RESEARCH ON THE CHANGES OF SOME PHYSIOLOGICAL PARAMETERS IN PRUSSIAN CARP UNDER THE ACTION OF THE FASTER INSECTICIDE

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Abstract. The production and use of pesticides is a serious threat to the hydrosphere due to the discharge of residual water from factories of antiparasitic products or to the action of rainfalls, which remove these substances from the treated agricultural lands (MOHAN & ARDELEAN, 1993). The spread of pesticides has become a global issue, as they are present in all inland waters and carried by the rivers and streams in seas and oceans (BREZEANU & SIMON-GRUIȚĂ, 2002). The existence and evolution of aquatic organisms decisively depend on the environment they live in - water. In case of water bodies contaminated with pesticides, the environmental impact is mainly induced by the capacity of bioaccumulation and biomagnification of pesticides in the organisms living in water (GAVRILESCU, 2008). Of all the pesticides, the most important in terms of the danger of water contamination are particularly insecticides (MĂLĂCEA, 1969). This paper presents the results of the investigations on the changes of certain physiological indices in fish under the action of the insecticide Faster 10 CE, the active substance of which is cypermethrin at a concentration of 100 g/l. Cypermethrin is a synthetic non-systemic pyrethroid, with contact action and through ingestion. It is toxic to aquatic organisms and may cause long-term adverse effects in the aquatic environment. According to our investigations, it was found that the insecticide Faster 10 CE exerts an inhibitory effect on the oxygen consumption and respiratory rate in the investigated species (*Carassius auratus gibelio* Bloch); at the same time, it increased the number of erythrocytes and blood glucose levels having a hyperglycaemic effect. The performed experiments show that the insecticide Faster 10 CE is toxic to fish at very low concentrations.

Keywords: faster, fish, energetic metabolism, erythrocytes, glycaemia.

Rezumat. Cercetări asupra modificărilor unor parametri fiziologici la caras sub acţiunea insecticidului Faster. Producerea şi folosirea pesticidelor constituie o gravă ameninţare pentru hidrosferă în care ajung datorită deversărilor de reziduuri de la fabricile de produse antiparazitare sau datorită spălării acestor substanţe de apele de ploaie de pe terenurile agricole tratate (MOHAN & ARDELEAN, 1993). Răspândirea pesticidelor a devenit globală, fiind prezente în toate apele continentale şi duse de apele râurilor şi fluviilor în mări şi oceane (BREZEANU & SIMON-GRUIȚĂ, 2002). Existenţa şi evoluţia organismelor acvatice depind în mod hotărâtor de mediul în care evoluează - apa. În cazul apelor contaminate cu pesticide, impactul ecologic se datorează în principal capacităţii de bioconcentrare şi biomagnificare a pesticidelor din apă în organisme (GAVRILESCU, 2008). Dintre toate pesticidele, cele mai importante din punctul de vedere al pericolului de contaminare al apelor sunt îndeosebi insecticidele (MĂLĂCEA, 1969). Lucrarea prezintă rezultatele investigaţiilor referitoare la modificările unor indici fiziologici la peşti sub acţiunea insecticidului Faster 10 CE, ce are ca substanţă activă cipermetrinul în concentraţie de 100 g/l. Cipermetrinul este un piretroid de sinteză nesistemic, cu acţiune de contact şi prin ingestie. Este toxic pentru organismele acvatice, putând provoca efecte adverse pe termen lung asupra mediului acvatic. În urma cercetărilor efectuate s-a constatat că insecticidul Faster 10 CE are efect inhibitor asupra consumului de oxigen şi ritmului respirator la specia de peşti investigată (*Carassius auratus gibelio* Bloch); de asemenea a determinat creşterea numărului de eritrocite şi a nivelului glicemic, având efect hiperglicemiant. Experienţele efectuate ilustrează faptul că insecticidul Faster 10 CE este toxic pentru peşti în concentraţii foarte mici.

Cuvinte cheie: faster, peşte, metabolism energetic, eritrocite, glicemie.

INTRODUCTION

Numerous research studies on the biological effects of environmental pollution are geared toward the detection of the functional changes of the animal organism resulting from the action of certain chemical agents used in the industrial technological processes or agriculture, which are then collected by inland waters (DRĂGHICI, 1976; 1979).

The contamination of freshwater bodies with a wide range of pollutants has become a matter of concern in recent decades (VINODHINI & NARAYANAN, 2008).

As they reach the aquatic environment, pesticides affect a wide range of non-target organisms, such as invertebrates and fish (OTLUDIL et al., 2004). Because of the accumulation of pesticides in the tissues and organs of fish and other hydrobionts, they intoxicate and, in their turn, become sources of pollution.

ZAMFIR (1974), synthesizing a rich specialized literature, shows that pesticides detected in some food products today are incriminated to cause reproductive impairment of the body and induce mutagenic, teratogenic and carcinogenic effects.

Faster 10 CE is an insecticide in the form of an emulsifiable concentrate, having cypermethrin as the active ingredient in a concentration of 100 g/l. Cypermethrin is a synthesis non-systemic pyrethroid of Type II - alpha-cyano pyrethroid, which acts at contact and through ingestion.

Pyrethroid insecticides are highly toxic for fish. They damage the nervous system of invertebrates, inducing paralysis (MILLER & SALGADO, 1985). It has a broad spectrum of action, particularly on the species of Lepidoptera, Coleoptera and Diptera; it blocks the transmission of nerve impulses (BRADBURY & COATS, 1989).

Even low levels of cypermethrin in water can have significant effects on the reproduction and development in carp (AYDIN et al., 2005).

It inhibits the production of ATP-ase enzyme (CLARK & MATSUMURA, 1982). This is of great importance to understand why aquatic organisms are more sensitive to pyrethroid insecticides than terrestrial organisms.

It has a good residual action on treated plants, with inhibitory effect on the feeding process of phytophagous insects. It is very toxic to aquatic organisms and may cause long-term adverse effects in the aquatic environment.

Cypermethrin has relatively low toxicity to mammals and birds, although it is extremely toxic to fish and aquatic invertebrates (BRADBURY et al., 1989). It is classified in group III of toxicity.

MATERIAL AND METHOD

The research was conducted on the Prussian carp (*Carassius auratus gibelio* Bloch), which we sampled from the Olt River (Fig. 1). When choosing the biological material we have considered the physiological state, body integrity and size of the specimens, the sensitivity to toxic substances, as well as the fact that the species easily adapts to the conditions of retention in the aquarium, tolerates high temperatures and shows good resistance to oxygen deficiency having a low lethal hypoxic limit.

Figure 1. *Carassius auratus gibelio* (Bloch, 1783).

The introduction of fish in the aquariums was done about a month before the start of the experiments, to live them enough time to adapt to the new conditions. The aquariums used during the experiments had a capacity of poisoning of 20-30 litres and were equipped with lighting, stirring and aeration systems. These tanks have been cleaned and sanitized. In case of the variants made at 5-7ºC, fish were placed in refrigerators, lighting being artificial. The solutions of toxic substances from the aquariums have been renewed at intervals of 24 hours.

The specimens of Prussian carp used in different experimental variants were selected and sorted according to the weight category in order to avoid or, on the contrary, enhance, the effect of the individual factor represented by body weight. We used fish lots consisting in ten specimens each.

Of the three methods for determining the amount of dissolved oxygen (iodometric, gasometric and polarographic method), we used Winkler iodometric method in the achieved experiments, as described by the author at the end of the last century (1888). This method is considered the safest way to quantitatively assess the content of dissolved oxygen in water, under various temperature conditions. To assess the oxygen consumption it was used the technique of confined space described by Stroganov (1962), which allows to easily determine the exact quantity of the oxygen consumed by fish within a known time, based on the difference between the amount of oxygen measured in a control tank and the amount determined at the end of the period (quoted by PICOŞ & NĂSTĂSESCU, 1988).

The measurement of the respiratory rate was performed by successive determinations by means of a timer (their arithmetic mean representing the respiratory rate at the time).

The experiments that required series performed in different days were always made within a nyctemeral as narrow as possible (same time each day), in order to avoid any possible influence of the circadian variations of the oxygen consumption and respiratory rate.

As we aimed at achieving experimental conditions as close as possible to natural conditions, there have been kept and used in the experiment only the fish specimens found in the conditions of natural photoperiodism. Although it was not demonstrated a direct relationship between energy metabolism and light, it was taken into account the importance of the light factor in triggering certain functional manifestations.

Much attention was paid to the effect of fish handling during the pre-experimental stage, which may alter the results of the determinations (FRY, 1957; MARINESCU, 1972). For the experiments carried out by the technique based on the principle of confined space, where it is not possible to entirely avoid perturbing fish due to manipulation, we tried to limit its action. Consequently, for the experiment, we chose fish species characterized by low mobility (gibel carp) and we always performed control samples before the actual experiment.

For the experiments carried out to determine the change in the number of erythrocytes and blood glucose levels under the action of the insecticide Faster 10 CE, there were taken blood samples from the fish specimens. Blood samples were taken from the caudal artery. The number of erythrocytes was determined using Thoma counting chamber (PICOŞ & NĂSTĂSESCU, 1988).

The determination of the blood glucose level was performed using Accutrend GCT device that allows the measurement of its value in a drop of blood on a testing strip in a very short time. All the determinations performed on fish were carried out under strict control. There were eight experimental variants designed to determine the four physiological indices.

Measurements were made after 14 days of treatment at two thermal levels: 5-7ºC and 20-22ºC.

The concentrations of Faster 10 CE used in this work are 0.000005, 0.00001, 0.00002 and 0.00004 ml Faster 10 CE/l, respectively 0.5, 1, 2 and 4 µg cypermethrin/l.

RESULTS AND DISCUSSIONS

1. The action of the insecticide Faster 10 CE upon the oxygen consumption

The insecticide Faster 10 CE caused the intensification of the oxygen consumption in fish, within the first 72 hours after placing the fish in the toxic solution at lower concentrations (0.000005 and 0.00001 ml Faster 10 CE/l), the most pronounced increase in the energy metabolism occurring 24 hours after the exposure to the insecticide (114%, respectively 117% compared to the control group) (Fig. 2).

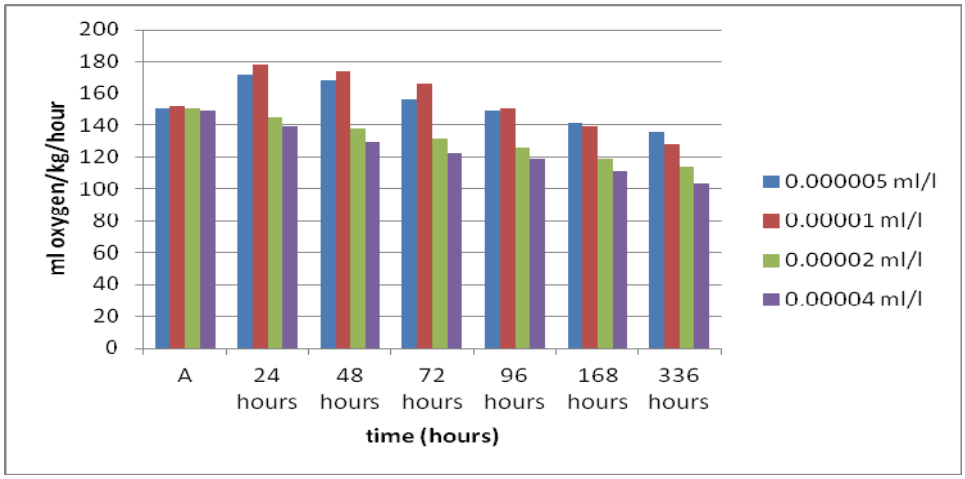

Figure 2. Variation of the average oxygen consumption at the Prussian carp specimens exposed to the action of the insecticide Faster 10 CE at different concentrations at 20-22°C.

The insecticide Faster 10 CE determined the decrease of the oxygen consumption at the end of the experiment (14 days), at all concentrations. The highest decrease was registered at the concentration of 0.00004 ml/l of Faster CE (30% less compared to the value recorded before the start of the experiment). In case of the first three concentrations, the reduction of the oxygen consumption was 10, 16 and 24% (in order of increasing concentration) compared to the control sample.

The temperature of 5-7°C determined the increase of the oxygen consumption at the Prussian carp specimens, during the first 72 hours after the exposure at lower concentrations (0.000005 ml and 0.00001 Faster 10 CE/l), as it was registered in case of the temperature of 20-22°C. The most pronounced increase in the energy metabolism was registered 48 hours after the exposure to the insecticide (110.3%, respectively 115.3% compared to the control group) (Fig. 3).

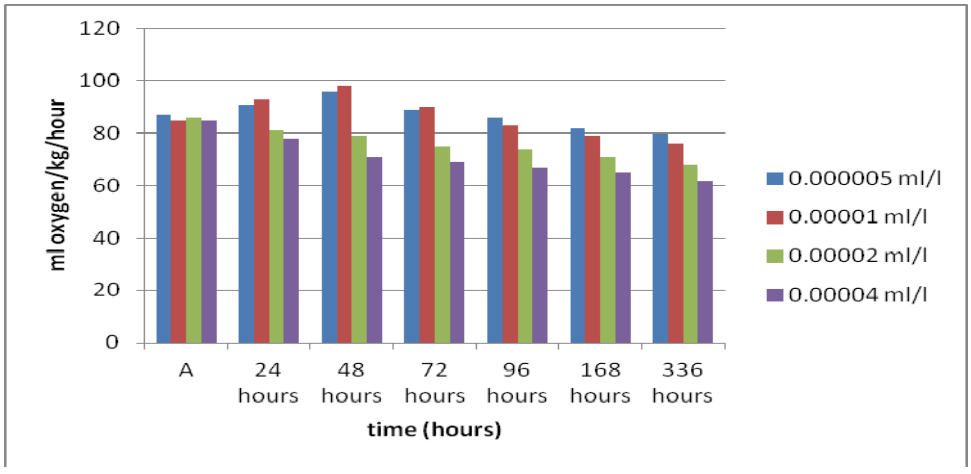

Figure 3. Variation of the average oxygen consumption at the Prussian carp specimens exposed to the action of the insecticide Faster 10 CE at different concentrations at 5-7°C.

The values registered at the end of the experiment are lower than the initial values in all the studied variants (8%, 11.5%, 21% and 27%). The reduction of the oxygen consumption is more pronounced at the temperature of 20-22°C.

2. Action of the insecticide Faster 10 CE upon the respiratory rate

The variation of the respiratory rate at the Prussian carp specimens exposed to the action of the insecticide Faster 10 CE at different concentrations at 20-22°C is rendered in figure 4.

In the first 24-48 hours, it can be noticed a slight increase in the respiratory rate and then, it begins to decrease, so that at the end of the experiment, the average values of this index are 20-30% lower compared to those registered before introducing the fish specimens in the toxic solution at all tested concentrations.

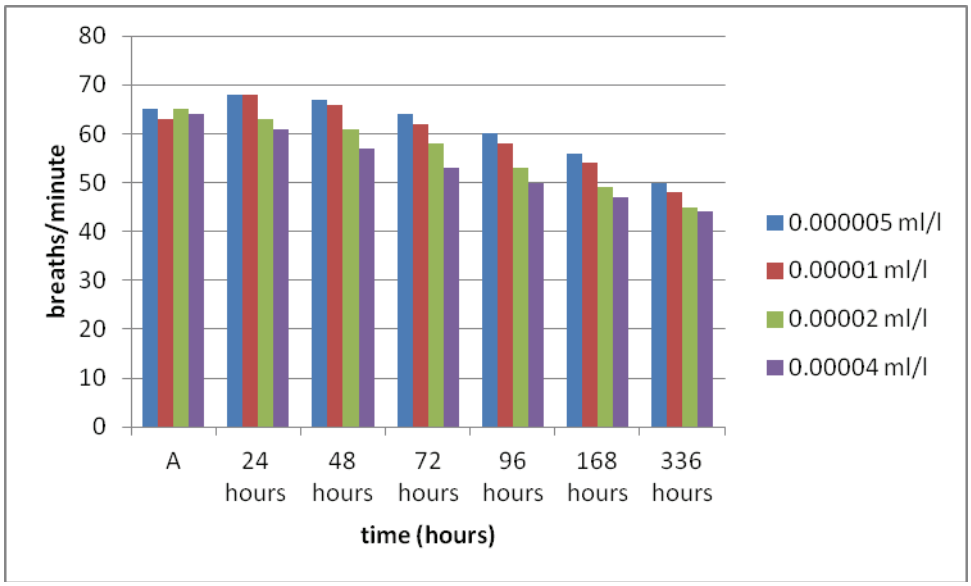

Figure 4. Variation of the respiratory rate at the Prussian carp specimens exposed to the action of the insecticide Faster 10 CE at different concentrations at 20-22°C.

Low temperature (5-7°C) reduced the respiratory rate throughout the experiment (Fig. 5), without registering an intensification of the index in the first hours after exposure, as it happened in case of the variants achieved at 20-22°C. The values determined at the end of the experiment are 17-26% lower compared to those registered before the fish exposure to the toxic substance. The reduction of the respiratory rate is more pronounced at 20-22°C.

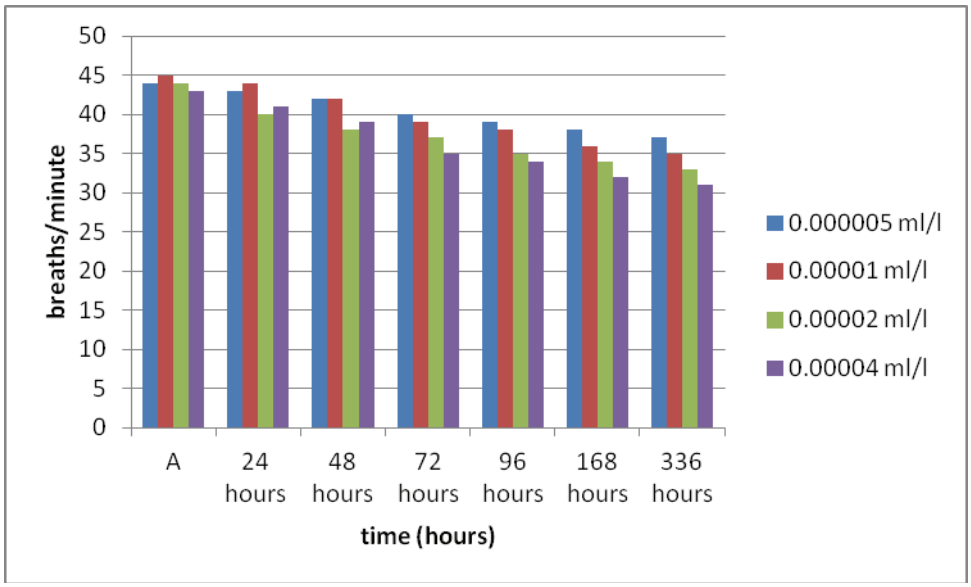

Figure 5. Variation of the respiratory rate at the Prussian carp specimens exposed to the action of the insecticide Faster 10 CE at different concentrations at 5-7°C.

The obtained results (oxygen consumption and respiratory rate) suggest a higher toxicity of the insecticide Faster 10 CE at the temperature of 20-22°C.

The decrease of the oxygen consumption can be explained by the histological changes in the gills occurred because of the toxic action of the insecticide Faster 10 CE.

COSTIN et al. (2007) and CENGHIZ (2006) reported changes at the level of the gills in the *Carassius auratus gibelio*, respectively, *Cyprinus carpio* after exposure to deltamethrin (hyperaemia, desquamation, necrosis, edema, lifting of lamellar epithelial, hyperplasia, aneurysm in the secondary lamellae, and fusion of secondary lamellae).

3. Action of the insecticide Faster 10 CE upon the number of erythrocytes

The number of erythrocytes at the Prussian carp specimens intoxicated with Faster 10 CE at a concentration of 0.000005 ml / l for 14 days, in case of both thermal levels, are rendered in figures 6 and 7.

At the temperature of 20-22°C, the insecticide Faster 10 CE at a concentration of 0.000005 ml / l determined the increase in the number of erythrocytes in exposed fish compared to the control group, where the increase was of only 4.44%.

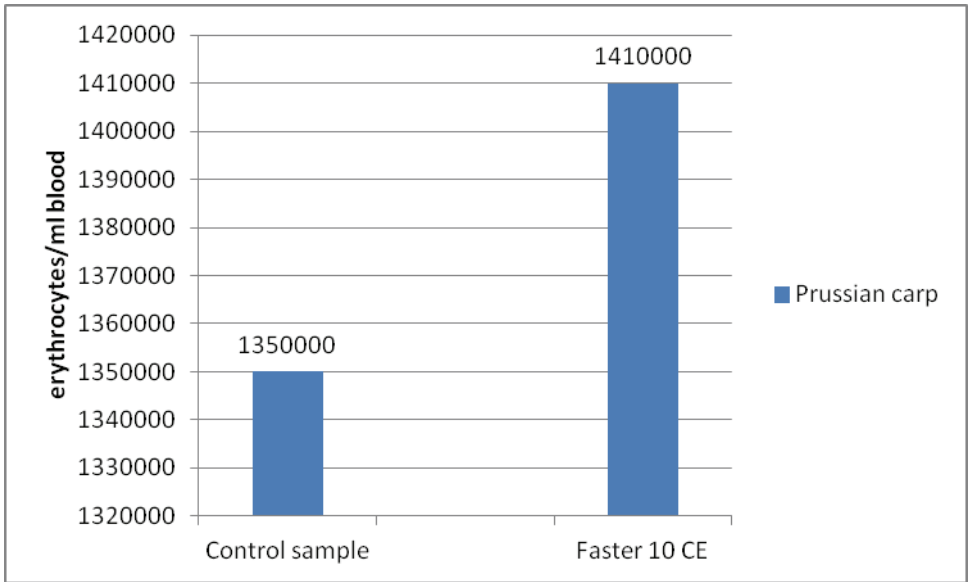

Figure 6. Action of the insecticide Faster 10 CE at a concentration of 0.000005 ml/l upon the number of erythrocytes at the Prussian carp at 20-22°C.

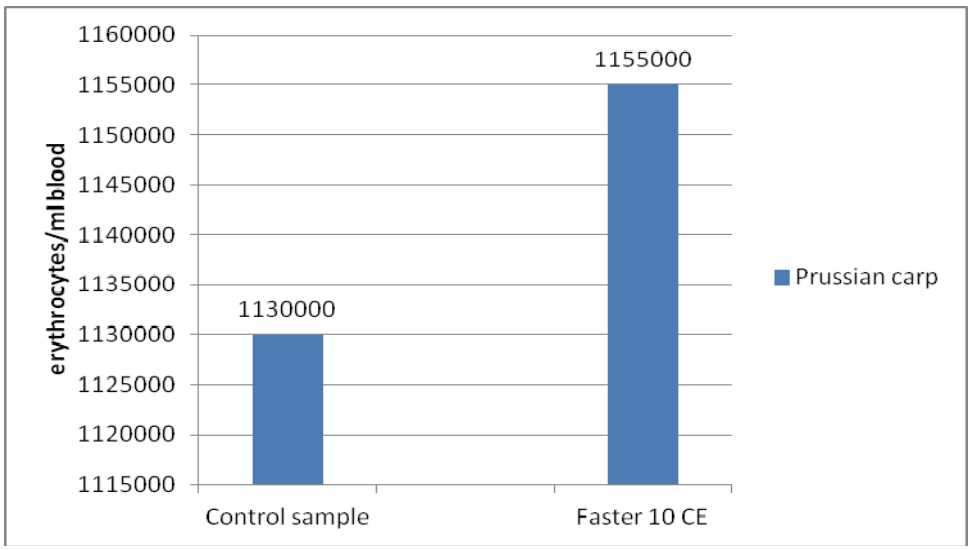

Figure 7. Action of the insecticide Faster 10 CE at a concentration of 0.000005 ml/l upon the number of erythrocytes at the Prussian carp at 5-7°C.

At the end of the experiment, at a temperature of 5-7°C, the number of erythrocytes increased by 2.21% as compared to the control group, the increase in this index being lower as compared to that registered at the temperature of 20-22°C. As the concentration of cypermethrin increased, it was also reported the increase in the number of erythrocytes, haemoglobin concentration, number of thrombocytes and sedimentation rate of erythrocyte in *Oncorhyncus mykiss* (ATAMANALP et al., 2002).

4. Action of the insecticide Faster 10 EC upon the blood glucose level

The temperature of 20-22°C increased the blood glucose level by 19.35% at the Prussian carp specimens intoxicated for 14 days with the insecticide Faster 10 CE at a concentration of 0.000005 ml / l compared to the control values (Fig. 8).

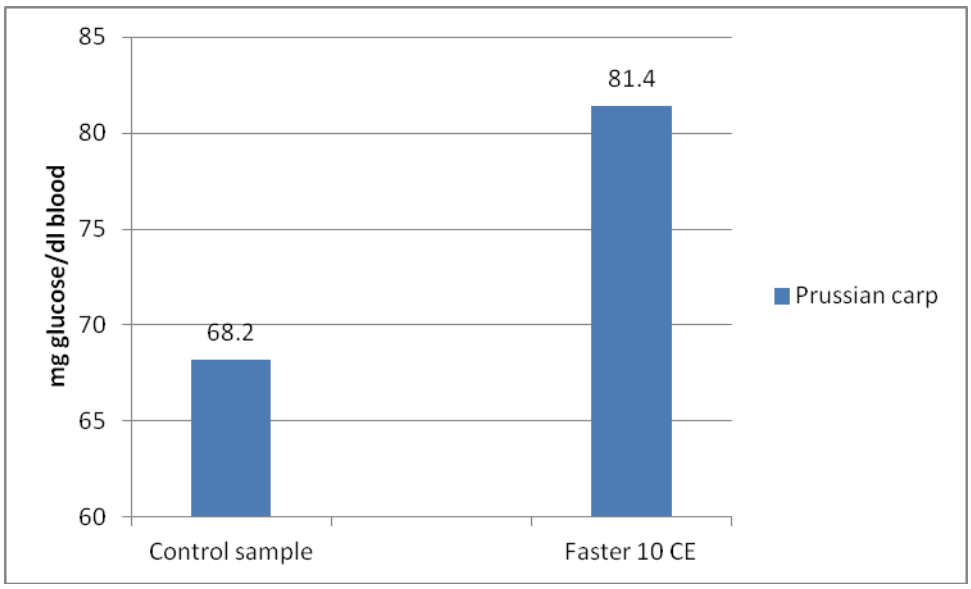

Figure 8. Action of the insecticide Faster 10 CE at a concentration of 0.000005 ml/l upon blood glucose level at the Prussian carp at 20-22°C.

The increase in the blood glucose level was also recorded at the temperature of 5-7°C (21.2% as compared to the control values) (Fig. 9).

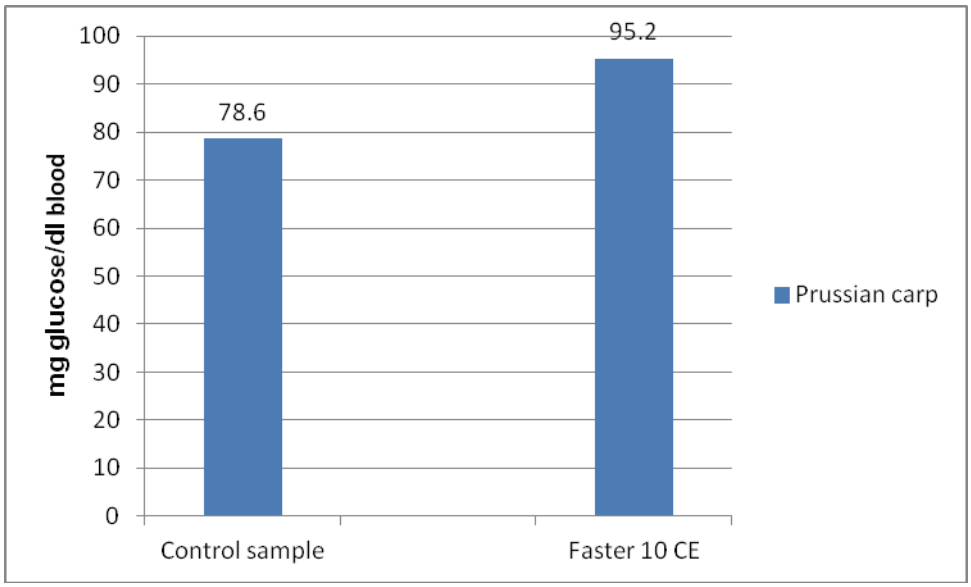

Figure 9. Action of the insecticide Faster 10 CE at a concentration of 0.000005 ml/l upon blood glucose level at the Prussian carp at 5-7°C.

A significantly higher level of blood glucose level was reported at the species *Korean rockfish* and *Sebastes schlegeli* by JEE et al. (2005) due to the long-term exposure to high concentrations of cypermethrin. This hyperglycaemia seems to determine an increase in the energy demand in fish under stress, in order to cope with the negative conditions imposed by the chronic exposure to toxic substances.

CONCLUSIONS

Low temperatures (5-7°C) determined the decrease in the oxygen consumption and respiratory rate, the reduction of the number of erythrocytes and the increase of the blood glucose level compared with the values registered at 20-22°C. The insecticide Faster 10 CE reduces the oxygen consumption and the respiratory rate in the Prussian carp (*Carassius auratus gibelio* Bloch). Initially, it caused a slight increase of these two physiological indices in the first 72 hours after the exposure to lower concentrations (0.000005 and 0.00001 ml Faster 10 CE/l); then, the values began to decrease, so that after 14 days of exposure to the insecticide, in all the used variants, the average values of these indices were lower than those registered before the exposure of fish to the toxic substance. The number of erythrocytes and

blood glucose levels increase when fish are exposed to the insecticide Faster 10 CE, as this insecticide has a hyperglycaemic effect.

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