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ANNOTATED LIST OF EPIPHYTIC DIATOMS FROM MACROPHYTES OF THE AQUATIC-ROCK COMPLEX KAZANTIP CAPE (THE CRIMEA, SEA OF AZOV)

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The paper is the first one to summarize original data on the diatom species composition for 129 samples of 14 macrophyte species from the upper sublittoral of the aquatic-rock complex Kazantip Cape (the Sea of Azov). The sampling was carried out in 2022–2023 at depths of 0.1–1.0 m at a temperature of +3.7…+29.0 °C and water salinity of 13.6–15.6 $g \cdot L^{-1}$. The annotated list covers 98 Bacillariophyta species and intraspecific taxa from 3 classes, 21 orders, 27 families, and 45 genera. Data on their sizes, occurrence, ecological and phytogeographic characteristics, abundance, and biomass are provided. High species diversity of diatoms was noted for the class Bacillariophyceae (72 taxa) and for three genera: *Nitzschia* (12 species), *Navicula* (11), and *Licmophora* (6). The highest number of diatom species was recorded on thalli of a green alga *Ulva linza* (61 species), red alga *Carradoriella denudata* (45), and brown alga *Ericaria crinita* (40), as well as in epiphyton of the seagrass *Zostera marina* (37) and its association with *Zannichellia palustris* subsp. *major* (35). The lowest number of species was revealed for epiphyton of a red alga *Pyropia leucosticta* (9). A benthic diatom *Berkeleya scopulorum* was found in the Sea of Azov for the first time. In epiphytic communities, benthic (83%), marine (51%), and cosmopolitan species (35%) predominate. In total, 30 colonial species were identified. Out of them, the following diatoms occur year-round: *Achnanthes brevipes*, *Berkelya rutilans*, *Grammatophora marina*, *Melosira moniliformis*, *Navicula ramosissima*, *Rhoicosphenia marina*, and *Tabularia tabulata*. Cosmopolitan species *Cocconeis scutellum*, *Gr. marina*, *Licmophora abbreviata*, *Rh. marina*, and *T. tabulata* dominate in abundance. We recorded 11 species capable of causing water blooms and 45 serving as indicators of saprobity of water organic pollution.

Keywords: diatoms, macrophytes, epiphyton, aquatic-rock complex Kazantip Cape, Sea of Azov

The coastal zone of the Kazantip Cape has long attracted the attention of algologists as an area with high biodiversity and minimum anthropogenic impact. Since the mid-20th century, aquatic macroflora has been periodically studied there, and annotated lists of macroalgae, aquatic flowering plants, and benthic cyanobacteria have been compiled and updated [Belich et al., 2019; Gromov, 1999; Maslov, 2004; Sadogurskaya, 2001; Sadogurskaya et al., 2006]. The surveys of this zone are important given the conservation status of terrestrial sites and coastal waters within the aquatic-rock complex Kazantip Cape [Litvinyuk, 2016].

In 2005–2010, the species composition and quantitative characteristics of benthic microalgae from various ecotopes of the Crimean coast of the Sea of Azov, including the Kazantip Nature Reserve and the cape water area, were studied [Bondarenko, 2017; Bondarenko, Ryabushko, 2010; Bondarenko et al., 2018; Ryabushko, Bondarenko, 2016]. In the shallow Sea of Azov, the interaction between benthic and pelagic zones is especially noticeable; planktonic and benthic microalgae represent a single ecological and floristic species complex there. At the initial stage of the survey, in order to clarify the species list, a search of literature sources was carried out (summaries, species guidelines, reference books, serials, abstracts, algal database, and also our own data) for the inventory and revision of the taxonomic composition of planktonic and benthic microalgae of the Sea of Azov as a whole [Ryabushko, Bondarenko, 2011]. As a result, a checklist of 1,060 taxa comprising 9 divisions was compiled, with a predominance of Bacillariophyta (538 taxa). In plankton, 621 taxa were revealed, and in benthos, 370; 69 taxa were common to the two biotopes. From the presented data, a lag in the study of microphytobenthos is evident.

Diatoms are known to be the key group of marine microphytobenthos in terms of their diversity and abundance. These microalgae feature high primary productivity and make a significant contribution to the formation of biodiversity in the benthic community. Along with other benthic organisms, diatoms are sensitive to environmental changes: fluctuations in water temperature, its salinity, and pollution with toxicants. These microalgae can be used as indicators of the quality of their habitat when assessing the environmental situation, *inter alia* in protected areas [Barinova et al., 2019b; Ryabushko, 2013].

In seas, diatoms inhabit all natural and artificial substrates. However, according to many authors, macrophytes are a preferred substrate for colonization by microalgae [Bondarenko, 2017; Korotkevich, 1960; Lee et al., 1975; Nikolaev, 1970; Ryabushko, 2013; Shiroyan, 2022; Siqueiros-Beltrones, Hernández-Almeida, 2006]. Therefore, in 2022–2023, we expanded our research on diatoms in the epiphyton of aquatic vegetation in the coastal waters of the Kazantip Cape. The study of the epiphyton of 11 macroalgal species allowed revealing 97 taxa of Bacillariophyta; out of those, 51 species were recorded in the area for the first time. Moreover, dominant species were identified, the qualitative and quantitative composition was analyzed, and also the structure of diatom communities depending on the season and the species of colonized macroalgae was surveyed [Bondarenko et al., 2024].

When we finished processing the material from the one-year studies, information on diatoms of the epiphyton of other macrophytes was added to the previously published results [Bondarenko et al., 2024]. Also, comprehensive data on the ecological, phytogeographic, and quantitative characteristics of diatoms were accumulated. Generalized information of this kind is valuable for understanding the ecology of species and the environmental effects on the formation of microphytobenthic communities. Moreover, such material is helpful when compiling databases covering information on microalgal habitats, ranges, morphological traits, *etc.* In addition, revealing the trends in the qualitative and quantitative distribution of diatoms in benthic macrophyte communities, especially in coastal ecosystems, is required to establish general patterns of interaction between diatoms, macrophytes, and their habitat.

The aim of this work is to summarize the original results of studying the species composition and autecology of diatoms of macrophyte epiphyton of the coastal waters of Kazantip Cape in order to compile an annotated list covering data on diatom dimensions, their occurrence, ecological and phytogeographic characteristics, abundance, and biomass.

MATERIAL AND METHODS

The material for the summarization was 129 samples of epiphyton of 12 macroalgal species:

- brown algae – *Ericaria crinita* (Duby) Molinari et Guiry, 2020, *Gongolaria barbata* (Stackhouse) Kuntze, 1891, and *Cladosiphon mediterraneus* Kützing, 1843;
- red algae – *Callithamnion corymbosum* (Smith) Lyngbye, 1819, *Ceramium arborescens* J. Agardh, 1894, *Pyropia leucosticta* (Thuret) Neefus et J. Brodie, 2011, and *Carradoriella denudata* (Dillwyn) Savoie et G. W. Saunders, 2018;
- green algae – *Bryopsis hypnoides* J. V. Lamouroux, 1809, *Cladophora liniformis* Kützing, 1849, *Cl. sericea* (Hudson) Kützing, 1843, *Ulva intestinalis* Linnaeus, 1753, and *U. linza* Linnaeus, 1753;

and also, the seagrass *Zostera marina* Linnaeus, 1753 alone and in its association with another flowering plant, *Zannichellia palustris* subsp. *major* (Hartman) Ooststroom et Reichgelt, 1964.

Sampling was carried out from October 2022 to September 2023 in the upper sublittoral zone of four bays of the Kazantip Cape (the Sea of Azov): the Shirokaya and Kunushkay (both within the Kazantip Nature Reserve area), Russkaya, and Tatarskaya (Fig. 1).

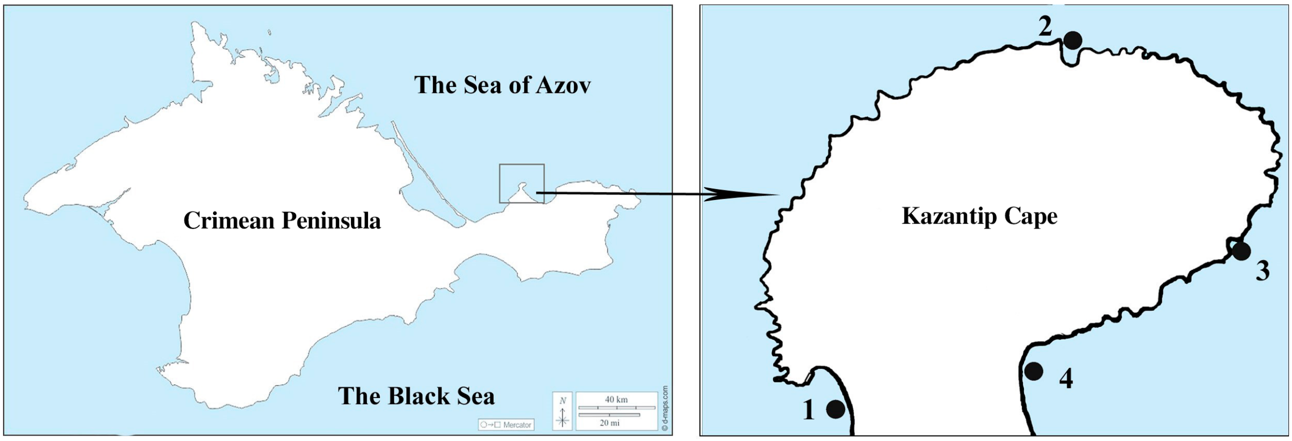

Fig. 1. Sites of the algological sampling in the Kazantip Cape area: 1, the Russkaya Bay; 2, the Shirokaya Bay; 3, the Kunushkay Bay; 4, the Tatarskaya Bay

Sampling was carried out at depths of 0.1–1.0 m and at temperatures from +3.7 to +29.0 °C. Water salinity varied from 13.6 to 15.6 g·L^{-1} (Table 1).

Table 1. Hydrological conditions of epiphyton sampling in various months of 2022–2023 in the Kazantip Cape bays (the Sea of Azov)

Parameter	2022		2023								
	13.10	29.11	31.01	28.02	29.03	25.04	31.05	22.06	27.07	29.08	25.09
Depth, m	0.5	0.2	0.5	0.2	0.5	0.2	0.5	0.1–1	0.1–1	0.1	0.1–0.5
Temperature, °C	+17.0	+10.0	+3.7	+4.9	+8.1	+14.0	+19.2	+26.2	+27.3	+29.0	+23.9
Salinity, g·L^{-1}	15.0	15.6	15.0	15.6	15.1	15.0	14.9	14.9	13.6	13.6	15.0

In the study area, samples occasionally contained various macroalgal species floating freely at the water's surface; their thalli were tightly intertwined. These samples were used only for qualitative analysis of the diatom composition.

For taxonomic identification, aqueous and permanent preparations were used. Diatom valves were cleaned by a standard acid-based technique [The Diatoms of the USSR, 1974]. After washing, frustules were embedded in Naphrax LTD resin (refractive index was 1.74). Living cells and frustules were studied under an Axioskop 40 light microscope (Carl Zeiss, Germany) at magnifications of 10 × 20 and 10 × 40, with a ×100 objective, and with immersion oil added (Carl Zeiss) (refractive index $n_e = 1.518$). Also, frustules were examined under a Hitachi SU3500 scanning electron microscope (Japan).

When analyzing the taxonomic structure of diatoms, we mostly used the system developed by F. Round *et al.* [1990], with amendments and additions of new data [AlgaeBase, 2025; Ryabushko, 2013; Ryabushko, Begun, 2015, 2016]. The taxonomic affiliation was determined based on literature sources [Diatomovyi analiz, 1950; Guslyakov et al., 1992; Kuylenstierna, 1989, 1990; Ryabushko, 2013; Ryabushko, Begun, 2015, 2016; Smith, 1853; Witkowski et al., 2000]. The ecological and phytogeographic characteristics of diatoms were analyzed using a number of publications [AlgaeBase, 2025; Barinova et al., 2019a, b; Bondarenko et al., 2024; Guslyakov et al., 1992; Ryabushko, 2013; Ryabushko, Begun, 2016; Sládeček, 1986]. Quantitative characteristics of diatoms (abundance and biomass) were determined according to the method [Ryabushko, 2013].

RESULTS AND DISCUSSION

As a result of the study of the diatom biodiversity in the epiphyton of coastal macrophyte thickets in the Kazantip Cape water area in 2022–2023, an annotated list was compiled; it includes 98 species and intraspecific taxa from 3 classes, 21 orders, 27 families, and 45 genera and provides the description of their taxonomic affiliation. For the first time, autecology data are presented for each taxon, including phytogeographic and quantitative characteristics (see Supplement: https://marine-biology.ru/mbj/article/view/507).

Two genera, *Nitzschia* and *Navicula*, are characterized by high taxonomic diversity (12 and 11 species accordingly). *Licmophora* is less represented (6 species). This genus is followed by *Amphora*, *Diploneis*, *Lyrella*, and *Melosira* (4 species each). Frequently encountered species were identified: *Achnanthes brevipes*, *Berkeleya rutilans*, *Cocconeis scutellum*, *Cylindrotheca closterium*, *Grammatophora marina*, *Licmophora abbreviata*, *Navicula cancellata*, *N. ramosissima*, *Nitzschia sigma*, *N. sigmoidea*, *Rhoicosphenia marina*, *Tabularia tabulata*, and *Trachyneis aspera*. Several species were found singly: *Amphora laevis*, *Grammatophora angulosa*, *Licmophora rostrata*, *Lyrella lyra*, *L. lyroides*, *Navicula cancellata* var. *gregoryi*, *Navicula dumontiae*, *Nitzschia dissipata*, *N. inconspicua*, *Planothidium delicatulum*, and *Petroneis humerosa*.

For the first time, *Berkeleya scopulorum* (Fig. 2F) was recorded in the Sea of Azov. It was registered in the Russkaya Bay of the Kazantip Cape in August, in epiphyton of *Z. marina*.

In epiphyton communities, 30 colonial species were noted. Out of those, 7 occurred year-round: *A. brevipes*, *B. rutilans*, *Gr. marina*, *Melosira moniliformis*, *N. ramosissima*, *Rh. marina*, and *T. tabulata*. In various months, 5 cosmopolitan species dominated: *C. scutellum*, *Gr. marina*, *L. abbreviata*, *Rh. marina*, and *T. tabulata* (Fig. 2A–E).

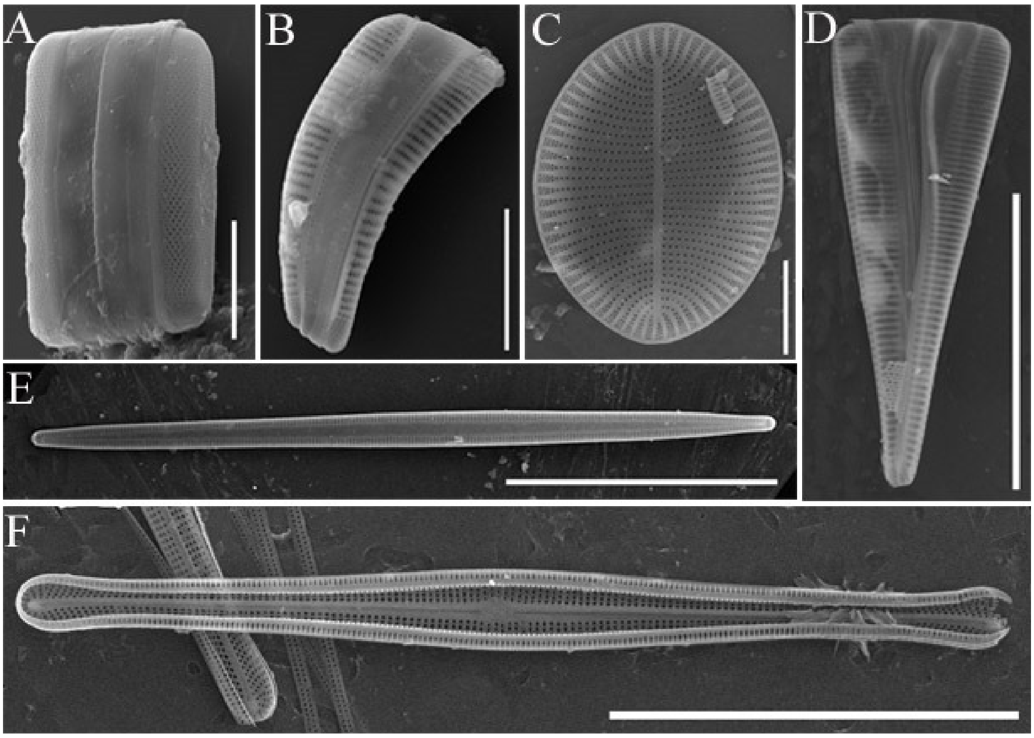

Fig. 2. Benthic diatom species in epiphyton of macrophytes (a scanning electron microscope). A, *Grammatophora marina*; B, *Rhoicosphenia marina*; C, *Cocconeis scutellum*; D, *Licmophora abbreviata*; E, *Tabularia tabulata*; F, *Berkeleya scopulorum*. Scale bars are 10 μm (A–C), 20 μm (F), 30 μm (D), and 50 μm (E)

In coastal mycrophyte thickets, 11 bloom-forming species were recorded: *B. rutilans*, *C. closterium*, *Gr. marina*, *L. abbreviata*, *Licmophora flabellata*, *Parlibellus delognei*, *Striatella unipunctata*, *Tabularia fasciculata*, and *T. tabulata*, as well as planktonic ones *Skeletonema costatum* and *Sk. subsalsum*. Also, *Halamphora coffeiformis* was registered: a potentially toxic species. As a rule, it occurs in epiphyton communities throughout the year; however, its abundance does not reach high values.

The highest number of diatom species was recorded on thalli of macroalgae: *U. linza* (61), *C. denudata* (45), and *E. crinita* (40), and also in the epiphyton of the seagrass *Z. marina* alone (37) and in its association with *Zannichellia palustris* subsp. *major* (35). The lowest number of taxa was noted in the epiphyton of a red alga *P. leucosticta* (9). Interestingly, the epiphyton of *U. linza* featured a high diatom species richness in August and September only (in other months, thalli of *Ulva* were colonized much less frequently). The minimum values of diatom abundance and biomass were often registered in its epiphyton (see Supplement: https://marine-biology.ru/mbj/article/view/507). Apparently, thalli of *U. linza*, which grew by the end of summer, served as a kind of "nets" on which diatoms settled, reaching the peak of their development by the end of summer.

Ecological analysis of the qualitative composition of the epiphyton community of diatoms showed the predominance of benthic forms (83%), marine ones (51%) and, to a lesser extent, brackish-marine ones (36%). Proportions of brackish, freshwater-brackish, and freshwater forms were small (5, 5, and 3%, respectively).

Saprobic indices were established for 45 diatom species. According to the system [Barinova et al., 2019a; Sládeček, 1986] and to our data [Barinova et al., 2019b; Bondarenko et al., 2024], 32 species out of these 45 are indicators of moderate organic water pollution (water quality class 3), and the leading role is played by a group of beta-mesosaprobes. We revealed 10 species inhabiting oligotrophic water bodies, and 6 species typical of eutrophic waters. The occurrence of oligosaprobes in communities, despite the constant influx of organic matter, including anthropogenic one, is likely to indicate the ability of coastal ecosystems to neutralize excess organic matter *via* self-purification processes, in which microalgae also participate.

In benthic communities, the proportion of marine species increased, and the proportion of freshwater species significantly decreased. Shifts in species composition and size structure were also reported for diatoms from upper sediments sampled in 1998 and 2019 at 32 stations in various areas of the Sea of Azov [Kovaleva, Gavrilova, 2021]. The authors highlight a noticeable drop in the number of frustules of the size fraction 5–10 μm and an elevated number of large-celled species. Those were related to the taxonomic restructuring of the complex of dominant diatoms, among which medium- and large-sized taxa, chiefly marine and brackish-marine forms, began to prevail in terms of their frequency of occurrence and abundance.

Taking into account our and literature data, it can be concluded that the revealed restructuring of the algal flora reflects a trend of increasing salinity of the Sea of Azov in recent decades [Berdnikov, 2023; Kovaleva, Gavrilova, 2021].

As established, by the end of the biological summer (August and September), in the coastal area of the Kazantip Cape, there is a noticeable rise in the quantitative characteristics of some algal bloom-forming species [Bondarenko et al., 2024]. One of the risk factors for the occurrence of this phenomenon is the summer-associated elevated recreational load on the Sea of Azov coast in the Crimea and, as a consequence, the influx of organic matter with untreated sewage and household wastewater into the coastal zone [Ryabushko, 2003]. On the one hand, various organic matter, being mineralized, becomes a source of biogenic elements available for microalgae and stimulating a boost in their abundance. On the other hand, organic matter causes an outbreak of heterotrophs, which include some diatoms (representatives of *Amphora*, *Cocconeis*, *Licmophora*, *Navicula*, *Striatella*, and *Tabularia*). Notably, algal bloom is a regular problem of the Sea of Azov, since humus and dissolved organic matter constantly enter this initially eutrophic water body along with the runoff of the Don, Kuban and numerous small rivers, and the concentration of biogenic elements does not reach analytical zero [Aleksandrova et al., 1998; Gidrometeorologiya i gidrokhimiya morei SSSR, 1991]. In shallow water, algal bloom is caused by the mass development of both planktonic and benthic microalgae. Therefore, information on the species composition and quantitative characteristics of diatoms of macrophyte epiphyton is an important component in a comprehensive assessment of the state of coastal ecosystems.

Conclusion. The original data on the study of epiphytic diatom biodiversity from macrophytes of the upper sublittoral zone of the aquatic-rock complex Kazantip Cape in 2022–2023 are summarized; information on diatom ecology (relation of species to habitat, water salinity, and saprobity), phytogeography, abundance, and biomass is given. An annotated list was compiled. It includes 98 Bacillariophyta species and intraspecific taxa from 3 classes, 21 orders, 27 families, and 45 genera, and provides the description of their taxonomic affiliation. For the first time, autecology data for each taxon are presented, including phytogeographic and quantitative characteristics. In the surveyed algal flora, a benthic diatom *Berkeleya scopulorum* was registered, new for the Sea of Azov; its scanning electron micrograph is presented. The highest taxonomic diversity was recorded for three genera: *Nitzschia* (12 species), *Navicula* (11), and *Licmophora* (6). In epiphyton communities, 30 colonial species were found; out of those, 7 occur year-round: *Achnanthes brevipes*, *Berkeleya rutilans*, *Grammatophora marina*, *Melosira moniliformis*, *Navicula ramosissima*, *Rhoicosphenia marina*, and *Tabularia tabulata*. In terms of abundance, the cosmopolitan species are dominant ones: *Cocconeis scutellum*, *Gr. marina*, *Licmophora abbreviata*, *Rh. marina*, and *T. tabulata*. The bioindicator properties of the identified saprobic diatoms can be used in further monitoring of the dynamics of anthropogenic load in the Sea of Azov.

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АННОТИРОВАННЫЙ СПИСОК ДИАТОМОВЫХ ВОДОРОСЛЕЙ ЭПИФИТОНА МАКРОФИТОВ АКВАЛЬНО-СКАЛЬНОГО КОМПЛЕКСА «МЫС КАЗАНТИП» (КРЫМ, АЗОВСКОЕ МОРЕ)

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Впервые представлены обобщённые оригинальные данные о видовом составе диатомовых водорослей в 129 пробах эпифитона 14 видов макрофитов из верхней сублиторали аквально-скального комплекса «Мыс Казантип» Азовского моря. Пробы отобраны в 2022–2023 гг. на глубинах 0,1–1,0 м при температуре от +3,7 до +29,0 °С и солёности воды от 13,6 до 15,6 $г{\cdot}л^{-1}$. Аннотированный список включает 98 видов и внутривидовых таксонов отдела Bacillariophyta из 3 классов, 21 порядка, 27 семейств и 45 родов; указаны их размеры, встречаемость, экологические и фитогеографические характеристики, численность и биомасса. Высокое видовое разнообразие диатомовых водорослей отмечено для класса Bacillariophyceae (72 таксона) и родов *Nitzschia* (12 видов), *Navicula* (11) и *Licmophora* (6). В эпифитоне зелёной водоросли *Ulva linza* обнаружен 61 вид, красной *Carradoriella denudata* — 45, бурой *Ericaria crinita* — 40, морской травы *Zostera marina* — 37, её ассоциации с *Zannichellia palustris* subsp. *major* — 35. Наименьшее число видов найдено в эпифитоне красной водоросли *Pyropia leucosticta* — 9. Обнаружен новый для Азовского моря бентосный вид *Berkeleya scopulorum*. В сообществах эпифитона преобладают бентосные (83 %), морские (51 %) и космополитные виды (35 %) диатомовых водорослей. Выявлено 30 колониальных видов, из них круглогодично присутствуют *Achnanthes brevipes*, *Berkeleya rutilans*, *Grammatophora marina*, *Melosira moniliformis*, *Navicula ramosissima*, *Rhoicosphenia marina* и *Tabularia tabulata*. По численности доминируют космополиты *Cocconeis scutellum*, *Gr. marina*, *Licmophora abbreviata*, *Rh. marina* и *T. tabulata*. Выявлено 11 видов, вызывающих «цветение» воды, и 45 видов — индикаторов сапробности органического загрязнения вод.

Ключевые слова: диатомовые водоросли, макрофиты, эпифитон, аквально-скальный комплекс «Мыс Казантип», Азовское море