THE RELATION BETWEEN MICROPLANKTON ADENOSINE TRIPHOSPHATE AND CHLOROPHYLL-*A* IN THE NORTH-WESTERN PART OF THE BLACK SEA IN THE AUTUMN AND SPRING SEASONS

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ABSTRACT

The study is based on the data collected during research cruises of R/V "Professor Vodyanitskiy" in October 2016 and March-April 2017. The variations of adenosine triphosphate (ATP) and chlorophyll-*a* distributions were considered as indicators of microplankton metabolically active biomass and production capabilities respectively. Heterotrophic and photoautotrophic microplankton biomass ratios were estimated.

Keywords: *the Black Sea, microplankton, ATP, chlorophyll-a, heterotrophic-photoautotrophic index.*

INTRODUCTION

In the practice of hydrobiological study, the productivity assessment of waters under study is one of the most important goals. One of the crucial tasks when studying the ecosystem of pelagial is an assessment of the key trophic link, microplankton [2]. Depending on the type of dominant processes, the biomass can grow or shrink [8] consequently changes the feed supply for higher-order consumers. In this regard, it seems appropriate for the study of these processes to apply the complex of biochemical methods taking into account the data on chlorophyll-a and ATP ratios in microplankton. Therefore such data reflects the production-destruction processes in the community. The suggested approach is different from the classical one due to its relative simplicity of use and efficiency in data processing.

MATERIALS AND METHODS

The water samples were collected with Sea-Bird CTD-probe with a carousel water sampler. Immediately after sampling 1.5 L of water was vacuum filtered through Sartorius™ cellulose nitrate membrane filters, 0.45μm pore size, for ATP and chlorophyll-*a* analyses. A vacuum pressure of 0.2-0.4 atm was used.

Filters with precipitated samples for chlorophyll-a analyses were dried in dark place. ATP extraction was carried out using the Holm-Hansen method [3]: filters with precipitated samples were placed in centrifuge tubes, immersed in 5 ml boiling TRIS buffer (pH 7.75) and kept in a boiling water bath for 5 minutes. Extracts then were poured into plastic tubes. Dried filters for chlorophyll-*a* analyses and tubes with ATP extracts were stored in a freezer at -18°C until further processing.

For the chlorophyll-*a* analyses nitrocellulose filters were dissolved with 90% acetone and centrifuged. The extinctions of the eluates were measured in the Speeol-11 (Carl Zeiss Jena) spectrophotometer. Chl-*a* concentrations were calculated using the formula of Jeffrey and Humphrey [4].

Samples were analyzed for ATP content with chemiluminescence method by the firefly luciferase-luciferin technique. Light emission was measured in ATP luminometer 1250 (LKB).

The trophic status of water bodies was estimated by microplankton ATP and chlorophyll-a concentrations according to Karl [5].

Heterotrophic-photoautotrophic index (HPI) was calculated by the formula: (Concentration_ATP/Concentration_Chl-*a*) · 100. HPI values 10-20 means the equal ratio of the biomass of heterotrophic and photoautotrophic organisms in microplankton community. HPI >20 indicates the dominance of heterotrophs, < 10 – photoautotrophs according to criteria developed by Chiaudani and Pagnotta [1].

The previously applied comparison of microplankton triphosphate (ATP) and chlorophyll-*a* concentration enabled to obtain relatively accurate estimate of production-destruction processes in the waters of the Black Sea and the Antarctic [6], [7].

RESULTS AND DISCUSSION

In the autumn season, based on the microplankton chlorophyll-*a* concentrations in surface waters and photic zone, it can be concluded that the most productive waters (communities) were located in the eastern zone of the investigated region. The increased local chlorophyll-a concentrations were registered in the coastal waters adjacent to the eastern part of the Crimean peninsula. In the open sea waters chlorophyll-a concentrations were typical for oligo-mesotrophic waters (0.1-1 $mg \cdot m^{-3}$), while in the near-shore Crimean waters concentrations were close to values that are associated with eutrophic waters (> 1 $mg \cdot m^{-3}$) (Fig . 1).

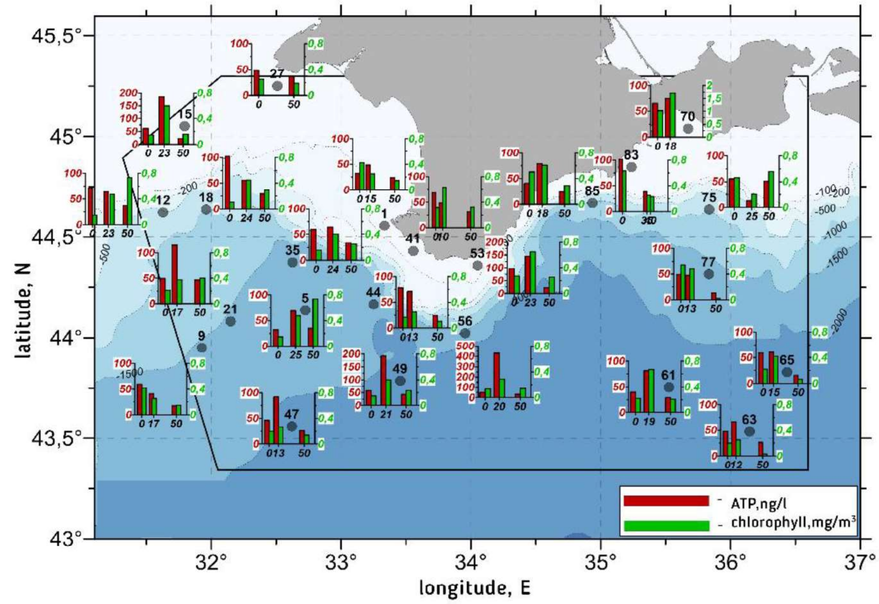

Figure 1. Distribution of chlorophyll-a and ATP concentrations of microplankton in Crimea coastal waters and deep-water northern part of the Black Sea in October 2016

Metabolically active microplankton biomass distribution was less dependent on proximity to the coast and sea depth than the microplankton production potential, according to the ATP concentrations observed. High ATP concentrations values did not have localization patterns. This distribution may be due to the fact that organic component of the food chain did not have a localized source. By ATP concentrations the study area waters can be attributed to mesotrophic (75-250 $ng \cdot l^{-1}$), with the inclusion of extensive eutrophic zones (> 250 $ng \cdot l^{-1}$).

In spring, considering the microplankton chlorophyll-*a* contents in the surface waters and photic zone, its most productive part was located in the eastern zone of the study area, and in the waters adjacent to the Kerch Strait. Green pigment concentrations in this regions reached values typical for eutrophicated waters (> 1 $mg \cdot m^{-3}$) (Fig. 2).

The distribution of ATP was slightly different from the one of chlorophyll-*a*. Values of surface ATP concentrations were, as a rule, typical of mesotrophic waters. In general, higher ATP concentrations in photic zone were identified in the peripheral regions of the investigated water area at a great distance from the Crimean peninsula.

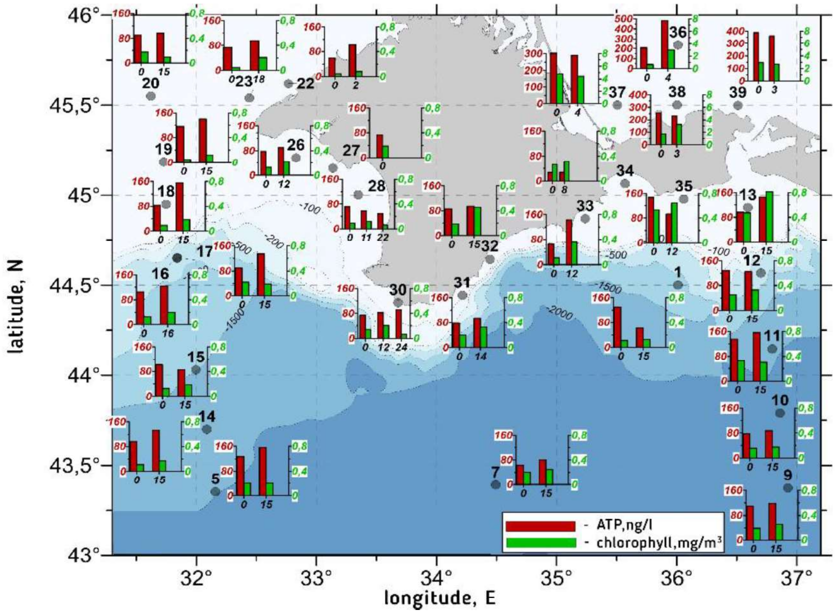

Figure 2. Distribution of chlorophyll-a and ATP concentrations of microplankton in the Crimean coastal waters and deep-water northern part of the Black Sea in April 2017

Estimation of heterotrophic - photoautotroph distribution

In the autumn season, according to identified heterotrophic–photoautotrophic index (HPI), heterotrophic forms of microplankton dominated on a greater part of the investigated region. The balance between heterotrophic and photoautotrophic microplankton was observed in waters adjacent to the Crimean peninsula and in the southwestern part of the investigated area from the east and west (Fig. 3). Heterotrophs were mainly dominant in the northwestern part of the investigated region, in the region adjacent to the shallow northwestern part of the Black Sea. Distribution trends in photic zone were similar to those in surface water layer. The exception was the significant excess of heterotrophic biomass of microplankton above the photoautotrophic biomass in the middle part of the investigated region (Fig. 4).

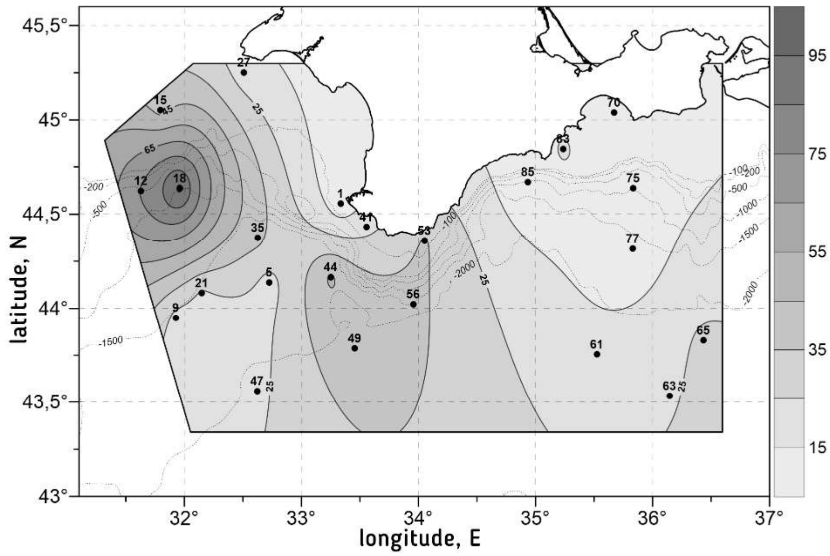

Figure 3. Distribution of microplankton HPI in the surface waters of the Crimean nearshore area and in the deep-water northern part of the Black Sea in October 2016

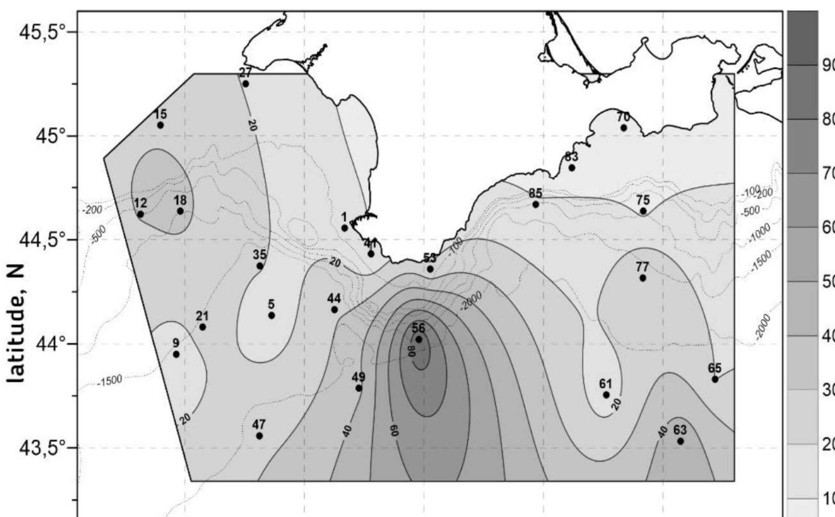

Figure 4. Distribution of HPI microplankton in the photic zone of the Crimean nearshore area and in the deep-water northern part of the Black Sea in October 2016

The distribution of microplankton HPI in spring is shown in Figures 5 and 6.

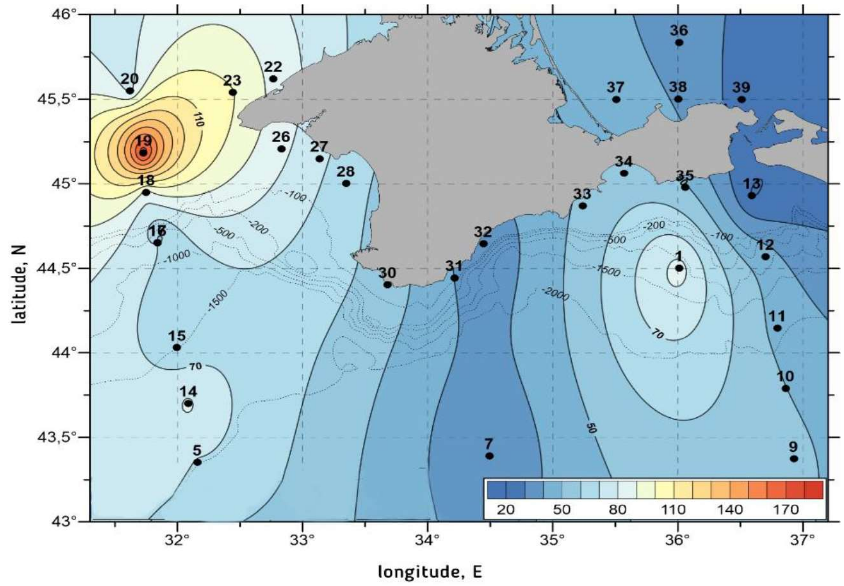

Figure 5. Distribution of microplankton HPI in the surface waters of the Crimean nearshore area and in the deep-water northern part of the Black Sea in April 2017

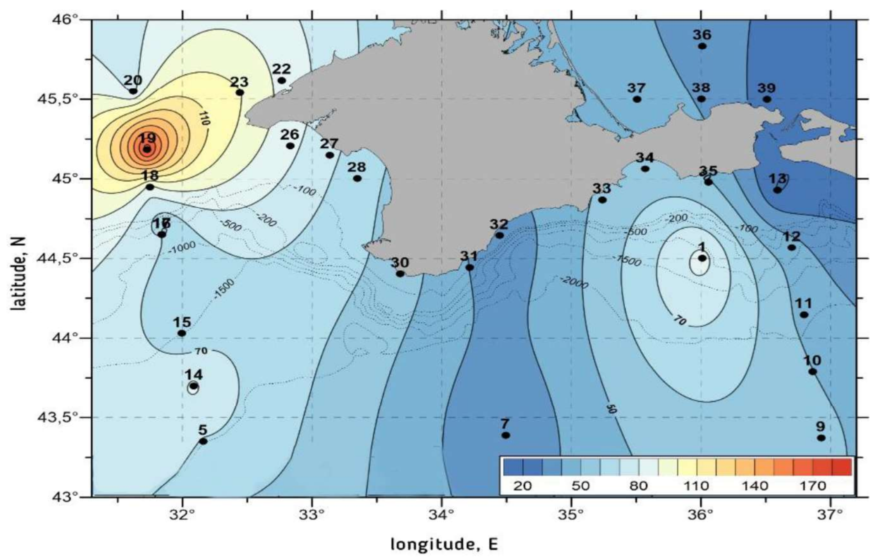

Figure 6. Distribution of HPI microplankton in the photic zone of the Crimean nearshore area and in the deep-water northern part of the Black Sea in April 2017

In surface waters, HPI values corresponding to balanced heterotrophic: autotrophic biomass ratios were registered in the waters adjacent to the Crimean peninsula in the northwestern, southern and eastern parts, particularly in the Kerch

Strait area (Fig. 5). Heterotrophic biomass was concentrated in the northwestern part of the investigated area, similar to that observed in the autumn season. Differences in the HPI distribution between surface waters and the photic layer, in general, were significantly less than in autumn. It could indicate a much smaller gradient of vertical stratification of water masses in the spring season, compared to the autumn one.

When compared the distribution of metabolically active biomass and HPI, there was a spatial similarity revealed between the high values of the total microplankton biomass and the increased inclusion of the photoautotrophic part of it.

CONCLUSIONS

The comparison of microplankton distribution in the Black Sea in spring and summer time is due primarily to the similarity of hydro-physical parameters of the aquatic environment. In general, if comparisons of distribution of metabolically active biomass and its productive part in the given seasons are made, the similarity in the values can be found in the Crimean shallow near-shore areas: the low ones, with oligo-mesotrophic value, are in waters at the western part of the Crimean peninsula and the higher ones, with meso-eutrophic value, are in the waters at the eastern shores of the Crimean peninsula, in the waters adjacent to the Kerch Strait. When comparing the distribution of metabolically active biomass and heterotrophic-photoautotrophic index, a spatial similarity in high values of the total microplankton biomass and the production capabilities rate of primary products has become evident during both seasons in the greatest part of the water area under study. Exceptions are the areas with high HPI. The largest local differences in the HPI distribution in terms of season have been identified in deep waters south of the central part of the peninsula: in autumn time HPI values indicated a far exceeding dominance of the heterotrophic forms of microplankton, whereas, in spring season, the equal values of heterotrophic and photoautotrophic microplankton have been registered in the same water area. By and large, the averaged HPI values in autumn turned out to be significantly higher than the ones in springtime. Therefore, it can be concluded that biotic factors played the dominant role in the process of forming a productive–destructive phase of succession in microplankton community in the given seasons when the similarity of hydro-physical parameters was not of great significance.

ACKNOWLEDGEMENTS

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