

## **Biochemical parameters of blood in fish from Zaporozhian Reservoir**

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### **ABSTRACT**

It had been determined the contents of total protein, albumin, glucose, urea and activity of alanine aminotransferase (ALT), aspartate aminotransferase (AST),  $\alpha$ -amylase in 4-years old individuals of both sexes of prussian carp (*Carassius gibelio* Bloch, 1782) and roach (*Rutilus rutilus* Linnaeus, 1758) in different parts of the Zaporozhian Reservoir (Ukraine). The level of biochemical parameters depended on the extent of anthropogenic impact on the aquatic environment. Comparing to the prussian carp, Zaporozhian Reservoir roach was established to have a higher vulnerability to the effects of pollutants, that is why it can serve as a kind of test bioindicator for testing hydroecological system ihtiotoxicity.

### **1. INTRODUCTION**

Blood, as the internal environment of the body, quickly and accurately responds to environmental change, permanently and accurately reflects the physiological state of the body, indicating the nature and severity of abnormalities. This is a sensitive and informative status indicator defenses animals [1; 2].

Along with the production parameters of fish products, great importance also has its health status [3–6].

In addition, researching of blood parameters will make it possible to understand the biological and ecological characteristics of certain species of fish depending on their age and gender [7–11].

It is possible to get information about the condition of hydroecosystem from the biochemical parameters of fish blood. Blood quickly responses to the effects of various adverse factors and can serve as one of the early indicators of breach of fish's condition under the terms of pollution. This allows not only to evaluate and predict the environmental consequences of breaching the quality of the water environment, but also to develop methods for optimizing fish production in ponds [1; 12–14]. Researching the blood allows to determine the adaptive capacity of fish in specific water bodies and blood picture can be used as a standard of ecological and physiological condition of the fish during the period of active anthropogenic impact on water [10; 13; 15; 16].

Understanding of the mechanisms of fish adaptation to living conditions promotes to hematological parameters adaptations of fish researching with different degrees of anthropogenic impact reservoirs.

### **2. MATERIALS AND METHODS**

Researches were conducted in Zaporozhian Reservoir – the multi-purpose reservoir, which occupies a leading place among Ukrainian fishery ponds. According to the results of long-term monitoring studies zones with varying degrees and types of anthropogenic impact were found in the waters of the reservoir [17; 18].

Researches were taken in two parts of the reservoir, which differ in their ecological and hydrological conditions, are the main spawning areas and the most industrial mastered reservoirs.

The lower section of the reservoir near the villages Liubymivka and Viyskove characterizes with satisfactory water exchange, located in the agricultural zone and barely feels the impact of toxic industrial waste. Samara Bay at the confluence of the Samara River in the reservoir is characterized with low flowage and a large area of shallow water, which leads to a water "bloom" and congestion. Its hydroecological mode is determined by the impact of highly mine wastewater. The main components of polluting mine water are suspended substances and heavy metals. This area is characterized by high reservoir water mineralization (2.8 g/l).

Both sexes of 4-year prussian carp (*Carassius gibelio* Bloch, 1782) and common roach (*Rutilus rutilus* Linnaeus, 1758) were the objects of research. Material for the research was collected during the research catching in the autumn using gill nets. Blood were collected from the tail vein. Blood samples were centrifuged at 3000 rpm for 10 min.

Biochemical researches were conducted on automatic analyzers OLYMPUS AU400 and AU480 of Beckman Coulter (US), by Beckman Coulter reagents in the laboratory "INVITRO" in Dnipropetrovsk. Methods for determining the activity of alanine aminotransferase (ALT), aspartate aminotransferase (AST),  $\alpha$ -amylase and total protein, albumin, glucose and urea are based on the recommendations of the International Federation of Clinical Chemistry (IFCC).

The data was analyzed as Mean  $\pm$  S.E.M. at reliability 95 % and significant level of  $P < 0.05$ . In order to test the significant of the test, t-test method was used.

### 3. RESULT AND DISCUSSION

The blood biochemistry indicators of prussian carp and roach caught from the two plots of Zaporizhian reservoir, indicating the water-salt, protein metabolism, which indicate the physiological state of the organism were researched.

The concentration of serum protein in fish's blood is an indicator of the general state of their health. According to the results of our research, level of total protein in serum of silver carp's blood from the lower part of Zaporizhian reservoir ranged from  $38.1 \pm 5.5$  g/l near Liubymivka to  $48.5 \pm 6.2$  g/l near Viyskove (Fig. 1). In roach protein content ranged from  $42.6 \pm 9.7$  to  $56.7 \pm 11.2$  g/l near Liubymivka and Viyskove respectively. A few researches of protein in the blood of fish show that its amount varies in large limits, not only among the whole group of fish, but also within the same species. These fluctuations are connected with metabolism and determined by the intensity and the nutrition. Physiological rate of protein blood serum contents, according to other authors, is 59.0 g/l for roach and 27–56 g/l for prussian carp. The high protein content within the established norms is a favorable sign; significant losses of protein are associated with a decrease in viability and may be accompanied by loss of fish. Low values indicate depletion, infectious diseases, kidney damage [14].

In Samara Bay total protein in serum were  $80.5 \pm 9.5$  g/l in prussian carp and  $83.1 \pm 13.5$  g/l in roach that prevailed normal value. The concentration of protein increased in both species respectively 46.5% and 66% compared with fish caught near the village Viyskove, and almost in two times comparing with Liubymivka ( $P < 0.05$ ). The cause of elevated level of protein in the fish blood from Samara Bay can be increased content of toxic substances in the area, including heavy metals that affect the liver, which synthesizes albumin and globulins. Increased concentrations of protein in blood plasma can be caused by structural changes in the liver, which lower the aminotransferase level with the simultaneous reduction of desamidization strength [11].

The main fraction of total protein is albumin. Normally albumin fraction of the total protein should be up to 55%. In the researched fish of the lower part of the reservoir, this fraction is up to 51%, and for fish from Samara Bay it is conversely lowered, which can indicate an excessive water content in tissues and a breach of the functional state of the liver, where the synthesis of albumin and is detoxification processes are held [19]. Due to toxic substances albumin is primarily used for energy purposes [20] (Fig. 2).

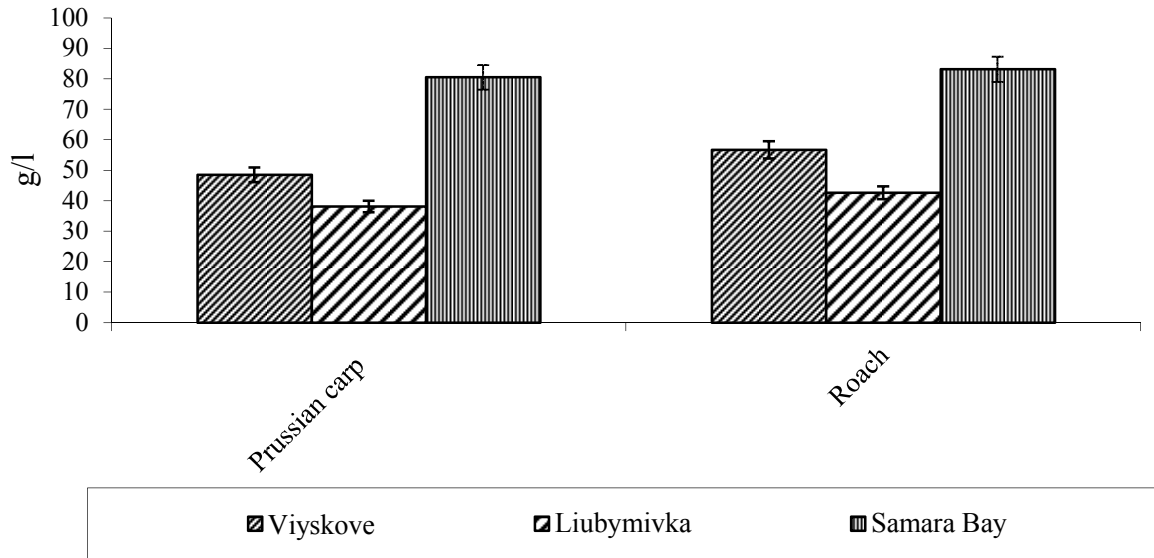


Fig.1. The protein content in blood serum of prussian carp and roach from Zaporozhian Reservoir

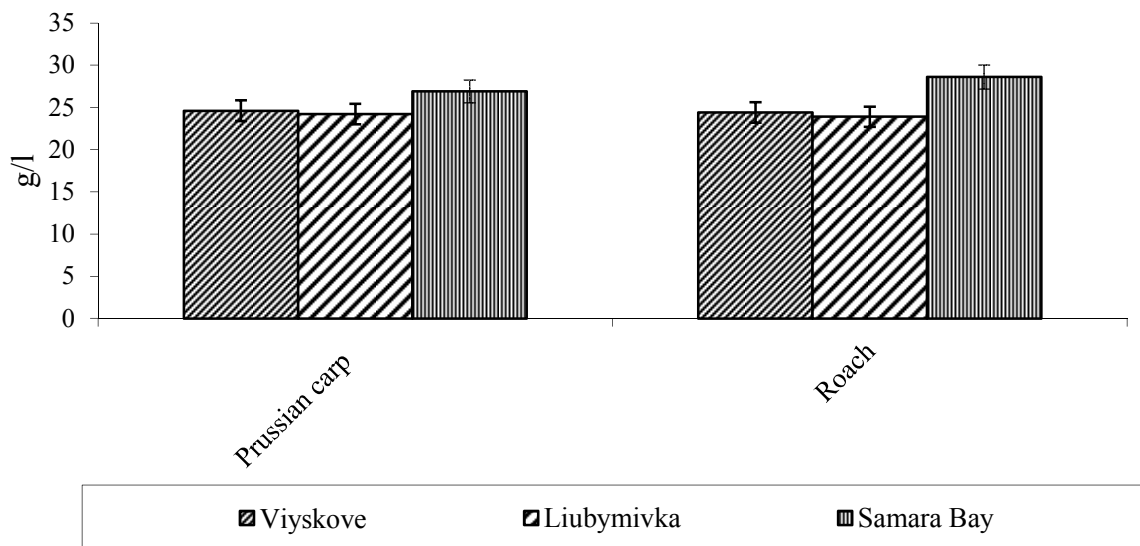


Fig. 2. The content of albumin in blood serum of prussian carp and roach from Zaporozhian Reservoir

High rates of glucose in blood serum of roach from Samara Bay were revealed. The fish caught from Samara Bay, glucose level exaggerated for 35.6% corresponding values obtained from roach near the village Viyskove, and 60.1% – near the village Liubymivka. Glucose level was higher in roach comparing to prussian carp for 30 and 69% in Viyskove and Samara Bay respectively ( $P < 0.05$ ) (Fig. 3). The increase of glucose in blood is observed with increasing glucocorticoids in body during stress conditions, with peculiarities of nutrition. This can be associated with hyperglycemia, which is caused by the heavy metals that were proved before experimentally [21].

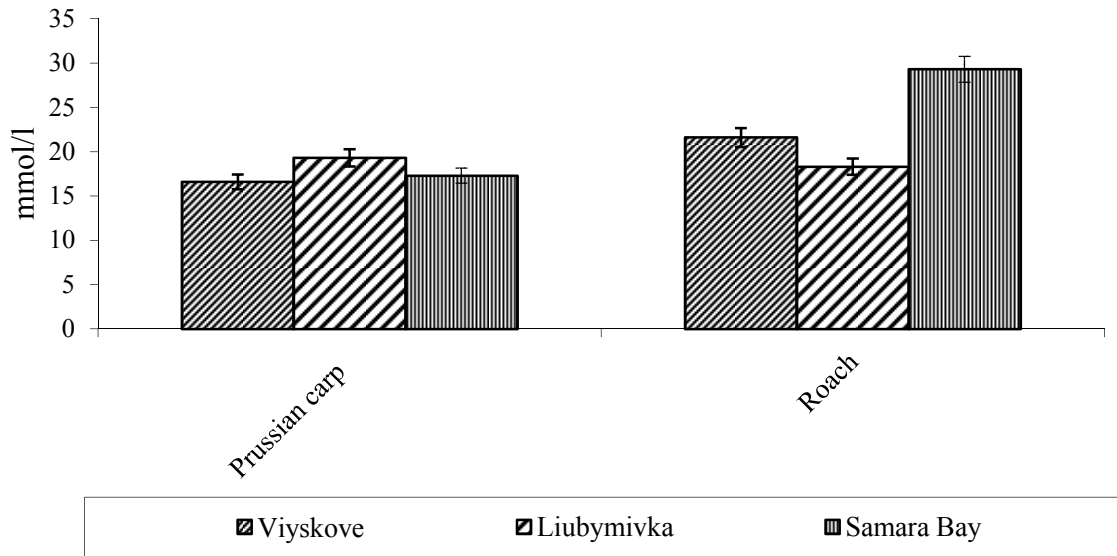


Fig.3. The content of glucose in the blood serum of prussian carp and roach from Zaporozhian Reservoir

The exchange of certain characteristics of nitrogen compounds was found in the researched fish. The content of urea in prussian carp from Samara Bay increased by 43% more than in fish from the lower part of the reservoir ( $P < 0.05$ ). A similar trend is observed in roach ( $P > 0.05$ ). Significantly bigger content (39%) in blood serum urea of prussian carp from Samara Bay compared to roach was set (Fig. 4). Increased urea level can indicate more charge on the kidneys in fish from the Samara Bay, which can be caused by bigger human pressure on the investigated area. Toxins reduce urea excretion by the kidneys and contribute to its accumulation in the blood [20].

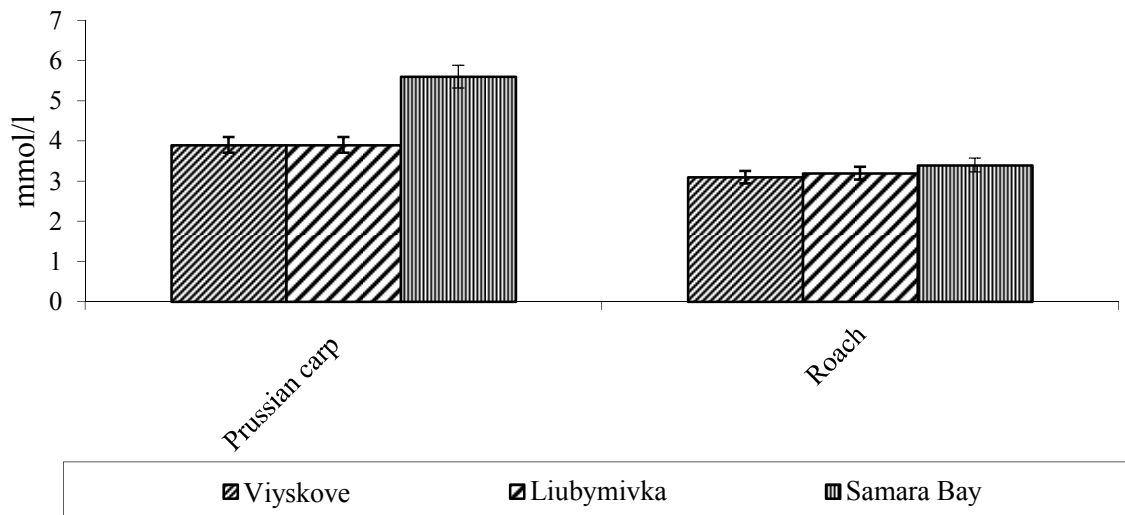


Fig. 4. The content of urea in the blood serum of prussian carp and roach Zaporozhian Reservoir

The activity of  $\alpha$ -amylase in serum of researched fish caught near the villages Viyskove and Liubymivka, varied considerable from  $268.8 \pm 45.3$  to  $303.9 \pm 35.1$  U/l for prussian carp and from  $549.8 \pm 46.9$  to  $986.2 \pm 32.6$  U/l for roach (Fig. 5). In Samara Bay  $\alpha$ -amylase activity level in blood of prussian carp and roach prevailed in 43.0–66.4% following indicators of fish from the lower part of the reservoir, which can indicate liver dysfunction due to significant toxic charge and adverse conditions. Also, roach has significantly higher amylase content than prussian carp to 44.0–69.0% ( $P < 0.05$ ).

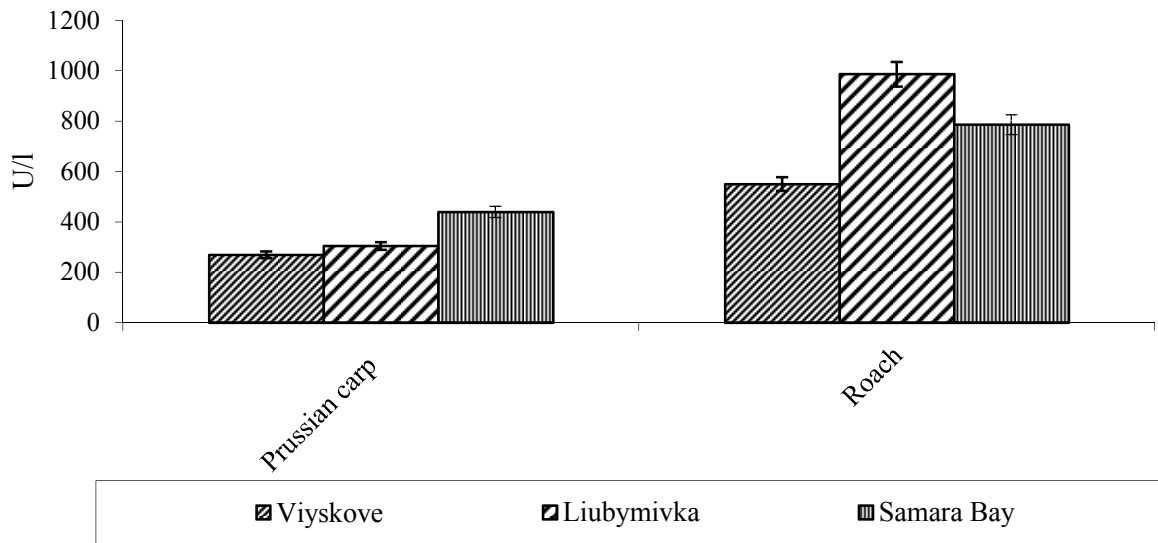


Fig. 5. Alpha-amylase activity in blood serum of prussian carp and roach from Zaporozhian Reservoir

Increased activity of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are the markers that indicate abuse and damage of muscles, liver and other internal organs [12; 23]. The results show that indicators of ALT activity in blood serum of prussian carp from Samara Bay exceeded for 47.3% the rate of fish caught near the village Viyskove, and for 72.4% than near the village Liubymivka. In roach indicators ALT activity in Samara Bay were higher than ALT activity level in fish near the village Viyskove and Liubymivka respectively for 33.9 and 57.8% (Fig. 6). AST activity of prussian carp from Samara Bay did not changed significantly compared with the lower section, but in roach indicators AST activity in serum exceeded the 57.4% rate of fish caught near the village Viyskove, and 76.9% – near the village Liubymivka (Fig. 7).

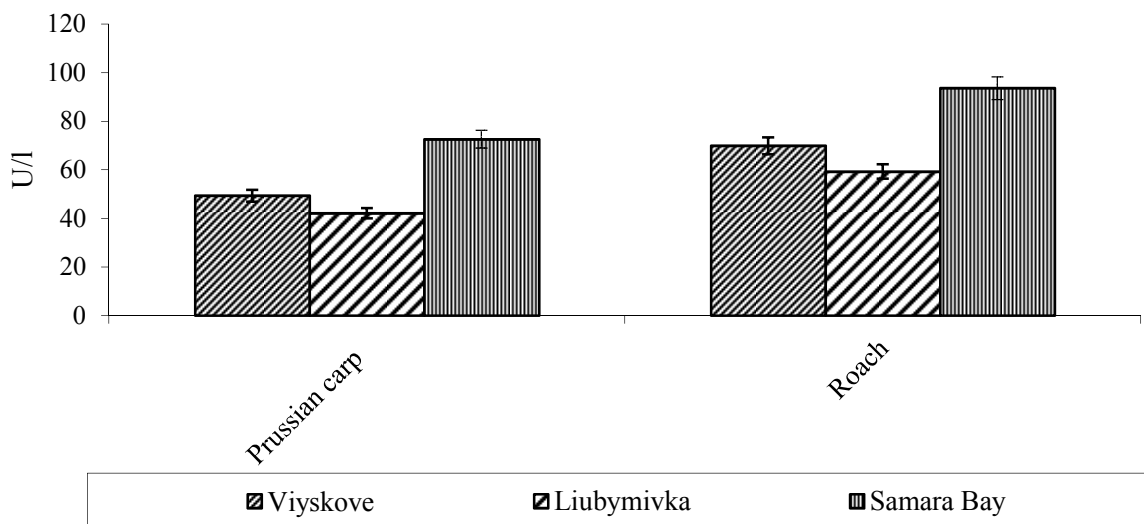


Fig. 6. Alanine aminotransferase activity (ALT) in blood serum of prussian carp and roach from Zaporozhian Reservoir

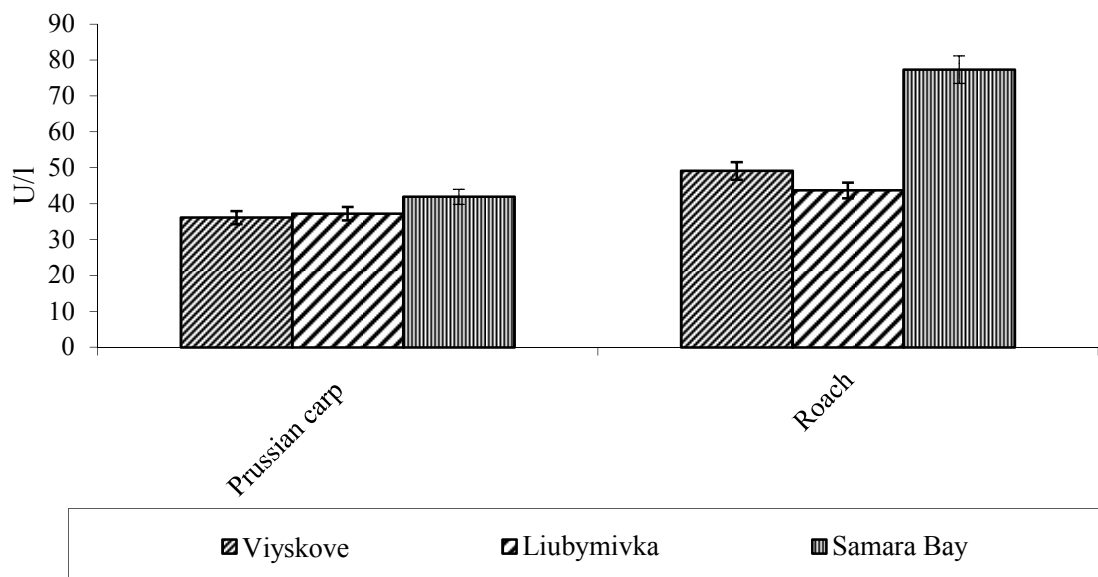


Fig. 7. Aspartate aminotransferase activity (AST) in blood serum of prussian carp and roach from Zaporozhian Reservoir

Specific features of ALT and AST activity in blood serum of researched fish was Established, as it is shown by other researchers [22]. Thus, the activity of these enzymes was significantly lower in for prussian than for carp roach – ALT 22 – 29% and AST – 15 – 46% in different parts of the reservoir.

#### 4. CONCLUSION

Thus, statistically significant deviations of the protein content, activity of  $\alpha$ -amylase, ALT, AST show a significant impact of toxicants on fish from the bay of Samara, primarily on the liver, which serves as a detoxification organ.

The increase in enzyme activity is a thin mechanism that is used for certain metabolic processes to adapt to adverse conditions.

Elevated rates of hepatic complex ( $\alpha$ -amylase, ALT, AST) and changes in protein fractions of fish caught in the bay of Samara were identified, indicating chronic intoxication of organism.

Based on these data we can determine greater vulnerability of common roach (*Rutilus rutilus*, L.) from Zaporozhian Reservoir comparing to prussian carp (*Carassius gibelio*, B.) to the effects of pollutants, that is why it can serve as a kind of test bioindicator for testing hydroecological system ihtiotoxicity.

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