

## Acute Toxicity of Cypermethrin on the Great Sturgeon (*Huso huso*) Juveniles

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**Abstract:** The acute toxicity of cypermethrin to Great sturgeon (*Huso huso*) juveniles was assessed in a static renewal bioassay for 96h. In addition, behavioral changes at each cypermethrin concentration were observed for the individual fish. Results of the present study indicate that cypermethrin is highly toxic to *H. huso*. The 24, 48, 72 and 96 h LC<sub>50</sub> values of cypermethrin for Great sturgeon juveniles were estimated at 6.860, 4.751, 2.677 and 0.952 µg/L, respectively. Obtained data from the cypermethrin acute toxicity tests were evaluated using the probit analysis statistical method.

**Key words:** Acute Toxicity % Cypermethrin % Great Sturgeon (*Huso Huso*) % Behavioral Changes

### INTRUDUCTION

Cypermethrin (a-cyano-3-phenoxybenzyl ester of 2, 2-dimethyl-3-(2, 2-dichlorovinyl)-2,2-dimethyl cyclopropane carboxylate) is a synthetic pyrethroid insecticide widely used for pest control programs in domestic, industrial and agricultural situations. The low effect of toxicity of pyrethroid insecticides on mammals and birds and their limited soil persistence has encouraged the widespread and increasing use of pyrethroids in agriculture, as potent agents against pests [1]. Unfortunately, application of these synthetic derivatives of pyrethrins is highly toxic to a number of non-target organisms such as bees [2], freshwater fish and other aquatic organisms even at very low concentration. Indiscriminate discharge of these pesticides from agricultural runoff and in aquaculture operation may be washed into nearby water bodies and affects non-target organisms such as fish and prawn which are of great economics importance to humans [3]. Among the aquatic animals fish are highly sensitive to the pyrethroids pesticides due to their neurotoxic effects and the pesticides are lethal to fish at a minimum concentration (10-1000 lower) than the corresponding values for other groups of mammals and birds [1]. Bradbury and Coats [4] have reviewed the toxicology of pyrethroids in mammals, birds, fish, amphibian and invertebrates (terrestrial and aquatic) and cited 96-h LC<sub>50</sub> of cypermethrin as 0.9-1.1 µg/L for carp (*Cyprinus*

*carpio*), 1.2 µg/L for brown trout (*Salmo trutta*), 0.5 µg/L for rainbow trout (*Salmo gairdneri*), 0.4 µg/L for *Scardinius erythrophthalmus* and 2.2 µg/L for *Tilapia nilotica*.

The Caspian Sea is the biggest land-locked body of water bordered by five countries: Azerbaijan, Iran, the Russian Federation, Kazakhstan and Turkmenistan. There are several important fisheries in the Caspian Sea, but the greatest emphasis has always been placed on the sturgeons. The sturgeons (Acipenseridae) are an ancient group of chondrosteian fish with fossil records dating back to the lower Jurassic period. Several anthropogenic factors, including both land-based and offshore pollution, threaten the survival of all fisheries, but especially sturgeon populations in the Caspian Sea [5]. Among these, chemical contamination seems to be one of the most significant factors influencing the population of sturgeons in the Caspian Sea [6]. Great sturgeon (*H. huso*) is one of the most important species of sturgeon in the Caspian Sea. The present study was performed to determine acute toxicity of cypermethrin, a synthetic pyrethroid pesticide and evaluated behavioral changes of great sturgeon juveniles exposed to different concentrations.

### MATERIALS AND METHODS

Juveniles of *H. huso* in the weight range of 183.20 ± 4.61 g and total length of 36.31 ± 1.27 cm were obtained

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from the Shahid Marjani proliferation and culture centre for sturgeon fish, Gorgan, Iran. Fish were transferred to Aquaculture Research Center of Gorgan University of Agricultural Sciences and Natural Resources and acclimated to the laboratory conditions for 2 weeks. Water was renewed (one third of the water) daily and fish were fed daily (2% of their body weight twice a day) with a formulated feed (containing 48% of crude protein and 4800 kcal of digestible energy). Fishes were randomly parceled in 21 fiberglass tanks (500 L) supplied with oxygenated water maintaining constant dissolved oxygen at  $7.8 \pm 0.5$ , temperature at  $23 \pm 1^\circ\text{C}$ , pH at  $7.89 \pm 0.9$ , total hardness  $295 \pm 10$  mg/L as  $\text{CaCO}_3$  and photoperiod at 14L/10D light: dark natural photoperiod. Technical grade Alpha-cypermethrin (95% pure) manufactured by Daga global chemicals, India and supplied by Gol Sam Co, Iran, was used for evaluation of its toxicity to fish. The stock solution of cypermethrin was prepared by dissolving in acetone (0.5%) and appropriate amount of tap water. Control group received acetone at the maximum acetone volume used in the dilution of the dosing concentrations. The acute toxicity test was conducted following the Organization for Economic Cooperation and Development guideline under static-renewal test conditions [7]. Test organisms (7 test fish for each concentration) were exposed to a logarithmic increasing range of concentrations such as 0.01, 0.1, 1.0 and 10.0  $\mu\text{g/L}$ . Percentage mortality was observed every 24 h interval at 24, 48, 72 and 96 h. In range finding test of cypermethrin on *H. huso* juveniles, it was found that mortality percentage of 0% and 100% lies in between 0.1 and 10.0  $\mu\text{g/L}$ . For definitive test seven cypermethrin concentrations 0, 0.5, 1, 2, 4, 6 and 8  $\mu\text{g/L}$  concentration was selected and test was conducted for each concentration containing 15 fishes. During the 96 h acute

toxicity experiment, water in each fiberglass tank was aerated and had the same conditions as the acclimation period. The static-renewal tests exposed the fish for 96 h with replacement of the test solution every 24 h (all stock solutions were immediately made prior to use). Fish mortality was recorded 24, 48, 72 and 96 h after exposure to cypermethrin. Fishes were considered dead when gill opercula and body movement ceased; and when these characteristics occurred fishes were immediately gathered by dip net. The behavioral pattern of fish was monitored regularly under above treatment conditions. For statistical analyses, the statistical software EPA Probit Analysis V. 1.5 was used. Data obtained from the cypermethrin acute toxicity tests were evaluated using Finney's probit analysis statistical method. All data were accepted if calculated chi square for heterogeneity was lower than the tabular value at the 0.05 level.

## RESULTS AND DISCUSSION

The 24, 48, 72 and 96 h  $\text{LC}_{50}$  values (with 95% confidence limits) of cypermethrin for Great sturgeon juveniles were estimated at 6.860 (5.333 - 11.189), 4.751 (3.615 - 6.608), 2.677 (2.015 - 3.473) and 0.952 (0.693 - 1.242)  $\mu\text{g/L}$ , respectively (Table 1 and Fig. 1). Results of the present study indicate that cypermethrin is highly toxic to *H. huso*. Fish mortality increased significantly when the concentration and the time of exposure were increased. No mortality was observed in the control group during the experiment. Behavioral changes are the most sensitive indicators of potential toxic effects. Fish in the control experiment appeared active and healthy throughout the test period. The changes in behavioral response started

Table 1: Lethal concentration (LC 1-99) of cypermethrin depending on time (24-96 h) for Great sturgeon (*H. huso*) juveniles

| Point                      | Concentration ( $\mu\text{g/L}$ ) (95 % of confidence limits) |                           |                         |                        |
|----------------------------|---|---------------------------|-------------------------|------------------------|
|                            | 24 h  | 48 h                      | 72 h                    | 96 h                   |
| LC 1.00                    | 1.347 (0.256 - 2.261)   | 0.722 (0.196 - 1.268)     | 0.449 (0.166 - 0.757)   | 0.213 (0.066 - 0.358)  |
| LC 5.00                    | 2.170 (0.715 - 3.145)   | 1.254 (0.496 - 1.908)     | 0.758 (0.358 - 1.139)   | 0.330 (0.137 - 0.497)  |
| LC 10.00                   | 2.798 (1.224 - 3.792)   | 1.683 (0.808 - 2.391)     | 1.002 (0.536 - 1.424)   | 0.417 (0.200 - 0.596)  |
| LC 15.00                   | 3.332 (1.740 - 4.348)   | 2.053 (1.116 - 2.804)     | 1.209 (0.702 - 1.662)   | 0.488 (0.257 - 0.675)  |
| LC 50.00                   | 6.860 (5.333 - 11.189)  | 4.751 (3.615 - 6.608)     | 2.677 (2.015 - 3.473)   | 0.952 (0.693 - 1.242)  |
| LC 85.00                   | 14.167 (9.397 - 50.098)                                       | 10.955 (7.610 - 24.063)   | 5.930 (4.441 - 9.450)   | 1.858 (1.405 - 3.029)  |
| LC 90.00                   | 16.818 (10.581 - 72.530)                                      | 13.410 (8.858 - 33.455)   | 7.157 (5.207 - 12.311)  | 2.176 (1.606 - 3.867)  |
| LC 95.00                   | 21.686 (12.576 - 125.891)                                     | 17.996 (11.037 - 54.796)  | 9.458 (6.540 - 18.365)  | 2.750 (1.940 - 5.610)  |
| LC 99.00                   | 34.935 (17.286 - 356.133)                                     | 31.245 (16.520 - 139.552) | 15.953 (9.884 - 39.450) | 4.267 (2.716 - 11.463) |
| Chi - Square (calculated)* | 1.285   | 5.379                     | 2.86                    | 0.681                  |
| Intercept $\pm$ SE         | 2.24 $\pm$ 0.68   | 3.07 $\pm$ 0.41           | 3.71 $\pm$ 0.29         | 5.07 $\pm$ 0.20        |

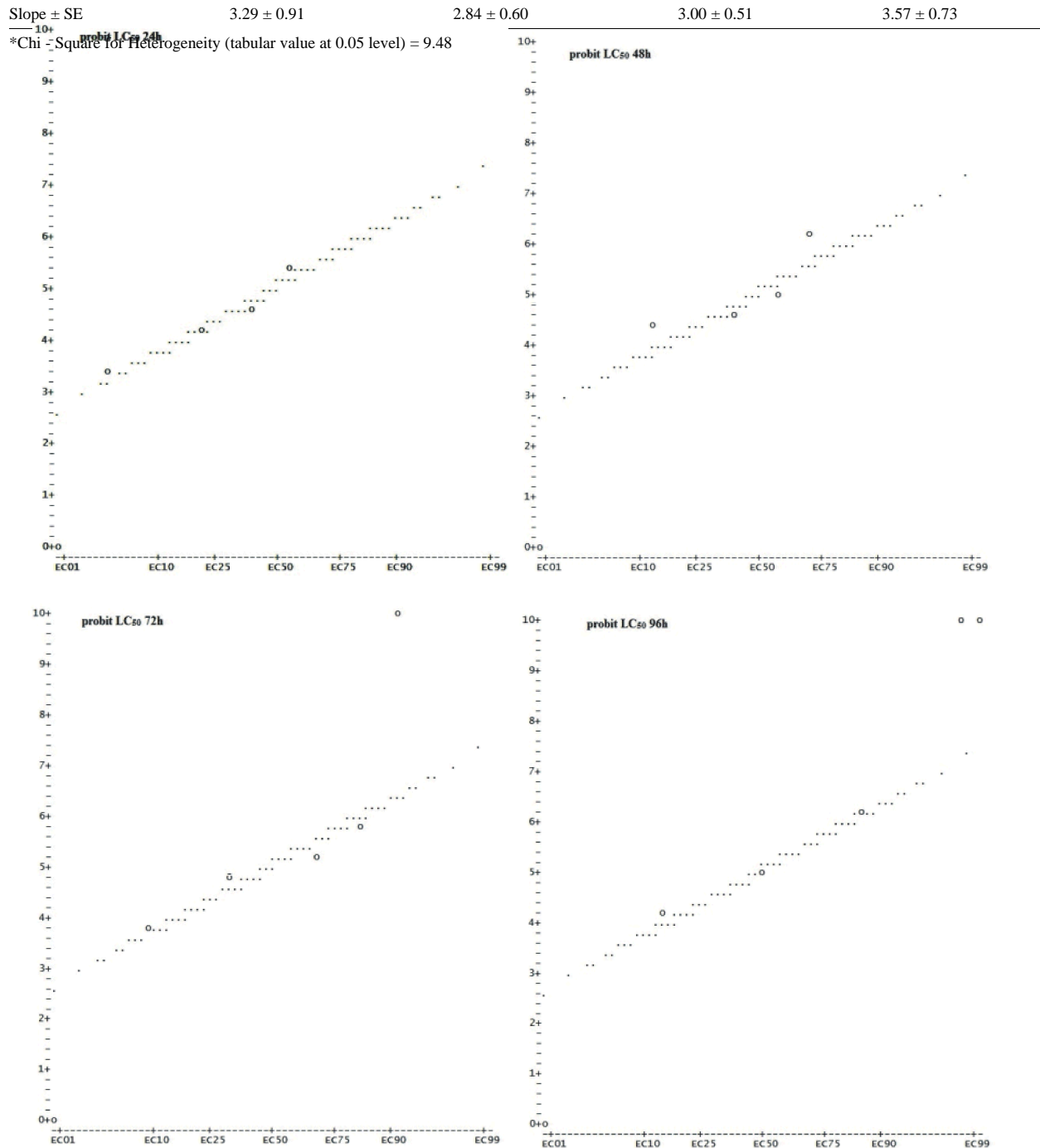


Fig. 1: Plot of adjusted probits and predicted regression line of cypermethrin for Great sturgeon (*H. huso*) juveniles

1 h after dosing. The abnormal behavioral responses (rapid gill movement, nervous manifestations, erratic swimming, loss of equilibrium and inability to remain upright) were observed in Great sturgeon juveniles at all

cypermethrin concentrations. The abnormal swimming behavior increased with increasing concentration and exposure time. Similar behavioral responses determined in this study have been observed with the guppy *Poecilia*

*reticulate* [8], freshwater catfish, *Heteropneustes fossilis* [9] and young mirror carp, *Cyprinus carpio* [10], exposed to various concentrations of the synthetic pyrethroids cypermethrin and deltamethrin. The acute toxicity results

of the present work are also in agreement with the results of other workers. Review of toxicity of pyrethroids by Bradbury and Coats [4] documents LC<sub>50</sub> value of most freshwater fish to range from 0.4 to 2.2 µg/L. Saha and Kaviraj [9] observed cypermethrin as highly toxic to freshwater catfish (72 h LC<sub>50</sub> value as 0.67 and 1.27 µg/L for water solubilized and acetone solubilized cypermethrin). Yilmaz *et al.* [8] studied the acute toxicity of alpha-cypermethrin to guppy and reported 96-h LC<sub>50</sub> value as 9.43 µg/L. Yilmaz [11] found the 96-h LC<sub>50</sub> value of alpha-cypermethrin for Tilapia as 3.42 µg/L. Examining cypermethrin toxicity to other aquatic organisms, the work of Clark *et al.* [12] reported the cypermethrin 96-h LC<sub>50</sub> for grass shrimp (*Palaemonetes pugio*) as 0.016 µg/L. The 24 h topical and aqueous LD<sub>50</sub> values for selected terrestrial and aquatic insects, when exposed to technical grade cypermethrin (99.4% purity), were in the range 0.30-49 ng/mg body weight and 1.3-9.8 µg/L, respectively [13]. Willis and Ling [14] observed EC<sub>50</sub> values of cypermethrin to the cyclopoid copepod, *Oithona similis* to range from 0.14 to 0.24 µg/L for nauplii and adults, respectively. Comparative pyrethroid studies in the lobster (*Homarus americanus*) and shrimp (*Crangon septemspinosa*) also indicated cypermethrin to be most toxic (96 h LC<sub>50</sub> value about 0.01 µg/L) [4]. Saha and Kaviraj [15] observed cypermethrin is highly toxic to aquatic insects (96 h LC<sub>50</sub> 0.06 µg/L) and 96h LC<sub>50</sub> values of aqueous cypermethrin ranged from 0.03 µg/L for the crustacean to 9.0 µg/L for the tadpole larva. Sreenivasan *et al.* [16] studied the acute toxicity of commercial cypermethrin to field crab (*Spiralothelphusa hydrodroma*) and reported 96-h LC<sub>50</sub> value as 1.35 ppm.

The results indicate that low levels of cypermethrin (0.5 µg/L) in the aquatic environment may have a significant effect on great sturgeon populations. In addition, Kumaragura and Beamish [17] reported that acute toxicity of synthetic pyrethroids to fish was negatively correlated to temperature. Therefore, the presence of pyrethroids in the aquatic environment when water temperature decreased may increase the toxic impact on fish [18]. In conclusion, cypermethrin contamination is dangerous to aquatic ecosystems and this fact should be taken into consideration when this insecticide is used in agriculture. Biological methods could be used for controlling mosquito and flies instead of cypermethrin in order to protect the natural environment.

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