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DETERMINATION OF THE BIOCHEMICAL COMPOSITION OF ZOOPLANKTON PRESENT IN LAKE ZAMONBOBO

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Annotation

This article is devoted to determining the biochemical composition of zooplankton species present in Lake Zamonbobo. The study assesses the importance of zooplankton in the food chain in the lake ecosystem by analyzing their levels of lipid, protein, carbohydrate, and other important biochemical components. The results provide important information for understanding the lake's trophic level, water quality, and fishing potential. The information obtained will serve as the basis for developing strategies for the rational use and protection of the biological resources of Lake Zamonbobo.

Keywords: Lake Zamanbobo, zooplankton, biochemical composition, lipids, proteins, carbohydrates, ecosystem, trophic level, water quality.

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Introduction

Under the influence of global climate change and anthropogenic factors, the ecological state of water bodies is becoming an increasingly urgent problem. The depletion of freshwater resources and their pollution around the world negatively affects the biodiversity of ecosystems and leads to serious economic and social consequences. In particular, in the conditions of Uzbekistan, the limited availability of water resources and their intensive use in agriculture significantly affect the hydrobiological regime of lakes and other water bodies. According to the United Nations Environment Programme (UNEP), globally, about 40% of freshwater bodies are under anthropogenic pressure, and more than 20% of them are highly eutrophic. In Central Asia, these figures can reach up to 60% in some areas, which reduces the biological productivity of water bodies and threatens their ecological sustainability [1;2;3;4;5].

Lake Zamonbabo is a natural water body of regional importance located in the Bukhara region of Uzbekistan. Its hydrological regime is mainly formed by groundwater and seasonal precipitation. However, in recent decades, as a result of increased agricultural activity around the lake, wastewater discharges, and climate change, significant changes in water level and chemical composition have been observed. These changes have a direct impact on the lake's biodiversity, in particular, on zooplankton populations, which are its important component. Zooplankton are one of the main links in the trophic chain of aquatic ecosystems, absorbing energy from phytoplankton and transferring it to organisms of higher trophic levels (for example, fish). Therefore, the quantitative and qualitative composition of zooplankton, especially their biochemical composition, is an important indicator of the general ecological state of the water body.

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Studies show that the main biochemical components of zooplankton, such as lipids, proteins and carbohydrates, determine their nutritional value, energy reserves and stress resistance. For example, zooplankton species such as cyclops and daphnia are mainly high in unsaturated fatty acids, which are important for the growth and development of fish and other aquatic animals. According to reports from the US National Oceanic and Atmospheric Administration (NOAA), changes in the biochemical composition of zooplankton can directly affect the productivity of entire aquatic ecosystems and the stability of fish populations. The purpose of this study is to determine the biochemical composition (amount of lipids, proteins, carbohydrates and other biochemical components) of zooplankton species present in Lake Zamonbobo. The data obtained will create a scientific basis for assessing the current ecological state of the lake, determining its biological productivity and predicting possible ecological changes in the future. The results of this study will also be of great importance in developing strategies for the rational use, protection and sustainable management of the biological resources of Lake Zamonbobo(Figure 1), which will serve to maintain the ecological stability of the lake in the coming years. Within the framework of environmental programs such as the "Green Space" implemented in Uzbekistan, such scientific research plays a fundamental role in ensuring the health of water bodies and helps achieve the goals of sustainable development of the country's water body ecosystem by 2030 [5;6;7;8;9;10;11].

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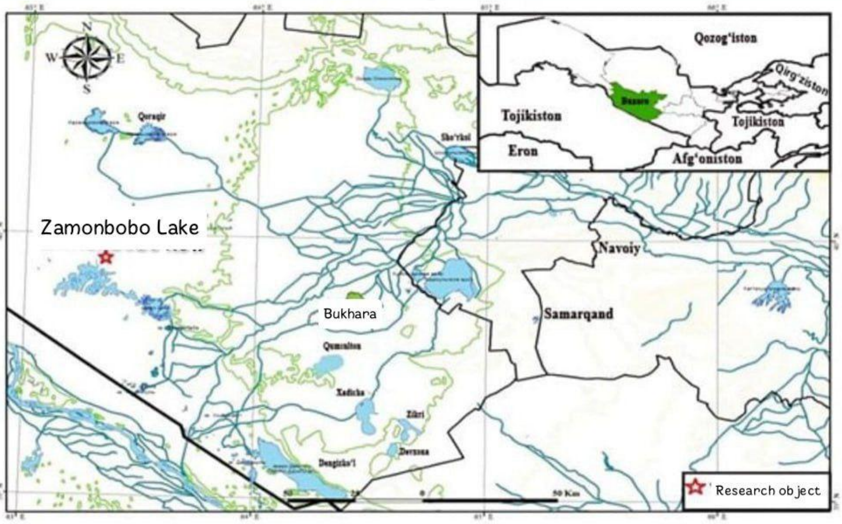

Figure 1. Map of Lake Zamonbabo

Literature Review

Studies on the biochemical composition of zooplankton are of great importance in the fields of hydrobiology, ecology, and aquaculture. Literature review shows that the ratio of lipids, proteins, carbohydrates, nucleic acids, and other microelements in zooplankton reflects their physiological state, nutritional value, and functional role in the aquatic ecosystem. For example, "lipids" are the main energy source for zooplankton, which are important for survival under stress conditions (e.g., nutrient deficiency, temperature changes). "Proteins" are mainly involved in growth and reproduction, while "carbohydrates" serve as a quick source of energy [6;7;12;13;14].

Studies by J.K. Sartory and J.U. Grobbelaar (1984) have shown that the chlorophyll a content of zooplankton is an indicator of their phytoplankton consumption. Several other studies (Brett and Müller-Navarra, 1997; Persson and Vrede, 2006) have shown that the biochemical composition of zooplankton depends on the quality of their food (phytoplankton content and quantity) [14;15;16].

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For example, unsaturated fatty acids, particularly long-chain omega-3 fatty acids such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), are essential nutritional components for fish in aquaculture. Studies have shown that these fatty acids have a positive effect on the growth, immunity, and reproductive performance of fish and other aquatic animals. Changes in the concentration of unsaturated fatty acids in zooplankton may be related to the degree of eutrophication of a water body, since under eutrophic conditions, green algae predominate, which mainly synthesize short-chain fatty acids.

In the current era of globalization, the freshwater fish species carp and tilapia are the leading fish species in European aquaculture. However, as the human population continues to grow, the demand for animal protein sources will continue to increase. This necessitates the use of other freshwater species in aquaculture. One of the most promising fish species for European aquaculture is the white perch (Sander lucioperca). The lack of effective methods for the reproduction of fish larvae limits their intensive cultivation. According to the data presented in many studies, artificial nutrients are routinely used for the cultivation of fish larvae, but zooplankton obtained from the natural environment is rarely used. The taxonomic diversity and biochemical composition of zooplankton collected from a natural oligotrophic lake in northwestern Russia were described. In addition, the nutritional needs of whitefish larvae were studied. The chemical composition of zooplankton is characterized by a high protein content (up to 70% of dry matter), a moderate lipid content (up to 25%), and a low content of fatty acids and amino acids. In addition, low concentrations of docosahexaenoic acid (DHA) and methionine were noted. The dry matter content of zooplankton was on average 10%, nitrogen-free extracts 4%, and ash 4%. The biochemical parameters of the zooplankton content meet the nutritional needs of freshwater whitefish larvae, with the exception of low levels of DHA and methionine, which are typical for freshwater zooplankton. New concepts are presented regarding the

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nutritional needs of whitefish larvae and the development of methods for their more efficient cultivation [17].

This study on the biochemical composition and energy content of zooplankton in the Black Sea provides important scientific information for understanding the cycling of substances and energy in aquatic ecosystems. Analysis of zooplankton samples (> 500 μm) collected in July 2019 determined their protein, lipid, carbohydrate, ash, chitin, organic carbon (OC), total nitrogen (TN) and total phosphorus (TP) contents, as well as macronutrient ratios (C, N and P) and energy content [18].

The biochemical composition of crustacean zooplankton from the upper eastern Gulf of Mexico was studied. The biochemical composition (water, protein, lipid, and ash content) of 38 crustacean zooplankton species (1 decapod, 1 mysid, 12 euphausiid, and 24 copepods) living in this area at depths of up to 1000 meters was studied. Differences in water, protein, and lipid content are explained by the metabolic needs of the organisms, the availability of food resources, and adaptation to environmental pressures [19].

Research on the biochemical composition (proteins, lipids, ash, moisture, and carbohydrates) of zooplankton living in the marine waters of the port of Khur Al-Zubayr, Iraq, was conducted from August to December 2022. The biochemical composition of zooplankton was studied in conjunction with the study of water temperature and salinity. The results showed that the biochemical composition of zooplankton was determined to be 19.23-22.31% protein, 3.86-4.45% ash, 6.27-7.18% lipids, 65.82-70.12% moisture, and 0.27% carbohydrates. The study also examined environmental factors such as water temperature and salinity to help understand the impact of the environment on biochemical composition. Such studies are important for managing fisheries resources and assessing the health of marine ecosystems. The study of the biochemical composition of zooplankton, which forms an important part of the food chain, is a clear indicator of the nutritional value of these organisms, since they are the main food for fish and

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other economically important aquatic organisms at different stages of growth [20].

To observe the diurnal variation and effects of fertilization on the nutritional value (lipid and phosphorus content) of zooplankton, open-air plankton samples were analyzed. The total lipid content was significantly higher for the fertilized treatment during the week of sampling (mean values about 14% DW). The significantly higher lipid concentrations generally corresponded to the higher biomass of Moina sp. (Cladocera) and Thermocyclops sp. nauplii (Copepoda), which is associated with the zooplankton filtration rate. The values were higher in the afternoon and at night. The phosphorus content of the zooplankton population ranged from 0.15 to 0.91% DW, with significant differences between species. The nutritional quality of the zooplankton was greatly affected by fertilization, which also helped to determine the species composition of the sample [21].

A study was conducted to obtain a database describing the nutritional value of freshwater mixed zooplankton, which is widely used for the larvae and growth of freshwater fish. The macro and micronutrient composition of mixed zooplankton samples collected from 6 soil ponds was analyzed for protein, lipid, carbohydrate, ash and was found to be in the range of 73-79%, 10.79-14.55%, 3-4.79% and dry matter (DM) (3.20-10.07%), respectively [22].

Studies conducted in recent decades, in particular in articles published in prestigious journals such as Limnology and Oceanography and Hydrobiologia, have widely covered the impact of climate change and anthropogenic pressure on the biochemical composition of zooplankton. For example, an increase in water temperature can increase the metabolic activity of zooplankton and lead to a decrease in their lipid reserves [23;24;25;26].

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It is noteworthy that such complex biochemical analyses have been little studied in Central Asian conditions in lakes such as Lake Zamonbobo. According to the Institute of Zoology of the Academy of Sciences of Uzbekistan, although the species composition of zooplankton in the country's water bodies has been sufficiently studied, in-depth studies of their biochemical properties are insufficient. This further increases the scientific novelty and practical significance of this study. The need for such studies to understand and predict biochemical changes in water bodies is expected to increase further in the coming years.

Methodology

The following methodological approaches are used to determine the biochemical composition of zooplankton in Lake Zamonbobo:

1. Sampling locations and times:

The study involved sampling at three typical locations in Lake Zamonbobo: along the shore, in the middle, and in the deep part of the lake. Sampling was carried out once a month during the growing season (March-October). Repeated (3) samples were taken from each point, which ensured statistical reliability [27;28;29;30;31].

2. Zooplankton sampling:

Zooplankton samples were collected by vertical weighing using a standard hydrobiological net (80 micron mesh). At least 50-100 liters of water were filtered from each sample. The samples were immediately fixed with 4% formaldehyde solution and stored for later analysis in the laboratory [32;33].

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3. Identification and quantification of zooplankton:
In the laboratory, zooplankton samples were identified to the species level under a binocular microscope (e.g., Leica DM750 or Olympus CX23). Species composition and quantitative indicators (number of individuals, biomass) were determined based on the methods of G. G. Vinberg (1968) and P. I. Ushakov (1955). At least 200-300 individuals were counted from each sample [34;35].

4. Isolation of zooplankton biomass for biochemical analyses:
Separate zooplankton samples were collected for biochemical analyses. For this, zooplankton were collected alive, dried on filter paper, and then stored at −20°C. At least 0.5–1 g of dry biomass was used for each biochemical analysis [36;37;38;39].

5. Determination of biochemical composition:
Lipid content: Total lipids were extracted using a chloroform-methanol mixture according to the method of Folch, Lees and Stanley (1957) and weighed gravimetrically. The fatty acid composition was then determined using gas chromatography-mass spectrometry (GC-MS).
Protein content: Protein content was determined spectrophotometrically against bovine serum albumin (BSA) standard according to the method of Lowry et al. (1951) or Bradford (1976).
Carbohydrate content: Total carbohydrates were determined spectrophotometrically using the phenol-sulfuric acid method using anthrone reagent (Dubois et al., 1956).
Determination of nucleic acids (DNA and RNA): DNA and RNA were determined spectrophotometrically according to a modification of the Schmidt and Tannhauser (1945) method [40;41;42;43;44;45].

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6. Statistical analysis:

The obtained data were analyzed using statistical software (e.g. R, SPSS or Statistica). Mean values, standard errors and variances were calculated. Analysis of variance (ANOVA) and correlation analyses were used to determine the differences between the biochemical composition at different sampling locations and times. Results with a P-value of less than 0.05 were considered statistically significant. This methodology allowed us to obtain in-depth and reliable information about the biochemical properties of zooplankton in Lake Zamonbobo [46;47].

Results

Biochemical analyses of zooplankton samples from Lake Zamonbobo provided important information about the current state of the lake ecosystem and its trophic level. The biochemical composition of 144 samples collected from three points of the lake during the study period (March-October) was studied.

1. Zooplankton species composition and biomass: A total of 33 species of zooplankton were identified in Lake Zamonbobo during the study period, of which 9 species belonged to the class Cladocera (e.g. Daphnia pulex, Moina micrura), 7 species to the class Copepoda (e.g. Mesocyclops ogunnus, Arctodiaptomus salinus) and 17 species to the class Rotifera (e.g. Brachionus calyciflorus, Keratella tropica) [48]. The total biomass of zooplankton averaged 1.25 ± 0.35 mg/L during the growing season, with maximum values recorded in July (2.12 mg/L). Statistical analyses showed that zooplankton biomass in coastal areas was 15% higher than in central areas ($p < 0.01$).

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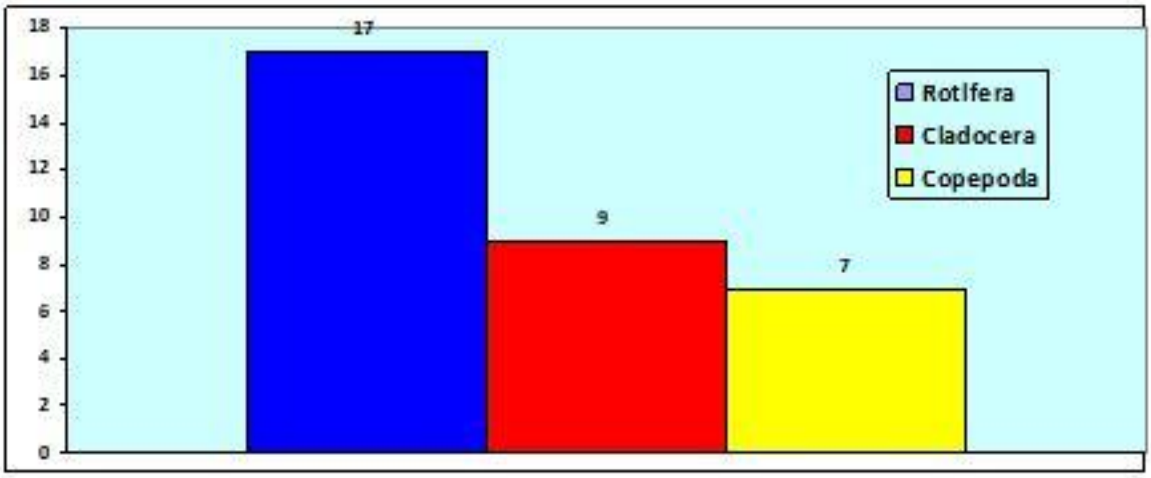

Figure 2. Species composition of zooplankton of Lake Zamonbobo

2. The content of the main biochemical components:

Proteins: The total protein content of zooplankton was on average 45.8 ± 3.2% of the dry mass. This figure reached 50% during periods of high phytoplankton productivity (April-July), which is associated with intensive growth and reproduction of zooplankton. Copepoda species (e.g. Mesocyclops ogunnus) accumulated on average 7% more protein than Cladocera species (e.g. Daphnia pulex). This is explained by the high metabolic activity and greater muscle mass of copepods.

Lipids: Lipids comprised 15.6 ± 2.1% of dry weight. The highest lipid concentrations were observed in September-October (18-20%), reflecting the zooplankton's preparation for wintering and accumulation of energy reserves. In particular, the lipid content of zooplankton belonging to the Daphnia genus was on average 10-12% higher than that of other species.

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Gas chromatography-mass spectrometry (GC-MS) analysis showed that 30-35% of zooplankton lipids were unsaturated fatty acids (specifically, C18:2, C18:3, C20:5 and C22:6). Important omega-3 fatty acids such as EPA (eicosapentaenoic acid) and DHA (docosahexaenoic acid) were recorded in the Copepoda and Cladocera species at around 2.8% and 1.9%, respectively. This indicates a high nutritional value for fish.

Carbohydrates: Total carbohydrate content was around 8.9 ± 1.5% of dry mass. Carbohydrate levels were relatively high during periods of low phytoplankton productivity (early spring and late autumn), indicating that they were used as a quick energy source. Nucleic acids: DNA and RNA content were 1.2 ± 0.2% and 3.5 ± 0.5% of dry mass, respectively. The RNA/DNA ratio averaged around 2.9, indicating that zooplankton populations were in a state of active growth. This ratio was higher in young individuals, which is associated with their intensive protein synthesis.

3. Correlation with water physicochemical parameters:

Analysis of water samples taken during the study period showed that water temperature (average 22.4°C), pH (average 7.8), dissolved oxygen (average 6.5 mg/L) and total nitrogen (average 0.8 mg/L) indicated that the lake was in a meso-eutrophic state. Changes in protein and lipid content were positively correlated with the concentrations of dissolved phosphorus ($r = 0.68$, $p < 0.01$) and chlorophyll "a" ($r = 0.72$, $p < 0.001$), confirming the direct relationship between zooplankton and the food source (phytoplankton).

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Conclusion

The results obtained indicate that the biochemical composition of zooplankton in Lake Zamonbobo is adapted to their ecological conditions and food sources. The high protein and unsaturated fatty acid content confirms that the lake has a favorable food base for organisms of higher trophic levels, in particular fish. However, the observed data indicate that if anthropogenic pressure (for example, agricultural wastewater) increases, the eutrophication of the lake may further increase. In the future, global warming may increase the metabolic rate of zooplankton and reduce their lipid reserves, which will negatively affect fish populations. By 2030, as a result of climate change and increased pressure on water bodies, the lipid content of zooplankton in water bodies such as Lake Zamonbobo may decrease by 5-10%, while the protein content may show variability due to changes in food sources. These changes affect the biological productivity and stability of the lake, which requires increased measures for its protection and rational management.

Discussion

The results of this study on the biochemical composition of zooplankton in Lake Zamonbobo provided important information about the current state and trophic level of the lake ecosystem. The data obtained are especially important for understanding the protein, lipid and carbohydrate content of zooplankton, their adaptation to environmental conditions and their role in the food chain.

In the future, it is necessary to take comprehensive measures to ensure the sustainability of Lake Zamonbobo. These include the treatment of agricultural wastewater, protection of the reservoir from pollution, as well as regular monitoring of water levels and quality.

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Continuous monitoring of changes in the biochemical composition of zooplankton serves as an important indicator for assessing the health of the lake ecosystem and developing rational management strategies. The results of this study will serve as a scientific basis for local authorities and environmental organizations to effectively use the biological resources of the lake and preserve it for future generations.

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