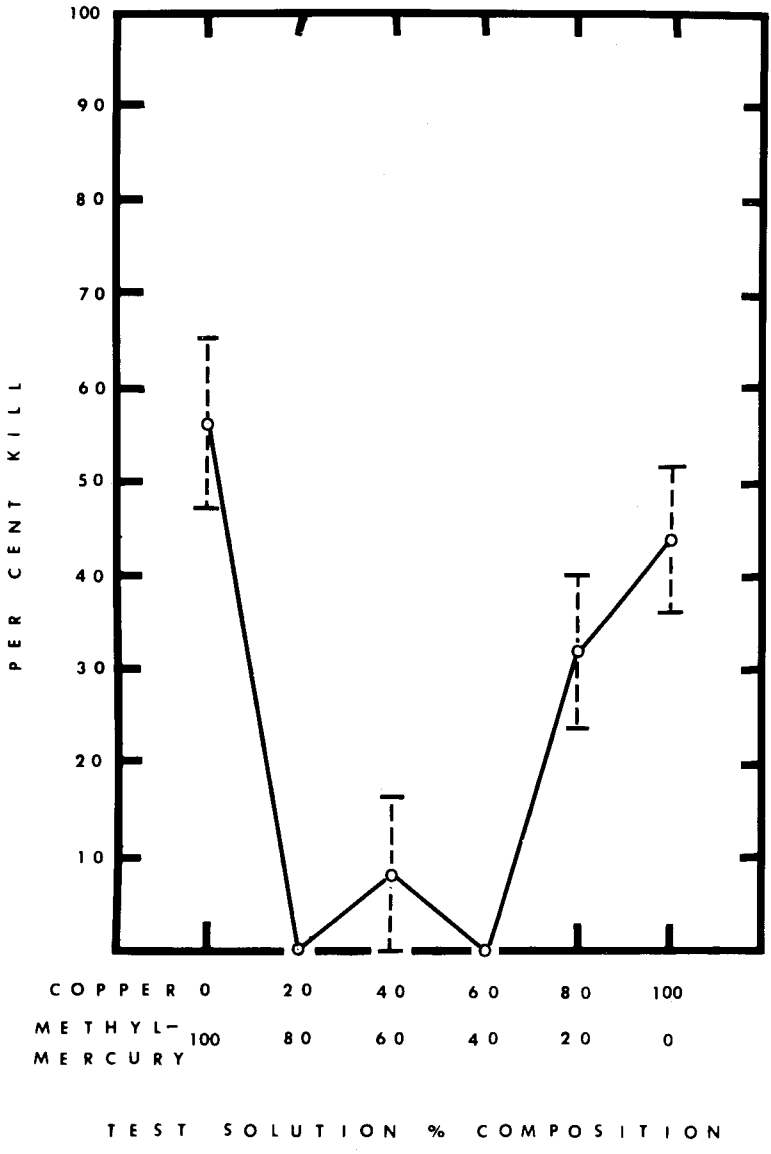


Figure 1. Dosage-mortality curve for blue gouramis exposed to varying percentages of the respective 96 hour $Tl_m$ dosage for copper and methylmercury.

TABLE III

Significance of difference between toxicants in the methylmercury/copper interaction tests

| Between | t value | Probability |
|---|---|---|
| 100% Methylmercury & 100% Copper | 1.029 | $>0.3<0.4$ |
| 0% Copper + 100% Methylmercury & 20% Copper + 80% Methylmercury | 6.574 | $>0.001<0.01$ |
| 20% Copper + 80% Methylmercury & 40% Copper + 60% Methylmercury | 0.963 | $>0.3<0.4$ |
| 40% Copper + 60% Methylmercury & 60% Copper + 40% Methylmercury | 0.963 | $>0.3<0.4$ |
| 60% Copper + 40% Methylmercury & 80% Copper + 20% Methylmercury | 4.050 | $>0.01<0.02$ |
| 80% Copper + 20% Methylmercury & 100% Copper + 0% Methylmercury | 1.082 | $>0.3<0.4$ |

N= 4; P = 0.05 when t = 2.776

the other two doses (80% methylmercury/20% copper and 40% methylmercury/60% copper) probably are due to antagonism between the two toxicants, copper protecting the fish from the toxic effects of methylmercury. This action seems to be

TABLE IV

Expected mortality due to dilution of the toxicants in the interaction bioassay (computed from the 96 hour $Tl_m$ data for each toxicant)

| Test solution composition | | Expected % kill due to dilution | |
|---|---|---|---|
| % Methylmercury | % Copper | Methylmercury | Copper |
| 80 | 20 | 13.4 | 0.0 |
| 60 | 40 | 0.0 | 4.2 |
| 40 | 60 | 0.0 | 15.6 |

consistent with the protective action of zinc against the pathological action of mercury in mammalian embryos (GALE 1973). Precisely what mechanism is involved in this phenomenon, however, remains to be determined.

## Summary

The 24, 48 and 96 hour $Tl_m$ for methylmercury and copper were determined for adult blue gouramis. In addition, it was found that copper at low to moderately-high doses appears to be antagonistic to the toxic action of methylmercury in these fish.

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