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The Effects of Nanosilver (Nanocid[®]) on Survival Percentage of Rainbow Trout (*Oncorhynchus mykiss*)

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Abstract: Nano materials have unique antimicrobial properties. The toxic level of nanosilver, with the commercial name of Nanocid was studied. Rainbow trout (*Oncorhynchus mykiss*) with median weight of 1.049 g, was treated with 1, 2, 5, 10 and 20 ppm of nanosilver. The results were analyzed after 24, 48, 72, and 96 h and the lethality dose obtained between 1.25-10 ppm. In conclusion, the 48, 72 and 96-h median lethal concentrations (LC50) of nanosilver tests were obtained at 3.5, 3 and 2.3 mg/L, respectively, indicating moderate toxicity of nanocid in rainbow trout fish.

Key words: Rainbow trout, nanosilver, toxicity test

INTRODUCTION

Nanotechnology is the science, art and the power breaking elements into nano-sized particles and controlling them. In other words, it is the technology and research in the atomic and molecular sizes and utilizing systems which have new functions and properties because of their small sizes. This size range is 1-100 nm. Silver is one of those materials that has been used for many years and can have important effects at the nanotechnology level. Silver nanoparticles show a broad band of antimicrobial effectiveness. The Pars Nano Nasb Company in Iran has been developing the technology of applying silver nano-particles to impose the efficiency of reducing bacterial contamination in the production of fish and other animals. In this paper the toxic lethal dose of nanocid in salmon trout was investigated.

MATERIALS AND METHODS

Animal: Healthy rainbow trout with average weight of 1.049 g were purchased from a local hatchery. The fish were held in a 300-L polyethylene tank for at least one week prior to experimentation. Aeration and three times feeding per day, 12 h day lighting were performed. For the experiment, the fish were transferred to 20-L glass aquariums. Five aquariums were used for each concentration in triplets with added 30 fish. The same condition was performed on the control aquarium. Fish were fed dry trout pellets to satiation three times a week. No food was given during the last 2 days prior to experimentation.

Water quality: The quality of water was maintained constant in whole period of the experiment. The aquariums were supplied with tap water at 16±1°C,

oxygen above 8 mg/L, dioxide carbon less than 6 mg/L, ammoniac less than 0.01 mg/L, nitrite less than 0.1 mg/L, hardness of water less than 200 mg/L, electrical conductivity E.C. 780 micro zimens and pH with 7.5-8.4.

Nanocid: The Nanocid L-series colloidal product of Nano Nasb Pars Co., containing 4000 ppm nano-silver particles is water-based and enabling application by mixing with other water-based ingredients.

Statistics: Median lethal time (LT50) values were obtained from mortality plots by standard log-probit analysis (Sprague, 1990), employing the nomographic method by Litchfield and Wilcoxon (1949) to calculate 95% confidence limits. Toxicity curves were plotted as log LT50 versus log concentration (e.g., Fig. 1) to check whether or not acute mortality had ceased (i.e., whether true incipient LC50 values were achieved) (Sprague, 1990). The partial mortality data at 24 h and 96 h were used to calculate the 48, 72 and 96-h LC50 values of 96% confidence limits, again by log-probit (p. value) analysis. All calculations were based on mean concentrations measured in the aquariums, not nominal values.

RESULTS

LC50: The experiment was performed based on determining the maximum and minimum concentration of Nanocid with no effects and 100% lethality, respectively. The calculation was done by ratio test and in the preliminary data the primary concentration limit was obtained. In the experiments, five dilutions, each one with triple repeats and each repetition with 10 fishes was performed. Treatment was done with 1, 3, 5, 7 and 9 ppm of Nanocid and the lethality dose was determined



Fig. 1: LC50 determination after 48 h



Fig. 2: LC50 determination after 72 h



Fig. 3: LC50 determination after 96 h

in 24, 48, 72 and 96 h. The minimum limitation was obtained between 1.25-10 ppm, as shown in Table 1. The mortality rate in 48, 72 and 96 h shown in Table 2, and Fig. 1-3. Indicating the moderate toxicity of Nanocid in salmin trout and more safer disinfectant than other chemical materials ever known.

| trout based on concentration and timing course | | | | | | | |
|------------------------------------------------|---------------------|-------------------|-------------------|-------------------|------------|--|--|
| | Concentration (ppm) | | | | | | |
| Time | 4 th h | 5 th h | 6 th h | 7 th h | After 24 h | | |
| 1.25 | - | - | - | - | 1 | | |
| 2.5 | - | - | - | - | 3 | | |
| 5 | - | 1 | 3 | - | 17 | | |
| 10 | - | 2 | 3 | - | 30 | | |
| 20 | 2 | 7 | 15 | 18 | 30 | | |
| 30-50 | - | - | - | 30 | - | | |
| 60-70 | - | 30 | - | - | - | | |

Table 1: Determination of toxicity range of nanosilver in rainbow

Table 2: LC50 determination of nanocid in different time course

| | Concentration (ppm) | | | | | |
|-----------|---------------------|------|------|------|--|--|
| No. of | | | | | | |
| mortality | 24 h | 48 h | 72 h | 96 h | | |
| 1 | - | - | - | 3 | | |
| 3 | - | 5 | 9 | 13 | | |
| 5 | - | 7 | 10 | 14 | | |
| 7 | - | 12 | 18 | 19 | | |
| 9 | 2 | 17 | 22 | 30 | | |

Toxicity: Fish exposed to nanocid showed some clinical signs including hypoxia, lethargy, unusual swimming behaviour, elvated ventilation and darkening of body. The severity of these signs were dependent on the nanocid concentration"

DISCUSSION

Nanotecnology, which deals with devices typically less than 100 nanometer in size, is expected to make a significant contribution to the fields of computer storage, semiconductors, biotechnology, manufacturing and energy.

Nanocid which is a liquid product of Nano Nasb Pars Co., in Iran has produced Nanosilver (Nanocid®) which is a potent disinfectant in fish production. Comparing to other antibacterial reagents such as malachite green, formalin, leteux-meyer mixture, chloramine-T and copper sulfate which are used in high doses in fish production by using nanocid in ppm quantity is quite cost effective. Salmonids are susceptible.

REFERENCES

- Sprague, J.B., 1990. Aquatic toxicology. In C.B. Schreck and P.B. Moyle, Eds., Methods for Fish Biology. American Fisheries Society, Bethesda, MD, USA, pp: 491-528.
- Litchfield, J.T. and F. Wilcoxon, 1949. A simplified method of evaluating dose-effect experiments. J. Pharmacol. Exp. Ther., 96: 99-113.