## SENSITIVITY OF THE MONOGENETIC TREMATODE DACTYLOGYRUS LAMELLATUS AND THE HOST GRASS CARP CTENOPHARYNGODON IDELLA TO DIFFERENT CONCENTRATIONS OF EARTHTEC<sup>TM</sup>

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# ABSTRACT

The effectiveness of EarthTec<sup>TM</sup> compound against the monogenetic trematode *Dactylogyrus lamellatus* was assessed, using 14 different concentrations, ranging from 0.03 to 60.0 mg Cu/L. The result indicated that short bath treatments at concentrations ranging from 42.0 to 60.0 mg Cu/L could kill the parasite, however, they might be toxic to the stressed grass carp. Concerning long bath treatments, it was found that the concentrations of 0.03, 0.06, 0.09, 0.12 and 0.15 mg Cu/L can eradicate the parasite after 120, 96, 48, 48 and 24 hours respectively. These subacute levels of the test compound showed no toxic effect on the grass carp.

### INTRODUCTION

The herbivorous fish or grass carp *Ctenopharyngodon idella* had been introduced into Egypt for biological control of aquatic weeds harboring the waterways. This fish can not reproduce naturally under common Egyptian climatological circumstances. As a result, a fish hatchery at the Delta Breeding Station was constructed by Channel Maintenance Research Institute. This phytophagous fish was noticed to be a target for many parasitic forms (Szakolerzai and Molnar, 1966; Musselius and Strelkov, 1968, Molnar, 1971; Stepanova, 1971; Riley, 1978; Shirman and Smith, 1983; Abdel-Meguid, 1989, 1995). One of those parasites is the monogenean ectoparasite, *Dactylogyrus lamellatus*. This parasite has been regarded as a main cause of death

among crowded fish in the hatcheries. It can infect the grass carp and attack both the skin and the gill filaments. It can induce severe damages to the gill filaments by causing hyperplastic proliferation, vacuolar degeneration, edema and necrosis (Molnar, 1972; Abdel-Meguid, 1989; Ramadan *et al.*, 1995).

Control of monogenean parasites have been tested by some investigators using copper sulphate by different concentrations (Wellborn, 1967; Abdel-Meguid, 1989; Ramadan *et al.*, 1995).

The objective of the present study was to control the monogenean ectoparasite *Dactylogyrus lamellatus* using different concentrations of EarthTec<sup>Tm</sup> with minimal or even no effect on the fish host.

### MATERIAL AND METHODS

Parasite control experiments consisted of exposing infected fish to various concentrations of EarthTec<sup>TM</sup> (active ingredients: copper sulphate pentahydrate 20%, inert ingredients 80%, metallic copper equivalent 5%). Test solutions of EarthTec<sup>TM</sup> were prepared using filtered Nile water. Treatments were characterized as a short term bath or a long term bath, depending on the time of fish exposure to each treatment. In the short bath treatment, infected fish were exposed to 4 concentrations (1:1429; 1:1250; 1:1111 and 1:1000). The infection status was examined every 5 minutes. For the long bath treatment, infected fish were exposed to 10 concentrations (1: 2'000'000; 1: 1'000'000; 1: 666'666; 1: 500'000; 1: 400'000; 1: 200'000; 1: 80'000; 1: 40'000; 1: 20'000 and 1: 10'000). The infection status was tested every 24 hours. Ten infected fish, weighing 10 grams each, were used for each treatment, after placing them in an aquarium of 20 liters capacity provided with aeration.

In the mean time, healthy fish, weighing 10 grams each, were inspected to assess their sensitivity to the same concentrations of EarthTec<sup>Tm</sup>. Such experiments were carried out in an aquarium of 20 liters capacity. Each aquarium was stocked with 10 healthy fish collected from a pond at Delta Breeding Station.

All experiments were carried out at a water temperature ranging from 19 to  $21^{\circ}$  C.

# RESULTS

The result presented in Table (I) shows that baths of treatment could be applied to kill the monogenean ectoparasite D. lamellatus. At the concentrations of 42.0, 48.0, 54.0 and 60.0 mg Cu/L, the parasite was killed after 45, 35, 25 and 15 minutes respectively. However, long baths' treatment could be used to eradicate the parasite from the gills after 24 hours at the concentrations of 0.15, 0.30, 0.75, 1.50 and 3.00 mg Cu/L. Also, the concentrations of 0,03, 0.06, 0.09 and 0.12 mg Cu/L could be used to eradicate the parasite after 5, 4, 4, and 3 days respectively.

It is clear from Table (I) that the fish started to suffer after 25 minutes at the concentrations of 48.0, 54.0 and 60.0 mg Cu/L. Total fish mortality (Lc100) was recorded after 80, 90 and 90 minutes respectively. At the concentration of 42.0 mg Cu/L, the fish started to suffer after 50 minutes and (L<sub>C</sub>100) was observed after 120 minutes. When the concentration of 6.0 mg Cu/L was applied, few fish died after 24 hours and all of them were killed after 2 days. At the concentrations of 0.75, 1.5 and 3.0 mg Cu/L, total mortality was observed after 3 days. At the concentration of 0.3 ppm Cu/L, all fish died after 4 days. On the other hand, when the fish were exposed to the concentrations of 0.03, 0.06, 0.09, 0.12 and 0.15 mg Cu/L, the fish tolerated these concentrations and remained in a rather healthy condition for more than 15 days post exposure.

# DISCUSSION

Treatment against ectoparasites by bathing the infected fish into water soluble compounds is the most common method of administering therapeutic agents. This type of application can be divided into 3 categories (dipping, short bath and long bath). Copper sulphate in short and long bath treatments has been recommended by many investigators (Wellborn, 1967; Abdel-Meguid, 1989, 1999; Ramadan *et al.*, 1995, Abdel-Meguid and Eure, 1996). However, there are problems associated with the use of copper sulphate because the  $Cu^{+2}$  quickly recombines into less soluble forms which precipitate out of the water column (McNight, 1983). Thus, it is necessary to apply large quantities of copper sulphate to achieve a concentration of dissolved Cu<sup>+2</sup> sufficient to control parasites. These large applications may lead to accumulations of Cu<sup>+2</sup> in the environment and cause toxic impacts on non-target organisms (Ramadan and Abdel-Salam, 1986; Abdel-Meguid, 1989, 1999; Eisler, 1998). The present study used EarthTec<sup>Tm</sup> compound that contains copper sulpate pentahydrate in a chelate-like carrier. This carrier is characterized by the phenomenon of holding the copper in suspension for a long time. This phenomenon offers the precise control of parasites at low concentrations. Different concentrations of EarthTec<sup>TM</sup> were used to eradicate the monogenean parasite within a short or a long period of time. In the present study, a short bath treatment at concentrations ranging from 42.0 to 60.0 mg Cu/L could be used to kill the parasite. However, it is not recommended using these concentrations since they might be toxic to the host fish.

Concerning a long bath treatment, the present study showed that D. lamellatus could be eradicated from the gill filaments of the grass carp within 24 hours at the concentrations that ranged between 0.15 and 3.00 mg Cu/L. However, it is recommended using the concentration of 0.15 mg Cu/L in order to save the fish and to minimize the cost of treatment. Interestingly, the present study showed that the monogenetic trematodes were more resistant to EarthTec<sup>Tm</sup> than the protozoan trichodinids (Abdel-Meguid, 2001). At the concentrations of 0.03, 0.06 and 0.09 mg Cu/L, D. lamellatus was exterminated from the gill filaments of the grass carp after 120, 96 and 96 hours respectively. On the other hand, Paratrichodina africana and Trichodina centrostigeata were completely eradicated from the gill filaments of Tilapia zillii after 48, 48 and 24 hours at the same concentrations. The recommended copper concentrations ranging from 0.03 to 0.15 mg Cu/L have no toxic effect on the fish, even after 15 days post exposure. Also, they reflect the requirement of maintaining copper concentration in compliance with Quality Standard of Water by Law 48/1982 for discharges of water bodies in Egypt.

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Sensitivity of the trematode *Dactylogyrus lamellatus* and the Grass 127 Carp *Ctenopharyngodon idella* to different concentrations of Earthtec<sup>tm</sup>

Table (1): Sensitivity of *Dactylogyrus lamellatus* and *Ctenopharyngodon idella* to different concentrations of EarthTec<sup>TM</sup>

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Earthtec <sup>TM</sup>	Concentration	D.lamellatus	C.idella
(V/V)	(mg Cu/L)	(L <sub>C</sub> 100)	(L <sub>c</sub> 100)
1:2'000'000	0.03	120 hours	no effect
1:1'000'000	0.06	96 hours	no effect
1:666'666	0.09	96 hours	no effect
1:500'000	0.12	72 hours	no effect
1:400'000	0.15	24 hours	no effect
1:200'000	0.30	24 hours	120 hours
1:80'000	0.75	24 hours	96 hours
1:40'000	1.50	24 hours	96 hours
1:20'000	3.00	24 hours	96 hours
1:10'000	6.00	24 hours	48 hours
1:1429	42.00	45 minutes	120 minutes
1:1250	48.00	35 minutes	90 minutes
1:1111	54.00	25 minutes	90 minutes
1:1000	60.00	15 minutes	80 minutes