SCIENTIFIC HORIZONS

Journal homepage: https://sciencehorizon.com.ua
***Scientific Horizons*, 26(5), 21-36**

UDC 639.37:612.015.3:597.551.2
DOI: 10.48077/scihor5.2023.21

Influence of various phosphoric concentrations on tissue and intracellular metabolism of *Cyprinus Carpio* L. in aquatic habitat

Nataliia Hrynevych*
Doctor of Veterinary Sciences, Professor
Bila Tserkva National Agrarian University
09111, 3a Heroes of Chernobyl Str., Bila Tserkva, Ukraine
https://orcid.org/0000-0001-7430-9498
Mykola Svitelskyi
Candidate of Agricultural Sciences, Associate Professor
Polissia National University
10008, 7 Staryi Blvd., Zhytomyr, Ukraine
https://orcid.org/0000-0003-1501-4168
Oleksandr Khomiak
Candidate of Agricultural Sciences, Associate Professor
Bila Tserkva National Agrarian University
09111, 3a Heroes of Chernobyl Str., Bila Tserkva, Ukraine
https://orcid.org/0000-0003-3010-6757
Oksana Ishchuk
Candidate of Agricultural Sciences, Associate Professor
Polissia National University
10008, 7 Staryi Blvd., Zhytomyr, Ukraine
https://orcid.org/0000-0002-8993-8366
Svitlana Matkovska
Candidate of Agricultural Sciences, Associate Professor
Polissia National University
10008, 7 Staryi Blvd., Zhytomyr, Ukraine
https://orcid.org/0000-0002-8019-5498

Article's History:
Received: 5.03.2023
Revised: 10.04.2023
Accepted: 6.05.2023

Suggested Citation:
Hrynevych, N., Svitelskyi, M., Khomiak, O., Ishchuk, O., & Matkovska, S. (2023). Influence of various phosphoric concentrations on tissue and intracellular metabolism of *Cyprinus Carpio* L. in aquatic habitat. *Scientific Horizons*, 26(5), 21-36.

Abstract. The research relevance is predefined by the fact that under the influence of various factors of the aquatic environment, changes in the speed and direction of the metabolic processes of hydrobionts are recorded. The research aims to study the influence of different inorganic phosphorus content in the aquatic environment on the indicators of phosphorus-calcium exchange in fish. The methods of thin-layer chromatography and variational statistics were used. Glandular tissues of the gills, liver and kidneys of fish were analyzed. To study the influence of inorganic phosphorus in the water environment on some indicators of intracellular metabolism, mitochondria were isolated in osmoregulatory organs. It was found that when the concentration of phosphorus increases to 0.3-0.6 mg/L in the water environment,

*Corresponding author

it accumulates in the liver, and gills and decreases in the kidneys, while the concentration is maintained at a relatively constant level in the blood serum of fish. An increase in the concentration of inorganic phosphorus in the water environment significantly affects the processes of energy generation in the mitochondria of fish livers. A change in the phosphorus content in the habitat of fish has a significant impact on tissue bioenergetic processes, which is manifested in a change in the adenosine triphosphate content, and alkaline phosphatase activity. Significant changes in these indicators are observed in the gills and kidneys. The research results can be used for the formation of adaptive and compensatory regulatory mechanisms in the organism of hydrobionts during their adaptation to certain conditions of cultivation and reproduction

Keywords: *Cyprinus carpio*; fish liver; gill; kidney; blood serum; bile

INTRODUCTION

All aquatic animals require minerals for their vital physiological and biochemical functions and their normal life processes. Regulation of phosphate is considered more critical than that of calcium because fish must effectively absorb and conserve phosphate in both freshwater and seawater environments.

A study by (Costa *et al.,* 2018; Boyd *et al.,* 2020) found that phosphorus is an essential nutrient in all aquatic ecosystems. The solubility of inorganic phosphorus in water systems is regulated by the physical and chemical characteristics of the water column. Zhao *et al.* (2019), and Yang *et al.* (2021) believe that, unlike terrestrial animals, which receive phosphorus compounds mainly with food, hydrobionts can assimilate it from the aquatic environment.

According to (Sugiura *et al.,* 2018; Lei *et al.,* 2021) phosphorus is one of the components of nucleic acids DNA (deoxyribonucleic acid) and RNA (ribonucleic acid) and phosphoproteins ADP (adenosine diphosphate) and ATP (adenosine triphosphate), therefore it is a necessary element for the existence of biological systems.

Ai *et al.* (2019), Zhao *et al.* (2021), and Yu *et al.* (2021), note that phosphorus is an important component of the endoskeleton of fish. More than a third of phosphorus in the body is contained in phospholipids, cell membranes and energy-rich compounds. Thus, phosphorus plays an important role in carbohydrate, lipid and nitrogen metabolism, in the metabolism of muscle and nerve tissues, as well as in various metabolic processes involving buffers in body fluids.

The effect of increased levels of inorganic phosphorus in water on metabolic processes in the body of fish was studied by (Jothy *et al.,* 2019; Zhao *et al.,* 2019; Wang *et al.,* 2022). At the same time, special attention was paid to the orientation of bioenergetic processes, since phosphate, which is utilized from water and food, can be a part of high-energy compounds that play an important role in the life support of all living organisms without exception.

According to the data obtained by (Solomatina *et al.,* 2018; Pouille *et al.,* 2018; Saurette *et al.,* 2019), the lack of mineral phosphorus compounds is a limiting factor for all hydrobionts. Phosphorus not only affects bone mineralization but also metabolic processes in fish, such as nutrient digestion and lipid catabolism in the liver. According to (Souders *et al.,* 2018; Musharraf *et al.*, 2019; Wang *et al.,* 2022), phosphorus is present in water bodies in the form of insoluble mineral compounds – tricalcium phosphate or as part of organic compounds contained in plant and animal remains and excrement of living organisms. Organic and inorganic phosphorus compounds are mineralized and solubilized by microorganisms (bacteria and fungi).

Huang *et al.* (2019), Svitelskyi *et al.* (2020), and Zhang *et al.* (2022) established that the content of mineral phosphorus in the water environment ranges from 0.02 to 0.6 mg/L. Phosphorus mineral compounds reach their maximum concentration in water in summer, as their amount formed in the process of regeneration of organic substances exceeds consumption by phytoplankton. The level of phosphates in reservoirs changes significantly as a result of anthropogenic influence.

Fedonyuk *et al.* (2019), and Xu *et al.* (2021) note, establishing the optimal concentration of phosphorus in the water is very important for the vital activity of fish. The role of phosphorus in hydrobionts is determined by the features of its entry into the body. Phosphorus ions entering the body of fish from the environment are found in large quantities in the places of penetration and absorption – gills, mucous membrane of the mouth, intestinal walls, and skin, but are also absorbed during metabolic processes. It should be noted that (Lall, 2022) experimental studies that reveal the peculiarities of the use of phosphorus by fish were carried out using radioactive and elemental phosphorus.

A review of literature sources indicates a lack of information regarding the impact of inorganic phosphorus in the aquatic environment on its accumulation in organs and tissues, as well as related changes in tissue metabolism. Therefore, the goal of this research was to establish the influence of different inorganic phosphorus content in water on metabolic processes in the body of fish.

MATERIALS AND METHODS

The research was conducted during the autumn period of 2022 in the conditions of Limited Liability Company "Skvyraplemrybhosp" of Bilotserkiv district, Kyiv region. The study analysed 50 specimens of two-year-old carp (*Cyprinus carpio* L.) with an average weight of 255.0±9.7 g. The study of the effect of different concentrations of phosphorus in the aquatic environment on *Cyprinus carpio* L. was carried out in the glandular tissues of the gills, kidneys, liver, serum and bile after 1 and 7 days of exposure. Before the experiment, the caught fish were kept in stationary containers with a volume of 4 m^3. Aquariums were filled with settled tap water with the main mineral components' concentration: Na^+–11.7; K^+–6.4; Ca^{2+}–50-100.0; Mg^{2+}–120.0 mg/L.

Determination of the ATP (adenosine triphosphate), ADP (adenosine diphosphate) and AMP (adenosine monophosphate) levels were carried out by thin layer chromatography on Merck plates. The activity of Na^+ K^+ Mg^{2+}– adenosine triphosphatase and alkaline phosphatase was judged by the increase of inorganic phosphorus in the incubation medium, consisting of adenosine triphosphatase – 0.025 M tris – HCl; 0.1 M NaCl; 0.005 M $MgCl_2$; 0.02 M KCl, 0.001 M ATP. Incubation period – 1 hour. For alkaline phosphatase, the incubation medium consisted of 2 ml of 1% Na-β-glycerophosphate solution, 0.5 ml 0.001 M $MgCl_2$. Incubation period – 2 hours. Enzymatic activity was expressed in μg phosphorus/mg protein/1 hour. Lipid and protein phosphorus had been determined after their extraction from tissues of homogenates with chloroform-ethanol mixture and combustion in a mixture of sulfuric and nitric acids, inorganic phosphorus, and total phosphorus according. To study the impact of phosphorus in the aquatic habitat on some indicators of their intracellular metabolism in osmoregulatory organs, subcellular particles were isolated, particularly mitochondria, where the main biosynthetic and bioenergetic processes of the cell took place. To obtain mitochondria, the tissue was homogenized in a homogenizer with a Teflon pestle diluted with the environment at 1:7. Debris of the cell and nucleus were precipitated at 3000 rpm/min – 5 minutes. Isolation of mitochondria had been carried out at 12-14 thousand rpm/min. After washing twice, the mitochondria were diluted based on a ratio of 0.2 ml of environment per original gram of tissue. Mitochondria, isolated from fish liver, contained 36-40 mg of protein per millilitre and 10-15 mg from gills.

During the experiment, microscopic research methods and the thin-layer chromatography method were used. Study results were calculated as average ± standard deviation (SD). The obtained digital data were processed using standard methods of variational statistics and special computer programs MS Excel and Statsoft Statistica 6.0.

The Ethical Committee approved the use of animals in this study of the Bila Tserkva National Agrarian University on the treatment of animals in research and the educational process (protocol No. 9 of October 1, 2020) following the Law of Ukraine "On the Protection of Animals from Cruelty" (Law of Ukraine…, 2006) and Directive 2010/63/EC of the European Parliament and of the Council of 22 September 2010 on the protection of animals used for scientific purposes (Directive 2010/63/EU…, 2010).

RESULTS

The data received on the tissue content of total phosphorus confirms the conclusion about the great significance of the gills in their absorption from water. Thus, the quantity of total phosphorus in the glandular tissue of the fish's gills, contained in a habitat with 0.3 and 0.6 mg/L of phosphorus, had increased sharply, especially after keeping them for 7 days in such an environment (Table 1).

Table 1. *Influence of phosphorus various concentrations in aquatic habitat on the total phosphorus content in tissues (% dry tissue) and liquids (mg %) of the carp body*

Phosphorus concentration in water, mg/L	Day of experiments			
	1		7	
	M±m	Deviation from control, %	M±m	Deviation from control, %
Liver				
0.06 (conditional control)	1.16±0.17	–	1.43±0.07	–
0.3	0.85±0.06	-26.72	1.85±0.14*	+29.37
0.6	1.42 0.21	+22.41	1.42±0.12	–
Gills				
0.06 (conditional control)	3.36±0.24	–	3.36±0.24	–
0.3	3.32±0.27	+13.69	5.14±0.46*	+52.38
0.6	2.84±0.09	-15.48	4.02±0.20*	+19.14
Kidneys				
0.06 (conditional control)	1.37±0.18	–	2.16±0.08	–
0.3	1.65±0.04	+20.44	1.32±0*	-38.89

Table 1, Continued

Phosphorus concentration in water, mg/L	Day of experiments			
	1		7	
	M±m	Deviation from control, %	M±m	Deviation from control, %
0.6	1.29±0.06	-5.84	0.86±0.02*	-60.19
Blood serum				
0.06 (conditional control)	18.50±1.81	–	19.00±1.08	–
0.3	22.25±2.35	+20.27	19.00±1.09*	–
0.6	30.50±0.54*	+64.86	20.00±1.45	+5.26
Bile				
0.06 (conditional control)	16.00±1.09	–	10.50±1.09	–
0.3	32.50±1.27*	+103.13	7.50±0.77*	-28.57
0.6	13.00±1.09	-18.75	12.50±0.54	+19.05

Note: ** reliable result*
Source: *compiled by the authors*

With a duration increase of staying fish in water with a lower phosphorus concentration (0.3 mg/L), more phosphates are found in the gill tissue than when they had been adapted to a higher concentration (0.6 mg/L). It provides the reason to believe that with an excessive increase of phosphorus level in the water to a certain level (0.3 mg/L), the glandular apparatus of the gills not only absorb but also removes phosphates in large quantities from the body. Quite unexpectedly, the total phosphorus level in the renal tissue was reduced at its higher concentrations in the aquatic habitat. A particularly significant drop in the total phosphorus level in the fish's kidneys was observed during their prolonged stay in a high phosphate content environment. At the same time, the quantity of total phosphorus in the fish's blood serum, kept in a habitat with its level increased to 0.3-0.6 mg/L, increased in a short-term period. During long-term (7 days) carp's acclimation to such conditions, the phosphorus level in the blood serum was close to the control value. The mentioned changes in total phosphorus content in blood serum may be explained by the occurrence of such mechanisms in carp that make it possible to regulate this element quite effectively. An increase of inorganic phosphorus content in the fish' habitat to 0.3-0.6 mg/L also has caused certain changes in calcium metabolism (Table 2).

Table 2. *Influence of phosphorus in aquatic habitat on the calcium content in tissues (% dry tissue) and liquids (mg %) of the fish's body*

Phosphorus concentration in water, mg	Day of experiments			
	1		7	
	M±m	Deviation from control, %	M±m	Deviation from control, %
Liver				
0.06 (conditional control)	127.00±9.00	–	147.00±8.00	–
0.3	232.00±21.00*	+82.68	119.00±11.00	-19.05
0.6	188.00±4.00*	+48.03	71.00±5.00	-51.70
Gills				
0.06 (conditional control)	1512.00±90.00	–	1098.00±40.00	–
0.3	831.00±77.00*	-45.03	843.00±63.00*	-23.22
0.6	1280.00±23.00	-15.34	1024.00±73.00	-6.73
Kidneys				
0.06 (conditional control)	122.00±3.00	–	106.00±1.00	–
0.3	111.00±7.00	-9.02	87.00±6.00*	-17.92
0.6	159.00±16.00*	+30.33	109.00±1.00	+2.83
Blood serum				
0.06 (conditional control)	29.40±2.70	–	25.40±2.40	–

Table 2, Continued

Phosphorus concentration in water, mg	Day of experiments			
	1		7	
	M±m	Deviation from control, %	M±m	Deviation from control, %
0.3	27.90±2.10	-5.10	26.40±1.10	+3.94
0.6	32.90±2.90	+11.90	18.50±1.30*	-27.17
Bile				
0.06 (conditional control)	42.00±2.10	–	136.00±1.40	–
0.3	100.00±4.00*	+138.10	199.00±7.90*	+44.20
0.6	78.80±7.90*	+87.62	198.00±19.70*	+43.48

Note: ** reliable result*
Source: *compiled by the authors*

Thus, in the liver tissue, the calcium content was increased in one, decreased during the seven-day time day, and decreased during the seven days of the fish acclimation to the increased phosphorus levels environment. The calcium content decrease in the fish's gill tissue was noted, which was more pronounced during the fish's acclimation to a relatively low phosphorus content in water (0.3 mg/L). As for the blood serum, the calcium concentration in it practically remained at the control values level. The only exceptions are the received data during a long-term (7 days) of carp' keeping in a 0.6 mg/L phosphorus habitat when the calcium content in the blood serum was an average of 27.2%.

It should be noted, that there is not a clear dependence of the calcium content in the fish's kidneys on the phosphorus present in their habitat. Thus, during one fish acclimation in a habitat of 0.6 mg/L phosphorus, the calcium content in the renal tissue exceeds the control level by more than 30.0%. With long-term carp acclimation to a lower (0.3 mg/L) concentration of phosphorus in water, the calcium content in the renal tissue was reduced.

An analysis of received data shows that during the seven days of carp acclimation to an enriched with inorganic phosphorus environment, it accumulates in the glandular tissues of the liver and gills, and a slight decrease in the kidneys. As for the blood serum, its level is kept at a relatively constant level. The noted changes in the total phosphorus content and calcium in a carp's body during its acclimation to the increased phosphorus concentration may be due to the activation of organ regulation mechanisms, in particular, an excretory processes increase. The intensity of the phosphorus excretion process with excretion products depends on its concentration in water and the fish's residence time in such conditions. Attention should be paid to the fact that quantitatively more phosphorus and calcium are excreted from the fish body with the faeces than in the urine (Table 3).

Table 3. *Influence of phosphorus in aquatic habitat on the calcium and phosphorus excretion with fish excrement (mg/kg of weight/day)*

Phosphorus concentration in water, mg/L	Day of experiments			
	1		7	
	M±m	Deviation from control, %	M±m	Deviation from control, %
Daily urinary phosphorus excretion				
0.06 (conditional control)	2.07±0.25	–	1.34±0	–
0.3	2.03±0.31	–	1.62±0.09*	+20.90
0.6	1.19±0.23*	-42.52	2.28±0.22*	+70.15
Daily phosphorus excretion in faeces				
0.06 (conditional control)	4.05±0.33	–	3.08±0.33	–
0.3	7.81±0.14*	+92.84	2.85±0.32	-7.47
0.6	3.00±0.22*	-25.93	0.56±0.06*	-81.82
Daily urinary calcium excretion				
0.06 (conditional control)	2.85±0.32	–	2.30±0.17	–
0.3	4.04±0.27*	+47.75	2.39±0.09	+3.91
0.6	1.54±0.12*	-45.96	2.23±0.26	-3.04

Table 3, Continued

Phosphorus concentration in water, mg/L	Day of experiments			
	1		7	
	M±m	Deviation from control, %	M±m	Deviation from control, %
Daily calcium excretion in faeces				
0.06 (conditional control)	6.89±0.21	–	7.09±0.37	–
0.3	9.33±0.16*	+35.42	9.32±0.05*	+31.45
0.6	8.20±0.20*	+19.01	9.59±0.05*	+35.26

Note: ** reliable result*
Source: *compiled by the authors*

Thus, the phosphorus excretion with urine in control fish, kept in a habitat with a 0.06 mg/L (conditional control) phosphorus level was 1.34-2.07 mg/kg of body weight/day, with faeces – 3.08-4.05 mg/kg day. With the phosphorus concentration increase in the aquatic environment to 0.3 and 0.6 mg/L, its excretion naturally increases mainly with urine, and this is especially revealed during a long period of acclimation.

It should be noted that the higher the phosphorus level in the water, the more it is excreted in the urine. During seven days of fish acclimation in a phosphorus 0.3 mg/L habitat, its excretion in urine increased by 20.9% and in 0.6 mg/L – by 70.15%. These results again confirm that the kidneys play a significant role in a fish's phosphate metabolism. The correlation between urinary phosphorus excretion and its feed intake has been shown by (Dilelis *et al.*, 2021). The phosphorus excretion dynamics through the fish's digestive system kept for a long time in an environment with an increased phosphorus level (0.6 mg/L) was reduced. In these experiments, the phosphorus excretion with urine and with fish faeces was lower than the control one during their short-term (1 day) acclimation and similar environmental conditions. However, with the same exposure, but keeping fish in a habitat with a lower (0.3 mg/L) phosphorus concentration, its excretion with the faeces increased sharply (by 92.8%). Thus, the digestive system takes an active part in the elimination of the excess phosphorus, entering the fish's body, with a short fish kept in an environment with a relatively low level of phosphorus (0.3 mg/L). It was also established that by keeping fish for 1 day in a habitat with an increased phosphorus level (0.3 mg/L), the total phosphorus content in bile sharply increases (by 103.13%). With a longer (7 days) exposure of the experiment, the excretion of phosphorus in the bile was not so high.

The fish's acclimation to a habitat with an increased phosphorus level increases calcium excretion (Table 3) through the digestive system (by 19.01-35.42%). Herewith, calcium concentration in the bile of experimental fish, increased by 43.48-138.10. It should be noted that the excretion of calcium in the urine changes to a lesser extent in fish kept in increased water phosphorus concentrations.

A significant change in the daily calcium contents in the urine was noted only in fish, and they acclimated for 1 day to an increased phosphorus content in water. So, if the phosphorus concentration in water equal to 0.3 mg/L caused an increase in the daily calcium excretion with carp urine by 47.75%, then keeping them in water with 0.6 mg/L of the element, its excretion was 45.96% lower than in the control fish. Based on the received data, it can be concluded that the increase in excess phosphorus excretion from the fish body, kept in water with its increased level of 0.3-0.6 mg/L, is accompanied by significant calcium losses, especially through the digestive system.

Features' research of phosphorus metabolism in the fish' glandular organs, involved in osmoregulation (liver, kidneys, gills) has revealed several regularities not only in the total phosphorus distribution but also in the adenyl nucleotide exchange. At the same time, changes in the high-energy phosphorus compounds level in the fish' glandular tissues, exposed to high (0.3-0.6 mg/L) phosphorus concentrations in water, are especially noticeable. Thus, the ATP content in the kidneys was reduced by 39-56%, when the fish were kept in a habitat with 0.3 mg/L of inorganic phosphorus, and the liver, by 29-42.0% at both element concentrations. A significant ATP value decrease in the gill tissue occurred by the end of the 7th day keeping fish in water with a total phosphorus concentration of 0.3 mg/L, and with an increase in its level to 0.6 mg/L – on the first day.

The influence of inorganic phosphorus increased level on adenylates in experimental fish is expressed by a large ATP concentration decrease without a significant ADP and AMP increase, which leads to their decrease. In contrast, the content of these (ADP, AMP) adenyl system components has been reduced in fish tissues, exposed to increased phosphorus levels in the water. Thus, the content of ADP decreases to the greatest extent in the carp's gill tissue, which has been kept in the habitat with both 0.3 and 0.6 mg/L of inorganic phosphorus in water (by 26.9-69.5%). At the same time, in the fish's kidneys and liver, a significant decrease in the ADP level has been noted only when a habitat is exposed to 0.6 mg/L of phosphorus. As for AMP in the researched tissues, it did not differ from the control level during fish acclimation to a lower phosphorus concentration in water (0.3 mg/L). However, with the phosphorus concentration increase in the aquatic habitat to 0.6 mg/L, the AMP level decreases only in the carp's kidneys. In the gills and liver, the AMP level was significantly higher than the initial one.

Based on the research results, it can be concluded that during the fish' acclimation to the increased level of phosphorus in the aquatic habitat, significant changes develop in the glandular fish organs' exchange of the high-energy organophosphate compounds. This conclusion is confirmed by the energy charge calculations of the experimental fish tissue adenylate system (Table 4).

***Table 4**. Influence of increased phosphorus level in water on the content of adenine nucleotides in carp tissues (μM adenine/g dry tissue)*

Phosphorus concentration in water, mg/L	Indicators	Control	Day of experiment			
			1		7	
				Deviation from control, %		Deviation from control, %
			Liver			
0.3	ATP	7.39±0.47	4.28±0.71*	-42.08	5.20±0.36*	-29.63
0.6		5.68±0.61	3.92±0.25*	-30.99	3.37±0.40*	-40.67
0.3	ADP	2.93±0.35	3.16±0.35	+7.85	2.86±0.28	–
0.6		4.50±0.33	2.70±0.35*	-40.00	3.28±0.27*	-27.11
0.3	AMP	2.27±0.23	2.09±0.20	-7.93	1.31±0.14*	-42.29
0.6		2.93±0.10	1.63±0.15*	-44.37	3.60±0.31*	+22.87
0.3	AN sum	12.52±1.28	9.74±0.87*	-22.20	9.27±0.52*	-25.96
0.6		13.01±0.81	8.37±0.51*	-35.66	10.32±0.95*	-20.68
0.3	AEC	0.70±0.03	0.60±0.02*	-14.29	0.72±0.02	+2.85
0.6		0.59±0.01	0.64±0.02*	+25.42	0.49±0.012*	-16.95
			Gills			
0.3	ATP	9.43±1.40	10.12±0.44	+7.30	3.59±0.23*	-61.93
0.6		10.84±0.35	5.97±0.69*	-44.92	7.50±0.87*	-30.81
0.3	ADP	7.18±1.12	3.58±0.18*	-50.14	2.19±0.32*	-69.90
0.6		9.43±0.50	5.28±0.44*	-44.01	6.89±0.27*	-26.94
0.3	AMP	2.07±0.22	2.32±0.12	+12.08	1.91±0.32	-7.73
0.6		2.07±0.22	2.40±0.14	+5.94	2.47±0.24*	+43.48
0.3	AN sum	18.70±1.69	12.01±0.69*	-35.78	7.85±0.23*	-58.02
0.6		22.68±0.42	17.67±0.25*	-22.09	17.36±1.15*	-23.46
0.3	AEC	0.69±0.01	0.66±0.03	-4.35	0.60±0.03*	-13.04
0.6		0.69±0.01	0.70±0.012	+1.45	0.63±0.02*	-8.70
			Kidneys			
0.3	ATP	11.00±0.64	6.70±0.45*	-39.00	4.84±0.26*	-56.00
0.6		8.80±0.63	8.97±0.62	+1.93	7.61±0.90	-13.52
0.3	ADP	3.42±0.71	3.52±0.56	+2.92	2.96±0.26	-13.45
0.6		6.06±0.42	4.37±0.28*	-27.89	5.28±0.22	-12.87
0.3	AMP	2.78±0.35	2.84±0.34	–	3.43±0.26	+23.40
0.6		4.38±0.35	2.50±0.28*	- 42.92	3.35±0.17*	-24.07
0.3	AN sum	17.29±1.07	13.12±0.51*	- 24.12	11.35±0.52*	-34.36
0.6		18.31±0.56	15.45±0.85	- 15.62	16.30±0.97	-10.97
0.3	AEC	0.74±0.02	0.65±0.02*	-12.16	0.56±0.02*	-24.32
0.6		0.67±0.02	0.72±0.04	+7.46	0.62±0.02	-7.46

***Note:** AN sum – the sum of adenyl nucleotides; AEC – adenylate energy charge. * – the reliable result*
***Source:** compiled by the authors*

Thus, the adenylate system energy charge in the kidneys of carp, which acclimated to a phosphorus concentration of 0.6 mg/L in water, has been maintained at the level of control fish. In the liver of these fish, with a short-term experiment exposure, the adenylate energy charge has been for 25.4% higher than the control level, while simultaneously, the AMP content in it decreased. During a seven-day carp' acclimation period to a containing phosphorus 0.6 mg/L habitat, the adenylate energy charge in the carp's liver and gills was reduced.

Dynamics change comparison in the content of adenyl nucleotides and inorganic phosphorus in the fish' tissues acclimated in a habitat with an increased phosphorus level (Table 5) gives reason to assume the activation of glycolytic processes and oppression of tissue respiration.

Table 5. *Influence of increased phosphorus level in water on the phosphorus compounds content in the fish tissues (mg% dry tissue)*

Phosphorus concentration in water, mg/L	**Day of experiments**			
	1		**7**	
	M±m	**Deviation from control, %**	**M±m**	**Deviation from control, %**
Liver				
Inorganic phosphorus				
0.06	527.80±11.66	–	446.92±45.302	–
0.3	394.15±14.88*	-25.30	506.10±36.62	+13.20
0.6	536.59±1.20	+1.70	698.38±18.84*	+35.00
Protein phosphorus				
0.06	248.88±12.11	–	132.70±4.74	–
0.3	347.32±11.22*	+39.60	344.27±13.15*	+159.40
0.6	366.67±3.70*	+47.30	299.52±4.80*	+125.70
Lipid phosphorus				
0.06	139.01±13.00	–	88.63±3.82	–
0.3	303.41±11.22*	+118.30	188.73±27.70*	+112.90
0.6	271.76±15.74*	+95.50	234.30±25.12*	+164.40
Gills				
Inorganic phosphorus				
0.06	689.08±74.14	–	559.22±25.29	–
0.3	629.31±4.37	-8.70	640.91±90.26	+14.60
0.6	976.70±41.50*	+41.70	571.52±88.48	+2.20
Protein phosphorus				
0.06	360.34±20.11	–	231.62±16.76	–
0.3	451.15±27.01*	+25.20	504.55±22.72*	+117.80
0.6	545.89±26.03*	+51.50	377.58±27.27*	+63.00
Lipid phosphorus				
0.06	155.17±8.04	–	118.72±7.26	–
0.3	190.80±16.67	+23.00	137.95±8.44	+16.20
0.6	237.67±14.38*	+53.20	113.33±7.88	-4.50

Note: ** reliable result*
Source: *compiled by the authors*

Switching energy metabolism in the fish's body, exposed to an increased phosphorus content in the aquatic habitat to the glycolytic path, leads to less fat usage for energy purposes, which the lipid phosphorus content has confirmed.

It was established that the lipid phosphorus level significantly increases in the carp's liver, acclimated to the content of phosphorus in water equal to 0.3 and 0.6 mg/L. The lipid phosphorus content in experimental fish's gills was increased only with a short-term (1 day) keeping fish in a habitat with 0.6 mg/L of phosphorus. The lipid phosphorus noted an increase in fish tissues, kept in an increased level of the element environment, which can occur due to its fast integration into organic compounds Huser *et al.* (2021).

An inorganic phosphorus increase in water has affected the activity of Na^+, and K^+, activated by Mg^{2+} – dependent adenosine triphosphatase of the studied fish organs (Fig. 1).

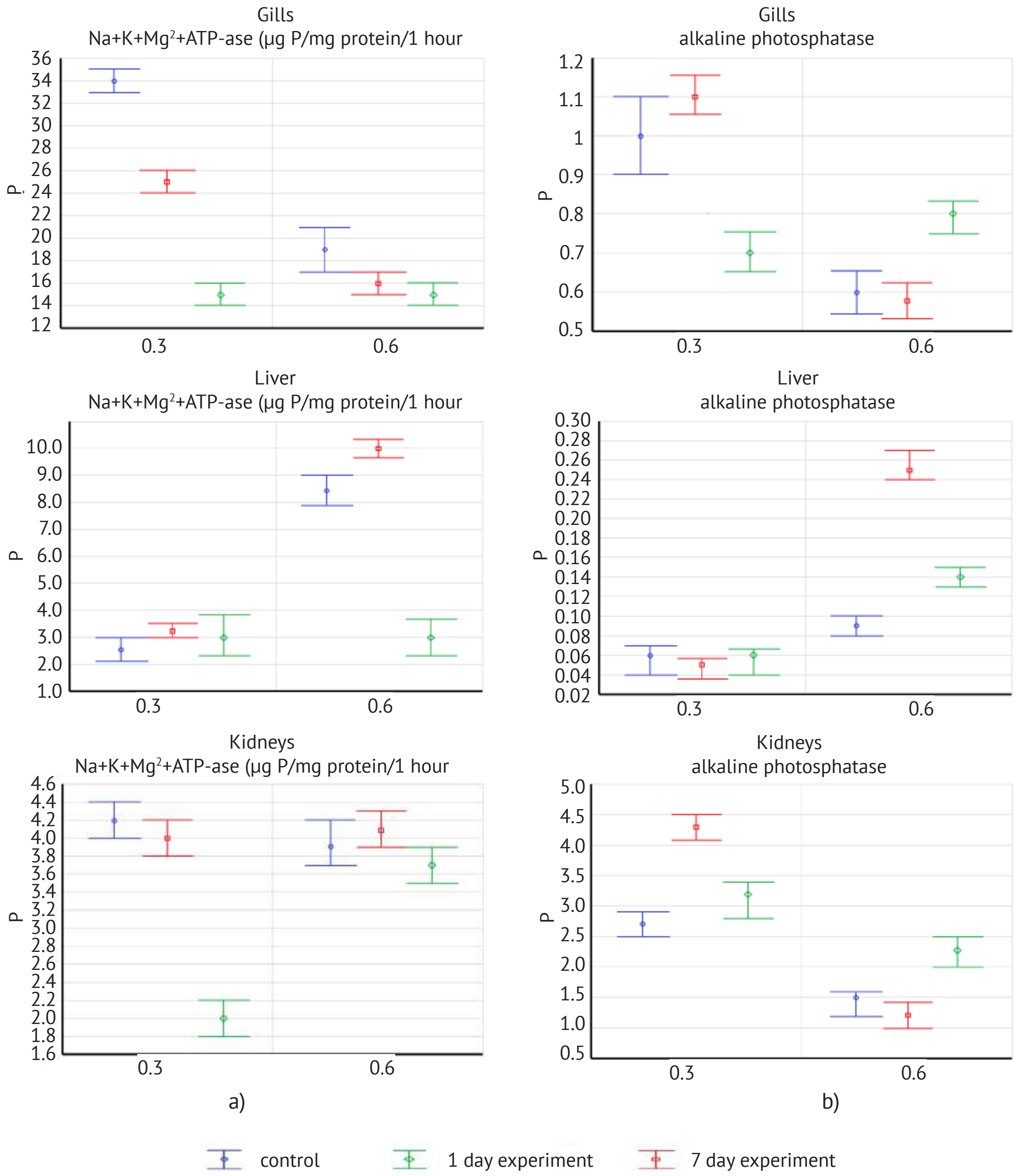

Figure 1. *The activity of Na^+ K^+ Mg^{2+}-ATP-ase (µg P/mg protein/1 hour) (a) and alkaline phosphatase (b) in carp's tissues during keeping in a habitat with an increased inorganic phosphorus concentration*

Herewith, the reactions of different tissues to the phosphates action are not the same. So, if the adenosine triphosphatase activity was reduced in the fish's gills and kidneys when they were kept in water with 0.3 mg/L of inorganic phosphorus, but in the liver, vice versa, it was much higher. At a higher inorganic phosphorus concentration (0.6 mg/L), the gills and kidneys' ATP-ase activity stayed as that at the control fish level and sharply decreased in the liver. The changes in adenosine triphosphatase activity may indicate the phosphates' transport decreases through the fish' gills at a sufficiently high content of it in the aquatic habitat. It consists of the value data of total phosphorus and phosphorus ATP in the tissues (Table 1, 4).

Under conditions of a significant phosphorus content increase in water, the tissue activity of alkaline phosphatase also changes (Fig. 1). Hence, in the gills' glandular tissue, the activity of the enzyme changed significantly only by the end of the seven-day experiment at both studied phosphorus concentrations in water. The alkaline phosphatase activity did not change in the liver during the fish was kept in a habitat with 0.3 mg/L of phosphorus. With an increase of inorganic phosphorus concentration in water up to 0.6 mg/L, the highest enzymatic activity (almost twice the control one) of glandular tissues was observed on the first day of keeping fish in such habitat. With an increase in time acclimation, the activity of alkaline phosphatase in the liver has continued to remain high. The most noticeable changes in the activity of alkaline phosphatase were registered in the renal tissue. The enzyme activity was significantly increased in the renal tissue at 0.3 and 0.6 mg/L of inorganic phosphorus in the water.

The enzymatic activity of the liver, kidneys, and gill tissues shows that its regular increase is observed only in the kidneys while keeping fish in an environment with 0.3 and 0.6 mg/L of phosphorus. In other organs, phosphatase activity increases only on particular days. An increase in alkaline phosphatase tissue activity in the kidneys may be related to eliminating excess phosphates from the fish's body, kept in a habitat with its high content (Table 3).

Thus, the research showed that with a phosphorus concentration increase in the aquatic habitat up to 0.3-0.6 mg/L, it accumulated in the liver and gills, and decreased in the kidneys, while in the fish's blood serum, its concentration was maintained. at a relatively constant level. A greater extent causes the accumulation of phosphates in fish tissues due to organic compounds, including lipid and protein fractions. In the excretion from the body phosphates, along with the kidneys, the liver plays a significant role, as evidenced by the increased calcium and phosphorus excretion with the fish's bile during their acclimation to an increased phosphorus level in the water. The phosphorus content change in the fish habitat significantly influences the tissue bioenergetics processes, which shows as the change in ATP content, the activity of ATP-ase and alkaline phosphatase. The most profound changes in these indicators have been noticed in the gills – that is the responsible organ for the intake and excretion of phosphates from the body, as well as in the kidneys, which provide a high rate of phosphates excretion.

As illustrated above, a phosphorus content change in the fish habitat has a significant influence on its tissue accumulation, as well as the high-energy phosphorus compounds supply, which has an extremely important role in bioenergetics processes. Following these data, there are also the results of the cellular processes research in the glandular organs of fish, acclimated to an increased inorganic phosphorus level in the water. At the same time, the total phosphorus content in the carp's mitochondria was in a certain dependent on this element amount in water and the time of its influence on the fish organism (Table 6).

Table 6. *Influence of increased phosphorus level, in the aquatic habitat, on the total and inorganic phosphorus content in the mitochondria of carp' gills and liver (μg/mg protein)*

Indicators	Day of experiment	0.06 (control)	Phosphorus concentrations in water (mg/L)			
			0.3		0.6	
				Deviation from control, %		Deviation from control, %
Gills						
Total phosphorus	1	6.22±0.62	5.78±1.27	-7.1	5.30±0.73	-17.4
Inorganic phosphorus		0.52±0.06	0.65±0.08	+25.0	0.35±0.03*	-32.69
Total phosphorus	7	5.50±0.38	7.63±0.50*	+38.7	2.71±0.28*	-50.7
Inorganic phosphorus		0.93±0.07	0.36±0.04*	-61.29	0.33±0.07*	-64.52
Liver						
Total phosphorus	1	5.42±0.69	3.78±0.26*	-31.7	3.70±0.36*	-31.7
Inorganic phosphorus		0.55±0.05	0.64±0.05	+16.36	0.39±0.03*	-29.09

Table 6, Continued

Indicators	Day of experiment	0.06 (control)	Phosphorus concentrations in water (mg/L)			
			0.3		0.6	
				Deviation from control, %		Deviation from control, %
Total phosphorus	7	6.98±0.91	5.91±0.37	-15.3	4.12±0.16*	-41.0
Inorganic phosphorus		0.52±0.06	0.19±0.02*	-63.46	0.29±0.01*	-44.23

Note: ** – reliable result*
Source: *compiled by the authors*

Thus, in the mitochondria of the fish's gills glandular apparatus, when they were kept for 3 days in a phosphorus habitat with 0.3 mg/L, the total phosphorus content remained unchanged and only by the end of the seventh day it has been slightly exceeded the control value. With the inorganic phosphorus level increase in water to 0.6 mg/L, the total phosphorus concentration in the mitochondria of the gills has decreased more than two times, which was especially clearly shown on the 7^{th} acclimation day.

Although phosphorus content increases in the fish habitat, mitochondrial accumulation of phosphates has not been observed. Nevertheless, in the mitochondria of the liver and gills' glandular cells, significant changes in the energy metabolism indicators were registered, particularly in the exchange of adenyl nucleotides and the activity of Na^+, K^+, and Mg^{2+}-ATP-ase.

Thus, the amount of ATP (Table 7) in the glandular cells' mitochondria of the carp's gill apparatus, during its daily acclimation to 0.3 mg/L of phosphorus, was sharply increased in the aquatic habitat. Further, fish acclimation to the increased inorganic phosphorus concentrations in the aquatic habitat was accompanied by significant energy consumption necessary to support homeostatic balance in their body. At the same time, the high fish' requirement for metabolic energy has been provided by the reserves of ATP. This is confirmed by its content decrease in the glandular organs' mitochondria, especially during prolonged (7 days) acclimation to an environment with 0.6 mg/L of phosphorus, with a simultaneous increase of the ATP-ase activity in these cellular structures. This direction of the mitochondria' bioenergetic processes in the fish's liver and gills ensures the adaptive capacities of their body to a sharp phosphorus content increase in water.

Table 7. *Influence of increased phosphorus level in the aquatic habitat on the exchange of adenine nucleotides in the fish' mitochondria tissues (μM adenine/100 mg protein)*

Indicators	Day of experiment	0.06 (control)	Phosphorus concentrations in water (mg/L)			
			0.3		0.6	
				% concerning control		% concerning control
Gills						
ATP	1	2.90±0.18	5.40±0.65	+86.21	2.60±0.20*	-10.34
ADP		2.60±0.21	2.70±0.23*	+3.85	1.30±0.20	-50.0
AMP		0.80±0.02	1.30±0.23	+62.5	2.50±0.12	+212.5
The adenylates' amount		6.20±0.50	9.40±0,63	+51.61	6.40±0.69*	+3.23
AEC		0.66±0.015	0.71±0.031*	+7.5	0.50±01.006	-24.24
ATP	7	2.50±0.09	3.50±0.21	+40.0	1.08±0.18	-56.8
ADP		1.00±0.11	2.50±0.23	+150.0	2.67±0.033	+167.0
AMP		0.80±0.01	1.30±0.12	+62.5	1.04±0.079	+30.0
The adenylates' amount		4.30±0.11	7.45±0.33	+73.26	4.80±0.29*	+11.62
AEC		0.70±0.011	0.65±0.002	-7.14	0.50±0.015	-28.57
Liver						
ATP	1	1.50±0.017	1.30±0.08*	-13.33	1.50±0.12*	–
ADP		1.10±0.15	2.50±0.18	+127.27	1.50±0.12	+36.36
AMP		0.90±0.09	1.60±0.11	+77.78	2.06±0.20	+128.89

Table 7, Continued

Indicators	Day of experiment	0.06 (control)	Phosphorus concentrations in water (mg/L)			
			0.3		0.6	
				% concerning control		% concerning control
The adenylates' amount		3.70±0.36	5.40±0.25	+45.95	5.10±0.29	+37.84
AEC		0.56±0.019	0.48±0.036	-14.29	0.46±0.016	-17.86
ATP	7	1.70±0.08	2.20±0.06	+29.41	1.17±0.013	-31.18
ADP		0.80±0.10	1.60±0.16	+100.0	1.17±0.043	+46.25
AMP		0.60±0.04	1.20±0.10	+100.0	1.13±0.031	+88.33
The adenylates' amount		3.12±0.09	5.02±0.31	+60.0	3.44±0.037*	+10.26
AEC		0.70±0.009	0.60±0.019	-14.29	0.51±0.003	-27.14

Note: ** AEC - adenylate energy charge; * – the reliable result*
Source: *compiled by the authors*

Analysis of other adenyl system components shows that the level of ADP, and AMP is influenced by an increased phosphorus content in the aquatic habitat, the adenyl nucleotide amount has been increased. It should be mentioned that more pronounced changes in these indicators were noted during acclimation in a habitat containing phosphorus of 0.6 mg/L. The energy charge of the adenylate system, which characterizes the metabolically available energy presence in the carp's mitochondria of the glandular tissues, exposed to an increased phosphorus level in the water, has been kept at a level lower than the control level (by 7.5-31.18%). Such a decrease in the adenylate charge level contributes to the activation of various energy generation mechanisms, as a result, the ATP content in the mitochondria of the carp' gills and liver, while they were kept in a habitat with a lower (0.3 mg/L) phosphorus concentration in water, or with a short (1 day) phosphorus exposure to 0.6 mg/L, is maintained at a close to the control level.

Thus, the change in phosphorus content in the habitat of fish significantly affects its tissue accumulation, as well as the content of macroregion phosphorus compounds, which ensure the high intensity of phosphate excretion. Wang *et al.,* (2022) found that about half of the absorbed phosphorus is retained in the fish liver. A similar pattern is revealed in the fish's phosphate content indicators in their liver tissue during their acclimation to these inorganic phosphorus concentrations. However, it should be noted that the opinion of researchers regarding the dependence between the absorption of phosphorus from the aquatic habitat and its level in it is somewhat contradictory. Thus, (Dilelis *et al.,* 2021; Hrynevych *et al.,* 2022) had been shown that with phosphorus concentration increases in the aquatic environment, its accumulation by fish also increases. At the same time, a high number of phosphorus can adversely influence the branchial membranes, thereby reducing the actual absorption and phosphorus transport (Villeger *et al.,* 2017). The unequal influence of the different phosphorus concentrations that come with feed on the processes of its absorption was also shown by the works of (Costa *et al.,* 2018; Knöpfel *et al.,* 2019; Gao *et al.,* 2023).

The renal system plays a fundamental role in the phosphates' excretion from the fish organism (Wang *et al.,* 2021). The data has also confirmed the high phosphorus lability content in the fish's kidney tissue, during their acclimation to its increased concentrations in water.

The deviations found in the exchange of calcium in the carp's body acclimated to an increased phosphorus content in the aquatic habitat, show the interrelation of tissue metabolism in these elements in terrestrial and aquatic animals. In experiments on birds, an increased calcium content in the serum of chickens who get a low phosphorus content feed was observed (Fedoniuk *et al.,* 2019). With an increase in the content of phosphorus in the diet of poultry, changes in the content of calcium in the blood became less noticeable. A negative linear relationship between the amount of phosphorus absorbed by carp and the calcium content of the feed has been shown (Huser *et al.,* 2021; Lall, 2022). According to other data, the level of calcium in the blood of carp did not depend on the amount of phosphorus in the food (Saurette *et al.,* 2019). Musharraf *et al.* (2019) have observed the violation of tissue calcium metabolism with enhanced carp phosphorus excretion, fed with a phosphorus diet enriched.

The data received does not align with the previously obtained results of a study by Pouil *et al.* (2018), which showed that the phosphorus excretion from the fish's body happened mainly through the kidneys, gills and, to a lesser extent, with faeces. However, in the research (Fedonyuk *et al.,* 2019; He *et al.,* 2022), the digestive system, under normal keeping fish conditions, is assigned a significant role in phosphorus elimination from the body.

The confirmation of the glycolytic processes' activation and inhibition of such fish tissue respiration reactions are established by Wang *et al.* (2022) decreased activity of respiratory enzymes, glycogen level and accumulation of glycolysis products in the liver and gills.

Other researchers (Solomatina *et al.,* 2018; Sugiura *et al.,* 2018; Huang *et al.,* 2019), showed that significant phosphorus reserves in aquatic organisms can be found in the form of lipid and protein compounds in the hepatopancreas. This is confirmed by the protein phosphorus amount significant (by 25.2-159.4%) increase in the liver and gills of fish, kept in an inorganic phosphorus high-level environment.

The researchers (Romanchuk *et al.,* 2018; Pinkina *et al.,* 2019; Solomatina *et al.,* 2019), also found that the mitochondrial content of inorganic phosphorus and total phosphorus was reduced in fish kept in water with elevated levels. This happens in contrast to calcium, the accumulation of which increased sharply in the mitochondria of the liver and especially in the gill apparatus with the increase of this element in the habitat. Researchers (Ai *et al.,* 2019; Wang *et al.,* 2022) found phosphorus in the aquatic habitat reduces the adsorption and transport of ions, negatively affecting the membranes of gill cells. This can explain the decrease in the accumulation of phosphates in the mitochondria of the glandular organs of the liver, especially the gills, during the maintenance of carp in a high-phosphate environment with a high level of phosphates.

Comparison of the tissue and cellular changes in the liver and glandular gills apparatus content of calcium and phosphorus, during the fish acclimation to an increased level of this element in water, reveals the unequal mitochondria participation in ensuring the adaptive organism's reactions to the ionic effects of the aquatic habitat. If, after an increased calcium intake into the fish's body from the aquatic habitat, it increases in the mitochondria of the glandular cells, then during the fish acclimation to a high phosphorus content, and vice versa, its mitochondrial content decreases. Therefore, it can be concluded that, if mitochondria play a significant role in the fish calcium metabolism, cytoplasmic structures are more important in phosphate metabolism. This proposal corresponds with the data received on the cytosol and mitochondria of the warm-blooded animals' hepatocytes when their body is loaded with inorganic phosphorus (Sun *et al.,* 2018; Wang *et al.,* 2021). Thus, after a 20-minute hepatocytes incubation with 620 mg of inorganic phosphorus, its content in isolated mitochondria has not changed, but it increased almost 2 times in the cytosol.

The above-noted deviations in the energy supply for the fish' acclimation process to an increased phosphorus level in the aquatic habitat happened due to the significant deviations in the links of mitochondrial respiration. Caused by swelling of mitochondria and uncoupling of oxidative phosphorylation Souders *et al.* (2018).

Described orientation of metabolic processes provides adaptive capabilities of the fish organism to a sharp increase in the phosphate content in the water. Thanks to this, *Cyprinus carpio* L. fish can tolerate high levels of phosphorus fluctuations in the aquatic environment.

CONCLUSIONS

As a result of the study, it was established, that with a duration increase of staying fish in water with a lower phosphorus concentration (0.3 mg/L), more phosphates are found in the gill tissue than when they had been adapted to a higher concentration (0.6 mg/L).

A particularly significant drop in the total phosphorus level in the fish's kidneys was observed during their prolonged stay in a high phosphate content environment. At the same time, the quantity of total phosphorus in the fish's blood serum, kept in a habitat with its level increased to 0.3-0.6 mg/L, increased in a short-term period.

The calcium content decrease in the fish's gill tissue was noted, which was more pronounced during the fish's acclimation to a relatively low phosphorus content in water (0.3 mg/L). As for the blood serum, the calcium concentration in it practically remained at the control values level. With long-term carp acclimation to a lower (0.3 mg/L) concentration of phosphorus in water, the calcium content in the renal tissue was reduced.

It was also established that by keeping fish for 1 day in a habitat with an increased phosphorus level (0.3 mg/L), the total phosphorus content in bile sharply increases (by 103.13%). The influence of inorganic phosphorus increased level on adenylates in experimental fish is expressed by a large ATP concentration decrease without a significant ADP and AMP increase, which leads to their amount decrease. In contrast, the content of these (ADP, AMP) adenyl system components has been reduced in fish tissues, exposed to increased phosphorus levels in the water. Thus, the content of ADP decreases to the greatest extent in the carp's gill tissue, which has been kept in the habitat with both 0.3 and 0.6 mg/L of inorganic phosphorus in water (by 26.9-69.5%).

Switching energy metabolism in the fish's body, exposed to an increased phosphorus content in the aquatic habitat to the glycolytic path, leads to less fat use for energy purposes, which the lipid phosphorus content has confirmed. In the mitochondria of the liver and gills' glandular cells, significant changes in the energy metabolism indicators were registered, particularly in the exchange of adenyl nucleotides and the activity of Na^+, K^+, and Mg^{2+}-ATP-ase. The perspective of further research is the influence of different concentrations of phosphorus in the aquatic environment on the tissue and intracellular metabolism of predatory fish species.

ACKNOWLEDGEMENTS

Bila Tserkva National Agrarian University, represented by the rector, Professor Shust O.A.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

[1] Ai, F., Wang, L., Li, J., & Xu, Q. (2019). Effects of a-ketoglutarate (AKG) supplementation in low phosphorous diets on the growth, phosphorus metabolism and skeletal development of juvenile mirror carp *(Cyprinus carpio)*. *Aquaculture*, 507, 393-401. doi: 10.1016/j.aquaculture.2019.03.047.

[2] Boyd, C.E., D'Abramo, L.R., Glencross, B.D., Huyben, D.C., Juarez, L.M., Lockwood, G.S., McNevin, A.A., Tacon, A.G.J., Teletchea, F., Tomasso, J.R., Jr, Tucker, C.S., & Valenti, W.C. (2020). Achieving sustainable aquaculture: Historical and current perspectives and future needs and challenges. *Journal of the World Aquaculture Society,* 51(3), 578-633. doi: 10.1111/jwas.12714.

[3] Costa, J.M., Sartori, M.M.P., Nascimento, N.F.D., Kadri, S.M., Ribolla, P.E.M., Pinhal, D., & Pezzato, L.E. (2018). Inadequate dietary phosphorus levels cause skeletal anomalies and alter osteocalcin gene expression in zebrafish. *International Journal of Molecular Sciences,* 19, article number 364. doi: 10.3390/ijms19020364.

[4] Dilelis, F., Freitas, L.W., Quaresma, D.V., Reis, T.L., Souza, C.S., & Lima, C.A.R. (2021). Determination of true ileal digestibility of phosphorus of fish meal in broiler diets. *Animal Feed Science and Technology,* 272, article number 114742. doi: 10.1016/j.anifeedsci.2020.114742.

[5] Directive 2010/63/EU of the European Parliament and of the Council of 22 September 2010 on the protection of animals used for scientific purposes. (2010). Retrieved from https://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2010:276:0033:0079:En:PDF.

[6] Fedoniuk, T.P., Fedoniuk, R.H., Romanchuk, L.D., Petruk, A.A., & Pazych, V.M. (2019). The influence of landscape structure on the quality index of surface waters. *Journal of Water and Land Development,* 43(1), 56-63. doi: 10.2478/jwld-2019-0063.

[7] Fedonyuk, T.P., Fedoniuk, R.H., Zymaroieva, A.A., Pazych, V.M., & Aristarkhova, E.O. (2019). Phytocenological approach in biomonitoring of the state of aquatic ecosystems in Ukrainian Polesie. *Journal of Water and Land Development,* 44(I-III), 65-74. doi: 10.24425/JWLD.2019.127047.

[8] Gao, S., Sun, P., Ren, H., Chen, J., Shen, Y., Wang, Z., Huang, Y., & Chen, W. (2023). Effects of dietary phosphorus deficiency on the growth performance, hepatic lipid metabolism, and antioxidant capacity of Yellow River *Carp Cyprinus carpio haematopterus*. *Journal of Aquatic Animal Health,* 35(1), 41-49. doi: 10.1002/aah.10177.

[9] He, Y., Song, Z., Dong, X., Zheng, Q., Peng, X., & Jia, X. (2022). Candida tropicalis prompted effectively simultaneous removal of carbon, nitrogen and phosphorus in activated sludge reactor: Microbial community succession and functional characteristics. *Bioresource Technology,* 348, article number 126820. doi: 10.1016/j.biortech.2022.126820.

[10] Hrynevych, N., Svitelskyi, M., Solomatina, V., Ishchuk, O., Matkovska, S., Sliusarenko, A., Khomiak, O., Trofymchuk, A., Pukalo, P., & Zharchynska, V. (2022). Acclimatization of fish to the higher calcium levels in the water environment. *Potravinarstvo Slovak Journal of Food Sciences,* 16, 101-113. doi: 10.5219/1732.

[11] Huang, C-L., Gao, B., Xu, S., Huang, Y., Yan, X., & Cui, S. (2019). Changing phosphorus metabolism of a global aquaculture city. *Journal of Cleaner Production,* 225, 1118-1133. doi: 10.1016/j.jclepro.2019.03.298.

[12] Huser, B.J., Bajer, P.G., Kittelson, S., Christenson, S., & Menken, K. (2021). Changes to water quality and sediment phosphorus forms in a shallow, eutrophic lake after removal of common carp *(Cyprinus carpio)*. *Inland Waters,* 12(1), 33-46. doi: 10.1080/20442041.2020.1850096.

[13] Knöpfel, T., Himmerkus, N., Günzel, D., Bleich, M., Hernando, N., & Wagner, C.A. (2019). Paracellular transport of phosphate along the intestine. *American Journal of Physiology-Gastrointestinal and Liver Physiology,* 317, 233-241. doi: 10.1152/ajpgi.00032.2019.

[14] Lall, S.P. (2022). The minerals. In *Fish Nutrition* (469-554). Cambridge: Academic Press.

[15] Law of Ukraine No. 27 "On the Protection of Animals from Cruelty". (2006, February). Retrieved from https://zakon.rada.gov.ua/laws/show/3447-15#Text.

[16] Lei, Y., Sun, Y., Wang, X., Lin, Z., Bu, X., Wang, N., Du, Z., Qin, J., & Chen, L. (2021). Effect of dietary phosphorus on growth performance, body composition, antioxidant activities and lipid metabolism of juvenile Chinese mitten crab *(Eriocheir sinensis)*. *Aquaculture,* 531, article number 735856. doi: 10.1016/j.aquaculture.2020.735856.

[17] Musharraf, M., & Khan, M.A. (2019). Dietary phosphorus requirement of fingerling Indian major carp, *Labeo rohita* (Hamilton). *Journal of the World Aquaculture Society,* 50, 469-484. doi: 10.1111/jwas.12521.

[18] Pinkina, T., Zymaroieva, A., Matkovska, S., Svitelskyi, M., Ishchuk, O., & Fediuchka, M. (2019). Trophic characteristics of *Lymnaea stagnalis* (Mollusca: Gastropoda: Lymnaeidae) in toxic environment. *Ekologia,* 38(3), 292-300. doi: 10.2478/eko-2019-0022.

[19] Pouil, S., Bustamante, P., Warnau, M., & Metian, M. (2018). Overview of trace element trophic transfer in fish through the concept of assimilation efficiency. *Marine Ecology Progress Series,* 588, 243-254. doi: 10.3354/meps12452.

[20] Romanchuk, L., Fedonyuk, T., Pazych, V., Fedonyuk, R., Khant, G., & Petruk, A. (2018). Assessment of the stability of aquatic ecosystems development on the basis of indicators of the macrophytes fluctuating asymmetry. *Eastern-European Journal of Enterprise Technologies,* 4(10), (94), 54-61. doi: 10.15587/1729-4061.2018.141055.

[21] Saurette, M., & Alexander, R.T. (2019). Intestinal phosphate absorption: The paracellular pathway predominates? *Experimental Biology and Medicine,* 244, 646-654. doi: 10.1177/1535370219831220.
[22] Solomatina, V.D., Pinkina, T.V., & Svitelskyi, M.M. (2018). The effect of the temperature of rearing young fish of different species on the content of organophosphorus high-energy compounds in their tissues. *Water Bioresources and Aquaculture,* 2, 79-88.
[23] Solomatina, V.D., Pinkina, T.V., Svitelskyi, M.M., & Feduchka, M.I. (2019). Changes in calcium and phosphorus metabolism in fish with their warm-water growing. *Water Bioresources and Aquaculture,* 1, 51-62. doi: 10.32851/wba.2019.1.5.
[24] Souders 2nd, C.L., Liang, X., Wang, X., Ector, N., Zhao, Y.H., & Martyniuk, C.J. (2018). High-throughput assessment of oxidative respiration in fish embryos: Advancing adverse outcome pathways for mitochondrial dysfunction. *Aquatic Toxicology,* 199, 162-173. doi: 10.1016/j.aquatox.2018.03.031.
[25] Sugiura, S.H. (2018). Phosphorus, Aquaculture, and the Environment. In *Reviews in Fisheries Science &amp. Aquaculture,* 26(4), 515-521. doi: 10.1080/23308249.2018.1471040.
[26] Sun, Y., Li, B., Zhang, X., Chen, M., Tang, H., & Yu, X. (2018). Dietary available phosphorus requirement of crucian carp *(Carassius auratus). Aquaculture Nutrition,* 24(5), 1494-1501. doi: 10.1111/anu.12686.
[27] Svitelskyi, M.M., Ishchuk, O.V., Fediuchka, M.I., Klymchyk, O.M., Pryshchepa, M.A., & Pylypchuk, N.V. (2020). Intracellular calcium metabolism of fish under hypercalcemic conditions of the aquatic environment. *Bulletin National University of Water and Environmental Engineering,* 1(89), 63-72.
[28] Wang, L., Fan, Z., Zhang, Y., Wu, D., Li, J., & Xu, Q. (2022). Effect of phosphorus on growt h performance, intestinal tight junctions, Nrf2 signaling pathway and immune response of juvenile mirror carp *(Cyprinus carpio)* fed different α-ketoglutarate levels. *Fish & Shellfish Immunology,* 120, 271-279. doi: 10.1016/j.fsi.2021.11.040.
[29] Wang, P., Li, X., Xu, Z., Ji, D., He, M., Dang, J., & Leng, X. (2021). The digestible phosphorus requirement in practical diet for largemouth bass *(Micropterus salmoides)* based on growth and feed utilization. *Aquaculture and Fisheries,* 7(6), 632-638. doi: 10.1016/j.aaf.2020.11.002.
[30] Wang, Y., Geng, Y., Shi, X., Wang, S., Yang, Z., Zhang, P., & Liu, H. (2022). Effects of dietary phosphorus levels on growth performance, phosphorus utilization and intestinal calcium and phosphorus transport-related genes expression of juvenile chinese soft-shelled turtle *(Pelodiscus sinensis). Animals,* 12, article number 3101. doi: 10.3390/ani12223101.
[31] Xu, C.M., Yu, H.R., Zhang, Q., Chen, B.B., Li, L.Y., Qiu, X.Y., Qi, T., Liu, J.Q., & Shan, L.L. (2021). Dietary phosphorus requirement of coho salmon *(Oncorhynchus kisutch)* alevins cultured in freshwater. *Aquaculture Nutrition,* 27, 2427-2435. doi: 10.1111/anu.13374.
[32] Yang, Q., Liang, H., Maulu, S., Ge, X., Ren, M., Xie, J., & Xi, B. (2021). Dietary phosphorus affects growth, glucolipid metabolism, antioxidant activity and immune status of juvenile blunt snout bream *(Megalobrama amblycephala). Animal Feed Science and Technology,* 274, article number 114896. doi: 10.1016/j.anifeedsci.2021.114896.
[33] Yu, H., Yang, Q., Liang, H., Ren, M., Ge, X., & Ji, K. (2021). Effects of stocking density and dietary phosphorus levels on the growth performance, antioxidant capacity, and nitrogen and phosphorus emissions of juvenile blunt snout bream *(Megalobrama amblycephala). Aquaculture Nutrition,* 27, 581-591. doi: 10.1111/anu.13208.
[34] Zhang, J., Zhang, S., Lu, K., Wang, L., Song, K., Li, X., Zhang, C., & Rahimnejad, S. (2022). Effects of dietary phosphorus level on growth, body composition, liver histology and lipid metabolism of spotted seabass *(Lateolabrax maculatus)* reared in freshwater. *Aquaculture and Fisheries,* 8(5), 528-537. doi: 10.1016/j.aaf.2022.02.004.
[35] Zhao, J., Liu, M., & Chen, M. (2019). Analytical errors in the determination of inorganic and organic phosphorus in waters based on laboratory experiences. *Journal of Water Resource and Protection,* 11(1), 53-67. doi: 10.4236/jwarp.2019.111004.
[36] Zhao, M., Luo, J., Zhou, Q., Yuan, Y., Shi, B., Zhu, T., Lu, J., Hu, X., Jiao, L., Sun, P., & Jin, M. (2021). Influence of dietary phosphorus on growth performance, phosphorus accumulation in tissue and energy metabolism of juvenile swimming crab *(Portunus trituberculatus). Aquaculture Reports,* 20, article number 100654. doi: 10.1016/j.aqrep.2021.100654.

Вплив різних концентрацій фосфору на тканинний та внутрішньоклітинний метаболізм *Cyprinus Carpio* L. у водному середовищі існування

Наталія Євгеніївна Гриневич

Доктор ветеринарних наук, професор
Білоцерківський національний аграрний університет
09111, вул. Героїв Чорнобиля, 3а, м. Біла Церква, Україна
https://orcid.org/0000-0001-7430-9498

Микола Михайлович Світельський

Кандидат сільськогосподарських наук, доцент
Поліський національний університет
10008, Старий бульвар, 7, м. Житомир, Україна
https://orcid.org/0000-0003-1501-4168

Олександр Андрійович Хом'як

Кандидат сільськогосподарських наук, доцент
Білоцерківський національний аграрний університет
09111, вул. Героїв Чорнобиля, 3а, м. Біла Церква, Україна
https://orcid.org/0000-0003-3010-6757

Оксана Василівна Іщук

Кандидат сільськогосподарських наук, доцент
Поліський національний університет
10008, Старий бульвар, 7, м. Житомир, Україна
https://orcid.org/0000-0002-8993-8366

Світлана Іванівна Матковська

Кандидат сільськогосподарських наук, доцент
Поліський національний університет
10008, Старий бульвар, 7, м. Житомир, Україна
https://orcid.org/0000-0002-8019-5498

Анотація. Актуальність дослідження зумовлена тим, що під впливом різних чинників водного середовища фіксуються зміни швидкості та спрямованості метаболічних процесів гідробіонтів. Метою дослідження є вивчення впливу різного вмісту неорганічного фосфору у водному середовищі на показники фосфорно-кальцієвого обміну у риб. Використовували методи тонкошарової хроматографії та варіаційної статистики. Аналізували залозисті тканини зябер, печінки та нирок риб. Для вивчення впливу неорганічного фосфору водного середовища на деякі показники внутрішньоклітинного метаболізму виділяли мітохондрії в осморегуляторних органах. Встановлено, що при підвищенні концентрації фосфору у водному середовищі до 0,3-0,6 мг/л відбувається його накопичення в печінці, зябрах і зменшення в нирках, при цьому концентрація в сироватці крові риб підтримується на відносно постійному рівні. Підвищення концентрації неорганічного фосфору у водному середовищі суттєво впливає на процеси енергоутворення в мітохондріях печінки риб. Зміна вмісту фосфору в середовищі існування риб має значний вплив на біоенергетичні процеси в тканинах, що проявляється у зміні вмісту аденозинтрифосфату, активності лужної фосфатази. Значні зміни цих показників спостерігаються в зябрах та нирках. Результати досліджень можуть бути використані для формування адаптаційно-компенсаторних регуляторних механізмів в організмі гідробіонтів при їх пристосуванні до певних умов вирощування та розмноження

Ключові слова: *Cyprinus carpio*; печінка; зябра; нирки; сироватка крові; жовч