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ESTIMATION OF GENOTOXIC EFFECT OF INSECTICIDE CHROMAGOR AT GOLDFISH (CARASSIUS AURATUS) AFTER 5 DAYS OF TREATMENT

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Abstract. The genotoxic effects of insecticide chromagor(dimethoate) were evaluated in goldfish Carassius auratus using the micronucleus test. The aim of this study is to estimate the damage of DNA, after treatment of goldfish Carassius auratus for five days with insecticide chromagor. The micronucleus assays have been used increasingly to evaluate genotoxicity of many compounds in polluted aquatic ecossystems. In order to investigate the frequencies of micronuclei and to assess the sensitivity of species, the results were compared with samples taken at the reference site and maintained in the laboratory, and fish treated with insecticide chromagor. Our results show significant increase of number of micronuclei in erythrocytes of goldfish.

Keywords: insecticide; chromagor; DNA; fish

Introduction

Environmental quality control requires the monitoring of various indicators, including the assessment of pesticide residues. Research on bioindicators is instrumental in detecting the toxic effects caused by these. Processes in the bioconcentration and transformation of pesticides have been studied mainly in fishes. This is due to these animals living in direct contact with aquatic sediments adjacent to areas where pesticides are commonly used. In these water bodies, many poorly water-soluble compounds eventually settle, with the consequential increase in the degree of local contamination, when compared to the water column as a whole (Grisolia, 2005; Umbuziero et al., 2006). Erythrocyte MN test in fish was also widely applied for genotoxicity assessment of freshwater (95, Marlasca, 1998) and marine (Napierska, 2009) environments *in situ* using native or caged animals following different periods of exposure.

Material and methods

We used the species of fish goldfish (Carassius auratus). In each aquarium put ten (10) fish, total number of fish is 50 fish.

The fish were treated in aquarium with insecticide chromagor for 5 days.

The frequency of micronuclei were estimated by counting 1000 cells. Concetration of insecticide chromagor , it was in first aquarium 2 ml insecticide chromagor / 40 liter water, in second aquarium 1.5 ml insecticide chromagor /40 liter water, in third aquarium 1 ml insecticide chromagor / 40 liter water, in fourth aquarium 0.5 ml insecticide chromagor / 40 liter water. Fifth aquarium use as control, without insecticide chromagor , contain only drinking water.

For each fish prepare three slides. Slides were coded, for each fish. The smears are air-dried and fixed in absolute ethanol for 25 minute.. After fixation, the slides were stained in aqueous Giemsa (diluted in destiled water ratio 1:5) for 50 minute.

Results and discussion

The frequencies of MN in peripheral blood erythrocytes after exposure to the insecticide chromagor treated for 5 days, are presented in Table 1, Figs. 1 and 2.

Spontaneous formation of micronuclei in fish is normally very rare. In our study, however, significant frequencies were observed in the captured specimens from each aqauria, with micronucleus testing revealing the rate of micronucleated erythrocytes to be high, with a minimum of 48 and a maximum of 74 per 1,000 erythrocytes evaluated. While at control group was 5 MN.

The frequencies of micronucleated (MN) erythrocytes were estimated for each fish in each aqauria. At first aquaria we detect the 74 micronuclei(MN), which is higher compared with other aquaria and with control group.

At second aquaria we determine 65 MN, while the third has 57 MN and fourth aquaria has 48 MN, at 1000 erythrocyte.

The average number of MN at all groups treated with insecticide are **61** MN, statistically are significantly higher compared with control group. At control group we determine 5 MN/ 1000 erythrocytes.

Table 1. Average number (per aquarium) of micronuclei(MN) in 1000 erythrocytes of peripheral blood of fish goldfish (Carassius auratus) after 72 hours treatment in different concentration of insecticide chromagor

Aquarium/ treated for 72 hours	Average number of MN/1000 erythrocytes per aquarium	Significancy -P
Aquarium 1(2 ml insecticide /40 l water): Aquarium controll	74	S , P = <0.001
Aquarium 2 (1.5 ml insecticide /40 l water): Aquarium controll	65	S , P = <0.001

Aquarium 3(1 ml insecticide /40 l water) : Aquarium controll	57	S, P = <0.001
Aquarium 4 (0.5 ml insecticide /40 l water) : Aquarium controll	48	S , P = <0.001
Aquarium controll	5	
Average number of MN at treaed fish, without control group	244: 4 = 61	

Legend: s – Significancy, Ns – Not Significancy

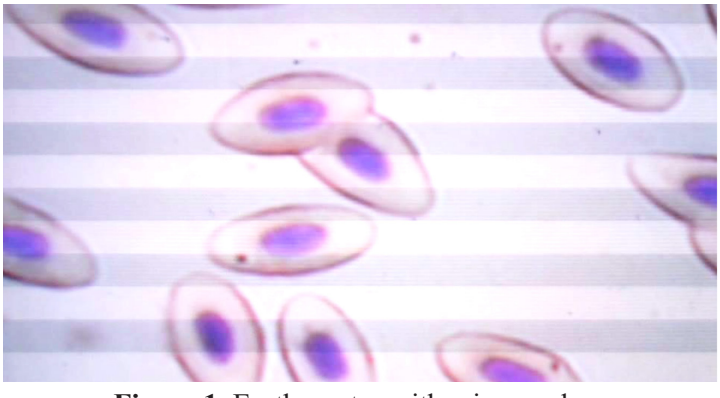

Figure 1. Erythrocytes with micronucleus

The results show that with decreasing of concentration of insecticide decrease and number of micronuclei. The present study reports dose and time dependent increase in MN induction in the peripheral blood erythrocytes of fish (Oreochromis mossambicus) which is in line with authors (Hooftman & De Raat, 1982; Abdul-Farah et al., 2003; Ali et al., 2008).

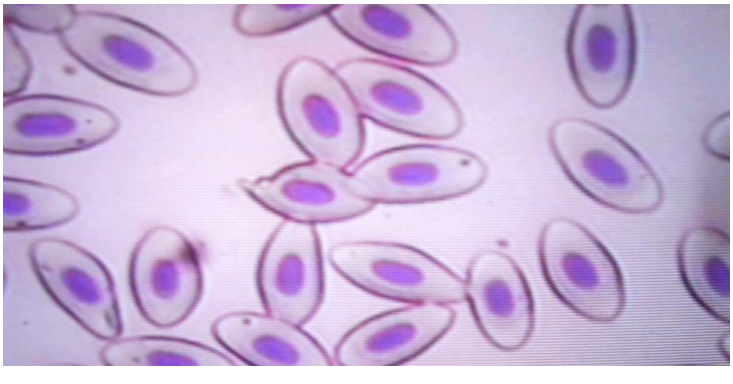

Figure 2. Erythrocytes with micronucleus

The results of the present study are similar to those of Campana et al. (1999), who reported that LCT is a genotoxic agent in erythrocytes of the fish Cheirodon interruptus interruptus, and are in accordance with those of Cavaş & Ergene-Gözükara (2003), who showed that LCT treatment caused an increase in the frequency of micronucleated erythrocytes in the fish Garra rufa at concentrations of 0.01 and 0.05 μg/l. Although discordant results have been reported, they demonstrate the ability of the insecticide chromagor and several glyphosate based products to induce DNA single-strand breaks evaluated by the SCGE bioassay in several fish. Positive results have been reported in circulating erythrocytes after laboratory exposure of Carassius auratus when not only the MN but also the comet assay was employed as an end point (Cavaş & Könen, 2007).

Conclusion

Based on this investigation we can conclude that insecticide chromagor is genotoxic, as it were damage the chromosome of fish, seeing that chromosome are fragmented, and formed the small nucleic body called micronuclei. The averge number of micronuclei it was higher, at treated fish, statistically significant compared with control group (P = <0.001).

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