

A PROTECTIVE EFFECT OF THIOUREA AGAINST LEAD TOXICITY OF THE EUROPEAN CHUB, *LEUCISCUS CEPHALUS*

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REZUMAT. Scopul acestei lucrări este acela de a urmări posibilele efecte benefice ale tioureei privind toxicitatea subacută indusă de plumb peștilor. Intoxicația cu plumb a determinat o creștere a glicemiei și a numărului de eritrocite, consumului de oxigen și frecvenței respiratorii. Am observat că tioureea a avut un efect protector împotriva intoxicației cu plumb.

Cuvinte cheie: clean, plumb, tiouree, glicemie, număr de eritrocite.

ABSTRACT. The main purpose of this work was to investigate possible beneficial effects of thiourea on lead-induced acute toxicity in fish. Intoxication with lead caused an increase a glycemia and a number of erythrocytes, oxygen consumption and breathing frequency. We observed that the thiourea had a protective effect against lead intoxication.

Keywords: european chub, lead, thiourea, glycemia, number of erythrocytes.

1. INTRODUCTION

Living in an environment that has been altered considerably by anthropogenic activities, fish are often exposed to a multitude of stressors including heavy metals.

Environmental pollutants such as metals, pesticides and other organics pose serious risks to many aquatic organisms including fish.

Water pollution with heavy metals, affects various physiological processes in fish, including blood cells. In fish, toxic substances taken up from the water enter the blood and therefore, blood cells are among the first targets of toxicity, immediately after the gill epithelium.

Lead, one of the oldest known metals, is not necessary for the biological functions of animals, even at low concentration. It is being discharged to aquatic systems, mainly from petroleum, chemistry, dye and mining industries, which has toxic effects and can cause mortality to aquatic animals [1-3].

Thiourea is an organic compounds consisting of carbon, nitrogen, sulphur and hydrogen with the formula that is CSN_2H_4 . Thiourea is a white and sparkling crystal in structure. The function of thiourea in aqueous solution is expected to be the protector from hydroxyl radicals [4]. Thiourea derivatives and polyazomethines constitute important classes of chelating agents. Thiourea derivatives have been extensively

studied as a potential anti-TB, anti- HIV, anti-tumor and plant growth regulators [5-7]. Consideration of the high activity of a new thiourea derivative chelating macrocyclic ligand N, N--pyridine-2,6-diyl-bis [N--phenyl(thiourea)] (PDPT) has prompted us to undertake systematic studies on the complexation tendency [8].

The aim of the present study was to compare the effect of lead and thiourea used in single and mixed solutions on the oxygen consumption and breathing frequency, glycemia, erythrocytes, leucocytes of european chub (*Leuciscus cephalus*).

2. METHODOLOGY

Determinations were made between September and November 2012 on european chub (*Leuciscus cephalus*), caught in the surrounding lakes and rivers of Pitești city. We choose this species of fish because these are the most frequent in Arges River. After 10 days of adaptation in the lab, when they were fed *ad libitum* once a day, the fish were separated in lots, which were used separately for the following experiments:

The first experiment was carried out with european chub individuals having an average weight of 17g, which were subjected to thiourea 0.1g/l water.

The second experiment was carried out with ten european chub individuals having an average weight

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of 15 g, which were subjected to lead concentrations of 0,1 mg/l water.

The third experiment was carried out with ten european chub individuals having an average weight of 13g, which were subjected to lead concentrations of 0,1mg/l water and thiourea 0.1g/l water.

The lead and thiourea concentrations that have been used have been established by preliminary survival test. The immersion of fish in these solutions has been made after they have been well stirred and aired for five minutes. The water temperature has been between 18° and 19° Celsius and the immersion solution has been changed every 24 hours and the water has been continuously aired; the fish have not been fed during the experiments, in order to avoid the intervention of this factor [9].

Determination of oxygen consumption was done by means of the oximetre and Winkler method and erythrocytes were counted with Thoma chamber, using a small amount of blood from the caudal artery on the optic microscope [9]. Glucose in the animals' blood was determinate by using two different techniques: the colorimetric dosage with o-toluidine, and by means of the *Glucotrend* glucometer.

There have been made determinations of oxygen consumption and frequency of respiratory movements at intervals of 24, 48, 72, 96 and 336 hours on all samples of these lots.

The statistical interpretation of the results was performed with ANOVA (LSD) test.

3. RESULTS AND DISCUSSION

In studied concentrations, the lead 0.1 mg/l water and thiourea 0.1 g/l water modified the values of breathing frequency as shown in Figure 1. At the concentration of 0.1g lead/l water, and mixture the lead and thiourea, lead had a significant effect on respiratory rhythm on european chub for the 336 hours.

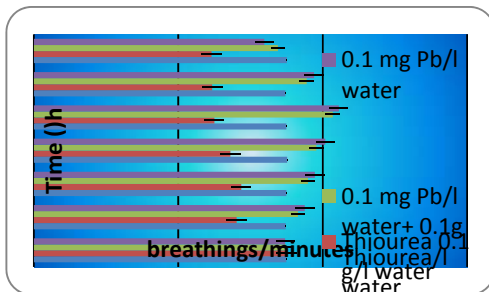


Fig.1. The influence of lead and thiourea upon breathing frequency on european chub (*Leuciscus cephalus*).

The oxygen consumption was found to be significantly influenced by the concentration of the used of lead into the water. Thus, as shown in Figure 2, at a

concentration of 0,1mg Pb /l water, this index increases 96 hours of immersion, after 96 hours oxygen consumption decreases significantly.

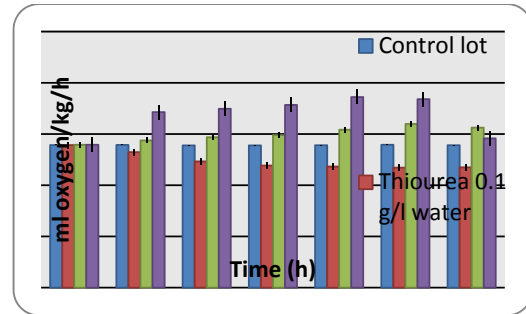


Fig. 2. The influence of the lead and thiourea upon oxygen consumption on european chub (*Leuciscus cephalus*).

The lead has changed the fish respiratory rhythm and oxygen consumption, but, the mixture between lead and thiourea determined a smaller increase. The thiourea 0.1g/l water decreased oxygen consumption and respiratory rhythm significantly after 24 hours. The lead's toxic effect was proven to be more powerful in the 96 hours from the fish's immersion. However, after this period of time, a certain degree of 'adaptation' was observed, and the mixture between lead and thiourea determined a established the respiratory rhythm and oxygen consumption. The mixture because the SH – groupings of tissual tioenzymes are protected against action of toxic substances, and the enzymes under discussion remain totally or partially unaltered according to the quantity of isothiourea [10-11].

The effects of lead on fish observed during this experiment showed that caution should be exercised in allowing lead into the aquatic environment. These effects included loss of balance, skin bleaching and weakness.

The results of the present study showed that, heavy metals induce morphological changes in fish blood cells.

Fish erythrocyte morphology is one of the most sensitive indicators of toxic impact of various environmental factors on fish. Many fish species are susceptible to the deleterious effects of heavy metals, as reflected in the blood changes, including anemia, eosinophilia, lymphocytosis, alterations in erythrocyte morphology and branchial and renal lesions [12].

After two weeks of exposure to the lead concentrations of 0.1 mg Pb /l water, 0.1 mg Pb /l water +0.1 g thiourea/l water, number of erythrocytes in european chub increase significantly compared to the control groups (Fig. 3).

The blood of european chub showed significant increase in glucose during 14 days of heavy metal intoxication. This might be due to the vulnerable stress induced by the heavy metals resulted in hyperglycemia (Fig.4).

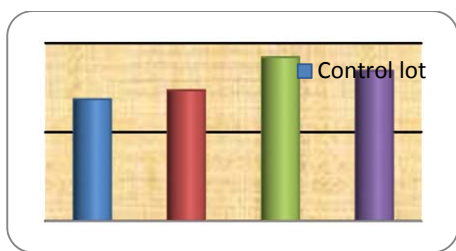


Fig. 3. Number of erythrocytes of european chub (*Leuciscus cephalus*) for 14 days of exposure lead and thiourea.

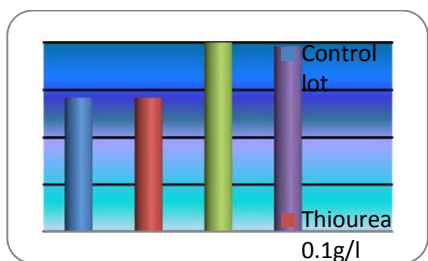


Fig. 4. The influence of lead and thiourea upon glycaemia level on european chub (*Leuciscus cephalus*).

Previous investigation proved that, lead modulate the metabolism of carbohydrates, causing hyperglycemia by stimulating the glycogenolysis in some marine and fresh water fish species [13-14]. Similar trend with characteristic hyperglycemia was observed in european chub throughout our experiment. Heavy metals increase the glucose content in blood, because of intensive glycogenolysis and the synthesis of glucose from extra hepatic tissue proteins and amino acids [15].

The fact that our experiments recorded a smaller increase in concentrations on 0.1 mg Pb /l water +0.1 g thiourea/l water of oxygen consumption, breathing frequency, erythrocytes, glycaemia, may be explained by the fact that the toxic effect of lead is diminished by daily administration of thiourea having protective effects.

4. CONCLUSIONS

The lead has changed the respiratory rhythm and oxygen consumption of european chub in concentration 0.1mg/l water.

The antitoxic action of thiourea manifests itself by the fact that heavy metals are blocked by SH-groupings isothiurea.

Our results confirm the sublethal effect of heavy metals on european chub by using a set of biochemical parameters. The increased level of RBCs, glycemia revealed the hematotoxic effect of heavy metals.

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