RENAL EFFECTS OF DELTAMETHRIN INDUCED INTOXICATION IN Carassius auratus gibelio (Pisces cyprinidae)**

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Abstract: Freshwater goldfish Carassius auratus gibelio were exposed to $2\mu g/l$ delthametrin for one, 2, 3, 7 and 14 days. Activities of kidney catalase (EC 1.11.1.6), glutathione reductase (EC 1.6.4.2) and glutathione-Stransferase (EC 2.5.1.18) were affected in a time-dependent manner by the pesticide exposure compared to controls. The results indicate that C. auratus gibelio kidney resisted to oxidative stress by antioxidant mechanisms and developed an adaptative response.

Keywords: Carassius auratus gibelio, MgCl₂, oxidative stress, antioxidant enzyme

Introduction and Literature Review

In agriculture, due to the excessive use of pesticides disequilibrium in the ecological balance was created, organisms which are not their target, like fish, becoming affected. Aquatic medium pollution generates histopathological and behaviour changes in fish.

Generally, the pesticides toxicity is directly correlated with the concentration and the exposure time. Delthamethrin and other pirethroids toxicity was reviewed by *Aldridge* (1990), *Vijverberg şi Van den Bercken* (1990), *Appel şi Gericke* (1993). Pesticides metabolization generates reactive oxigen species (*Gram*, 1997). A severe oxidative stress causes cellular lessions and can provoke *in extremis*, cell death.

Taking in account the importance of fish in human nutrition, our aim was to study the effects of delthametrin on the activity of some enzymes oxidative stress related.

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Materials and Methods

Specimens of *Carassius auratus gibelio* weighing 20-30 g were obtained from the Nucet Fishery Research Station, Romania. Before the experiments, the fish were kept in glass aquaria containing aerated dechlorinated tap water at 25° C. After 3 days of adaptation, the fish were randomly divided into two groups of ten each and were not fed. Group 1, the control group, was kept in tap water. Group 2 was exposed for one, 2, 3, 7 and 14 days, respectively, to 2 µg delthametrin per L water. The pesticide was added only in the first day of experiment. After the mentioned periods of time, the fish of each group were sacrificed, and kidneys were excised and frozen at -80° C.

The frozen tissues were homogenized with a Potter-Elvejhem homogenizer in 0.1 M Tris-EDTA buffer, pH 7.4, and then centrifuged at 8, 000 g for 30 min at 4° C. Aliquots of the supernatants were used for analysis.

Gluthatione reductase (GR) activity was recorded by Golberg and Spooner method (12), using GSSG and NADPH as substrates. The convertion of NADPH to NADP+ was followed by recording the change in absorbance intensity at 340 nm. One unit of GR activity was calculated as 1 nmol of NADPH consumed per minute, using a molar extinction coefficient of $6.22 \times 10^3 \, \text{M}^{-1} \, \text{cm}^{-1}$.

Glutathione S-transferase (GST) activity was determined by the method of Habig et al. (1974). The change in absorbance was recorded at 340 nm and the enzyme activity calculated as nmol CDNB conjugate formed/min/mg protein using a molar extinction coefficient of $9.6 \times 10^3 \,\mathrm{M}^{-1} \,\mathrm{cm}^{-1}$.

The catalase (CAT) activity was assayed by monitoring the disappearance of H_2O_2 at 240 nm, according to the method of Aebi (11). One unit (U) of catalase activity represents the decrease of a 1 nmol of H_2O_2 per minute.

All the enzymatic activities were expressed as specific ones, namely in units per mg of protein. The protein content was determinated using the method of Lowry (1951) with bovine serum albumin (BSA) as standard.

Results of Investigations and Discussion

The fish intoxication with $2\mu g/L$ delthametrin produces a significant increase of kidney glutathione reductase specific activity (Fig.1) in the first two days of exposure. Between three and 14 days of intoxication, this activity significantly decreased.

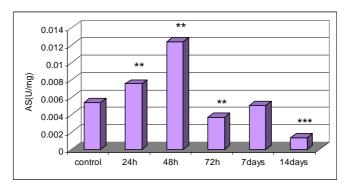


Fig.1. The glutathione reductase specific activity induced by the intoxication with $2\mu g/L$ delthametrin in the kidney of *Carassius auratus gibelio* (*-p<0.05; **-p<0.01; ***-p<0.001).

The profile of the specific activity of GST is almost the same with that of GR (Figure 2).

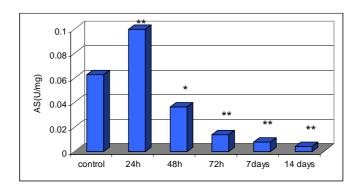


Fig.2. The glutathione S transferase specific activity induced by the intoxication with $2\mu g/L$ delthametrin in the kidney of *Carassius auratus gibelio* (*-p<0.05; **-p<0.01; ***-p<0.001).

In the case of the catalase specific activity, an increase can be noticed after 2 and 3 days. After 7 and 14 days this decreased (Figure 3).

Our results show that $2\mu g/L$ delthametrin exposure generates an important oxidative stress. In order to counteract this effect a high level of GSH was necessary and the GR specific activity increased in the first two days of intoxication. So, a higher concentration of GSH is present in kidney.

At longer time of exposure, the oxidative stress affects this enzyme so much that the GSH generation is significantly decreased. The variation of the GST specific activity depending on time presents the same pattern as the GR one, because GSH is one of the GST substrates.



Fig.3. The catalase specific activity induced by the intoxication with $2\mu g/L$ delthametrin in the kidney of *Carassius auratus gibelio* (*-p<0.05; **-p<0.01; ***-p<0.001).

Probably, during the delthametrin metabolism hydrogen peroxide is produced. As a result, the level of catalase specific activity is increased after 2 and 3 days of exposure. Its later decrease is due possibly to the generation of a low concentration of hydrogen peroxide, which is correlated with the increase of glutathione peroxidase specific activity (not shown data).

Conclusions

Our study have put in evidence the lack of an adaptive response in fi kidney of *Carassius auratus gibelio* to 7 and 14 days exposure to 2 μ g/L delthametrin.

UTICAJ INTOKSIKACIJE IZAZVANE DELTAMETRINOM NA BUBREGE CARASSIUS AURATUS GIBELIO (Pisces cyprinidae)

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Rezime

Slatkovodna riba *Carassius auratus gibelio* je bila izložena uticaju 2µg/l deltametrin u trajanju od 1, 2, 3, 7 i 14 dana. Antioksidativna reakcija bubrega je ispitivana. Registrovano je signifikantno povećanje aktivnosti glutation reduktaze u prva dva dana, nakon čega je došlo do signifikantnog smanjenja do 14 dana intoksikacije. Aktivnost glutation S transferaze se povećala nakon jednog dana i signifikantno smanjila do 14 dana izlaganja, što navodi na podatak o niskom nivou redukovanog glutationa. Aktivnost katalaze se povećala u prva tri dana i kasnije smanjila.

Naše istraživanje je potvrdilo nedostatak adaptivne reakcije bubrega *Carassius auratus gibelio* u slučajevima izlaganja od 7 i 14 dana dejstvu 2 μg/L deltametrina.

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