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RESEARCH ARTICLE

Tolerance of roach for modified water mineralization levels and temperatures expressed by biochemical indicators

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Abstract. Changes in the activity of Na^+/K^+-ATPase, lactate dehydrogenase, and succinate dehydrogenase in the white muscles and gill petals of roach, *Rutilus rutilus* (L), were studied for the combined actions of water mineralization and water temperature. It was established that decreased mineralization to 260.0 mg dm^{-3} and increased water temperature to 32°C increased aerobic processes in roach tissues. A sharp increase in water temperature and changes in water mineralization significantly affected the tolerance and physiological state of fish and could impact their abundance in waterbodies.

Keywords: roach, water temperature, mineralization, ion exchange, enzyme activity, sensitivity, adaptation

Introduction

Significant climatic changes are currently occurring (Schiedeck et al. 2007, Janauer 2012, Abraham et al. 2013), and they are manifested in increasing average annual water temperatures in natural reservoirs, in significant fluctuations of temperature regimes, and changes in seasonal cycles (Shcherbak et al. 2014). This has a most pronounced impact on shallow waters (Grebin 2015). Additionally, amounts of annual atmospheric precipitation are constantly changing, which, in turn, affects water mineralization levels (Kurilo 2016). Anthropogenic factors and the external contamination of reservoirs contribute significantly to these processes (Schiedeck et al. 2007).

The mineral composition of water is an important ecological factor that determines the structural and functional features of aquatic animal groups. It can act as a limiting factor in the distribution of certain groups of hydrobionts. The value of total mineralization determines the peculiarities of the growth, development, stability, and reproduction of aquatic animals, and particularly fish (Lukyanenko 1987). Fluctuations in water mineralization in natural reservoirs are accompanied by increased stress on systems providing osmotic and ion balance in organisms. After certain minimum concentrations of certain ions in water are reached, violations of vital functions in organisms occur, in particular, ionic homeostasis. Thus, the boundaries of the ranges of the species in reservoirs are determined by the concentrations of ions in the water at which hydrobionts are able to maintain normal osmotic equilibrium between their internal environments and the external environment.

Temperature plays a decisive role in the lives of hydrobionts. It should be noted that aquatic

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ecosystems are characterized by high temperature buffers. Most aquatic animals, including fish, are cold-blooded animals, and their metabolic activity depends directly on the temperature of the surrounding environment. However, even minor fluctuations in temperature regimes can cause significant changes in fish metabolic processes (Romanenko et al. 1991).

One of the features of the species composition of fish is their wide variety and the ability of individual species to adapt to the effects of adverse factors thanks to polymorphism. Fish tolerance forms over a significant period of time. Species adapt to specific environmental conditions, but, because of significant environmental changes, optimum conditions for fish are disrupted. Significant environmental changes cause heterogeneous reactions in groups of hydrobionts, which, in the future, could lead to decreased population numbers. The physiological state of fish and the possibility of the existence of individual species in the face of changes in climatic conditions, namely substantial changes in temperature regimes, atypically high temperatures, total water mineralization levels, and annual cycles can be estimated based on the activity of enzymatic systems (Hochachka and Somero 2002, Shakhmatova 2012). Physiological reactions are crucial to the processes of fish adapting to changes in temperature regimes and other environmental factors in natural reservoirs.

It is possible to use enzyme activity indices of energy metabolism as indicators of the physiological state of fish under the influence of abiotic factors, particularly water temperature and changes in water mineralization. Thus, the activities of lactate dehydrogenase (LDH) and succinate dehydrogenase (SDH) are important indicators of the adaptive abilities of various fish species (Alexander et al. 2013, Chan et al. 2013). SDH is an important enzyme in the tricarboxylic acid cycle, which catalyzes the oxidation of succinate to furamate. The activity of this enzyme depends largely on the presence of oxygen in the water. LDG is an enzyme that catalyzes the conversion of pyruvic acid into lactic acid (Alexander et al. 2013).

Changes in the functioning of cell membranes play significant roles in temperature adaptation (Nemova 1996, Ozernyuk 2000). Thus, the roles of Na^+/K^+-ATPase can be key not only in maintaining the structural integrity of membranes, but also in ensuring their functioning, primarily of ion exchange (Zaprudnova 1991). It is known that cell membrane Na^+/K^+-ATPase is one of the first that provides for the formation of primary reactions to the action of factors and triggers mechanisms for the formation of a long-term adaptation (Boyko et al. 2002, Kyaivjaryainen et al. 2010). Thus, the study of the stability/tolerance of aquatic animals, including fish, at this stage is one of the most important problems in modern hydrobiology, because with these results it will be possible to predict the ecological state of water systems in the future (Rudneva 2013).

The choice of roach, *Rutilus rutilus* (L), stems from the fact that its historical range is significantly different from its modern one that differs particularly with regard to hydrological and hydrochemical characteristics. Roach is a freshwater fish that is common in the rivers, lakes, and reservoirs of Eastern Europe (Moiseev et al. 1981). The purpose of our study was to establish the rate of reactions of the activity of enzymes in the energy metabolism of roach under the influence of different degrees of water mineralization and under fluctuating water temperature regimes that exceed the optimal 24°C for this fish species (Golovanov et al. 2012). These conditions have recently been observed frequently in the shallow water areas of Ukraine.

Material and methods

The research was conducted at the Bila Tserkva experimental hydrobiological station. The object of the study was roach aged 3+. To achieve the set aims, a series of experiments in 60 dm^3 aquariums with different degrees of water mineralization was conducted at 240, 350, and 520 mg dm^{-3} and fluctuating daily water temperature regimes of 21-23, 25-30, and 27-32°C. It should be noted that the water

temperature was maintained at a given level for 12 hours by water heaters, and in the next 12 hours it was allowed to cool to 21°C. Each aquarium was stocked with 8 fish specimens. The oxygen regime was maintained using a micro-compressor. Since oxygen solubility depends directly on temperature, the concentrations of it in the experiments differed. Thus, the water oxygen content in the temperature range of 21-23°C was 6-7 mg dm^{-3}, while in that at temperature ranges of 25-30 and 27-32°C was 5-6 and 4.5-5.5 mg dm^{-3}, respectively, which are not critical for this species (Alabaster and Lloyd 1980). One-third of the total water volume was exchanged daily to reduce the quantities of metabolites in the water.

On day 14 of the experiment samples of fish tissues were collected for biochemical tests from the liver, gill lobes, and white muscle. The tissues (200 mg) were homogenized in 0.2 M KCl (5 mL) in a homogenizer. Then, the homogenate was centrifuged at 3000 rpm for 15 min. The activity of K^+/Na^+-ATPase was measured by increasing the content of inorganic phosphorus (Prohorova 1982). The 2 mL reaction mixture consisted of Tris HCl (2.5 mg), NaCl (11 mg), KCl (3 mg), $MgCl_2 \times 6\ H_2O$ (2 mg), ATP (2.5 mg), and enzyme extract (0.5 mL). The reaction mixture was incubated for 60 min at 25°C. The reaction was stopped by the addition of 1 ml of 10% TCA. Subsequently, 0.1 ml of the extract obtained and 0.4 ml of 20% ammonium molybdate in 8N sulfuric acid was added to 3 ml of ethyl alcohol. Absorbance was measured at 390 nm with a spectrophotometer. The activity of lactate dehydrogenase (LDH) was established using the method by Prohorova (1982); 3 ml of phosphate buffer (pH 7.5) with pyruvate (0.06 mg) and NADH (0.3 mg) were added 0.1 ml of the enzyme extract. Absorbance was measured at 340 nm with a spectrophotometer every 30 s for 3 min. The activity of succinate dehydrogenase was determined with the ferricyanide method (Asatiani 1965); 0.5 ml of enzyme extract was added to 1.4 ml of phosphate buffer (pH 7.8) with succinic acid (1 mg), EDTA (0.005 mg), and sodium aside (1.0 mg). The reaction mixture was incubated for 15 min at 25°C. Then 0.1 mg of 25 mM potassium ferricyanide was added and the reaction mixture was incubated for 15 min at 25°C. The reaction was stopped by the addition of 2 ml of 20% TCA. Absorbance was measured at 420 nm with a spectrophotometer. The activity of enzymes was calculated per mg of protein in the sample. The protein content was determined using the Lowry method (Lowry et al. 1951). The number of fish (n) used in the experiments was 6 individuals per analysis. The significance of differences between the mean values of the groups was determined with using the T-criterion of Student's probability level ($P < 0.05$). The statistical data was processed with Statistica 5.5. All bioethical norms were observed throughout the experiment.

Results

ATPase activity in roach muscles depended largely on water temperature. For example, at 21-23°C, the activity of the enzyme in all the experimental groups was almost unchanged. As water temperature increased to 25-30°C, marked activation of ATPase was observed, especially in water with mineralizations of 260, 350, and 520 mg dm^{-3} at which enzyme activity in roach muscles was 2.1, 12.8, and 8 times higher, respectively, than that of fish that were in the same water mineralizations but in the temperature range of 21-23°C (Fig. 1).

Apparently, the high ATPase activity in the muscles of the experimental groups in a water temperature of 25-30°C was a consequence of the general increase of metabolic processes. In the water temperature regime of 27-32°C, the activity of the enzyme in roach muscle tissues decreased significantly compared to that in the experimental groups in the water with the same water mineralization conditions but with a temperature range of 25-30°C. Thus, ATPase activity decreased 3.4, 3.2, and 8.4 times at water mineralizations of 260, 350, and 520 mg dm^{-3}, respectively. The results obtained showed that the ATPase activity in gills at water mineralizations of

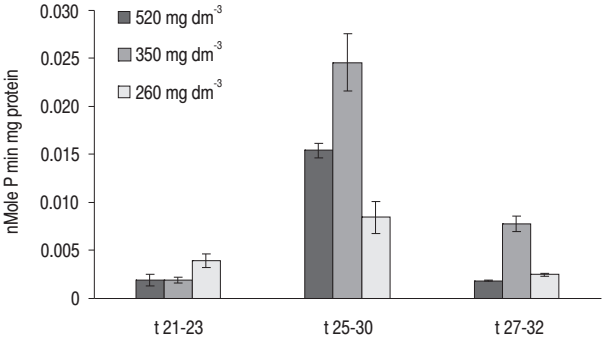

Figure 1. Mean (± SE, n = 6) level of activity Na^+/K^+ ATPase in roach (*R. rutilus*) muscle.

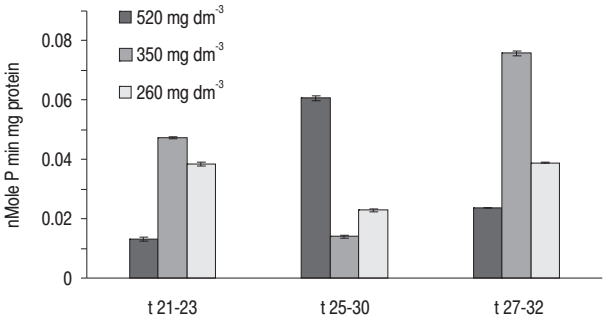

Figure 2. Mean (± SE, n = 6) level of activity Na^+/K^+ ATPase in roach (*R. rutilus*) gills.

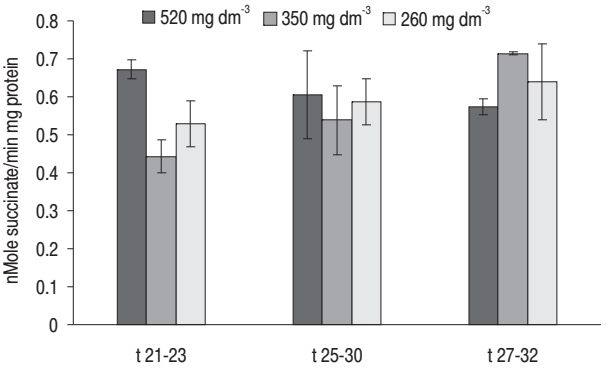

Figure 3. Mean (± SE, n = 6) level of succinate dehydrogenase activity in roach (*R. rutilus*) muscle.

260 and 350 mg dm^{-3} and at a water temperature range of 21-23°C was significantly higher by 2.9 and 3.6, respectively, than it was in fish that were in a water mineralization of 520 mg dm^{-3} (Fig. 2). As the temperature range of 25-30°C increased, another dependence of ATPase activity on the temperature factor was observed, and the highest enzyme activity was noted in the gills of roach that were in water with a mineralization of 560 mg dm^{-3}. It should be noted that the activity of ATPase in the gills of this experimental group was 4.6 times higher than that of fish that were in the same mineralization, but in a temperature range of 21-23°C.

In contrast, the enzymatic activity of ATPase was very low in the gills of the experimental groups that were in less mineralized water of 260 and 350 mg dm^{-3} and at a temperature range of 25-30°C. Thus, the activity of the enzyme in the gills of fish was 4.3 and 2.6 times lower than that in fish that were in water with a mineralization of 520 mg dm^{-3}. It was also noted that the activity of ATPase in gills decreased significantly by 1.7 and 3.4 times in fish that were in water with a mineralization of 260 and 350 mg dm^{-3} and an increased water temperature of 21-23 to 25-30°C. With an increase in water temperature from 25-30 to 27-32°C, the activity of ATPase in the gills of roach in water with a mineralization of 520 mg dm^{-3} decreased 2.7 times. In contrast, at lower water mineralizations of 260 and 350 mg dm^{-3} the enzyme activity increased 1.7 and 5.4 times, respectively (Fig. 1).

The activity of SDH in the muscle tissues of the fish that were in water with a mineralization of 520 mg dm^{-3} decreased as temperature increased. For example, with an increase in temperature from 21-23 to 27-32°C, enzyme activity decreased by 16% (Fig. 3). In the muscles of experimental groups that were in water with lower mineralizations of 260 and 350 mg dm^{-3}, the activity of SDH with the same increase in water temperature increased by 21.1 and 61.1%, respectively. The same tendency was observed in the gills of roach. In fish that were in water with a mineralization of 520 mg dm^{-3} SDH activity decreased 2.5 times with an increase in temperature from 21-23 to 27-32°C. In contrast, SDH activity increased 8, 4, and 4.2 times in experimental groups in water with mineralizations of 260 and 350 mg dm^{-3} (Fig. 4).

In fish that were in water with mineralizations of 350 and 520 mg dm^{-3} when the water temperature increased from 21-23 to 27-32°C the activity of the

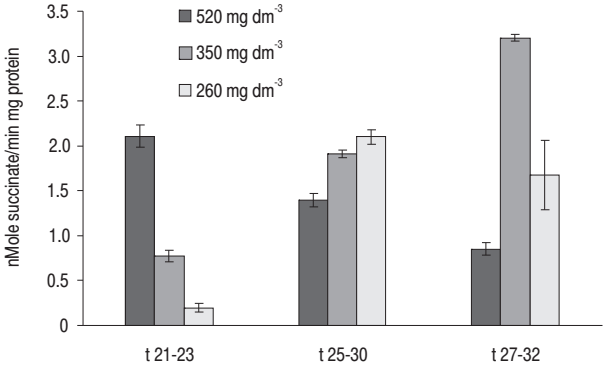

Figure 4. Mean (± SE, n = 6) level of succinate dehydrogenase activity in roach (*R. rutilus*) gills.

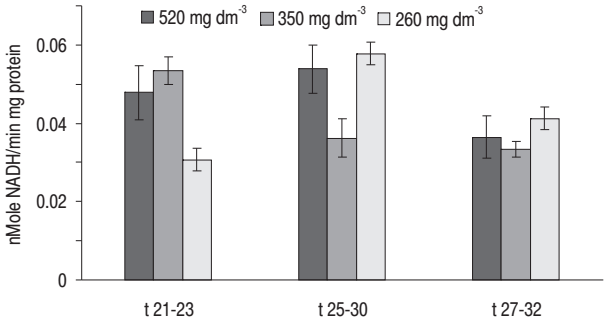

Figure 5. Mean (± SE, n = 6) level of lactate dehydrogenase in roach (*R. rutilus*) muscle.

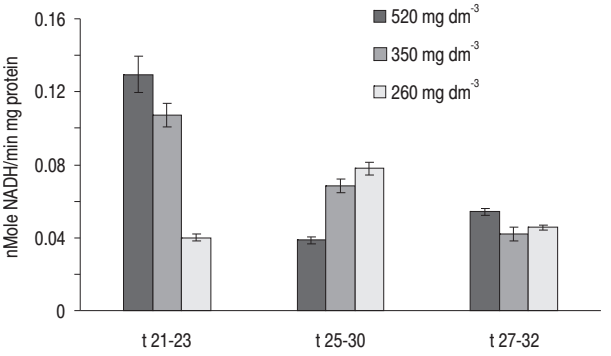

Figure 6. Mean (± SE, n = 6) level of lactate dehydrogenase in roach (*R. rutilus*) gills.

LDH in gills decreased 2.3 and 2.5 times, and in the muscles it decreased 1.6 and 1.3 times, respectively (Fig. 5 and 6). In fish that were in water with a mineralization of 260 mg dm^{-3} the dependence of LDH activity in roach tissues was somewhat different. When water temperature increased from 21-23 to 25-30°C LDH activity increased in muscles 1.8 times and in gills 1.9 times. In contrast, with a further increase in temperature to 27-32°C the enzyme activity in the gill and muscle tissues decreased 1.9 and 1.4 times, respectively. It should be noted that the activation of this enzyme in muscle and gill (13 and 34%) was higher than that of fish that were in water at a temperature of 21-23°C.

Discussion

Changes in water mineralization and temperature did not cause a homogeneous response in roach tissue metabolism. In particular, with changes in these parameters in the experiments different ATpase activities were observed in fish tissues. It is well known that fish are cold-blooded animals and their metabolic activity depends directly on the temperature of the external environment (Johnston and Dunn 1987). Apparently, the high activity of ATPase in the muscles of experimental groups that were in a water temperature of 25-30°C was a consequence of the general increase of metabolic processes. Evidently this phenomenon stems from the inclusion of certain compensatory mechanisms that lead to a decrease in the activity of general metabolic processes at high temperatures, which protect the fish from exhaustion (Berezov and Korovkin 1988). In contrast to the muscles, the dynamics of ATPase activity in gills was quite different. It should be noted that different water mineralizations had a significant impact on the activity of the enzyme in the gills. The greater sensitivity of gills than muscles to fluctuations in external environmental factors is likely to be because of adaptive reactions to the direct contact of these tissues under constantly changing environmental conditions (Martem'yanov 1989, Kyaivjaryainen et al. 2008). It should also be noted that one of the important functions of the gills is body ion regulation, namely maintaining the most favorable ratio of ions necessary for normal cell function. Thus, a significant increase in the activity of ATPase at water mineralizations of 260 and 350 mg dm^{-3} is likely to indicate the active

regulation of the ion concentration gradient in tissue cells thanks to this enzyme system. Obviously, a significant increase in ATPase activity in the gills of this experimental group stemmed from the general increase in metabolic processes, which we noted in the muscles from energy metabolism enzyme activity. Thus, to maintain homeostasis, namely the maintenance of stable ion content in fish bodies, there was an increased need for intensive ion exchange with the environment.

Most likely, against the backdrop of increasing ion exchange in the muscle tissues of these experimental groups to reduce the outflow of ions from the body, the activity of ATPase in their gills decreased. Obviously, this phenomenon is associated with significant changes in the permeability of the membranes (Kumosani 2004, Boldyrev et al. 2006). The significant decrease in the activity of ATPase in a water mineralization of 520 mg dm^{-3} is probably associated with a decrease in ion exchange in the fish, which we observed in white muscle. A significant increase in the activity of the enzyme in experimental groups, which were in water with mineralizations of 260 and 350 mg dm^{-3}, was observed with significant structural changes in their membranes.

Data from the literature indicate that during significant increases in water temperature lipid membrane reorganization occurs; namely, an abnormal membrane disintegration occurs because of increases in the pool of unsaturated lipids. Simultaneously there is an increase in the rate of ion diffusion through the membrane, because of the differentiation of the fatty acid chains (Kreps 1979). Perhaps, as a result of such changes, in fish that were in water with mineralizations of 260 and 350 mg dm^{-3}, ATPase activity increased to quickly restore the concentration gradient in cells. Consequently, increased ATPase activity probably suggests a forced amplification of ion exchange. Thus, a water temperature range of 27-32°C and mineralizations of 260 and 350 mg dm^{-3} were critical limits for roach at which certain violations were possible, which could ultimately lead to death. It is known that water with low ion concentrations leads to death because of desalting (Vinogradov and Komov 1988, Vinogradov 2000).

One of the main criteria for energy, in particular carbohydrate metabolism in tissues, is establishing LDH and SDH enzymatic activity. The evaluation of these enzymatic systems also makes it possible to identify the direction of energy processes. The overall decrease in LDH activity in all experimental groups in the water temperature range of 27-32°C probably indicated a decrease in glycolysis in roach and the transition to more beneficial energy-supplying processes, namely, aerobic respiration. The results of our studies show that the activity of these enzymes is completely dependent on changes in water temperature and mineralization.

Obviously, the decrease in the activity of SDH in the tissues of the experimental groups that were in a water mineralization of 520 mg dm^{-3} was due to certain adaptation processes. Chronic stress is often accompanied by metabolic depression, namely, energy savings are minimized by reducing concentration gradients and weakening ion transport flows (Zaprudnova 1991). Evidently, increasing the activity of SDH in fish suggests the activation of metabolic processes caused by water temperature and mineralization. Data from the literature indicate that changes in water mineralization lead to certain violations in ionic balance and causes certain changes in the water balance levels of cells. In particular, a significant reduction in water mineralization is accompanied by a significant increase in the gradients of ion concentrations between the internal and external environments of the fish. This situation increases the burden on the body's osmotic regulation system, which incurs additional energy costs (Martem'yanov and Mavrin 2012). It should also be emphasized that in freshwater fish the cytoplasm of the cells is hypertonic, that is, the concentration of most of the ions in it predominates in terms of their content in the external environment. This, in turn, leads to a constant flow of water into the cell. Thus, freshwater fish are constantly forced to cope with excess water. With a decrease in water mineralization, the intensity of water withdrawal increases. In addition, it should be noted that these

processes are energy intensive (Nemova 2010). Consequently, in water with mineralizations of 260 and 350 mg dm^{-3}, the activity of SDH in fish tissues increased as a result of increased energy needs for the processes of water balance in cells and in fish bodies.

The overall decrease in LDH activity in all experimental groups in the temperature range of 27-32°C probably indicated a decrease in glycolysis in roach and the transition to a more beneficial energy-supplying processes, namely, aerobic respiration. It is generally known that about 200 kJ of energy is released during glycolysis. Part of it (almost 84 kJ) is spent on the synthesis of two ATP molecules, and the rest (about 116 kJ) is dissipated in the form of heat. In the course of aerobic respiration (the respiratory cycle of tricarboxylic acids) about 2800 kJ of energy is released, of which 1596 kJ or 15% is stored (in the form of macroergic bonds of ATP), and 45% is dissipated in the form of heat. Thus, the glycolysis process is energetically ineffective, since only 35-40% of the energy is accumulated in macroergic ATP bonds. Consequently, the aerobic stage of energy metabolism plays a major role in providing cells with energy (Shulman and Tokarev 2006). Obviously, an increase in the activity of LDH in roach gills and muscles in a water mineralization of 260 mg dm^{-3} was caused by water temperature. This led to the intensification of the processes of osmoregulation and ion exchange, which, accordingly, led to increased energy costs to stabilize these processes.

Summary

It was established that under conditions of increased water temperatures and periodic decreases of water mineralization the biochemical parameters related to energy and ion exchange changed significantly in the fish investigated. Certain changes in the activity of a number of enzymes occurred with increasing water temperatures in all the experimental groups. It should be noted that adaptive processes were not identical in different water mineralizations. Thus, at the highest water mineralization of 520 mg dm^{-3} and with increased water temperature, the protective reaction of the roach bodies was to reduce energy processes. In the lower water mineralizations of 350 and 260 mg dm^{-3} and increased water temperatures increased SDH activity in muscles and gills was observed of 21.1 and 61.1% and 8.4 and 4.2 times. At the same time there was a decrease in LDH activity at 350 mg dm^{-3} of 2.3 and 1.9 times, which indicated the redirection of energy from glycolysis to the aerobic path. It should be noted that a water mineralization of 260 mg dm^{-3} was not comfortable for the roach. Such high enzymatic activity in the tissues of experimental groups could lead to negative consequences, namely, exhaustion and, ultimately, death. The results obtained can be used to estimate the physiological outlook of roach to predict the qualitative and quantitative composition of fish under conditions of climatic excess of water temperatures and changes in mineralization.

Author contribution. M.P., Y.Kh. – designed and performed the experiment, O.Z. – analyzed the data, O.P. – wrote the paper.

References

Abraham J.P., Baringer M., Bindoff N.L., Boyer T., Cheng L.J., Church J.A., Conroy J.L., Domingues C.M., Fasullo J.T., Gilson J., Goni G., Good S.A., Gorman J.M., Gouretski V., Ishii M., Johnson G.C., Kizu S., Lyman J.M., Macdonald A.M., Minkowycz W.J., Moffitt S.E., Palmer M.D., Piola A.R., Reseghetti F., Schuckmann K., Trenberth K.E., Velicogna I., Willis J.K. 2013 – A review of global ocean temperature observations: Implications for ocean heat content estimate sand climate change – Rev. Geophys. 51: 450-483.

Alabaster J.S., Lloyd R. 1980 – Water Quality Criteria for Freshwater Fish – London - Boston: Butterworth, 297 p.

Alexander A., Verma O.P., Mathur A., Sharan A., Varshne P.K. 2013 – Evaluation of changes in metabolic parameters and enzymes involved in metabolic pathways in *Clarias botrachus* after exposure to phenolic compounds – Asian J. Biomed. Pharm. Sci. 3: 60-67.

Asatiani V.S. 1965 – New methods of biochemical photometry – Nauka, Moscow 544 p. (in Russian).

Berezov T.T., Korovkin B.F. 1988 – Biological Chemistry – Meditsina, Moscow, 704 p. (in Russian).

Boldyrev A.A. 1998 – Na+/K+ ATPase. Properties and biological role – Soros Educational Journal, Biology. 4: 2-10 (in Russian).

Boldyrev A.A, Kaivarainen E.I., Ilykha V.A. 2006 – Biomembranology – Petrozavodsk: KarRC RAS, Petrozavodsk 226 p. (in Russian).

Boiko N., Celevycz M., Sanagurski D. 2002 – The heavy metal ion influence on the Na^+, K^+-ATPase activity and the dynamic of transmembrane potential of loach embryos – Visnyk of L'viv Univ., Biology Series 29: 25-32 (in Ukrainian).

Chan FK.M., Moriwaki K., De Rosa MJ. 2013 – Detection of necrosis by release of lactate dehydrogenase activity – Methods Mol. Biol. 979: 65-70.

Golovanov V.K., Smirnov A.K., Kapshaj D.S. 2012 – Final thermopreferendum and upper lethal temperature in juveniles of some freshwater fish species – Transactions of Karelian Research Center of the Russian Academy of Science, Experimental Biology, 2: 70-75 (in Russian).

Grebin V.V. 2010 – The current water regime of Ukraine's river (landscape-hydrological analysis) –Nika-Center, Kyiv. 316 p. (in Ukrainian).

Hochachka P.W., Somero G.N. 2002 – Biochemical adaptations mechanism and process in physiological evolution – Oxford: Oxford University Press. Princeton University Press, USA, 356 p.

Janauer G.A. 2012 – Aquatic Vegetation in River Floodplains: Climate Change Effects, River Restoration and Ecohydrology Aspects – In: Climate Change (Eds) A. Berger, F. Mesinger, D. Sijacki Springer, Vienna: 149-155.

Johnston I.A., Dunn J. 1987 – Temperature acclimation and metabolism in ectotherms with particular reference to teleost fish – Symp. Soc. Exp. Biol. 41: 67-93.

Kreps E.M. 1979 – Cellular lipids and their role in the adaptation of aquatic organism to the conditions of existence – In: Physiology and Biochemistry of Marine and Freshwater Animals (Ed.) E.M. Kreps, Nauka, Leningrad, 3-22 (in Russian).

Kumosani T.A. 2004 – Na+, K+-ATPase activity from various organs of Tilapia *(Oreochrom ismossambicus):*A bioindicator for environment monitoring – Proc. 2nd Saudi Sci. Conf., Fac. Sci. 29-41.

Kurilo S.M. 2016 – Main tendencies of long-term changes of water mineralization and content of main ions in rivers of Ukraine – Hydrochemistry. Hydrology. Hydroecology. 2 (41): 85-90 (in Ukrainian).

Kyaivjaryainen E.I., Borvinskaya E.V., Nemova N.N. et al. 2008 – Changes in the activity of the enzyme Na / K-ATPase sterlet (*Acipenser ruthenus*) with a change in salinity and acidity. Mat. VI Anniversary International Scientific Conference „Innovationsin Science and Education – 2008", Russia, Kaliningrad: 100-101 (in Russian).

Kaivarainen E.I., Borvinskaya E.V., Serupin G.G., Kuklina M.M., Nemova N.N. 2010 – Role of Na^+/K^+-ATPase in biochemical mechanisms of adaptation to biotic and abiotic environmental factors. – Problems of physiology and biochemistry and water organisms. Mat. III Intern. Conf. Petrozavodsk: 100-101 (in Russian).

Lowry O.H., Rosebrough N.J., Farr A.L., Randall R.J. 1951 – Protein measurement with the folin phenol reagent – J. Biol. Chem. 193: 265-275.

Lukyanenko V.I. 1987 – Ecological aspects of Ichthyotoxicology – Agropromizdat, Moscow. 240 p. (in Russian).

Martem'yanov V.I. 1989 – Effect of salinity on fresh water fish – Zool. Zh. 68: 72-81 (in Russian).

Martemyanov V.I., Mavrin A.S. 2012 – Threshold concentrations of cations in the experimental environment defining the spread boundaries of the minnow *Phoxinus phoxinus* L. in fresh waters – Water: Chemistry and Ecology, 7: 115-118 (in Russian).

Moiseyev P.A, Azizova N.A., Kuranova I.I. 1981 – Ichthyology – Legkaya i pishchevaya promishlennost, Moscow, 364 p. (in Russian).

Nemova H.N. 1996 – Intracellular proteolytic enzymes in fish – Karelian Research Center of the Russian Academy of Science, Petrozavodsk, 104 p. (in Russian).

Nemova N.N. 2010 – Mechanisms of Biochemical Adaptations in Aquatic Organisms: Ecological and Evolutionary Aspects – In: Current Problems of Physiology and Biochemistry of Aquatic Organisms. Volume I. Ecological Physiology and Biochemistry of Aquatic Organisms Collection of Scientific Articles – Karelian Research Center of the Russian Academy of Science, Petrozavodsk: 198-214 (in Russian).

Ozernyuk N.D. 2000 – Temperature adaptations – MSU Publishing House, Moscow, 205 p. (in Russian).

Prohorova M.I. 1982 – Methods of biochemical research (lipid and energy metabolism) – Publishing House of Leningrad University Press, Leningrad, p. 272 (in Russian).

Romanenko V.D., Arsan O.M., Solomatina V.D. 1991 – Mechanisms of temperature acclimation of fishes – Naukova Dumka Press, Kyiv, 192 p. (in Russian).

Rudneva I.I. 2013 – Applications of fish biomarkers for ofshore zones ecological state evaluation – Scientific notes of the Ternopil National Pedagogical University, Series Biology, 2 (55): 68-73 (in Russian).

Schiedeck D., Sundelin B., ReadmanJ. W., Macdonald R.W. 2007 – Interactions between climate change and contaminants – Mar. Pollut. Bull. 54: 1845-1856.

Shcherbak V.I., Zadorozhnaya A.M., Kalenichenko K.P. 2014 – Phytoplankton spatial and emporal dinamics in the additional net of the Kiev Section of the Kanev Reservoir – Hydrobiol. J. 50: 3-12 (in Ukrainian).

Shakhmatova O.A. 2012 – Response hydrobionts to stress factors of marine ecosystems – Optimization and protection of Ecosystems, TNU, Simferopol, 7: 98-113 (in Russian).

Shulman G.E, Tokarev Yu.N. 2006 – Functional diversity ask an important factor of the existence of biotic components of ecosystems – Marine Ecological Journal 5: 35-56.

Vinogradov G.A. Komov V.T. 1988 – Ion exchange in gold Crucian carp and Gold carp during acclimation in the low mineralization water – Vopr. Ikhtiol. 28: 124-131 (in Russian).

Vinogradov G.A. 2000 – Processes of ion regulation in freshwater fish and invertebrates – Nauka, Moscow, 216 p. (in Russian).

Zaprudnova R.A. 1991 – Concentration of ions in the blood plasma and muscles in the moribund freshwater fish – Vopr. Ikhtiol. 31: 520-524 (in Russian).