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Editorial Note

Editorial Note for Ukrainian Journal of Ecology

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Without continual growth and progress, such words as improvement, achievement, and success have no meaning. Founded in 2011, Ukrainian Journal of Ecology (ISSN: 2520-2138) is growing continuously. It is our pleasure to announce that during year 2019, all issues of volume 9 were published online on time and the print issues were also brought out and dispatched within 30 days of publishing the issue online.

All published articles of this journal are included in the indexing and abstracting coverage of Web of Science (Emerging Sources Citation Index), CAB Abstracts, ProQuest, DOAJ, Google Scholar, EBSCOhost, JournalTOCs, Scilit, WorldCat-OCLC, eLIBRARY.ru, Agricola, Agris, Academic Journals Database, Ulrich's Periodicals Directory, OAIsters Directory, HINARI, Bielefeld Academic Search Engine (BASE).

During the calendar year 2019, Ukrainian Journal of Ecology received a total of 201 papers, out of which 10 articles (10%) were rejected in the preliminary screening due to plagiarism or being out of the format and peer review process. During 2019 around 191 articles were subjected for publication after they are accepted in the peer review process. In the 4 issues of Volume 9 published during the year 2019, a total of 191 articles were published (at an average of 70 articles per issue) of which, articles were published from authors all around the world.

A total of 181 research scientists from all over the world reviewed the 191 articles published in volume 9. Average publication lag time of an article was further reduced to 3- 4 weeks. Journal will be running its website https://www.ujecology.com/ parallel for Editorial and review work process so as to maintain its highest standard of scientific work. Ukrainian Journal of Ecology has increased Readership Metrics (By Google Analytics) of Ecology which can be accessed at Google Analytics Metrics.

I take this opportunity to acknowledge the contribution of Associate Editor: Matsyura O.V (Ukraine), during the final editing of articles published in bringing out issues of Ukrainian Journal of Ecology in time. I would also like to express my gratitude to all the authors, reviewers, the publisher, the advisory and the editorial board of Ukrainian Journal of Ecology, the office bearers for their support in bringing out yet another volume of Ukrainian Journal of Ecology and look forward to their unrelenting support to bring out the Volume 10 of Ukralnian Journal of Ecology in scheduled time.

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ORIGINAL ARTICLE

Effect of reserviour temperature and oxygen conditions on the activity of Na-K pump in embrios and larvae of perch, roach, and ruffe

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The Na + / K + –ATPase of cell membranes is one of the first to provide the formation of a primary response to the action of factors and initiates the mechanisms of formation of long-term adaptation. That is why the purpose of the study was to study the activity of Na-K pump in embryos and caviar of perch, roach, and ruffe under the action of different temperature and oxygen modes of the reservoir. The biological material of the studies was caviar, embryos and larvae of perch (*Perca fluviatilis* L.), roach (*Rutilus rutilus* L.) and ruffe (*Gymnocephalus cernuus* L.). Three reservoirs (ponds) were selected, which, due to their location and degree of shading, differed in temperature conditions and, consequently, in the oxygen regime. The studies were conducted durlng April-May, at a time when perch, roach and ruffe spawning occur in natural water bodies. The maximum activity of the enzyme at the pre-cell stage was observed at a temperature of 16.3°C and amounted to 4.67 μmol Pi mg protein^{-1} h^{-1}, and when it reached an ambient temperature of 18.1°C it decreased by 46.4% compared to the maximum. With the increase in the temperature of the aqueous medium above the norm decreased the activity of Na-K pump, which is due to the weakening of redox and increased anaerobic processes at low oxygen content in water. The activity of Na-K pump for perch embryos clearly shows that the most favorable temperatures for embryonic development are its lower values, in particular for the stage of eye pigmentation it is 14.9°C. After hatching at the pre-stage stage, maximum enzyme activity was observed at 16.3°C. For embryonic development of ruffe in the middle of the shell, the optimum water temperature is 15–16°C, but already at the stage of pre-cellulose, its optimum increased significantly, reaching 19–20.9°C. Roach is a more thermophilic species compared to perch and prefers higher water temperatures during embryonic development. It was noted that the activity of ATPase increased significantly with increasing water temperature. This may indicate a high adaptation of the roach to the temperature of the reservoirs, both under climatic norms and somewhat exceeding this norm. The best temperature for embryonic development of the roach is 16–17°C, and during pre-embryonic stage its optimum increased to 20–21°C.

Keywords: Temperature and oxygen regime of the reservoir; Perch; Roach; Ruffe; Embryos

Introduction

Fresh waters are particularly vulnerable to climate change because many species within these fragmented habitats have limited abilities to disperse as the environment changes; water temperature and availability are climate-dependent; and many systems are already exposed to numerous anthropogenic stressors (Woodward et al., 2010; Vodianitskyi et al., 2017). Changes in temperature are known to have a significant effect on metabolic rate and overall metabolic rate (Awenius et al., 2001; Roesner et al., 2006; Grynevych et al., 2018; Boyko & Brygadyrenko, 2019; Rudenko et al., 2019).

Climate change is causing species to shift their phenology, or the timing of recurring life events such as migration and reproduction, in variable and complex ways. This can potentially result in mismatches or asynchronies in food and habitat resources that negatively impact individual fitness, population dynamics, and ecosystem function. Numerous studies have evaluated phenological shifts in terrestrial species, particularly birds and plants (Staudinger et al., 2019). The existence of a relationship between the level of metabolism and enzymatic activity makes it possible to characterize fish status by molecular biomarkers. The major molecular biomarkers of metabolism include: Na-K pump, Ca^{2+}, Mg^{2+} -ATPase and total ATPase activity, lactate dehydrogenase and succinate dehydrogenase (Dahlhoff, 2004). Na-K pump was detected in the plasma membrane of virtually all animal cells. It maintains K + in high and Na + in low concentrations inside the cell (compared to the environment) (Fiedler & Scheiner-Bobis, 1996; Blanco & Mercer, 1998). It is known that Na-K pump can hydrolyze GTP and UTF, and provide active transport of sodium and potassium ions during this reaction. However, ATP is not only a substrate, but also a modulator of Na-K pump, regulating its affinity for transporting

cations. Other nucleotides are not capable of modulating affinity for transporting ions, although their hydrolysis also leads to active transport (Blanco & Mercer, 1998; van der Meer et al., 2005; Poulsen et al., 2010). It should be emphasized that both the level of temperature changes and the speed of their changes are important for the normal course of exchange processes. A sharp decrease in temperature can lead to a slowdown in metabolic processes that can no longer ensure the normal course of life. The opposite significant increase in temperature can cause such an acceleration in the intensity of metabolic processes, which is difficult or impossible to provide with oxygen (Gracey et ai., 2001; Jezek & Hlavata, 2005; Roesner et al., 2006). Therefore, poikilothermic animals have mechanisms to control the intensity of metabollc processes, and, first of all, due to enzymatic regulation (Awenius et al., 2001).

The most noticeable changes in enzymatic activity are found in fish at embryonic and early post-embryonic stages of development (Awenius et ai., 2001). The enzyme plays a key role in the implementation of numerous cellular functions and processes related to ionic gradients (Souza et al., 2000; Woo et al., 2000; Therien & Blostein 2000). In the case of temperature adaptation, changes in the function of cell membranes can play an important role. The role of Na-K pump in this case can be crucial not only in maintaining the structural integrity of membranes, but also in ensuring their functional status, above ali, ion exchange. This enzyme participates in the processes of osmotic and ionic regulation, provides active transport. It is embedded in the outer plasma membrane of the cell, providing the transfer of Na + and K + ions against their concentration gradient. In addition, Na-K pump can regulate the transport of various metabolites, including sugars and amino acids across the cell membrane (Vodianitskyi et ai., 2017). It is known that the Na-K pump of cell membranes is one of the first to provide the formation of a primary response to the action of factors and initiates the mechanisms of formation of long-term adaptation.

The purpose of the study was to study the activity of Na-K pump in embryos and fish primates (perch, roach, ruffe) under the action of different temperature and oxygen modes of the reservoir.

Materials and Methods

The biological material of the studies was caviar, embryos and larvae of perch (*Perca fluviatilis* L.), roach (*Rutilus rutilus* L.), and ruffe (*Gymnocephalus cernuus* L.). Three reservoirs (ponds) were selected, which, due to their location and degree of shading, differed in temperature conditions and, consequently, in the oxygen regime. The water temperature was measured with a mercury thermometer during the day at 4, 12, and 20 h and as the embryonic stages of development of the experimental fish have passed. The dissolved oxygen content was measured at four o'clock in the morning by the Winkler method (Romanenko, 2006). All experimental reservoirs were filled with water from the river Ros, which was characterized by the following hydrochemical parameters: O_2 – 8.4–9.7 mg/dm³, pH – 8.3, hardness – 6.1 mg-eq/dm³, Ca^{2+} – 3.3 mg-eq/dm³, Mg^{2+} – 2.8 mg-eq/dm³, Cl^- – 0.85 mg-eq/dm³, NH_4^+ – 0.277 mg N/dm³, NO_2^- – 0.006 mg N/dm³, NO_3 – 0.080 mg N/dm³, PO_4^{3-} 0.062 mg P/dm³, PO – 8.0 mg O/dm³, BO – 18.48 mg O/dm³. The studies were conducted during April-May, at a time when perch, roach and ruffe spawning occur in natural water bodies. Fertilized spawn of experimental fish species was placed in mesh containers (Cont. = 169 cm²) in the reservoir. Caviar of all fish species was selected from three different females and placed in ponds in three replicates. The average number of eggs in each mesh container reached 100-150 eggs. Upon reaching the certain stages of caviar development: the end of gastrulation, iens primordium, and eyes pigmentation it was selected and frozen in the freezer at –18°C.

The activity of Na, K-activated, Mg-dependent ATPase (KF 3.6.1.4) was determined by the increase of inorganic phosphorus content in the incubation medium according to the method of M.N. Kondrashova and others (Asatiani, 1965; Kondrashova et al., 1965). The incubation environment (1 cm³ soiution) contained 100 µmol of NaCl, 20 µmol of KCl, 5 µmol of $MgCl_2$, 50 µmol of Tris – HCl (pH 7.4). To the environment was added 0.1 cm³ of 3 molar ATPase Na_2 solution on 50 molar Tris-HCl buffer (pH 7.4). ATPase activity was expressed as µmoi Pi mg protein^{-1} h^{-1}

Results and Discussion

Studies have been conducted on the embryos and caviar of perch, as a typical aboriginal fish species in our reservoirs. In the early stages of development (end of gastrulation) the maximum activity of Na-K pump was fixed at 10.2°C and amounted to 3.11 µmol Pi mg protein^{-1} h^{-1} (Figure 1A). Further, with the subsequent increase in ambient temperature by only 0.2°C the enzyme activity decreased by 47%, but already at a temperature of 11.2°C it again increased to 2.9 µmol Pi mg protein^{-1} h^{-1}n, which is 6.7% less than with the maximum. At the stage of development (Figure 1B), a similar tendency, which we saw in the previous stage of development, remains: with increasing water temperature the enzyme activity decreases. The maximum activity of Na-K pump in the lens primordium stage was observed at a minimum water temperature of 8.7°C and amounted to 5.00 µmol Pi mg protein^{-1} h^{-1}. Already with a decrease in the temperature of the aqueous medium by only 0.1°C, there was a tendency to decrease the activity of the enzyme by 7.2%, and its activity level was 4.64 µmol Pi mg protein^{-1} h^{-1}. The minlmum actlvity was recorded at a temperature of 9.1°C–4.23 µmol Pi mg protein^{-1} h^{-1}, which is 15.4% less than the maximum.

The pattern described in the previous two stages is also characteristic of the stage of eye pigmentation, but the overall activity at this stage of development was significantly lower. In Fig. 1B clearly shows that with increasing water temperature enzyme activity decreases. Maximum activity was observed at 14.9°C, which was 0.92 µmol Pi mg protein^{-1} h^{-1}, and a minimum of 0.86 µmol Pl mg protein^{-1} h^{-1} was at 15.7°C, which is 6.5% less than compared to with the maximum. According to the activity of Na-K pump for perch embryos it can be judged that the most favorable temperatures for embryonic development are its lower values, in particular, for the stage of eye pigmentation is 14.9°C.

After hatching at the stage of premolarization, the water temperature in the studied reservoirs significantly increased to 16.3–18.1°C, which also significantly affected the activity of the enzyme in embryonic tissues. It was also noted that during the embryonic and early post-embryonic development, the same inverse relationship was maintained between water temperature and Na-K pump activity. Since perch is a spring-spawning species of fish and embryonic development occurs at relatively low temperatures, we did not observe any problems with the oxygen regime durlng all the studies, its concentration was ln the range of 5.0–11.6 mg/dm³. The maximum activity of the enzyme at the pre-cell stage was observed at a temperature of 16.3°C and amounted to 4.67 µmol Pi mg proteln^{-1} h^{-1}, and when it reached an ambient temperature of 18.1°C it decreased by 46.4% compared to the maximum.

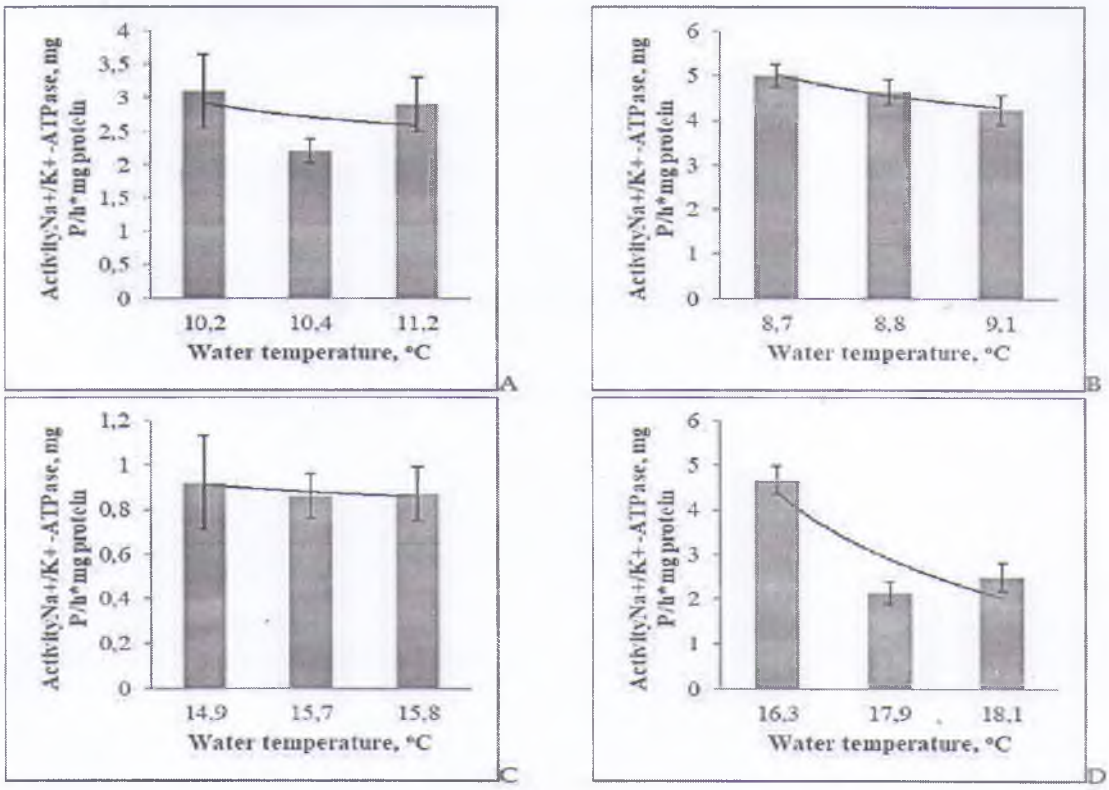

Figure 1. Changes in the activity of Na-K pump in caviar and embryos of perch under the influence of water temperature.
A -End of gastrulation, B - Lens primordium, C – Eyes Pigmentation of the eyes, D – Forebrain development.

Because roach is a more thermophilic species of fish than perch, it is characterized by significantly higher water temperatures during embryonic development, which can fluctuate within a rather wide range of 14–21°C.

At the end of gastruiation stage (Figure 2A), a pattern was observed that with increasing temperature, Na-K pump activity increased significantly. Its minimum activity was recorded at a temperature of 16.9°C and amounted to 2.84 μmol Pi mg protein^{-1} h^{-1}. Further, with increasing water temperature to 18°C, it tended to increase by 5.3%. Already at 18.5°C, enzyme activity reached its maximum (3.46 μmol Pi mg protein^{-1} h^{-1}), which is 17.9% higher than the minimum. This indicates that gambling embryos feel better at higher temperatures from the study range, but at lower values, development is within normal limits.

During the next stage of development – lens primordium formation, the temperature in the studied reservoirs slightly (by 1.5–2.0°C) decreased. However, despite this, the pattern that was characteristic of the previous stage of deveiopment, in this case remains. The minimum activity of Na-K pump was at 15°C and amounted to 1.59 μmol Pi mg protein^{-1} h^{-1}. With the subsequent increase in water temperature by 1.2°C, it increased by 37.1%. The maximum enzyme activity was observed at 16.9°C, namely 3.18 mg P/h × mg protein, which is 50% more than the minimum. The stage of development of roach (eye pigmentation) also took place with a general decrease in water temperature to 14.4–14.8°C, which, in turn, affected the enzyme activity (Figure 2B). Clearly, in the absence of differences in water temperature in different reservoirs, the activity of ATPase in embryos was almost at the same level. The minimum activity of Na-K pump at a temperature of 14.7°C was lower than at a temperature of 14.4 °C by only 6.9% and these differences ware not significant.

During the hatching of melanomacrophage centers from the shell, the temperature in the reservoirs significantly Increased to 19.6–20.9°C, which, as well as the loss of the shell and perivitellin fluid, significantly influenced the increase of enzyme activity. In addition, this stage of development is characterized by the preservation of a similar pattern of changes in the activity of ATPase, depending on the temperature of the aquatic environment, which we observed in the previous stages of embryonic development of roach. Its minimum activity was recorded at 19.6°C, which amounted to 8.94 μmol Pi mg protein^{-1} h^{-1}, and its maximum activity at 20.9°C was 15.63 μmol Pi mg protein^{-1} h^{-1}, which is 42.8% more than the minimum.

Based on our own studies on the activity of Na-K pump in embryos and melanomacrophage centers, it can be observed that it increases significantly with increasing water temperature. This may indicate a good adaptation of the roach to the temperature regime of the reservoirs, both under climatic norms and somewhat exceeding this norm. The best temperature for embryonic development of the roach is 16–17°C, and at its pre-stage stage its optimum increased to 20–21°C.

Changes in the activity of Na-K pump were performed depending on the ambient temperature of embryos and ruffe primers. The results of our own studies showed that in the early stages of embryo development – the end of gastruiation there was a clear tendency to increase the activity of the enzyme with increasing temperature, although there were no sharp jumps (Figure 3A). The minimum activity was recorded at 13.6°C, which was 4.54 μmol Pi mg protein^{-1} h^{-1}, and the maximum was observed at 14.5°C and it was 5.02 μmol Pi mg protein^{-1} h^{-1}, which was by 9.6% more compared to the minimum. This indicates that higher temperatures from the studied range are preferable for embryonic ruffe development.

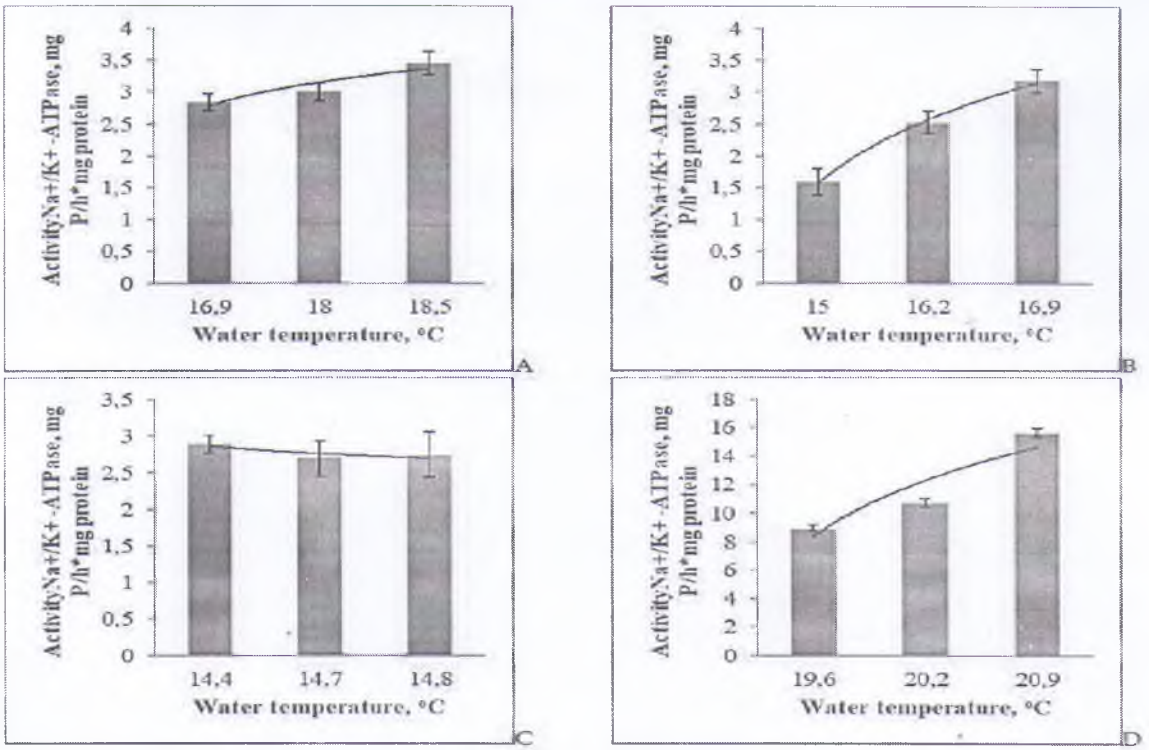

Figure 2. Changes in the activity of Na-K pump in roach caviar and melanomacrophage centers under the influence of water temperature. A - end of gastrulation, B – lens primordium, B – eyes pigmentation, D – forebrain development.

During the transition to the lens primordium formation there was a signlficant increase in water temperature in the studied reservoirs up to 16.3–18.8°C, which is what Na-K pump reacted with the change of its activity (Figure 3B). At this time observed a sharp decrease in the activity of the enzyme with a significant increase in ambient temperature. Its maximum activity was recorded at 16.3°C and amounted to 1.52 μmol Pi mg protein^{-1} h^{-1}, and the minlmum activity at 18.8°C was 0.88 μmol Pi mg protein^{-1} h^{-1}, which was by 42.0% less compared to the maximum. This indicates that there is a narrow corridor of optimal temperature for ruffe embryos, and when exiting the body, the body tries to adapt to the unfavorable environment, reducing the activity of Na-K pump.
During the pre-stage stage, the temperature in the reservoirs increased. However, at this stage of development, the body of the ruffe felt more comfortable, as evidenced by the consistently high activity of Na-K pump at all tested water temperatures from 19.4 to 21.3°C. An increase in enzyme activity was observed at a temperature of 20.9°C and reached its maximum –8.82 μmol Pi mg protein^{-1} h^{-1}. Further, it fell slightly and stood at 21.3°C - 7.64 μmol Pi mg protein^{-1} h^{-1}, which was by 13.3% less than the maximum.

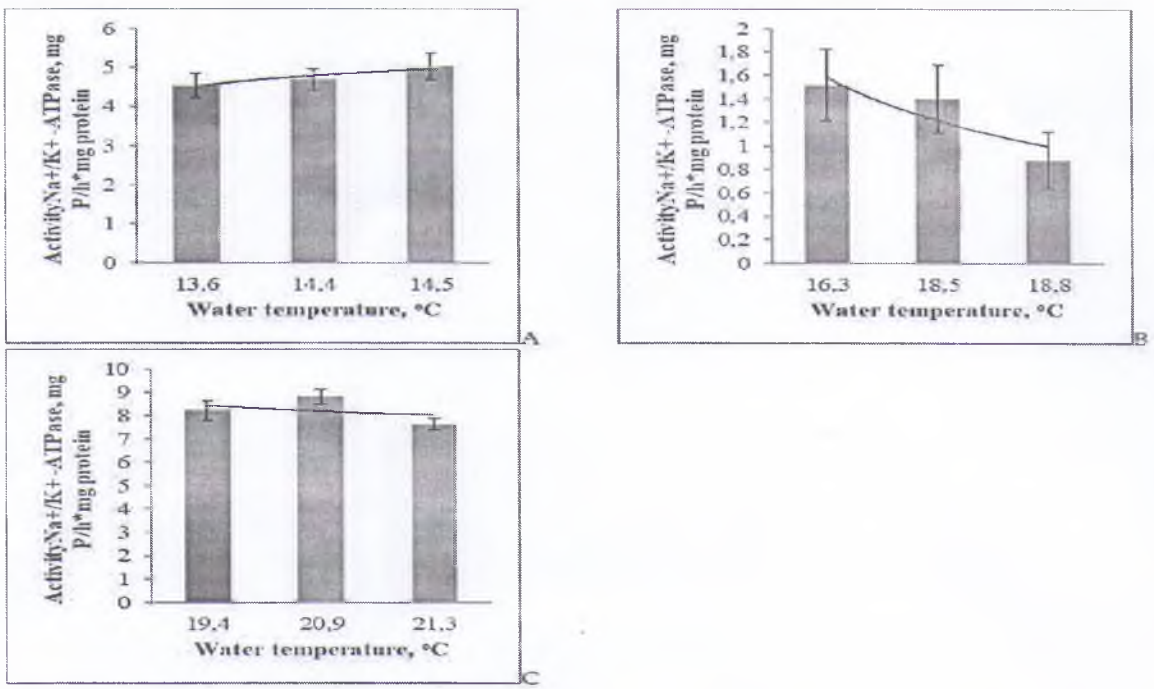

Figure 3. Changes in Na-K pump activity in caviar and ruffe melanomacrophage centers under the influence of water temperature. A -end of gastrulation, B – lens primordium, C – forebraln developmnet.

Conclusion

Therefore, the results of our studies on the activity of key enzymes of energy and plastic exchange showed that with increasing the temperature of the aqueous medium above normal decreased activity of Na-K pump, which is due to the weakening of redox and increased anaerobic processes in low oxygen content. Since the embryonic development of perch occurs at relativeiy low temperatures, we did not observe any problems with the oxygen regime during all the studies, its concentration was in the range of 5.0–11.6 mg/dm^3. The activity of Na+/K+ -ATPhase for perch embryos clearly shows that the most favorable temperatures for embryonic development are its lower values, in particular for the stage of eye pigmentation it is 14.9°C. After hatching at the pre-stage stage, maximum enzyme activity was observed at 16.3°C. For embryonic deveiopment of ruffe in the middle of the shell, the optimum water temperature is 15–16°C, but already at the stage of pre-cellulose, its optimum increased significantly, reaching 19–20.9°C. Roach is a more thermophilic species of fish compared to perch, which is characterized by significantly higher water temperatures during embryonic development. It was noted that the activity of ATPase increased significantiy with increasing water temperature. This may indicate a high adaptation of the roach to the temperature of the reservoirs, both under climatic norms and somewhat exceeding this norm. The best temperature for embryonic development of the roach is 16–17°C, and for the pre-stage the optimum is 20–21°C.

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