

## RESEARCH ON THE EFFECTS OF PENDIGAN 330 EC HERBICIDE ON FIZIOLOGICAL INDICATORS AT *Carassius gibelio*, Bloch, 1782 UNDER DIFFERENT EXPERIMENTAL CONDITIONS

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### Abstract

*The herbicides are chemical substances used to destroy the dark weeds in agriculture (they represent 70% of pesticides) in water, in the embankments of the railway lines in the industrial areas and they are used in small amounts in forest, in wild habitats and in meadows.*

*Pendigan 330 EC is a herbicide from the group of toluidines which has the aspect of a clear, homogenous, flexible, yellow-brown liquid with a specific smell (aromatic) used to combat the annual monocotyledonous dark weeds and some annual dycotyledonous dark weeds from the sunflower, corn and vegetable crops (Matolcsy et al., 1988).*

*In this study we analysed the effects of the concentrations of the Pendigan 330 EC (0,002 ml Pendigan 330 EC/l water, 0,004 ml Pendigan 330 EC/l water, 0,008 ml Pendigan 330 EC/l water) herbicide over some physiological indices of Prussian carp (*Carassius gibelio* Bloch, 1782).*

*The inhibitor action of the herbicide Pendigan 330 EC manifested for the duration of the experiments over all physiological studied indices (oxygen consumption, respiratory rate, number of erythrocytes), the toxic effect being worse in the case of the experimental variations realized at room temperature 18-20°C compared to those realized at the temperature 6-8°C.*

*Keywords: number of erythrocytes, oxygen consumption, Pendigan 330 EC, Prussian carp, respiratory rate.*

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### 1. INTRODUCTION

The losses made worldwide by the different damaging organisms rises yearly at 35% of crops, which corresponds to 100 milliard dollars (Jäntschi, 2003). From these losses, the insects take 13,8% back of them, the mushrooms take 11,6% back and the dark weeds take 9,5% back and other organisms take 0,1%. These casualties vary in different areas of the world under the climatic conditions and under many other ecological factors (Jäntschi, 2003).

The herbicides are chemical substances used to destroy the dark weeds in agriculture (they represent 70% of pesticides) in water, in the embankments of the railway lines, in the industrial areas and they are used in small amounts in forests, in wild habitats and in meadows (Matolcsy et al., 1988).

Pendigan 330 EC is a herbicide from the group of toluidines which has the aspect of a clear, homogenous, flexible and yellow-brown liquid with a specific smell (aromatic) used to combat the annual monocotyledonous dark weeds and some annual dycotyledonous dark weeds from the sunflower, corn and vegetable crops (Matolcsy et al., 1988).

The excessive use of this herbicide proves itself to be harmful not only for the targeted plants, but also for many species of algae, crustacean and fish (Dimitrov et. al., 2006 ).

The active substance of the herbicide is pendimethalin 330g/l.

Dimitrov et al. (2006) showed an induction of the aberrations of the chromosomes and of the micronuclei in the cells of the bone marrow at the mice treated with pendimethalin. Pendigan 330 EC belongs to the fourth class of toxicity; The LC 50 values after 96 hours are: 3,189 mg/kg at mice, 1,421 mg/kg at wild ducks, more than 5000 mg/kg at rats, 0,138 mg/l at *Oncorhynchus mykiss*, 0,28 mg/l at *Daphnia magna*, at rainbow trout and catfish were 0,138 mg/l water, respectively 0,418 mg/l (EXTOXNET, 2013).

## 2. MATERIALS AND METHODS

In all the experimental variants was used as test animals Prussian carp.

The experiments were realized at two thermic levels, at the temperature between 6-8°C in the case of the variants kept in the refrigerators and respectively 18 - 20°C in case of the variants kept at lab temperature. The acclimatisation of fish at lab temperature was realized during 14 days in glass aquariums with 100 l storage capacity under the conditions of the natural photointervals. The fish wasn't fed during the experiments, avoiding in this way the intervention of the food factor (Hoar, 1942).

The lots fulfilled the necessary criteria (mortality under 5% in the first week and without mortality in the second week of acclimatisation), being shared in experimental variants (10 specimens /experimental lot).

The concentration of Pendigan 330 EC used were: 0,002 ml Pendigan 330 EC/ 1 water, 0,004 ml Pendigan 330 EC/ 1 water, 0,008 ml Pendigan 330 EC/ 1 water.

The determination of the oxygen consumption was realized through the method of confined space known also as the method of the closed respiratory room or Winkler method; to do the determinations, the fish is obliged to breath in a limited water volume (Picoş and Năstăsescu, 1988). These determinations were realized at intervals of 24, 48, 72, 96, 168 and 336 hours.

The determination of the respiratory rate was realized during the maintenance of the fish to realize the method of Winkler, were performed successive determinations of this index with the help of a timer until three close values were obtained (the arithmetic mean of these representing the respiratory rate at that moment). The determination of this rate was realized at intervals of 24, 48, 72, 96, 168 and 336 hours.

Through the determination of the oxygen consumption and of the respiratory frequency of the fish kept in sublethal doses of toxic substances result the concentrations at which start respiratory modifications to appear.

The determination of the erythrocytes was done with the help of a calculating room Thoma through the method described by Picoş and Năstăsescu (1988), of blood collected from the caudal artery.

The number of erythrocytes was determined after two weeks of toxicity exposure.

The nuclear erythrocytes of fish can be considered real nuclear biosensors for the ecotoxicological study of the complex pollutant mixtures, but also an alternative system for the ecological monitorisation of the aquatic medium, in a perfect harmony and in accordance with the legislation in force (Bratosin et al., 2004).

Depending on the physiological conditions, the number of erythrocytes vary: it grows before the breeding period, it grows again through regeneration and drops in autumn only to be again high in the winter. The fish which lives in the bottom of water has more hemoglobin than the pelagic one.

The quantity of hemoglobin modifies during the year. The fish acclimated in waters poor in oxygen has a low number of erythrocytes (Mălăcea, 1969).

### 3. RESULTS AND DISCUSSIONS

At the changing of the medium temperature, the majority of the physiological processes of the poikilothermic organisms modifies. Generally, the rate of the physiological processes increases with the temperature heat, until maximum, afterwards it decreases more rapidly or more slowly (Precht et al., 1973).

In this study we analyzed the concentrations effects of the Pendigan 330 EC herbicide under some physiological indices of Prussian carps at two thermic levels. The tested concentrations were established after the prior realization of the survival-tests and after the consultation of scientific literature.

The oxygen consumption is a physiological index very much used to determine the stress level of the fish. This suffers significant modifications in contact with the Pendigan 330 EC herbicide in all the concentrations that were used.

Pendigan 330 EC produces significant modifications of the oxygen consumption at both thermic levels, the effect of the herbicide being more stressed at the variants realized at the temperature of 18 - 20°C.

The decrease of the energetic metabolism can be also explained through the haematological modifications appeared as a result of the herbicide action.

Ahmad et al. (1995), reported significant reductions of the hemoglobin at the trout exposed to a sublethal dose of mancozeb, which led to the decrease of the oxygen quantity which reaches the human tissues and to the drop of energy production of animals. The alternatives of this physiological index at the carp exposed to the action of Pendigan 330 EC herbicide in different concentrations at two thermic levels are presented in the first figure.

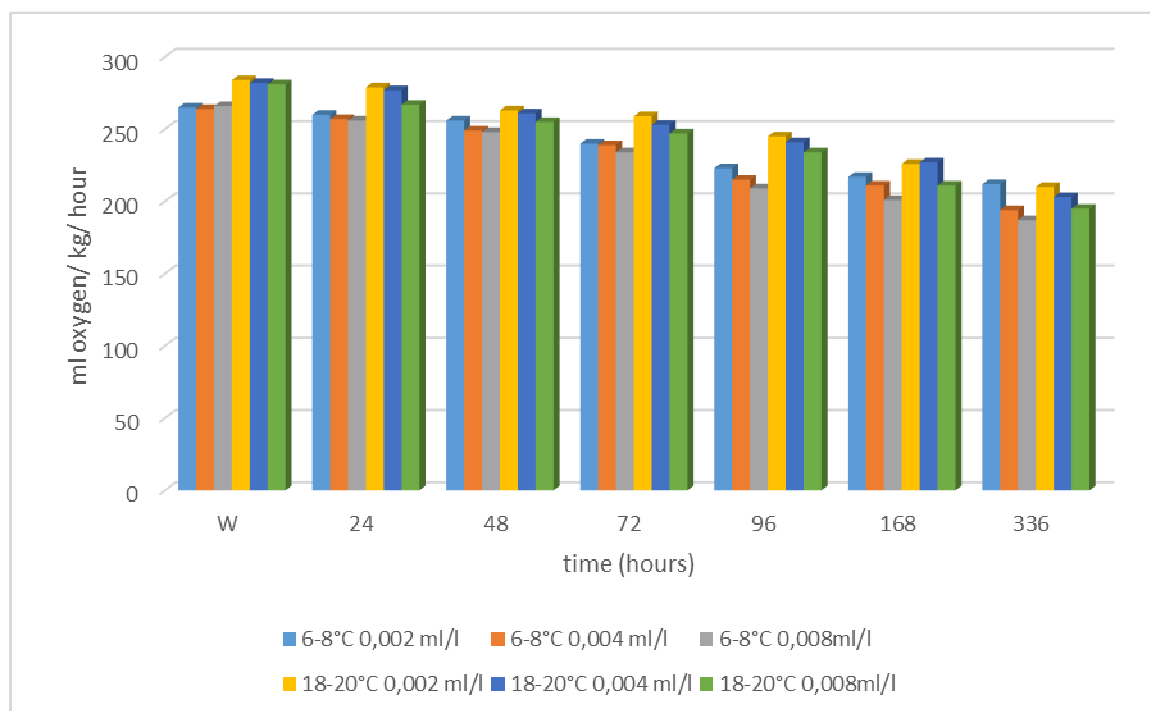


Figure 1. The variation of the oxygen consumption at the Prussian carps exposed to the action of Pendigan 330 EC herbicide in different concentrations at two thermic levels

Concerning the dynamic of the respiratory rate appear significant differences in both experimental variants, both at warm and at cold. The evolution of the respiratory rate should be associated with the evolution of the energetic metabolism presented in figure 1, the respiratory function being affected most likely due to the lesions which appear at branchial level.

The respiratory modifications are good indices of the animal's health, being related to the stress produced by factors as: the temperature, the salinity, the starving and the degree of pollution (Cebrián et al., 1992).

The variation of the respiratory rate at the carp exposed to the action of Pendigan 330 EC herbicide in different concentrations at two thermic levels is presented in figure 2.

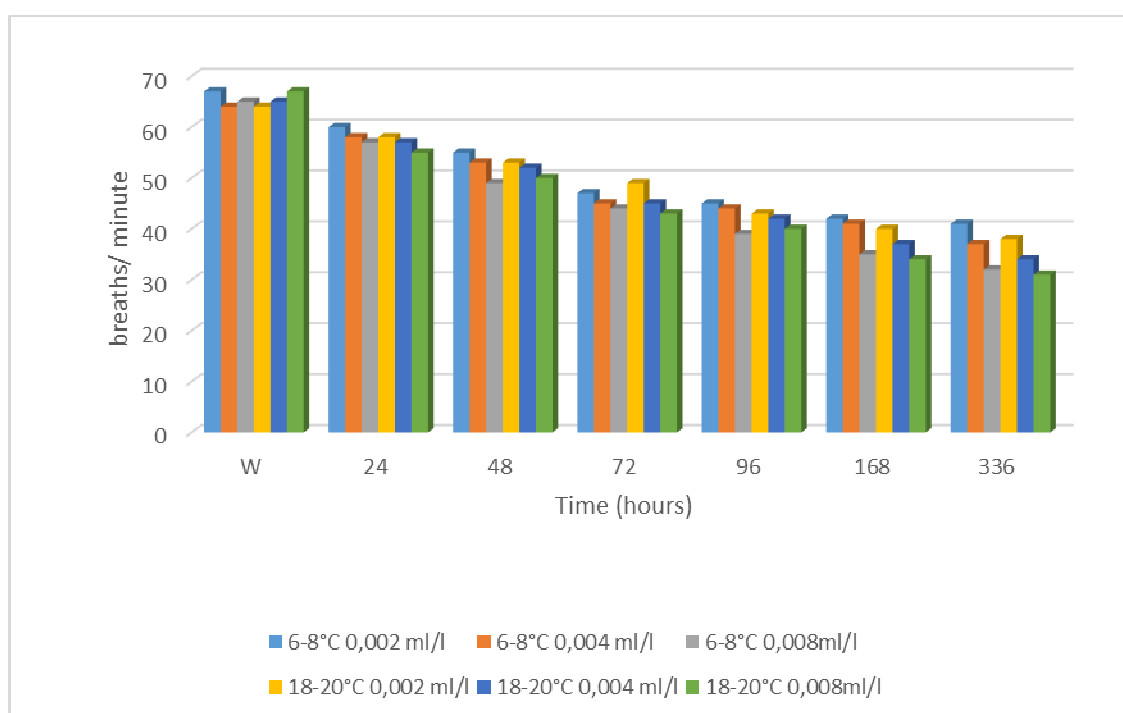


Figure 2. The variation of the respiratory rate at the Prussian carps exposed to the action of the Pendigan 330 EC herbicide in different concentrations at two thermic levels.

In figure 3 are represented the medium values of the erythrocytes number at Prussian carps exposed to the action of Pendigan 330 EC herbicide in two different concentrations at two thermic levels, after two weeks after the experiments installation.

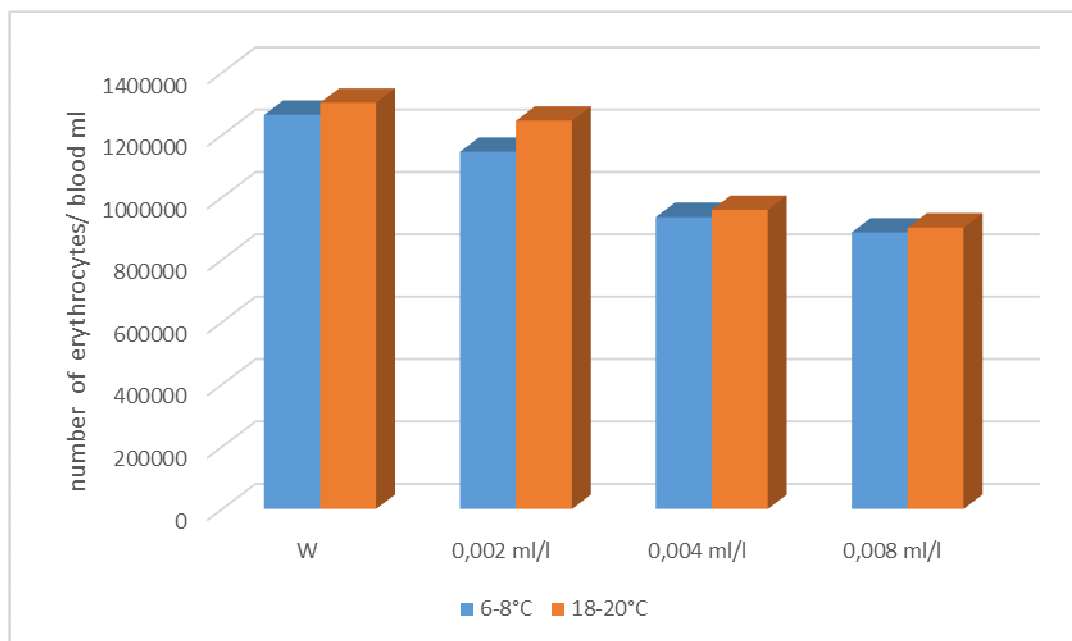
Significant modifications were registered at the haematological indices level, the herbicide Pendigan 330 EC being responsible of anemia installing, characterized through the drop of erythrocytes.

The red-pink color of the gills became red-white, and at the concentration of 0.008 ml/l the gills lost totally the red color, observing plenty mucus secretions.

The drop of erythrocyte number at the fish exposed to the low temperatures was reported by many researchers (Frankel et al., 1966; De Wilde and Houston, 1967; Houston and De Wilde, 1968; Huggurgs and De Wilde, 1969; Cameron, 1970 – cited by Precht et al., 1973).

Gabriel et al., mention that the drop of the erythrocyte number was can be the result of the imbalance between the haematofforming processe and in the hemolysis processes due to the toxicity

action, expressing through a decrease of erythrocyte synthesis or through a intensification of hemolysis.



**Figure 3.** The number of red blood cells for the Prussian carps (*Carassius gibelio* Bloch, 1782) 14 days after the exposure at different concentrations at two temperature levels

#### 4. CONCLUSIONS

Excepting the first concentration, which correspond to a quantity of Pendigan 330 EC of 0,002 ml/l, the herbicide modified significantly all the investigated physiological parameters.

In all the experimental variants was determined an inhibitor effect of the oxygen consumption, in the first 24 hours, the percentage-wise drops being more stressed at room temperature.

The effect of Pendigan 330 EC herbicide is inhibitor over the respiratory rate of Prussian carps at both thermic levels, the decrease of this physiological index is maximum after 14 days of contact with the herbicide in a concentration of 0,008 ml/l.

The medium number of erythrocytes register drops after 14 days from the fish immersion in different herbicide concentrations both at thye variants at room temperature as at the cold ones.

The anemia installed at the Prussian carps exposed to the herbicide Pendigan 330 EC may exist due to the hypoxia induced through the gill damage, because the red-pink color of the gills became red-white, and at the concentration of 0,008 ml/l the gills lost totally the red color, observing plenty mucus secretions.

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