THE VARIABILITY OF COASTAL PHYTOPLANKTON OF THE NORTH-WESTERN BLACK SEA REGION

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Goal. The inter-annual changes in the structural organization of the coastal phytoplankton of the North-Western part of the Black Sea in 2016, 2017 and 2019 were analyzed and compared with historical data in order to assess the direction of environmental processes.

Methods. Samples taken in coastal waters were processed under a light microscope, and subsequently the complex of phytoplankton structural parameters was analyzed.

Results. Unlike most of the Black Sea, where an increase in phytoplankton biomass was observed, the phytoplankton biomass significantly decreased in the North-Western region coastal waters in 2019.

Conclusions. The data indicates that the state of phytoplankton is stable for the North-Western region of Black Sea. This confirms the current trend in the recovery ecosystem of this region that has been observed after the eutrophication of the 1970–80s.

Keywords: phytoplankton, structural organization, ecological status, Black Sea.

Introduction. The Black Sea is a recipient of the anthropogenic pressure coming from vast European and Asian river basins, atmospheric deposition and urban discharge from numerous resorts. This results in a flow of nutrients and their long-term accumulation in the deep basin. This process is reflected in nutrient stock at depth and in the bioproductivity of the basin, which leads to changes in the structure and abundance of phytoplankton. Therefore, the state of phytoplankton is one of the main indicators of ecosystem health. That is why long-term study of phytoplankton is crucial for understanding of the trends in marine communities. Long-term observations are especially important in Black Sea coastal areas, where the load on the ecosystem is the greatest.

The EMBLAS (Improving Environmental Monitoring in the Black Sea) project has shown that joint field survey is a powerful tool for studying the Black Sea ecosystem as a whole, and in particular phytoplankton. Three large Black Sea cruises and a number of regional monitoring programs were carried out during the project. First of all, these studies made it possible to identify the processes occurring in phytoplankton communities of various water areas of the Black Sea. Secondly, they allowed the generalization of data on spatial distribution of its structural organization. And finally, these studies gave us the opportunity to track inter-annual dynamics over three years. Systematic and comprehensive studies allow us not only to continue with the analysis of the occurring changes direction, but also to consider the general trends in the dynamics of phytoplankton in accordance with the Water Framework Directive (Directive 2000/60/EC) and the Marine Strategy Framework Directive (Directive 2008/56/EC). These results allowed us to successfully implement the main objective of the project: to create common standards for assessing the quality of the marine environment, which are necessary for more effective water resources management.
Significant efforts were made to unify and improve methods of phytoplankton monitoring and to find the indicators of unicellular algae that are most suitable for assessing the quality of the aquatic environment. A methodology for calculating the integrated indices for assessing the quality of the aquatic environment was developed as well. Besides, the boundaries of the water areas were delimited based on a certain uniformity of environmental conditions. Finally, rating scales and threshold volumes were developed to assess the ecological status of the water environment.

The purpose of this work is to analyze the inter-annual changes in the structural organization of the coastal phytoplankton of the North-western Black Sea region in 2016, 2017 and 2019 and to compare them with historical data with the purpose to assess the direction of ecological processes.

Materials and methods

Phytoplankton of coastal zone (1 nautical mile from coast) of the Black Sea North-western region (CNWR) was sampled in 2016, 2017 and 2019. Sampling was performed at the coastal monitoring stations (CMS) in 2016 (123 samples) and 2017 (43 samples), as well as during the cruises NPMS (National pilot monitoring stations) UA 2017 (16 samples) and NPMS UA 2019 (31 samples) (Fig. 1; Table 1). In total, 213 phytoplankton samples were collected. Studies of 2016, 2017, and 2019 were carried out in different periods and corresponded to different stages of annual phytoplankton succession. The samples were taken from June to November (CMS 2016), from March to June (CMS 2017), in August (NPMS UA August 2017), and from August to October (NPMS UA 2019) (see Table 1). Thus, when analyzing the inter-annual variability of phytoplankton of the North-Western region, various segments of annual cycles are compared, mainly related to the warm period.

Fig. 1. An overview map of sampling stations in 2016, 2017 and 2019 (North-western Black Sea region, EMBLAS project).

The sampling strategies were similar in 2016, 2017 and 2019. Quantitative phytoplankton samples were collected by vertical series at a range of depths, so that the material could be collected from main hydrophysical layers. The layers at each NPMS station were identified according to CTD-sounding, conducted prior to the phytoplankton sampling. The samples in NPMS were collected from upper mixed (0–1 m) layer, upper thermocline layer, lower thermocline layer and near-bottom layer depending on the features of the station. We used 5L Niskis bottles, attached to the CTD rosette system for sampling at each depth; 1–2 L water samples were collected during NPMS.
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Table 1. Studies of 2016, 2017, and 2019 (North-western Black Sea region, EMBLAS project)

| CRUISE  | St.         | Time               | № of samples |
|---------|-------------|--------------------|--------------|
| CMS 2016| 2, 6, 9, 12, R | 01.06.2016-30.11.2016 | 123          |
| NPMS UA August 2017 | 1-7         | 15.08.2017-20.08.2017 | 16           |
| CMS 2017| 2, 6, 9, 12, R | 10.03.2017-29.06.2017 | 43           |
| NPMS UA 2019 | 1-15, 18-20 | 01.08.2019-16.10.2019 | 35           |

Phytoplankton samples were fixed with 4% buffered formaldehyde up to the final concentration of 2% and delivered to the laboratory. The samples were slowly decanted to 30–40 ml upon sedimentation of the phytoplankton cells during 2 weeks. Samples were concentrated on board by the funnel of inverted filtration to the volume of 50–100 ml and then also fixed with 4% buffered formaldehyde up to the final concentration of 2%. Samples were kept at temperature of 5–7°C during a month before further processing. The concentrated samples were concentrated one more time, down to 10–20 cm³ by slow decantation prior to counting. Identification of species and cells' counting were carried out under a light microscope LOMO with magnifications of 600 in the drop with the volume of 0.05 ml (Naguotte chamber). Species were identified mainly using Schiller (1937), Kisselew (1950), Carmelo (1997), Cronberg and Annadotter (2006) and the taxonomic nomenclature according to the on-line data-base of World Register of Marine Species (WoRMS). The phytoplankton threshold biomass values in Ukrainian waters of the Black Sea were used (Slobodnik et al., 2021) to determine the ecological status of the waters at the study area.

Results and discussion

The total number of species found in the coastal zone of the North-Western region in the warm period in 2016, 2017 and 2019 decreased. In 2016, 247 species of phytoplankton were identified. In 2017, their number decreased to 193 species, and in 2019 — to 160 species (Fig. 2). The relative contributions of the unicellular algae dominant taxonomic groups to the species richness changed insignificantly. The contribution of Dinophyceae to the total number of species in 2016 was 26%, in 2017 — 29%, and in 2019 — 26%. The contribution of Bacillariophyceae varied from 38% in 2016 to 41% in 2017 and to 39% in 2019 (Fig. 2).

Fig. 2. Phytoplankton taxonomic composition (coastal zone of North-western Black Sea region, 2016, 2017 and 2019).
The minimum phytoplankton diversity indices of the North-Western region coastal zone were identified in 2017, the maximum — in 2019. The average Shannon index for the region (2.17) was the highest for the entire of Black Sea study area (Fig. 3).

Fig. 3. Phytoplankton diversity indices of coastal zone of North-western Black Sea region in 2016, 2017 and 2019 (H — Shannon index for biomass, 1-Lambda — Simpson index for biomass).

The highest average abundance of species dominating in the warm period in the coastal zone of the North-Western region was detected in 2016. In 2017 and 2019, the values of this indicator were lower. So, in 2016, the average abundance of Dinophyceae algae Mesoporos perforates (19119,8 $10^5$ cells/l) was more than twice as high as that of the dominant species of 2017 — the cyanobacteria Limnothrix planctonica (8869,7 $10^5$ cells/l). The average abundance of the dominant species of 2019, Merismopedia minima, was even lower (5397,2 $10^5$ cells/l). In some years, the composition of the dominant species and their taxonomic affiliation was different (Table 2).

**Table 2.** Species of phytoplankton that dominated in abundance (N, $10^5$ cells/l) in the coastal zone of North-western Black Sea region (warm period 2016, 2017 and 2019)

| Species CW-NW | Nx  |
|---------------|-----|
| **2016**      |     |
| Mesoporos perforatus (Gran) Lillick, 1937 | 19119 |
| Cyclostephanos dubius (Hustedt) Round, 1988 | 10938 |
| Limnothrix planctonica (Woloszynska) Meffert, 1988 | 8968 |
| Micracanthodinium sp. | 6322 |
| Licmophora ehrenbergii (Kützing) Grunow, 1867 | 5465 |
| Microcystis viridis (A.Braun) Lemmermann, 1903 | 4865 |
| Microcystis aeruginosa (Kützing) Kützing, 1846 | 4771 |
| **2017**      |     |
| Limnothrix planctonica (Woloszynska) Meffert, 1988 | 8869 |
| Skeletonema costatum (Greville) Cleve, 1873 | 2790 |
| Planktolyngbya limnetica (Lemmermann) Komárová-Legnerová & Cronberg, 1992 | 1993 |
| Pseudo-nitzschia delicatissima (Cleve) Heiden, 1928 | 1739 |
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| Species                                      | 2016 |
|----------------------------------------------|------|
| **Merismopedia tenuissima** Lemmermann, 1898 | 1694 |
| *Emiliania huxleyi* (Lohmann) W.W.Hay & H.P.Mohler, 1967 | 1668 |
| Dactyliosolen fragilissimus (Bergon) Hasle, 1996 | 1170 |

2019

| Species                                      | 2019 |
|----------------------------------------------|------|
| Merismopedia minima Beck, 1897               | 5397 |
| Dolichospermum spongios Klebahn, 1895        | 1925 |
| Jaaginema kisselevii (Anissimova) Anagnostidis & Komárek, 1988 | 865 |
| Jaaginema sp.                                | 788  |
| Cyanophyceae gen.sp.                         | 404  |
| Crucigenia fenestrata (Schmidle) Schmidle, 1900 | 319 |
| Stephanodiscus hantzschii Grunow, 1880       | 219  |

* — species common to different years are highlighted.

The highest average biomass values for the CNWR in 2016 were found for *Pseudopedinella sp.* (Dictyochophyceae) and *Wolosynskia pascheri* (Dinophyceae) — 8876 and 7745 mg/m³ respectively. In 2017, Bacillariophyceae *Pseudo-solenia calcar-avis* and *Dactyliosolen fragilissimus* dominated. In 2019, the largest biomass was formed by *Ceratium tripos* (Dinophyceae) and *Nodularia spumigena* (Cyanophyceae). The average biomass of *C. tripos* in 2019 (1039 mg/m³) was for an order of magnitude lower than the biomass of *P. calcar-avis* in 2017 (16626 mg/m³) (Table 3).

Table 3. Species of phytoplankton that dominated in biomass (B, mg/m³) in the coastal zone of North-western Black Sea region (warm period 2016, 2017 and 2019)

| Species                                      | Bx   |
|----------------------------------------------|------|
| **2016**                                     |      |
| *Pseudopedinella sp.*                       | 8876 |
| Wolosynskia pascheri (Suchlandt) Stosch, 1973 | 7745 |
| Ulnaria sp.                                  | 5495 |
| **Pseudosolenia calcar-avis** (Schultze) B.G.Sundström, 1986 | 5296 |
| Cerataulina pelagica (Cleve) Hendey, 1937    | 1826 |
| Thalassiosira gravida Cleve, 1896             | 1258 |
| Pterosperma cristatum Schiller, 1925         | 1195 |
| **2017**                                     |      |
| *Pseudosolenia calcar-avis* (Schultze) B.G.Sundström, 1986 | 16626 |
| Dactyliosolen fragilissimus (Bergon) Hasle, 1996 | 7071 |
| Durinska dybowskii (Wolosynskia) S.Carty, 2014 | 5178 |
| Tovellia coronata (Wolosynskia) Moestrup, Lindberg & Daugbjerg, 2005 | 1302 |
| Cerataulina pelagica (Cleve) Hendey, 1937    | 630  |
| Oblea rotunda (Lebour) Balech ex Sournia, 1973 | 506  |
| Skeletonema costatum (Greville) Cleve, 1873  | 502  |
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| 2019 |
|---------------------------------|
| Ceratium tripos (O.F. Müller) Nitzsch, 1817 | 1039 |
| Nodularia spumigena Mertens ex Bornet & Flahault, 1886 | 582 |
| Dolichospermum spiroides Klebahn, 1895 | 456 |
| Akashiwo sanguinea (K. Hirasaka) G. Hansen, Ø. Moestrup | 309 |
| Protoperidinium divergens (Ehrenberg, 1841) Balech, 1974 | 247 |
| Pseudosolenia calcar-avis (Schultze) B. G. Sundström, 1986 | 156 |
| Dinophysis acuta Ehrenberg, 1841 | 154 |

* — species common to different years are highlighted.

The inter-annual changes in the relative contributions of taxonomic classes to phytoplankton biomass of the CNWKR were associated with redistribution of Bacillariophyceae (average contribution 62%) and Dinophyceae (average contribution 26%). The exception was 2019, when the contribution of Cyanophyceae has increased. The contribution of Dinophyceae to biomass has decreased to the minimum values in 2017. The contribution of Bacillariophyceae has increased proportionally. In 2019, the contribution of Dinophyceae has increased again (Fig. 4).

The maximum average abundance of phytoplankton communities in the coastal zone of the North-Western region was revealed in the warm period of 2016. This indicator decreased during the warm period of 2017 and 2019 (Fig. 5, a). The average phytoplankton biomass of the CNWKR during the study period was 3012.4 mg/m³ and varied from 26.3 to 43555.6 mg/m³ (Fig. 5, b). The average abundance and biomass of the CNWR phytoplankton communities in 2016 and 2017 did not differ significantly (Fig. 5). The average biomass of 2019 was significantly lower than these indicators in 2016 and 2017. The annual average diversity indices and phytoplankton biomass of the CNWKR varied in the opposite way. The average Shannon index varied from 1.96 (2017) to 2.38 (2019). Thus, a decrease in biomass in 2019 was accompanied by an increase.

Fig. 4. Contributions of taxonomic classes to the phytoplankton abundance (N %) and biomass (B %) (coastal zone of North-western Black Sea region, 2016, 2017, 2019).
in the values of the diversity indices (see Fig. 3; Fig. 5). The average concentration of chlorophyll "a" was the lowest in 2016 (1.5 μg/L). In contrast to biomass, the concentration of chlorophyll "a" significantly increased in 2019 reaching 3.1 μg/L (Fig. 6).

A peculiarity of the CNWR phytoplankton development is an increase in biomass in late winter (or early spring) and late summer (or early autumn). The biomass decreases from November to January (Krivenko, Parkhomenko, 2010; North-western part of Black Sea, 2006). This determines the impact of different monitoring periods of 2016, 2017 and 2019 on data comparability. However, a good knowledge of the CNWR allows us to trace the long-term dynamics of biomass. The biomass has increased from 0.7 to 18 g/m³ between 1973–1980 (North-western part of Black Sea, 2006). Since 1981, the phytoplankton biomass began to gradually decrease to 15 g/m³ (1980–1990) and 5 g/m³ (1990–1993).

Fig. 5. Average abundance (N, a) and biomass (B, b) of the phytoplankton communities (coastal zone of North-western Black Sea region, 2016, 2017 and 2019).

Fig. 6. Average chlorophyll a concentration (coastal zone of North-western Black Sea region, 2016, 2017 and 2019).
In 1999, the autumn biomass of phytoplankton of the Odessa Gulf was close to 3 g/m³ (North-western part of Black Sea, 2006). This corresponds to the values obtained in 2016, 2017 and 2019. An analysis of 1477 samples obtained by Institute of Marine Biology, National Academy of Sciences in the CNWR allows to track changes in biomass in the warm period of 2000–2012. The biomass dynamics had an undulating character and varied from 440.7 mg/m³ in 2001 to 5915.1 mg/m³ in 2010, when climatic factors caused the development of *Nodularia spumigena*. The average biomass for 2000—2012 (2195.6 mg/m³) is close to the average values during the study period (Fig. 7).

Wave-like changes are also typical for distribution of the Dinophyceae and Bacillariophyceae biomass. Both the largest (91%, 2017) and the smallest (33%, 2019) contributions of Bacillariophyceae were identified during the study period. The average contribution of Bacillariophyceae was 60% (in 2000-1012) and 62% (in 2016–2019) (Fig. 8).

Assessment of the ecological status based on biomass threshold values showed that most of the coastal stations of CNWR belong to the GES category (Good Ecological Status). In some years, areas of increased river (eg. Dnieper and Dniester) inflow have been mainly assigned as having NotGES (Fig. 9). This was due to mass summer-autumn development of Cyanophyceae, which reached bloom concentrations, observed in the coastal waters of the Danube-Dnieper interfluve both in 2017 and 2019. (The maps were colored in accordance with the principle of correspondence of the status of the district to the lowest revealed value of the indicator.)
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Thus, an analysis of the biomass and taxonomic structure of the CNWR phytoplankton in 2016, 2017, and 2019 did not reveal significant changes compared to the period of 2000-2012; and in 2019 the biomass significantly decreased. This confirms the current trend in the recovery of CNWR ecosystem that occur after the eutrophication of the 70–80s.

Conclusions

Studies of coastal phytoplankton communities conducted in the coastal zone of the Black Sea North-western region (CNWR) in 2016, 2017 and 2019 showed that 83% of species belonged to 4 taxonomic classes: Bacillariophyceae, Dinophyceae, Cyanophyceae and Chlorophyceae. The maximum contribution to the phytoplankton biodiversity in 2016, 2017 and 2019 was made by Bacillariophyceae. The largest biomass in 2016 was formed by the Pseudopedinella sp. In 2017, Pseudosolenia calcarea-avis dominated. In 2019, the maximum biomass of Ceratium tripos was detected. Unlike most of the Black Sea region, where an increase in phytoplankton biomass was observed in 2019, the phytoplankton biomass of North-Western region coastal waters significantly decreased. The average annual values of biomass and diversity indices in North-Western region coastal waters changed in the opposite way: the increase in biomass was associated with a decrease in diversity indices. The taxonomic structure of phytoplankton was determined by the variability in contributions of Bacillariophyceae and Dinophyceae to the biomass. The average contribution of these classes to biomass was respectively: 62 and 26 % for the study period of three years. Rapid increase in the contribution of Bacillariophyceae to the biomass in 2017 and its decrease in 2019 were revealed for the CNWR. Assessment of the ecological status based on biomass threshold values showed that most of the stations of CNWR belong to the GES category (Good Ecological Status). In a number of regions, a decrease in the ecological status to NotGES was revealed in some years. This was due to mass summer-autumn development of Cyanophyceae, which reached water bloom concentrations, observed in the coastal waters of the Danube-Dnieper interfluve both in 2017 and in 2019. This is consistent with historical data that demonstrate the wave-like nature of inter-annual changes in phytoplankton. The average annual values of the indicators could differ by orders of magnitude. In addition to the high variability of phytoplankton, which quickly responds to changes in environmental conditions, the factors that determined the inter-annual differences in 2016–2019 were different sampling times, as well as climatic conditions in individual years. Taking this into account, it can be concluded that the state of phytoplankton communities is stable for North-Western region of Black Sea. In general, the variability of phytoplankton indicators corresponded to the indicators of the period that began as a result of the end of eutrophication in the 1980s and 1990s.

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МІНЛИВІСТЬ ПРИБЕРЕЖНОГО ФІТОПЛАНКТОНУ ПІВНІЧНО-ЗАХІДНОГО РЕГІОНУ ЧОРНОГО МОРА

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Мета. З метою оцінки спрямованості екологічних процесів було проаналізовано міжрічні зміни структурної організації прибережного фітопланктону Північно-Західного частини Чорного моря в 2016, 2017 та 2019 роках, та проведено їх порівняння з історичними даними. Методи. Проби, відібрані в прибережних водах, оброблялись під світловим мікроскопом з наступним аналізом комплексу структурних показників фітопланктону. Результати. На відміну від більшої частини Чорноморського регіону, де спостерігалося збільшення біомаси фітопланктону, біомаса фітопланктону прибережних вод Північно-Західного регіону у 2019 році значно зменшилася. Висновки. Зроблено висновок, що стан угруповань фітопланктону Північно-Західного регіону в 2019 році значно зменшилася. Це підтверджує сучасну тенденцію відновлення екосистеми CNWR, що виникла після евтрофікації 70–80-х років. Ключові слова: фітопланктон, структурна організація, екологічний статус, Чорне море.