





Phytoplankton Dynamics in an Oligo-mesotrophic Environment along the Montenegrin Coast (South-East Adriatic Sea)

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Abstract

The dynamics of the phytoplankton community were studied along the Montenegrin coast, in the mesotrophic area of Boka Kotorska Bay and the oligotrophic coastal open sea area.

This two year study investigates differences in dynamics of phytoplankton assemblages and abundances in the Bay area, which is highly impacted from the land, and the open coastal part of the Montenegrin coast, which is highly influenced by Levantine water masses but less affected by anthropogenic influence."

Regarding phytoplankton diversity, the microplankton species detected are those that prefer areas rich in inorganic nutrients, such as *Chaetoceros affinis*, *Dactyliosolen fragilissimus*, *Leptocylindrus danicus*, *Pseudo-nitzschia* spp., and *Thalassionema nitzschioides*. These species used both new and regenerated nutrients which regenerated at each grazing level of the microbial loop and are thus made available to the primary producers.

The potentially toxic *Pseudo-nitzschia* spp. reached an abundance of 10^5 cells/L.

Among the potentially toxic dinoflagellates, *Dinophysis acuminata*, *D. acuta*, *D. caudata*, *D. fortii*, *D. tripos*, *Gonyaulax spinifera*, *Lingulodinium polyedra*, *Phalacroma rotundatum* were recorded (the abundance reached values up to 10^2 cells/L).

The detected presence of several potentially toxic and toxic phytoplankton species warrants the need to raise awareness for the necessity of continuous monitoring activities and preventive measures.

Introduction

The southern part of the Adriatic Sea is under the influence of two coastal currents, one flowing from the north along the western coast and the other from the south along the eastern coast (Gačić et al., 1996). The current along the western (Italian) side comes from the northern Adriatic, one of the most productive areas of the Mediterranean, while the current along the eastern (Balkan) coast comes from the central Mediterranean (the Ionian Sea), the most oligotrophic area worldwide (Yacobi et al., 1995). The southern Adriatic, due to the circulation of these currents, is characterized as an

extremely oligotrophic area (Orlić et al., 1992). Although the southern Adriatic has been considered highly oligotrophic, the coastal part is under increased anthropogenic impact and eutrophication (Viličić, 1983), which has led to pressure on algal populations, and such changes can result in periodically intense growth of microalgal species.

The distribution of the phytoplankton community and its dynamics in the open southern Adriatic Sea during two winter-spring seasons (2016 and 2017) were investigated by Jasprica et al. (2022). The authors emphasized a pronounced inflow of Levantine Intermediate Water into the Adriatic as the elementary

environmental factor that influenced the phytoplankton community.

Batistić et al., (2012; 2019) noticed phytoplankton blooms in the open South Adriatic during winter, where its intensity and occurrence depend on extreme winter conditions such as low temperatures, strong winds, heavy rainfalls. This was accompanied by appearance of Eastern Mediterranean Transient (EMT) that caused massive intrusion of nutrients enriched Atlantic Water (AW) into Adriatic Sea upper layers (50–200 m).

This can lead to higher interannual variability in phytoplankton abundance (Viličić, 1989; Ljubimir et al., 2017).

Great contributions to the distribution of the phytoplankton community in the southern Adriatic Sea were given by Viličić et al. (2011) across the Albanian shelf and continental slope, and there is a lack of information on the fine distribution of plankton.

The Montenegrin coast consists of the semi enclosed Boka Kotorska Bay and the open coastal part of the sea. Boka Kotorska Bay is a relatively small but very important transitional system that looks like fjords with a different biological composition from other adjacent ecosystems (Drakulović et al., 2017; Bosak et al., 2009; Sarno et al., 1993). The Boka Kotorska Bay is under higher influence from the land in comparison with the open coastal part.

Marine phytoplankton are a very diverse group of organisms (De Vargas et al., 2015; Naem, 2012) and the key component that determines and influences the functioning of pelagic ecosystems (Irwin & Finkel, 2018). These organisms are very sensitive to changes in their environment, and therefore, they are useful indicators of water quality (Brettum & Andersen, 2005). Therefore, it is very important to better understand the relationship between the variability in phytoplankton diversity and its effect on ecosystem processes (Otero et al., 2020).

Currently, we are witnessing increased human influence along the coast. To better assess the biological quality of the ecosystem, information on the phytoplankton assemblages is essential (Toming & Jaanus, 2007; Jaanus et al., 2009).

Investigation and comparison of the distribution and phytoplankton composition along the Montenegrin coast in two different areas throughout two years of research is the main focus of this article. Our scope is to determine whether there is a significant difference in the composition of phytoplankton communities in two ecologically different areas with focus on potentially toxic and toxic species. Bay area is a semi-closed system with lower discharge of water masses and higher pressure from the coast, while the open sea area is more influenced by higher streaming of oligotrophic water masses.

The data available in this research will be of interest, especially for the area outside of the bay, considering the lack of investigation and information when comparing that area with the Bay area.

Materials and Methods

Study Area

The Montenegrin coast, as part of the southern Adriatic, is located between Albania and Croatia and extends approximately 90 km in a straight line. The entire length of the coast, including small islands, is approximately 300 km (PPPNzMD, 2007). The Montenegrin coast consists of a semienclosed bay (area of 87.3 km²), Boka Kotorska, and an open coastal part of the sea (Figure 1).

Three parts form the Boka Kotorska bay: Kotor and Morinj -Risan Bays belong to the nethermost part, and they are connected with Tivat Bay by the Strait Verige, while Strait Kumbor connects central Tivat Bay with the outermost Herceg Novi Bay (Magaš, 2002). In nethermost area, there are numerous submarine springs and streams, and in a very short period, high discharge reaches two rivers, Sopot and Ljuta (Fig. 1) (Bellafiore et al., 2011). The dynamics of the water column are highly influenced by the higher input of submarine springs and streams (Milanović, 2007). The area of Boka Kotorska Bay is highly influenced by pressure from the land rather than the open sea, which also presents better dynamics and exchange of water masses.

Sampling Methods and Phytoplankton Analysis

Sampling of marine water was performed monthly from January 2019 to December 2020 (sampling was not performed in May or June 2019 and in June 2020) at twelve locations, eight in the Bay area and four in the coastal open part of the Montenegrin coast. Water samples for physico-chemical and phytoplankton analysis were taken from two depths: surface and bottom layers. The bottom depths of the investigated locations are as follows: BK-1 (14 m), BK-2 (28 m), BK-3 (16 m), BK-4 (24 m), BK-5 (38 m), BK-6 (42 m), BK-7 (10 m), OS-1 (74 m), OS-2 (30 m), OS-3 (35 m), OS-4 (10 m), and OS-5 (12 m). Samples were taken with 5 L Niskin bottles. Two parameters (temperature and salinity) were analyzed directly in situ using a universal meter (Multiline P4; WTW). On a Shimadzu UV/VIS 1900 spectrophotometer, nitrates and nitrites were determined according to Grasshoff, 1983 and phosphates and silicates were determined according to Korelff, 1983.

According to Winkler (1888), by using fixation with an appropriate reagent and titration, the oxygen concentration was measured. For the sampling of phytoplankton, 5-L Niskin bottles were used, and samples were dropped into 250 ml plastic bottles. Preservation of samples was performed with 4% neutralized formalin solution. Samples were transported in a refrigerator in the dark at a temperature of approximately 8±3°C. Sampling was performed according to ISO5667-9 (1992-Water quality



Figure 1. Area of investigation on which sampling of phytoplankton communities were performed in the Bay area (IBMK-BK-1, Kotor-BK-2, Risan-BK-3, Sveta nedjelja-BK-4, Tivat-BK-5, Herceg Novi-BK-6, Igalo-BK-7) and the area outside of the bay (Mamula-OS-1, Budva-OS-2, Bar-OS-3, Ulcinj-OS-4, Bojana-OS-5) in periods of investigation from 2019-2020 (QGIS⁴⁸) (<http://www.qgis.org>)

– Sampling Part 9: Guidance on sampling from marine waters).

Phytoplankton cells were counted using a Leica DMI4000 B inverted microscope (Heerbrugg, Switzerland) in accordance with the method of Utermöhl (1958) (MEST EN 15204: 2014-Water quality – Guidance standard on the enumeration of phytoplankton using inverted microscopy). In the laboratory, samples were settled in sedimentation chambers of 25 ml, and after a period of sedimentation of 24 h, we started with the processing of the determination. Enumeration was performed at the following magnifications: 200 ×, 400 ×, and 630 ×. Half of the bottom chamber (taxa larger than 30 μm) was crossed at a magnification of 200 ×, while at the same magnification, two transects were used for counting abundant microplankton (>20 μm). Small phytoplankton (nanophytoplankton 2–20 μm) were counted at magnifications of 400 × and 630 × using 10 randomly selected fields.

Determination of phytoplankton species was performed using an appropriate key (Cupp, 1943; Hustedt, 1930; 1962a; 1962b; H. Peragallo & M. Peragallo, 1965; Dodge, 1985; Schiller, 1933; 1937; Sournia, 1986; Stein, 1883). Nonidentified microalgae were classified into taxonomic categories: nanoflagellates, small dinoflagellates, small coccolithophores and chrysophytes.

Statistical Analysis

The programs (Microsoft Excel, Grafer 7), Statistica 7.0., Primer 6.0, and Ocean Data View software (version number ODV 5.6.5, 2023) were used for statistical analysis of the data.

For analyses of phytoplankton diversity, the Shannon–Wiener diversity index, $H' = -\sum p_i \log(p_i)$, (Shannon and Wiener, 1949) and Margalef's evenness index, $d = (S-1)/\log(N)$, were used. Statistica 7.0 and Box Whisker were used for the presentation of diversity indices, both Margalef's index (Margalef, 1958) and Shannon–Wiener's index (Shannon, 1948).

The Kruskal–Wallis test (K-W test) was used to analyze differences in the abundances of phytoplankton among the sites and months.

Spearman's rank correlation analysis was carried out between the environmental and biological data. Furthermore, Principal Component Analyses (PCA) was used to evaluate the linear and cause-effect relations between the abundance of the phytoplankton groups (as active variables) and the environmental parameters (as supporting variables).

Results

Environmental Data

During the research period, at surface and deep water, a variation in temperature in terms of maximum and minimum values was noticed. During 2019 and 2020, maximal temperatures were recorded in August (27.39°C and 28.2°C). After that, the temperature slowly decreased. Minimum of temperature were observed in November and December 2019 (3.6°C) and January 2020 (8.8°C) (Tables 1 and Table 2).

Regarding the highest values of salinity concentration, lower temporal variations were observed, while for minimum of salinity, the situation differed, and expressed variations were noted. The highest salinity concentrations were measured in August and September 2019 (38.9 psu) and in July 2020 (38.7 psu). The lowest salinity concentrations due to higher precipitation were recorded in November 2019 (3.4 psu) and March 2020 (3.8 psu) (Table 1 and Table 2).

Variations in nitrate concentration were not pronounced, and the highest concentrations were in December 2019 and 2020 (13.71 $\mu\text{mol/l}$ and 16.6 $\mu\text{mol/l}$), while the lowest were in November 2019 (0.01 $\mu\text{mol/l}$) and May 2020 (0.1 $\mu\text{mol/l}$).

The temporal distribution of nitrate concentrations reveals very low maximal values between January and July 2019 and from April to May 2020. This pattern is likely influenced by two factors: the impact of the river and the increase in vertical mixing processes. The presence of the river and its inflow on one side and the rise in vertical mixing processes on the other hand play a role in diffusing bottom-regenerated nutrients throughout the water column, leading to higher nitrate values during the fall-winter period (November-December 2019, 2020). During these months, the water column shows increased nitrate concentrations, as river inflow and vertical mixing processes bring up nutrients from the bottom layers to the surface.

The maximum concentrations of phosphate were 0.69 $\mu\text{mol/l}$ in July 2020 and 0.87 $\mu\text{mol/l}$ in December 2019.

Throughout most of the year, minimum phosphate concentrations were observed to be <0.01 $\mu\text{mol/l}$ at all locations. However, in December 2019 and July 2020, there were slightly higher minimal concentrations of phosphate present. These two periods stand out as exceptions when compared to the rest of the year, during which phosphate levels remain consistently close to zero.

Maximum concentrations of silicate were noticed in August in both investigated years (184.08 and 56.3 $\mu\text{mol/l}$). The lowest concentrations were recorded in July 2019 (0.13 $\mu\text{mol/l}$) and February 2020 (0.07 $\mu\text{mol/l}$).

Variations in the highest and lowest concentrations of dissolved oxygen (DO) were slight, with maximum values in February 2019 at surface and 2020 at deeper layer (10.1 mg/l and 9.6 mg/l) and

minimum values in November 2019 and March 2020 at surface layer (4.1 mg/l and 4.6 mg/l) (Table 1 and Table 2). Dissolved oxygen shows comparable concentrations at all locations, as well as good general oxygenation of the waters.

Additionally, the distribution of nutrients (NO_3^- ; PO_4^{3-} , SiO_4^-) along the area of Boka Kotorska Bay (Figure 2) was presented, as this area is under higher human pressure than the coastal open part. The results generally showed higher values of nutrients in the inner part of the bay, as expected. Nitrates at both depths (surface and bottom) showed slight increases in values in the inner part of the bay, and then the concentration decreased toward the outer part of Herceg Novi Bay (Figure 2 a, d). Silicates showed a similar distribution of concentrations as nitrates, while phosphates were the highest in the area of Tivat Bay and then values slightly decreased toward the outer part of Herceg Novi Bay (Figure 2 b, c, e, f). In the coastal part of open sea concentrations of nutrients (NO_3^- ; PO_4^{3-} , SiO_4^-) were the highest at location OS-5 what is expected as that location is under influence of Bojana river (Table 3).

The nutrients show specific characteristics with generally higher surface concentrations, with the exception of PO_4^{3-} concentrations, which demonstrate increased values in depth. At the bottom, nutrient concentrations show either uniformity (NO_3^- and SiO_4^-) or an increase with depth (PO_4^{3-}). These variations in nutrient distribution provide valuable insights into the dynamics of the aquatic ecosystem.

Phytoplankton Abundance

The distribution of phytoplankton abundance varied among locations situated in the bay area (especially refers on locations BK-1 and BK-2 in the Kotor Bay) and those in the open coastal area. Higher values were recorded in Boka Kotorska Bay than in the open coastal area. Values for silicoflagellates were low and were excluded from presentation.

The abundances of phytoplankton in Boka Kotorska Bay ranged from 10^4 to 10^6 cells/l. The highest recorded values at all locations were up to 10^5 cells/l, except at location Kotor (BK-2), where abundance was higher and reached values up to 10^6 cells/l.

The maximum abundance of phytoplankton during the investigation period in the area of Boka Kotorska Bay was noticed in Kotor Bay, location Kotor (BK-2), (2.89×10^6 cells/l) in May 2020. The lowest value of total phytoplankton was at location Sveta Nedjelja in September 2020 (BK-4) (6.89×10^4 cells/l). This maximum value mainly consisted of coccolithophores, which reached values of 1.95×10^6 cells/l. Except at the location at Kotor, where the maximal value consisted mostly of coccolithophores, diatoms contributed highly to the phytoplankton abundance at other locations. In the area outside of the bay of Montenegro, the value of total phytoplankton was generally lower. In the area outside of the bay, the highest abundance of

Table 1. Values of physico-chemical parameters in the area of Boka Kotorska Bay and area outside of the bay in 2019

		2020											
		Jan	Feb	Mar	Apr	May	Jul	Aug	Sep	Oct	Nov	Dec	
Temp.(°C)	Max	14.7	15.9	14.7	17.5	19.8	26	28.2	26.6	22.5	21	18.2	
	Min	8.8	11.4	10	13.9	15.9	16.2	19.1	15.4	12.7	14.2	11.4	
	AVG	12.92	14.18	13.19	15.57	18.08	21.32	23.31	23.3	19.31	19.11	15.64	
	SD	1.63	1.12	1.07	0.87	1.36	3.25	2.69	2.93	2.76	1.91	1.95	
Sal. (psu)	Max	35.2	38.6	37.5	37.3	37.8	38.7	38.5	38.3	38.5	37.7	37.6	
	Min	19.5	15	3.8	19.7	20	18.7	14.4	30.2	4.5	10.6	6.1	
	AVG	31.9	34.1	28.61	34.6	34.72	36.43	35.69	37.1	29.94	33.2	30.04	
	SD	4.16	6.04	11.83	4.01	5.13	4.03	5.64	1.76	12.06	8.32	12.15	
DO (mg/l)	Max	8.3	9.0	8.6	9.2	9.6	8.7	8.6	8.1	8.9	8.7	8.9	
	Min	5.1	5.80	4.6	7.5	6.6	7.1	4.7	5.2	6.7	5.4	5.2	
	AVG	6.65	7.71	7.44	8.49	8.19	7.93	7.63	7.12	7.64	6.89	6.68	
	SD	0.97	0.83	1.19	0.47	0.74	0.32	0.82	0.74	0.62	0.79	1.12	
NO ₃ (µmol/l)	Max	10.5	9.1	13.10	2.2	3.5	11.5	12.8	6.37	16.45	11.54	16.6	
	Min	0.3	0.9	0.6	0.2	0.1	0.38	0.13	0.14	0.45	0.21	0.54	
	AVG	2.41	2.84	4.24	0.84	1.15	1.39	1.42	0.93	4.78	1.88	4	
	SD	2.11	2.27	4.03	0.46	0.8	2.29	2.9	1.36	4.95	3.1	5.16	
NO ₂ (µmol/l)	Max	0.88	0.58	0.56	0.25	0.75	0.21	0.27	0.1	0.7	1.09	0.69	
	Min	0.004	0.09	0.098	0.01	0.05	0.017	0.007	0.009	0	0.02	0.04	
	AVG	0.15	0.23	0.27	0.08	0.21	0.05	0.06	0.04	0.17	0.17	0.18	
	SD	0.18	0.14	0.14	0.07	0.23	0.04	0.05	0.03	0.15	0.26	0.14	
PO ₄ ³⁻ (µmol/l)	Max	0.175	0.52	0.68	0.13	0.42	0.69	0.11	0.132	0.25	0.14	0.25	
	Min	0	0	0	0	0	0.003	0	0	0.03	0	0	
	AVG	0.05	0.06	0.19	0.04	0.08	0.11	0.05	0.04	0.09	0.03	0.11	
	SD	0.06	0.12	0.2	0.03	0.13	0.14	0.03	0.03	0.04	0.04	0.07	
SiO ₄ (µmol/l)	Max	22.3	16.8	20.1	6.41	18.3	15.5	56.3	10.7	26.19	20.2	28.28	
	Min	1.14	0.07	1.64	0.81	0.67	0.33	0.53	0.46	1.93	1.31	0.55	
	AVG	4.1	3.53	7.45	2.59	5.03	2.17	5.5	2.53	8.23	5.26	6.01	
	SD	4.47	4.21	5.68	1.41	4.86	3.21	12.44	2.55	7.04	5.27	7.76	

Max-maximum; Min-minimum; AVG-average, SD-standard deviation.

Temp. (°C) Temperature; Sal. (psu), Salinity; DO (mg/l), Dissolved Oxygen concentration; NO₃⁻ (µmol/l)

Nitrate concentration; NO₂⁻ (µmol/l), Nitrite concentration; PO₄³⁻ (µmol/l), Phosphate concentration; SiO₄⁻ (µmol/l), Silicate concentration

phytoplankton was recorded at location Mamula in January 2019 (OS-1) (2.59×10^5 cells/l), and the minimum abundance was recorded at location Budva in September 2020 (OS-2) (6.99×10^4 cells/l) (Figure 3 and Figure 4). As in the bay area, diatoms are a phytoplankton group that contributed highly to the phytoplankton abundance in the area outside of the bay.

During two years of research, spatial variation in the maximum and minimum values of phytoplankton and phytoplankton groups was noticed in both areas, the bay area and the open sea area and along two depths (surface and deep water) In the bay area, values of phytoplankton were higher in the much closer part of that area, in Kotor Bay, while in the more open part, Tivat and Herceg Novi Bay values were lower. Additionally, values in Boka Kotorska Bay were higher than values in the open coastal part (Figure 3 and Figure 4). The Kruskal–Wallis test showed that there was a statistically significant difference in the abundances of total phytoplankton (TP), diatoms, dinoflagellates and coccolithophores among the investigated sites (K-W test 6.285, 6.284, 6.284, 6.283; $P < 0.01$) (Figure 3). In the open coastal part of the sea, the Kruskal–Wallis test showed statistically significant differences in the abundances of TP and diatoms among the investigated sites (K-W test 4.209; $P < 0.01$) (Figure 4).

Additionally, the varying distributions of TP and dominant phytoplankton group-diatoms in the different parts of Boka Kotorska Bay were presented. The results showed a pronounced increase in phytoplankton abundances in the inner part and a decrease in abundances in the outer part (Figure 5). In the area outside of the bay, there was a slight increase in abundance in the northern part, then a slow decrease in the central part, and again a slight increase in abundance in the southern part (Figure 6).

Regarding the temporal distribution of phytoplankton, in the bay area, the value of phytoplankton showed a mostly uniform distribution during the two years, with the exception of 2020, when the highest abundance was noted in April and May. In the coastal open part of the sea, during two years of investigations, the abundance of phytoplankton showed slight variations. In 2019, the highest values were observed during winter and early spring, and in 2020, the highest values were observed in late winter and spring. Moreover, in 2020, there was a higher abundance of phytoplankton during the autumn period (Figure 7 and Figure 8). The Kruskal–Wallis test showed that there was a statistically significant difference during the investigated period in the Bay Area related to the abundances of TP, diatoms, dinoflagellates and coccolithophores (K-W test 20.285, 20.284, 20.283;

Table 2. Values of physico-chemical parameters in the area of Boka Kotorska Bay and area outside of the bay in 202

		2020											
		Jan	Feb	Mar	Apr	May	Jul	Aug	Sep	Oct	Nov	Dec	
Temp. (°C)	Max	14.7	15.9	14.7	17.5	19.8	26	28.2	26.6	22.5	21	18.2	
	Min	8.8	11.4	10	13.9	15.9	16.2	19.1	15.4	12.7	14.2	11.4	
	AVG	12.92	14.18	13.19	15.57	18.08	21.32	23.31	23.3	19.31	19.11	15.64	
	SD	1.63	1.12	1.07	0.87	1.36	3.25	2.69	2.93	2.76	1.91	1.95	
Sal. (psu)	Max	35.2	38.6	37.5	37.3	37.8	38.7	38.5	38.3	38.5	37.7	37.6	
	Min	19.5	15	3.8	19.7	20	18.7	14.4	30.2	4.5	10.6	6.1	
	AVG	31.9	34.1	28.61	34.6	34.72	36.43	35.69	37.1	29.94	33.2	30.04	
	SD	4.16	6.04	11.83	4.01	5.13	4.03	5.64	1.76	12.06	8.32	12.15	
DO (mg/l)	Max	8.3	9.0	8.6	9.2	9.6	8.7	8.6	8.1	8.9	8.7	8.9	
	Min	5.1	5.80	4.6	7.5	6.6	7.1	4.7	5.2	6.7	5.4	5.2	
	AVG	6.65	7.71	7.44	8.49	8.19	7.93	7.63	7.12	7.64	6.89	6.68	
	SD	0.97	0.83	1.19	0.47	0.74	0.32	0.82	0.74	0.62	0.79	1.12	
NO ₃ ⁻ (µmol/l)	Max	10.5	9.1	13.10	2.2	3.5	11.5	12.8	6.37	16.45	11.54	16.6	
	Min	0.3	0.9	0.6	0.2	0.1	0.38	0.13	0.14	0.45	0.21	0.54	
	AVG	2.41	2.84	4.24	0.84	1.15	1.39	1.42	0.93	4.78	1.88	4	
	SD	2.11	2.27	4.03	0.46	0.8	2.29	2.9	1.36	4.95	3.1	5.16	
NO ₂ ⁻ (µmol/l)	Max	0.88	0.58	0.56	0.25	0.75	0.21	0.27	0.1	0.7	1.09	0.69	
	Min	0.004	0.09	0.098	0.01	0.05	0.017	0.007	0.009	0	0.02	0.04	
	AVG	0.15	0.23	0.27	0.08	0.21	0.05	0.06	0.04	0.17	0.17	0.18	
	SD	0.18	0.14	0.14	0.07	0.23	0.04	0.05	0.03	0.15	0.26	0.14	
PO ₄ ³⁻ (µmol/l)	Max	0.175	0.52	0.68	0.13	0.42	0.69	0.11	0.132	0.25	0.14	0.25	
	Min	0	0	0	0	0	0.003	0	0	0.03	0	0	
	AVG	0.05	0.06	0.19	0.04	0.08	0.11	0.05	0.04	0.09	0.03	0.11	
	SD	0.06	0.12	0.2	0.03	0.13	0.14	0.03	0.03	0.04	0.04	0.07	
SiO ₄ (µmol/l)	Max	22.3	16.8	20.1	6.41	18.3	15.5	56.3	10.7	26.19	20.2	28.28	
	Min	1.14	0.07	1.64	0.81	0.67	0.33	0.53	0.46	1.93	1.31	0.55	
	AVG	4.1	3.53	7.45	2.59	5.03	2.17	5.5	2.53	8.23	5.26	6.01	
	SD	4.47	4.21	5.68	1.41	4.86	3.21	12.44	2.55	7.04	5.27	7.76	

Max-maximum; Min-minimum; AVG - average, SD - standard deviation Temp. (°C) - Temperature; Sal. (psu) - Salinity; DO (mg/l) - Dissolved Oxygen Concentration; NO₃⁻ (µmol/l) - Nitrate concentration; NO₂⁻ (µmol/l) Nitrite concentration; PO₄³⁻ (µmol/l) - Phosphate concentration; SiO₄ (µmol/l) - Silicate concentration

P<0.01) (Figure 7). In the open coastal part of the sea, the Kruskal–Wallis test showed statistically significant differences during the investigated period related to TP, diatoms, dinoflagellates and coccolithophores abundance (K-W test 20.209; P<0.01) (Figure 8).

Taxa Abundances and Composition

From January 2019 to December 2020 in Boka Kotorska Bay, 128 taxa were recorded, of which 63 taxa belonged to diatoms, 55 taxa to dinoflagellates, 6 taxa to coccolithophores, 2 taxa to silicoflagellates and 2 taxa to chlorophytes. Species from the diatoms group that were present with a frequency of more than 30% were *Asterionellopsis glacialis* (35.71%), *Bacteriastrium hyalinum* (57.14%), *Chaetoceros affinis* (33.33%), *Chaetoceros* spp. (92.86%), *Cocconeis scutellum* (38.09%), *Dactyliosolen fragilissimus* (40.48%), *Diploneis bombus* (41.18%), *Guinardia striata* (50.0%), *Hemiaulus hauckii* (40.48%), *Leptocylindrus danicus* (38.09%), *Lioloma pacificum* (35.71%), *Navicula* spp. (83.33%), *Nitzschia longissima* (45.24%), *Pleurosigma elongatum* (64.29%), *Proboscia alata* (92.86%), *Pseudo-nitzschia* spp. (100%), *Thalassionema frauenfeldii* (33.33%), and *T. nitzschoides* (95.24%). From the dinoflagellates group with a frequency of more than 30%, nine taxa were

observed: *Gonyaulax* spp. (90.48%), *Gyrodinium fusiforme* (55.88%), *Prorocentrum cordatum* (47.62%), *P. micans* (88.09%), *Scrippsiella* spp. (38.09%), *Tripos furca* (30.95%), *T. fusus* (32.35%), *T. kofoidii* (33.33%), and *T. muelleri* (45.24%). Coccolithophores observed more frequently were *Calyptrosphaera oblonga* (64.29%), *Rhabdosphaera tignifer* (54.76%), and *Syracosphaera pulchra* (64.29%) (Table 4).

Four taxa from the diatoms group were present with a frequency of more than 90%: *Chaetoceros* spp., *Pseudo-nitzschia* spp., *Proboscia alata*, and *Thalassionema nitzschoides*. *Pseudo-nitzschia* spp. Were the most represented in diatom abundance (the highest abundance was 2.79 × 10⁵ cells/l), followed by *Chaetoceros* spp. and *Proboscia alata* (1.13 × 10⁵ cells/l and 2.57 × 10⁴ cells/l), respectively. The highest value of the frequent diatom *Thalassionema nitzschoides* was 1.79 × 10⁴ cells/l.

Three taxa from the dinoflagellates group were present with a frequency of more than 50%: *Gonyaulax* spp., *Gyrodinium fusiforme*, and *Prorocentrum micans*. Of these, species from the genus *Gonyaulax* (maximal value of 2.86 × 10⁴ cells/l) were the most represented, followed by the second most represented and abundant species *Prorocentrum micans*, which reached the highest abundance of 1.29 × 10⁴ cells/l (Table 4).

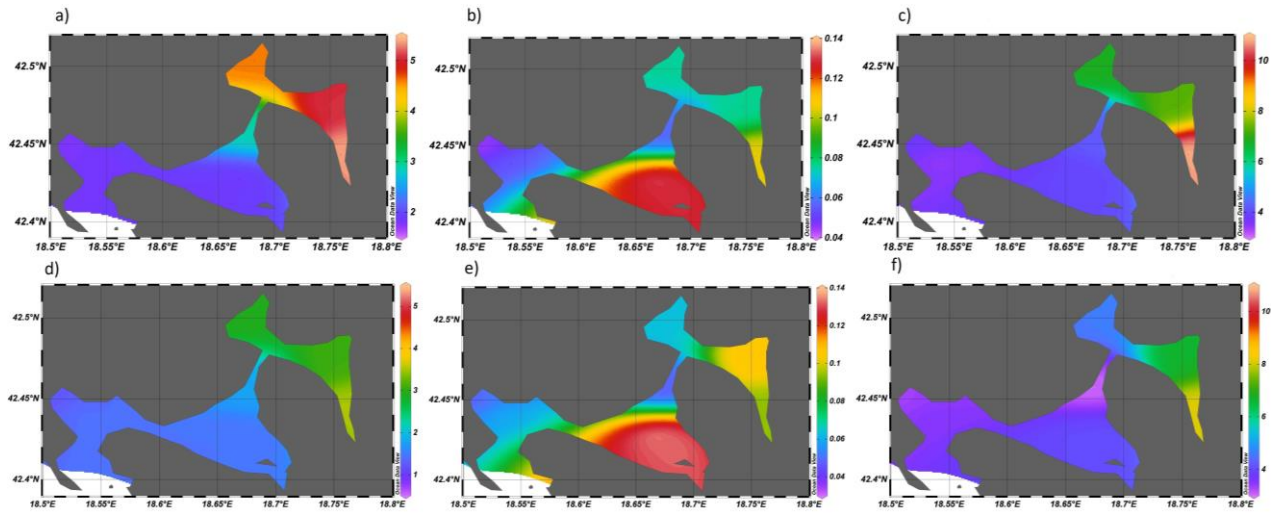


Figure 2. a and d: distribution of concentration of NO_3^- at surface and bottom; b and e: distribution of concentration of PO_4^{3-} at surface and bottom; c and f: distribution of concentration of SiO_4^- at surface and bottom during research period 2019-2020 in the area of Boka Kotorska Bay (BK-1, BK-2, BK-3, BK-4, BK-5, BK-6 and BK-7) (version number ODV 5.6.5)

Table 3. Average values of nutrients concentration (NO_3^- ; PO_4^{3-} ; SiO_4^-) during research period 2019-2020 in the area outside of the bay

Location	Average concentration ($\mu\text{mol/l}$)		
	NO_3^-	PO_4^{3-}	SiO_2^-
OS-1	0.9012	0.0650	2.4127
OS-2	1.0974	0.0535	3.9060
OS-3	1.6679	0.0734	3.4932
OS-4	1.6244	0.0853	7.9398
OS-5	4.2498	0.1030	18.4614

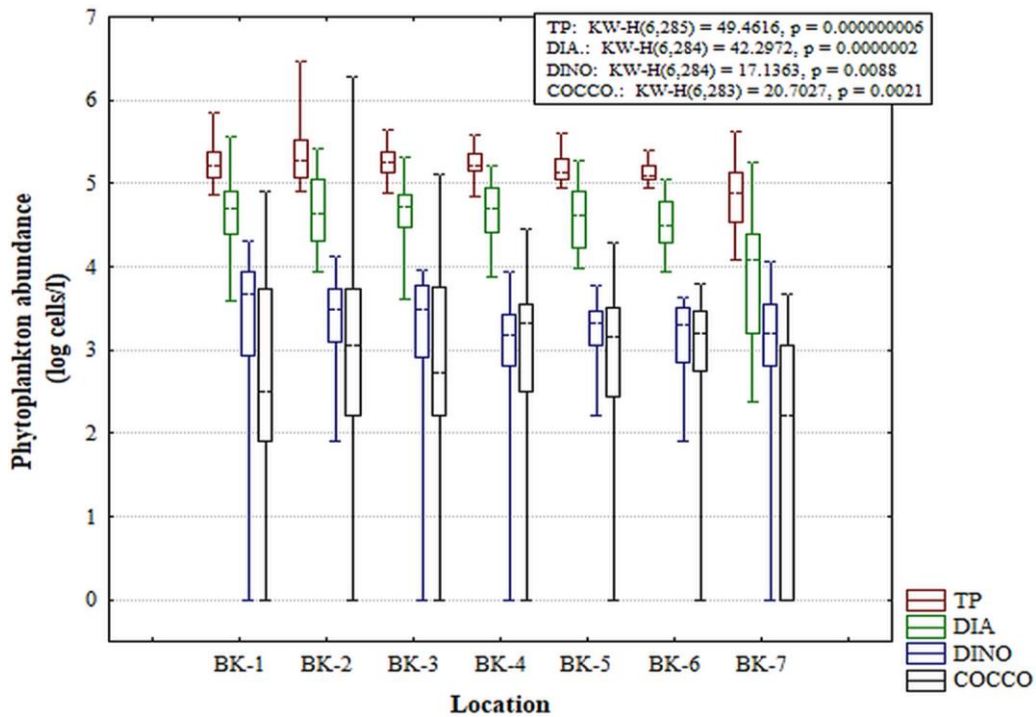


Figure 3. Distribution of abundances (log cells/L) of total phytoplankton (TP) and phytoplankton groups (diatoms, dinoflagellates, and coccolithophores) during research period 2019-2020 in the area of Boka Kotorska Bay (BK-1, BK-2, BK-3, BK-4, BK-5, BK-6 and BK-7)

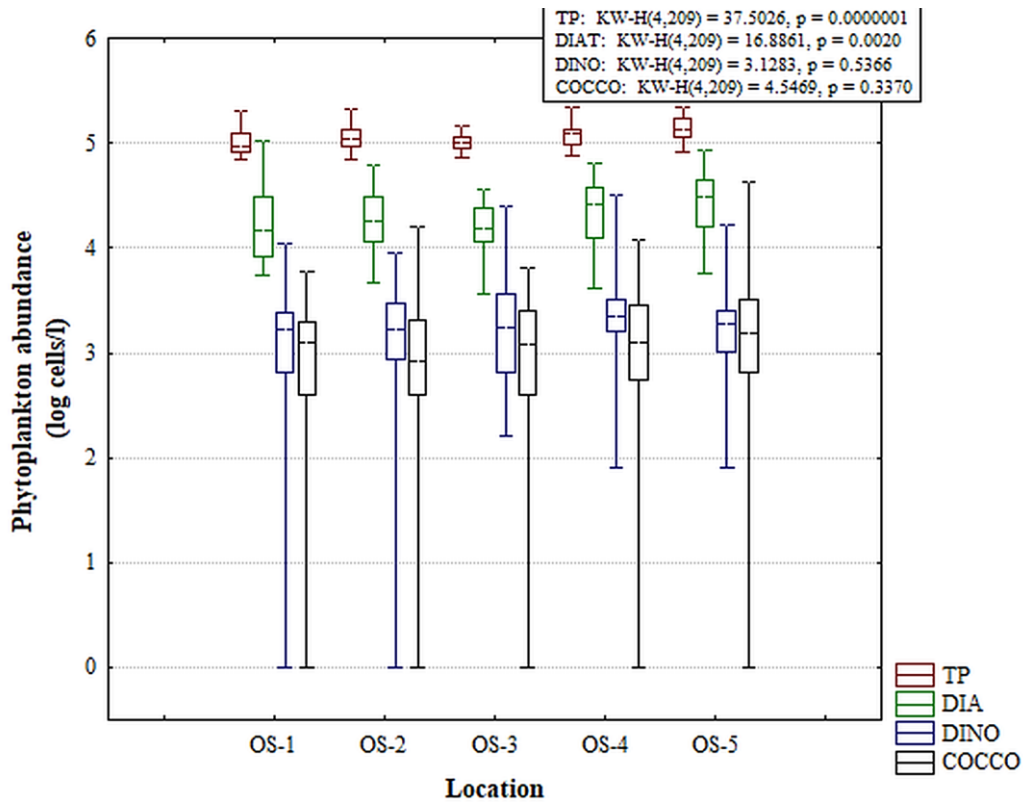


Figure 4. Distribution of abundances (log cells/L) of total phytoplankton (TP) and phytoplankton groups (diatoms, dinoflagellates, coccolithophores, and chlorophyte) during research period 2019-2020 in the area outside of the bay (OS1, OS-2, OS-3, OS-4, OS-5)

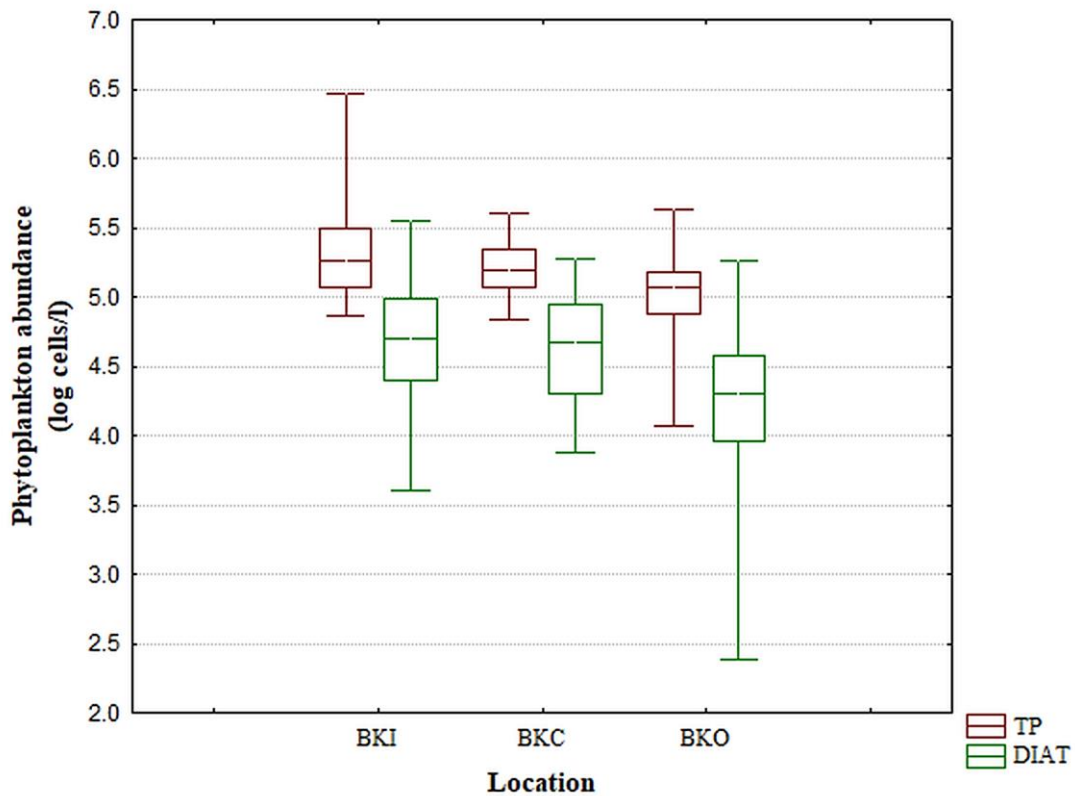


Figure 5 Distribution of abundances (log cells/L) of total phytoplankton (TP) and diatoms in the different parts of the Bay area (BKI - inner part; BKC - central part; BKO – outer part)

Of the 128 taxa that were recorded in the bay area, nine were potentially toxic and toxic taxa. Potentially toxic diatoms *Pseudo-nitzschia* spp. were noted (maximum abundance of 4.54×10^5 cells/l) and eight toxic dinoflagellate species from four genera: *Dinophysis* (*Dinophysis acuminata*, *D. acuta*, *D. caudata*, *D. fortii*, *D. tripos*), *Gonyaulax* (*Gonyaulax spinifera*), *Lingulodinium* (*Lingulodinium polyedra*), *Phalacroma* (*Phalacroma rotundatum*).

In the area of the outside of the bay, 118 taxa were recorded during the investigation from January 2019 to December 2020. In the coastal area of the open sea, the following taxa were recorded: 58 diatoms, 50 dinoflagellates, 5 coccolithophores, 2 silicoflagellates and 3 chlorophytes. Taxa from the diatom group that were present with a frequency of more than 30% were *Tetramphora ostrearia* (33.33%), *Asterionellopsis glacialis* (38.09%), *Asteromphalus flabellatus* (40.48%),

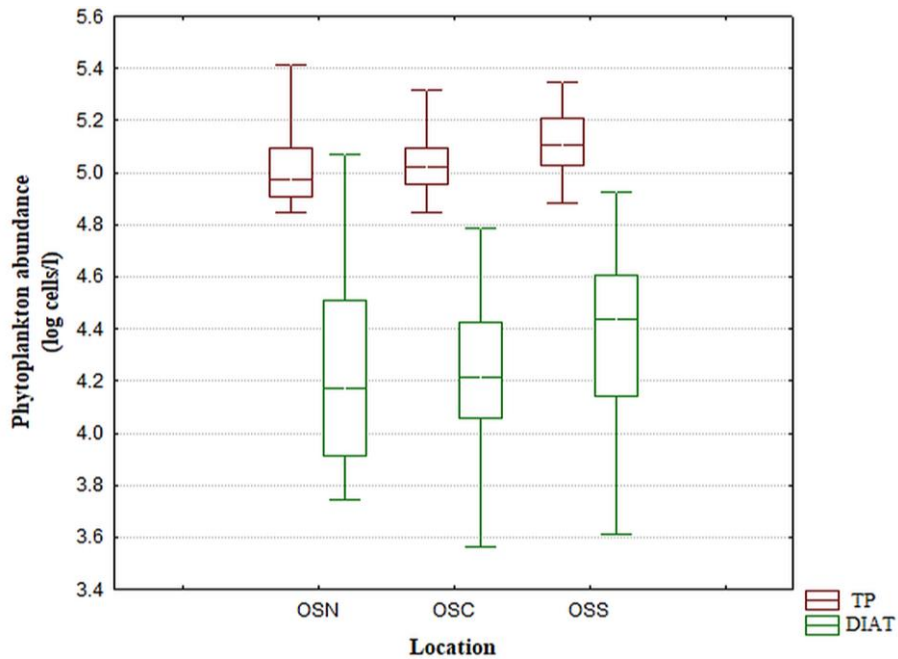


Figure 6. Distribution of abundances (log cells/L) of total phytoplankton (TP) and diatoms in the different parts of area outside of the bay (OSN - northern part; OSC-central part; OSS-southern part)

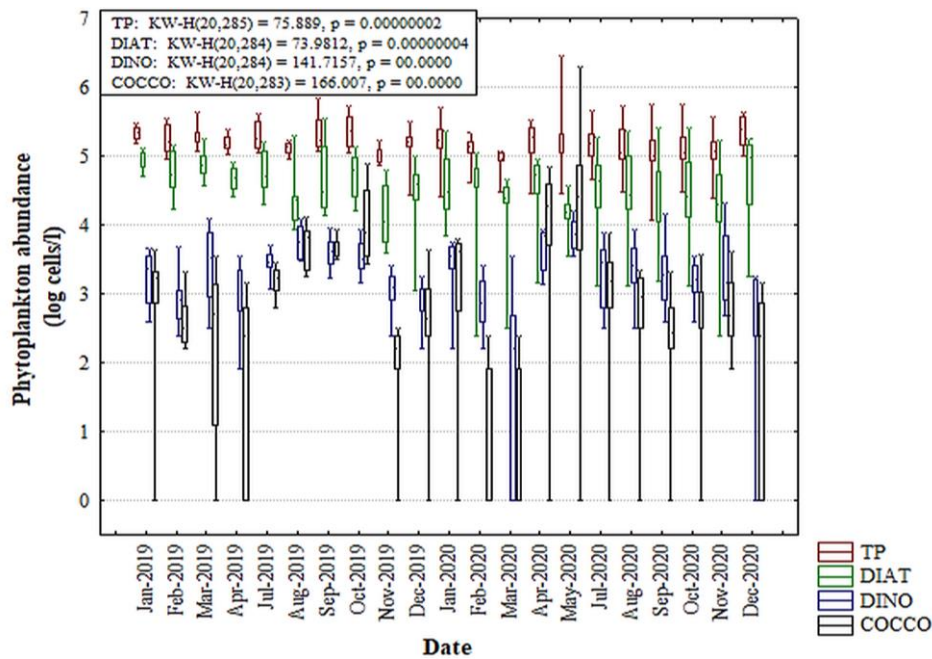


Figure 7. Temporal distribution of abundances (log cells/l) of total phytoplankton (TP) and phytoplankton groups (diatoms, dinoflagellates, and coccolithophores) during research period 2019-2020 in the area of Boka Kotorska Bay (BK-1, BK-2, BK-3, BK-4, BK-5, BK-6 and BK-7)

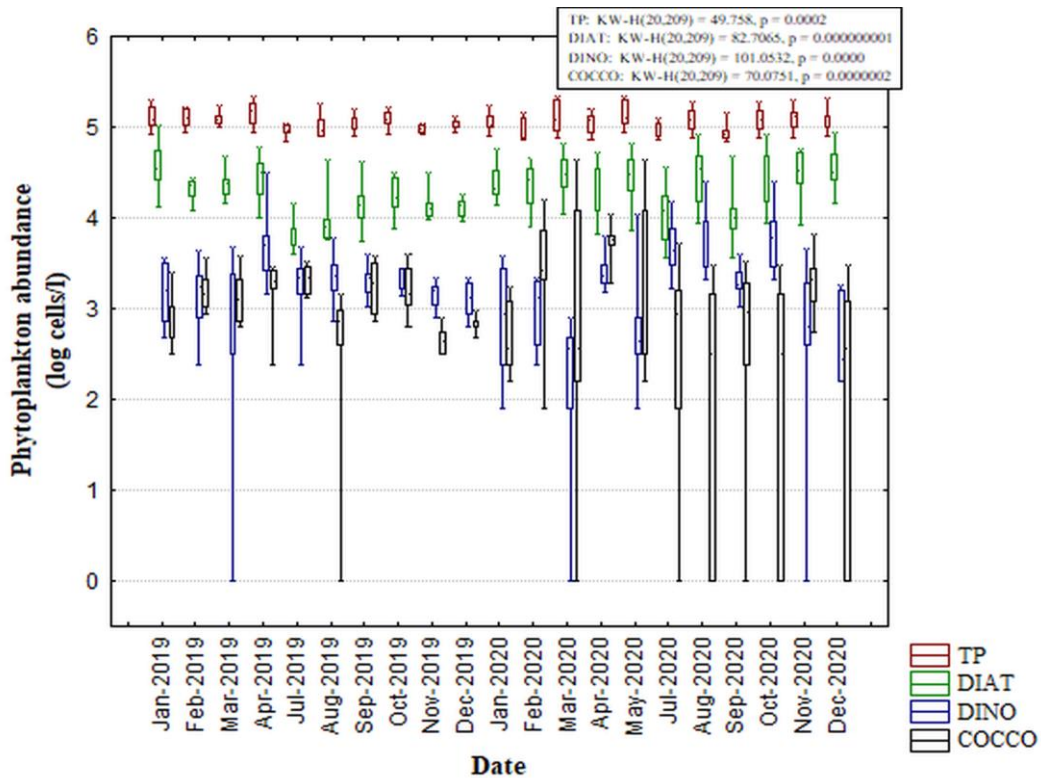


Figure 8. Temporal distribution of abundances (log cells/l) of total phytoplankton (TP) and phytoplankton groups (diatoms, dinoflagellates, coccolithophores, and chlorophyte) during research period 2019-2020 in the area outside of the bay (OS1, OS-2, OS-3, OS-4, OS-5)

Bacteriastrum hyalinum (30.95%), *Chaetoceros* spp. (90.48%), *Cocconeis scutellum* (50.0%), *Diploneis bombus* (30.95%), *Fragilaria* spp. (54.76%), *Guinardia striata* (33.33%), *Leptocylindrus danicus* (33.33%), *Licmophora paradoxa* (33.33%), *Navicula* spp. (88.09%), *Nitzschia longissima* (61.9%), *Pleurosigma elongatum* (61.9%), *Proboscia alata* (76.19%), *Pseudo-nitzschia* spp. (100%), *Ardissonea fulgens* (50.0%), *Thalassionema frauenfeldii* (35.71%), and *T. nitzschioides* (85.71%). From the dinoflagellates group, six taxa were observed: *Gonyaulax* spp. (88.09%), *Gyrodinium fusiforme* (47.61%), *Prorocentrum cordatum* (47.62%), *P. micans* (59.52%), *Scrippsiella* spp. (47.62%), and *Tripos furca* (33.33%). Coccolithophores observed more frequently were *Calyptrosphaera oblonga* (66.67%), *Rhabdosphaera tignifer* (61.9%), and *Syracosphaera pulchra* (73.81%). From chlorophytes, it was noticed that *Pediastrum duplex* had a frequency of 59.52% (Table 5).

Three species were the most common, with frequencies from 80% to 90%: *Chaetoceros* spp., *Pseudo-nitzschia* spp., and *Thalassionema nitzschioides*. *Pseudo-nitzschia* spp. were the most abundant diatoms (the highest abundance was 5.92×10^4 cells/l), followed by species from the genera *Chaetoceros* and *Thalassionema nitzschioides* (3.99×10^4 cells/l and 1.99×10^4 cells/l, respectively). Two species from the dinoflagellates group were the most abundant, with a frequency of more than 50%: *Gonyaulax* spp. and *Prorocentrum micans*. The most frequent species was

from the genus *Gonyaulax* (the highest abundance was 1.07×10^4 cells/l), followed by *Prorocentrum micans*, with a maximal abundance of 1.43×10^3 cells/l (Table 5).

In the area outside of the bay, among the 118 taxa, 5 were potentially toxic and toxic taxa. Among diatoms, *Pseudo-nitzschia* spp. (maximal abundance of 5.92×10^4 cells/l) were recorded, as well as four toxic dinoflagellate species from three genera: *Dinophysis* (*Dinophysis acuminata* and *D. acuta*), *Lingulodinium* (*Lingulodinium polyedra*), *Phalacroma* (*Phalacroma rotundatum*).

Relationships among Parameters

In the area of Boka Kotorska Bay, the highest positive correlations were determined between diatom and phosphate concentrations on one side and temperature and salinity on the other side. Also positive correlation was noticed between dinoflagellates and coccolithophores on one side and temperature and salinity on the other side. Total phytoplankton had a significantly negative correlation with salinity, diatoms with temperature and salinity and dinoflagellates and coccolithophores on one side with NO_3^- and PO_4^{3-} on the other side (Table 6).

In the coastal area of the open sea, the highest positive correlations were determined between total phytoplankton and diatoms on one side and nutrients on the other side, dinoflagellates with temperature and salinity, and coccolithophores with SiO_4^- . Total

Table 4. List of phytoplankton taxa recorded during research period 2019 - 2020 in the area of Boka Kotorska Bay (BK-1, BK-2, BK-3, BK-4, BK-5, BK-6 and BK-7).

	BK-1		BK-2		BK-3		BK-4		BK-5		BK-6		BK-7	
	MAX	FR.	MAX	FR.	MAX	FR.	MAX	FR.	MAX	FR.	MAX	FR.	MAX	FR.
Diatoms														
<i>Achnanthes brevipes</i> C. Agardh	1120	23.81	320	5.88	160	2.38	400	4.76	1428	4.76	160	2.38	160	2.38
<i>Ardissonea fulgens</i> (Greville) Kanjer, Kusber & Van de Vijver	80	2.38	160	2.94	320	7.14	240	7.14			880	4.76	800	2.38
<i>Asterionellopsis glacialis</i> (Castracane) Round	27846	19.04	4640	8.82	39984	19.05	19992	30.95	29988	26.19	16065	35.71	23562	35.71
<i>Asteromphalus flabellatus</i> (Brébisson) Greville			80	2.94	80	2.38	160	7.14	80	2.38	80	2.38		
<i>Bacteriastrium hyalinum</i> Lauder	10710	42.86	34272	32.35	27846	42.86	19278	47.62	12852	40.48	9996	57.14	38556	45.24
<i>Biddulphia biddulphiana</i> (J. E. Smith) Boyer	80	2.38												
<i>Cerataulina pelagica</i> (Cleve) Hendey	560	2.38	47838	5.88	1140	11.9	2856	14.29	8568	19.05	7854	19.05	2142	23.81
<i>Chaetoceros affinis</i> Lauder	11424	14.29	13566	14.71	11424	16.67	52836	23.81	13566	33.33	7854	23.81	8568	26.19
<i>Ch. convolutus</i> Castracane											1428	2.38		
<i>Ch. curvisetus</i> Cleve	13566	9.52	6426	2.94	4998	7.14	2856	4.76			4284	7.14	2142	4.76
<i>Ch. diversus</i> Cleve	4998	4.76	3927	2.94	2856	2.38	3570	9.52	12138	4.76	5712	21.43	3570	16.67
<i>C. messanense</i> Castracane	800	4.76												
<i>Chaetoceros</i> spp.	68544	92.86	88535	82.35	111384	92.86	107100	88.09	113526	88.09	52836	85.71	75684	90.48
<i>Cocconeis scutellum</i> Ehrenberg	240	21.43	400	20.59	1428	38.09	714	38.09	1428	38.09	714	33.33	560	35.71
<i>Coscinodiscus perforatus</i> Ehrenberg	80	2.38												
<i>Coscinodiscus</i> spp.	1428	16.67	80	2.94	1428	16.67	320	9.52	320	7.14	160	11.9	160	9.52
<i>Cyclotella striata</i> (Kützing) Grunow	20706	11.9	4998	5.88	4284	4.76	800	11.9			4998	16.67	4641	11.9
<i>Cylindrotheca closterium</i> (Ehrenberg) Reimann & J.C.Lewin	160	4.76	240	17.65	640	7.14	800	11.9	714	11.9	714	11.9	240	11.9
<i>Dactyliosolen blavyanus</i> (H. Peragallo) Hasle							400	4.76						
<i>D. fragilissimus</i> (Bergon) Hasle	6426	26.19	4998	26.47	12138	40.48	9996	26.19	9996	23.81	9996	33.33	6426	33.33
<i>Detonula pumila</i> (Castracane) Gran	4284	11.9	240	2.94	4284	9.52	2856	2.38	2856	9.52	640	9.52	720	4.76
<i>Diploneis bombus</i> (Ehrenberg) Ehrenberg	714	21.43	714	41.18	1071	23.81	714	33.33	2142	26.19	1428	23.81	240	21.43
<i>D. crabro</i> (Ehrenberg) Ehrenberg	160	4.76			160	2.38					80	2.38		
<i>Divergita toxoneides</i> (Castracane) Theriot											80	2.38		
<i>Diploneis</i> sp.											160	4.76		
<i>Entomoneis pulchra</i> (Bailey) Reimer	80	9.52					80	2.38			80	4.76		
<i>Eucampia cornuta</i> (Cleve) Grunow	640	2.38			1840	2.38	1200	2.38	240	2.38	4284	4.76	1600	4.76
<i>Guinardia flaccida</i> (Castracane) H. Peragallo	1428	11.9	1785	11.76	800	28.57	2142	23.81	1428	11.9	2499	14.29	3570	14.29
<i>G. striata</i> (Stolterfoth) Hasle	4284	42.86	3570	44.12	9282	47.62	4284	42.86	7140	23.81	4284	47.62	9996	50.0
<i>Grammatophora oceanica</i> Ehrenberg					400	2.38					1200	4.76	400	4.76
<i>Hemiaulus hauckii</i> Grunow ex Van Heureck	2142	23.81	3570	35.29	9639	30.95	7854	26.19	3570	23.81	1428	30.95	2142	40.48
<i>H. sinensis</i> Greville	3570	23.81	640	5.88	11424	19.05	1280	14.29	6426	21.43	3570	21.43	4284	14.29
<i>Lauderia annulata</i> Cleve									320	4.76				
<i>Leptocylindrus danicus</i> Cleve	17850	38.09	43554	29.41	12852	33.33	12138	23.81	12852	33.33	17850	23.81	9282	28.57
<i>L. mediterraneus</i> (H. Peragallo) Hasle			400	2.94			1360	2.38			1280	4.76	8568	7.14
<i>Licmophora flabellata</i> (Greville) C. Agardh	240	4.76			1428	4.76	1428	19.05	320	11.9	240	4.76	1428	14.29
<i>L. paradoxa</i> (Lyngbye) C. Agardh	80	2.38	80	2.94	714	11.9	714	2.38	1428	14.29	714	16.67	714	16.67
<i>Lioloma pacificum</i> (Cupp) Hasle	1040	30.95	1428	11.76	13566	28.57	4284	35.71	2142	23.81	5712	21.43	2856	11.9
<i>Lithodesmium undulatum</i> Ehrenberg	2856	7.14	1120	11.76	560	2.38	320	9.52	160	4.76	240	9.52	80	4.76
<i>Melosira nummuloides</i> C. Agardh	2400	2.38			1120	2.38	1680	2.38			1120	2.38		
<i>Navicula</i> spp.	4998	83.33	2142	35.29	9996	64.29	4284	69.05	4284	69.05	5712	71.43	1785	80.95
<i>Neocalyptrella robusta</i> (Norman ex Ralfs) Hernández-Becerril & Meave	160	7.14					160	2.38	80	2.38			80	2.38
<i>Nitzschia incerta</i> (Grunow) M. Peragallo													80	2.38
<i>Nitzschia longissima</i> (Brébisson) Ralfs	2142	21.43	2499	20.59	2856	40.48	2142	47.62	2142	38.09	1428	45.24	2856	21.43
<i>Trieres mobiliensis</i> (Bailey) Ashworth & E.C. Theriot	400	2.38	80	2.94	714	7.14			80	2.38				
<i>Paralia sulcata</i> (Ehrenberg) Cleve	6320	4.76					400	2.38	1600	2.38				
<i>Pinnularia viridis</i> (Nitzsch) Ehrenberg					560	2.38					160	2.38		
<i>Pleurosigma angulatum</i> (J.T. Quekett) W. Smith	240	14.29	714	14.71	160	16.67	714	14.29	160	14.29	714	23.81	160	9.52
<i>P. elongatum</i> W. Smith	8211	55.81	2856	44.123	5712	42.86	3570	64.29	2856	59.52	2856	59.52	3570	61.9
<i>P. formosum</i> W. Smith	160	4.76	80	2.94	80	2.38	240	4.76	240	7.14	160	4.76	1428	2.38
<i>Proboscia alata</i> (Brightwell) Sundström	8568	92.86	27132	23.53	20706	71.43	25704	71.43	7854	73.81	5712	80.95	8568	73.81
<i>Pseudosolenia calcar avis</i> (Schultze) B.G. Sundström	714	7.14	714	8.82	160	16.67	714	9.52	160	7.14	320	9.52	160	2.38
<i>Pseudo-nitzschia</i> spp.	311303	100	189210	97.06	75684	100	454103	100	127092	100	55692	100	279173	100
<i>Rhizosolenia imbricata</i> Brightwell					240	2.38	160	2.38	400	2.38	160	2.38		
<i>Rh. setigera</i> Brightwell	1428	9.52	640	20.59	80	7.14	320	26.19	320	14.29	2856	19.05	80	14.29
<i>Skeletonema</i> spp.	12138	4.76	22848	11.76	5712	4.76	12852	7.14	19992	11.9			4284	7.14
<i>Striatella unipunctata</i> (Lyngbye) C. Agardh							80	2.38					1428	2.38
<i>Synedra</i> spp.			160	2.94									400	2.38
<i>Tetramphora ostrearia</i> (Brébisson) Mereschkowsky	160	7.14	160	5.88	240	2.38	714	7.14	160	2.38	240	2.38	240	4.76
<i>Thalassionema nitzschioides</i> (Grunow) Mereschkowsky	12138	88.09	8925	70.59	17850	21.43	10710	88.09	17136	92.86	12852	95.24	14994	85.71
<i>T. frauenfeldii</i> (Grunow) Tempère & Peragallo	2142	19.05	1040	17.65	8568	14.29	5712	33.33	3570	33.33	1600	33.33	2856	28.57
<i>Thalassiosira eccentrica</i> (Ehrenberg) Cleve	4284	30.95												
<i>Thalassiosira rotula</i> Meunier			4284	20.59	3570	19.05	1600	23.81	800	11.9	720	11.9	400	9.52
Dinoflagellates														
<i>Amphisolenia globifera</i> F. Stein					80	2.38								
<i>Dinophysis acuminata</i> Claparede et Lachmann	1428	9.52	714	8.82	714	14.29	80	7.14	160	11.9			160	11.9
<i>D. acuta</i> Ehrenberg	400	19.05	2142	20.59	240	14.29	80	4.76	80	7.14	160	9.52	160	14.29
<i>D. caudata</i> Kent	714	7.14	320	11.76					80	2.38			240	2.38
<i>D. fortii</i> Pavillard	80	2.38	80	2.94					80	2.38	80	2.38		
<i>D. hastata</i> F. Stein					80	2.38								
<i>D. tripos</i> Gourret	160	4.76	80	2.94										
<i>Diplopsalis lenticula</i> Bergh	160	19.05	240	17.65	160	16.67	400	19.05	320	21.43	240	26.19	240	23.81
<i>Gonyaulax digitale</i> (Pouchet) Kofoid	714	21.43	1520	5.88	1428	11.9	240	7.14	80	7.14	80	4.76	80	4.76

Table 4. Continued

<i>Gonyaulax polygramma</i> F. Stein	240	7.14	160	2.94	80	2.38	80	2.38	160	4.76	240	4.76	240	9.52
<i>Gonyaulax spinifera</i> (Claparede & Lachmann) Diesing	80	2.38			80	2.38	80	2.38					80	2.38
<i>Gonyaulax</i> spp.	28560	85.71	3570	23.53	4998	76.19	2856	71.43	3570	78.57	2856	69.05	6426	90.48
<i>Gonyaulax verior</i> Sournia														
<i>Gymnodinium</i> spp.	7854	14.29	2856	20.59	2499	9.52	714	21.43	714	16.67	1428	16.67	240	23.81
<i>Gyrodinium fusiforme</i> Kofoid et Swezy	2856	28.57	2142	55.88	2856	40.48	3570	50.0	1071	54.76	320	61.9	800	47.62
<i>Gyrodinium</i> spp.														
<i>Hermesinum adriaticum</i> Zacharias	80	4.76	160	8.82	80	4.76	160	9.52			80	4.76	80	2.38
<i>Lingulodinium polyedra</i> (F. Stein) J.D. Dodge	320	4.76	320	23.53	240	14.29	1040	11.9	320	21.43	160	21.43	320	28.57
<i>Noctiluca scintillans</i> (Macartney) Kofoid & Swezy											80	4.76		
<i>Ornithocercus heteroporus</i> Kofoid			80	2.94										
<i>Oxytoxum sceptrum</i> (F. Stein) Schröder			80	5.88	240	2.38	160	4.76	80	2.38	80	2.34	80	2.38
<i>O. scolopax</i> F. Stein	1071	7.14	240	5.88	2142	4.76	80	2.38	160	14.29			80	2.38
<i>O. sphaeroideum</i> F.Stein	160	2.38			160	4.76	80	2.38			160	11.9	240	16.67
<i>O. tessellatum</i> (F.Stein) Schütt									80	4.76	80	2.38	160	4.76
<i>Phalacroma rotundatum</i> (Claparede et Lachmann) Kofoid et Michener	80	4.76	80	2.94			160	4.76	714	7.14	80	7.14	80	2.38
<i>Podolampas elegans</i> F. Schütt							80	2.38						
<i>P. palmipes</i> Stein	80	4.76			160	4.76	80	7.14			80	2.38	80	4.76
<i>Prorocentrum cordatum</i> (Ostenfeld) J. D. Dodge	4284	40.48	3570	41.18	2856	38.09	580	47.62	3570	47.62	2856	42.86	3570	40.48
<i>P. micans</i> Ehrenberg	12852	66.67	2142	61.76	2499	64.29	2856	88.09	2499	61.90	560	40.48	640	42.86
<i>P. scutellum</i> B.Schröder	80	2.38					80	2.38						
<i>P. triestinum</i> J. Schiller	9282	28.57	1428	23.53	1428	21.43	1428	21.43	1428	26.19	1428	26.19	2142	14.29
<i>Protoperidinium conicum</i> (Gran) Balech	80	2.38			80	2.38								
<i>P. crassipes</i> (Kofoid) Balech	480	23.81	400	11.76	160	11.9	160	21.43	160	16.67	320	7.14		
<i>P. diabolium</i> (Cleve) Balech	320	7.14	80	11.76	240	7.14	80	7.14			80	2.38	2142	9.52
<i>P. divergens</i> (Ehrenberg) Balech	80	2.38			160	9.52							80	2.38
<i>P. globulum</i> (F. Stein) Balech									80	2.38			160	2.38
<i>P. pallidum</i> (Ostenfeld) Balech							80	4.76						
<i>P. pellucidum</i> Bergh	80	7.14	80	2.94			160	2.38			320	11.9	160	11.9
<i>P. steinii</i> (Jørgensen) Balech	80	4.76	80	8.82	160	7.14	80	9.52			160	4.76	80	2.38
<i>P. tuba</i> (Schiller) Balech	714	26.09	240	23.53	240	9.52	160	11.9	714	14.29	480	16.67	320	26.19
<i>Protoperidinium</i> spp.			714	11.76	714	16.67	80	2.38	400	19.05	240	4.76	240	11.9
<i>Pselodinium fusus</i> (F. Schütt) F. Gómez			80	2.94			80	2.38					80	2.38
<i>Scrippsiella</i> spp.	1785	28.57	2856	35.29	714	30.95	720	23.81	560	33.33	1428	38.09	9996	33.33
<i>Tripes azoricus</i> (Cleve) F. Gómez							80	4.76	80	4.76				
<i>T. gibberus</i> (Gourret) F.Gómez											240	7.14	240	4.76
<i>T. furca</i> (Ehrenberg) Gómez	240	23.81	714	29.41	714	21.43	320	11.9	240	9.52	160	30.95	480	11.9
<i>T. fusus</i> (Ehrenberg) Gómez	560	21.43	560	32.35	714	21.43	320	30.95	240	16.67	714	14.29	160	14.29
<i>T. horridus</i> (Cleve) F. Gómez	240	9.52	240	8.82	714	14.29	80	7.14	160	7.14	80	9.52		
<i>T. setaceus</i> (Jørgesen) Gómez	2142	16.67	80	8.82	714	9.52	720	16.67	714	14.29	960	11.9	714	33.33
<i>T. macroceros</i> (Ehrenberg) Hallegraef & Huisman	160	4.76			160	4.76	160	2.38	80	4.76	80	2.38	80	4.76
<i>T. muelleri</i> Bory de Saint-Vincent	3570	45.24	3570	44.12	4284	40.48	2142	45.24	1428	23.81	160	9.52	714	14.29
<i>T. teres</i> (Kofoid) F.Gómez			80	2.94	80	4.76	80	2.38	80	4.76	80	2.38		
<i>Tripes</i> spp.	80	2.38												
<i>Tryblionella compressa</i> (Bailey) Poulin	320	11.9	320	11.76	240	9.52	480	23.81	160	11.9	714	19.05	714	23.81
Coccolithophores														
<i>Acanthoica quattropsina</i> Lohmann			714	2.94	80	2.38								
<i>Calcosolenia brasiliensis</i> (Lohmann) J. R. Young	240	7.14	1428	17.65			160	11.9	160	9.52	714	7.14	240	19.05
<i>Calyptrosphaera oblonga</i> Lohmann	69615	35.76	1944281	44.12	132090	40.48	28560	47.62	17493	45.24	4998	54.76	11424	64.29
<i>Helicosphaera walichii</i> (Lohmann) Okada & McIntyre	60690	21.43	28560	29.41	16065	16.67	7140	19.05	7854	23.81	720	14.29	5712	14.29
<i>Rhabdosphaera tignifer</i> J. Schiller	7854	19.05	5712	32.35	3570	28.57	2856	47.62	3570	40.48	4284	54.76	2856	42.86
<i>Syracosphaera pulchra</i> Lohmann	9996	40.48	7140	50.0	4284	54.76	4284	64.29	3570	54.76	2142	61.90	2856	30.95
Silicoflagellates														
<i>Dictyocha fibula</i> Ehrenberg	3213	28.57	4998	11.76	4998	9.52	714	14.29	240	11.9	714	14.29	714	7.14
<i>Octactis octonaria</i> (Ehrenberg) Hovasse	714	21.43	714	11.76	160	7.14	714	11.9	160	9.52	714	11.9	714	7.14
Chlorophytes														
<i>Pediastrum duplex</i> Meyen											5712	4.76	3570	4.76
<i>Scenedesmus quadricauda</i> (Turpin) Brébisson											5712	4.76	2142	4.76

phytoplankton and diatoms had a significantly negative correlation with temperature and salinity, and dinoflagellates had a significantly negative correlation with PO₄³⁻ (Table 7).

Observed relationships proved by Spearman's rank were also confirmed by PCA (Figure 9).

The highest positive correlations were determined between dinoflagellates and coccolithophores on one side and temperature and salinity on other side. TP, diatoms, dinoflagellates and coccolithophores showed negative correlation with inorganic matters (NO₃⁻ and PO₄³⁻). The bi-plot of the first two PCA components

accounts for 78.43% of the total variance. The first axis explains most of the variance (50.3%) and it is related to TP, diatoms, dinoflagellates and coccolithophores. The second axis, which explains much less of the total variance (28.13%), is mainly related to temperature and salinity. Thus second axis is seasonal, separating the spring/summer samples from the autumn/winter ones, whatever the station. Furthermore, PCA showed that temperature, NO₃⁻, and SiO₄⁴⁻ were the main factors influencing the vertical distribution and abundances of phytoplankton.

Diversity Indices: Margalef and Shannon & Wiener

Biodiversity indices (Margalef and Shannon–Wiener) were calculated for locations in Boka Kotorska Bay and for the area outside of the bay from 2019–2020. The Margalef's value ranged from 1.57 to 6.67, while the Shannon–Wiener value ranged from 0.11 to 2.56 for the bay (Figure 10). For the coastal part, the value of Margalef's index ranged from 0.62 to 6.62, and that of Shannon's index ranged from 0.87 to 2.88 (Figure 11). Analyses of the biodiversity indices (Margalef and Shannon–Wiener) showed relatively high biodiversity of the phytoplankton community (Figure 10 and Figure 11). The calculated diversity indices show small fluctuations between the investigated locations in the bay area and the coastal area of the open sea. Regarding the temporal distribution, the highest value of phytoplankton indices in the bay area was noticed during the summer period in July 2019 for the Margalef index and during late summer–early autumn in September 2019 for the Shannon–Wiener index. The lowest value for the indices was recorded during the late winter–early spring period in March 2020 and during the spring period in May 2020 (Figure 12). In the area outside of the bay, the highest value of phytoplankton indices was noticed during the winter period in February 2020 for both indices. The lowest values of the indices were recorded during the summer period in July 2020 and during the autumn period in September 2020 (Figure 13).

The graphical representation of clusters for Boka Kotorska Bay provided a differentiation between locations according to the abundances of phytoplankton groups (diatoms, dinoflagellates, coccolithophores, and silicoflagellates). The presentation revealed two main groups, where similarity was the highest (Figure 14). The first group included locations BK-6 and BK-7, and the second group, where the highest similarity was noticed, included locations BK-4 and BK-5.

The grouping of locations could be a result of their positions. Locations BK-4 and BK-5 were in the area of Tivat Bay, while locations BK-6 and BK-7 were in Herceg Novi Bay.

The graphical representation of clusters for the area outside of the bay provided a differentiation between locations according to abundances of phytoplankton groups (diatoms, dinoflagellates, coccolithophores and silicoflagellates). The presentation revealed two main groups, where similarity was the highest (Figure 14). The first group included locations OS-2 and OS-3, which showed the highest similarity, and the second highest similarity was noticed between locations OS-4 and OS-5. For the Bay area, the grouping of locations could be a result of their positions.

The graphical representation of clusters for both areas provided a differentiation between locations according to abundances of phytoplankton groups (diatoms, dinoflagellates, coccolithophores, and silicoflagellates). The presentation revealed four main groups, where similarity was the highest (Figure 14). The

first two groups included locations BK-4 and BK-5 and BK-6 and BK-7, and the second two groups included locations from the area outside of the bay, OS-2 and OS-3 and OS-4 and OS-5. The highest similarity was noticed between these groups.

Discussion

The area of the South Adriatic Sea is characterized by relatively low phosphate and nitrate concentrations, and primary production is often limited by phosphate (Viličić, 1989; Viličić et al., 1998). The area of research (especially the Bay area) is characterized by significant freshwater inputs from streams or underground springs during the precipitation season, in late winter and spring and then in autumn (Bellafiore et al. 2011). During this period, streams and springs influenced the physical, chemical and biological dynamics of the seawater and had a marked positive impact on productivity. During the summer period, the inflow of water was lower; therefore, remineralization processes and sewage discharge are considered the most important nutrient sources in this system. Comparison with previous data for this region mostly covered the area of Boka Kotorska Bay, as there is a lack of data for the area outside of the bay. In addition, data on the phytoplankton community in the open southern Adriatic consist mainly of episodic samplings, with long gaps in some periods. Recent investigations performed in the last decade have contributed useful knowledge on phytoplankton dynamics and structure, particularly in winter–spring seasons (Ljubimir et al., 2017; Batistić et al., 2019). The concentrations of nutrients (particularly silicates, maximal up to 184.08 $\mu\text{mol/l}$) during research were generally higher than the previous data (maximum of silicates 23.17 $\mu\text{mol/l}$) available for Boka Kotorska Bay (Drakulović et al., 2017) but similar to the data of Drakulović et al. (2012). Concentrations of nutrients generally followed the growth of phytoplankton. Higher values of nutrients and phytoplankton were noticed in the inner part of the bay (Kotor Bay and Tivat Bay) (Figure 2, Figure 3 and Figure 4). The nutrient concentrations observed in our research were similar to those found in the most oligotrophic area of the Lastovo archipelago, where extremes were recorded at the beginning of the study period. However, extremes recorded at the beginning of the study period in Lastovo are more typical for eutrophic systems, such as the Neretva Estuary (Jasprica et al., 2012), Mali Ston Bay (Matek et al., 2023), and Boka Kotorska Bay (Drakulović et al., 2013).

The hydrological and biological evolution shows that the locations in the bay area, especially in the inner part, are influenced by river outflows and influence from the coast. Conversely, the BK1 and BK3 locations are largely affected by small river runoff, irrespective of their magnitude. Notably, nutrient concentrations tend to be higher in the near-surface region. This observation aligns perfectly with the pivotal role played by rivers as

Table 5. List of phytoplankton taxa recorded during research period 2019 - 2020. in the area of coastal part of open sea of Montenegro (OS-1, OS-2, OS-3, OS-4, OS-5)

	OS-1		OS-2		OS-3		OS-4		OS-5	
	MAX	FR.	MAX	FR.	MAX	FR.	MAX	FR.	MAX	FR.
Diatoms										
<i>Achnanthes brevipes</i> C. Agardh			400	7.14	1785	9.52	400	2.38		
<i>Ardissonea fulgens</i> (Greville) Kanjer, Kusber & Van de Vijver	560	11.9	560	21.43	320	14.29	480	16.67	7140	50.0
<i>Asterionellopsis glacialis</i> (Castracane) Round	33558	38.09	4284	30.95	7140	26.19	16422	33.33	11424	33.33
<i>Asteromphalus flabellatus</i> (Brébisson) Greville			7854	40.48						
<i>Bacteriastrium hyalinum</i> Lauder	12138	28.57			4284	26.19	5712	30.95	5712	19.05
<i>Cerataulina pelagica</i> (Cleve) Hendey	1428	21.43	480	4.76	3570	9.52	1428	4.76	4284	16.67
<i>Chaetoceros affinis</i> Lauder	7854	19.05	7854	21.43	6783	14.29	6783	14.29	5712	23.81
<i>Ch. curvisetus</i> Cleve	1428	2.38								
<i>Ch. diversus</i> Cleve	1071	2.38	5712	7.14	4284	9.52	3570	9.52	2856	9.52
<i>Chaetoceros</i> spp.	39984	73.81	27846	83.33	26418	85.71	31059	71.43	28560	90.48
<i>Cocconeis scutellum</i> Ehrenberg	714	23.81	1428	40.48	1428	40.48	714	50.0	5712	35.71
<i>Cocconeis</i> spp.									240	2.38
<i>Coscinodiscus</i> spp.	160	14.29			714	7.14	640	2.38		
<i>Cyclotella striata</i> (Kützing) Grunow	2499	11.9			2856	7.14	320	7.14	1280	16.67
<i>Cylindrotheca closterium</i> (Ehrenberg) Reimann&J.C.Lewin	160	7.14	160	16.67	160	7.14	400	21.43	1040	9.52
<i>Dactyliosolen blavyanus</i> (H. Peragallo) Hasle									240	2.38
<i>D. fragilissimus</i> (Bergon) Hasle	2856	23.81	3570	11.9	2856	9.52	3213	14.29	2142	16.67
<i>Detonula pumila</i> (Castracane) Gran	320	2.38								
<i>Diploneis bombus</i> (Ehrenberg) Ehrenberg	2142	23.81	1071	30.95	1071	23.81	1428	28.57	714	23.81
<i>D. crabro</i> (Ehrenberg) Ehrenberg					160	2.38				
<i>Entomoneis pulchra</i> (Bailey) Reimer			240	9.52	160	2.38	240	16.67		
<i>Eucampia cornuta</i> (Cleve) Grunow	2856	4.76								
<i>Fragillaria</i> spp.							2856	9.52	67830	54.76
<i>Guinardia flaccida</i> (Castracane) H. Peragallo	1200	16.67	1428	14.29	1428	4.76	3213	16.67	4641	14.29
<i>G. striata</i> (Stolterfoth) Hasle	7140	26.19	7140	21.43	1428	26.19	3570	33.33	12138	26.19
<i>Grammatophora oceanica</i> Ehrenberg	80	2.38	3570	4.76	160	2.38				
<i>Hemiaulus hauckii</i> Grunow ex Van Heureck	2856	28.57	2142	21.43	1428	16.67	640	4.76	3213	14.29
<i>H. sinensis</i> Greville	5712	14.29	2499	4.76	560	9.52	640	11.9	3927	11.9
<i>Leptocylindrus danicus</i> Cleve	15708	16.67	5712	33.33	7854	21.43	4998	30.95	71400	23.81
<i>L. mediterraneus</i> (H. Peragallo) Hasle	320	4.76	800	2.38	3570	4.76			800	2.38
<i>Licmophora flabellata</i> (Greville) C. Agardh	714	19.05	714	23.81	720	11.9	880	21.43		
<i>L. paradoxa</i> (Lyngbye) C. Agardh	160	7.14	1428	23.81	1428	33.33	1760	21.43	160	7.14
<i>Lioloma pacificum</i> (Cupp) Hasle	4284	9.52	1428	16.67	1071	21.43	240	11.9	320	7.14
<i>Lithodesmium undulatum</i> Ehrenberg	240	4.76	80	4.76	400	2.38	1428	4.76	2856	9.52
<i>Melosira nummuloides</i> C. Agardh	640	4.76	880	2.38	320	2.38			800	2.38
<i>Navicula</i> spp.	2856	83.33	14278	88.09	2142	88.09	11781	83.33	7140	83.33
<i>Neocalyptrella robusta</i> (Norman ex Ralfs) Hernández-Becerril & Meave	80	2.38			80	2.38				
<i>Nitzschia incerta</i> (Grunow) M. Peragallo					160	2.38	240	4.76		
<i>Nitzschia longissima</i> (Brébisson) Ralfs	2142	26.19	1071	47.62	1428	45.24	8925	61.9	2856	50
<i>Tetramphora ostreararia</i> (Brébisson) Mereschkowsky	160	7.14	714	14.29	320	19.05	2142	28.57	1428	33.33
<i>Trieres mobiliensis</i> (Bailey) Ashworth & E.C.Theriot	80	2.38								
<i>Pinnularia viridis</i> (Nitzsch) Ehrenberg									2856	47.62
<i>Pleurosigma angulatum</i> (J. T. Quekett) W. Smith	714	23.81	1071	2.38	320	4.76	240	16.67	240	11.9
<i>P. elongatum</i> W. Smith	2856	45.24	1785	52.38	1785	42.86	2499	61.9	1428	42.86
<i>P. formosum</i> W. Smith	240	2.38	80	2.38	80	2.38	714	11.9		
<i>Proboscia alata</i> (Brightwell) Sundström	2856	71.43	6426	76.19	2856	64.29	2142	52.38	2142	45.24
<i>Pseudosolenia calcar - avis</i> (Schultze) B.G. Sundström Schultze	714	11.9	480	16.67	714	16.67	714	7.14	320	7.14
<i>Pseudo-nitzschia</i> spp.	59262	97.62	30702	95.24	19278	100	19992	90.48	42840	97.62
<i>Rhizosolenia imbricata</i> Brightwell	320	7.14			160	2.38	240	4.76	80	2.38
<i>Rh. setigera</i> Brightwell	714	11.9	160	4.76	714	4.76	80	2.38	80	4.76
<i>Skeletonema</i> spp.	1360	2.38	3570	2.38	3570	2.38	4284	4.76	5712	9.52
<i>Striatella unipunctata</i> (Lyngbye) C. Agardh	80	2.38	1428	11.9	714	4.76	240	4.76		
<i>Synedra</i> spp.									37128	2.38
<i>Thalassionema nitzschioides</i> (Grunow) Mereschkowsky	19992	80.95	9996	78.57	8925	78.57	9282	78.57	12138	85.71
<i>T. frauenfeldii</i> (Grunow) Tempère & Peragallo	2142	33.33	2499	26.19	2499	21.43	3570	28.57	2142	35.71
<i>Thalassiosira eccentrica</i> (Ehrenberg) Cleve	400	9.52								
<i>Thalassiosira rotula</i> Meunier					160	2.38	1428	4.76		
<i>Toxarium undulatum</i> Bailey	714	2.38								
Dinoflagellates										
<i>Cladopyxis caryophyllum</i> (Kofoid) Pavillard			80	2.38						
<i>Dinophysis acuminata</i> Claparede et Lachmann	160	7.14	80	2.38	160	2.38	160	9.52	160	4.76
<i>D. acuta</i> Ehrenberg	80	2.38	240	2.38	80	2.38	160	11.9	80	2.38
<i>Diplopsalis lenticula</i> Bergh	240	19.05	160	4.76	400	9.52	320	16.67	80	4.76
<i>Triadinium polyedricum</i> (Pouchet) J. D. Dodge					80	4.76				
<i>Gonyaulax digitale</i> (Pouchet) Kofoid	80	9.52	80	4.76	400	7.14	714	7.14	80	4.76
<i>Gonyaulax polygramma</i> F. Stein	240	4.76	80	9.52	480	9.52	320	9.52		
<i>Gonyaulax</i> spp.	9282	83.33	3570	88.09	3570	76.19	9282	80.95	10710	76.19
<i>Gonyaulax verior</i> Sournia										
<i>Gymnodinium</i> spp.	400	23.81	714	16.67	1428	21.43	1428	28.57	720	19.05
<i>Gyrodinium fusiforme</i> Kofoid et Swezy	1428	45.24	3570	47.61	1428	40.48	1428	40.48	1785	33.33
<i>Gyrodinium</i> spp.							160	2.38		
<i>Hermesinium adriaticum</i> Zacharias	80	2.38	80	2.38			240	4.76	160	2.38
<i>Lingulodinium polyedra</i> (F. Stein) J.D. Dodge	160	2.38	714	23.81	160	7.14	80	2.38		

Table 5. Continued

<i>Ornithocercus heteroporus</i> Kofoid					80	2.38				
<i>Oxytoxum sceptrum</i> (F. Stein) Schröder			80	7.14	80	2.38			80	4.76
<i>O. scolopax</i> F. Stein			160	7.14	160	4.76	80	4.76	160	2.38
<i>O. sphaeroidem</i> F. Stein	80	4.76	80	2.38	160	2.38			80	2.38
<i>O. tessellatum</i> (F.Stein) Schütt	80	2.38	160	2.38	80	2.38	80	2.38		
<i>Phalacroma rotundatum</i> (Claparede et Lachmann) Kofoid et Michener	80	2.38	80	4.76	80	9.52	80	4.76	80	2.36
<i>Podolampas palmipes</i> Stein 1883	80	4.76			80	2.38	80	2.38		
<i>Tryblionella compressa</i> (Bailey) Poulin	160	19.05	80	2.38	160	4.76			80	9.52
<i>Prorocentrum cordatum</i> (Ostenfeld) J.D. Dodge	1428	47.62	2142	33.33	5712	35.71	2142	42.86	2142	35.71
<i>P. micans</i> Ehrenberg	1071	35.71	714	40.48	1428	40.48	1428	33.33	1428	59.52
<i>P. triestinum</i> J. Schiller	400	2.38	4284	23.81	8568	26.19	2142	23.81	1428	21.43
<i>Protoceratium</i> spp.	80	2.38								
<i>Protoperidinium conicum</i> (Gran) Balech					80	4.76				
<i>P. crassipes</i> (Kofoid) Balech	160	2.38	240	11.9	714	7.14	80	4.76	160	4.76
<i>P. diabolium</i> (Cleve) Balech	80	7.14	80	4.76			160	2.38	400	7.14
<i>P. divergens</i> (Ehrenberg) Balech			80	2.38						
<i>P. globulum</i> (F. Stein) Balech	80	4.76	80	2.38	160	2.38				
<i>P. pallidum</i> (Ostenfeld) Balech							160	4.76		
<i>P. pellucidum</i> Bergh	80	11.9	714	9.52	160	4.76	160	4.76	160	2.38
<i>P. steinii</i> (Jørgensen) Balech	240	9.52	160	9.52	80	14.29	80	7.14		
<i>P. tuba</i> (J. Schiller) Balech	160	14.29	714	11.9	240	11.9	320	11.9	160	16.67
<i>Protoperidinium</i> spp.	80	4.76	240	19.05	714	19.05	714	14.29	160	14.29
<i>Scrippsiella</i> sp.	2142	40.48	2142	38.09	4998	21.43	4284	42.86	800	47.62
<i>Tripas azoricus</i> (Cleve), F. Gómez	80	2.38								
<i>T. candelabrum</i> (Ehrenberg) F. Gómez			160	4.76					80	2.38
<i>T. gibberus</i> (Gourret) F. Gómez			240	2.38	240	4.76	240	2.38	160	4.76
<i>T. furca</i> (Ehrenberg) Gómez	480	7.14	160	14.29	400	11.9	714	21.43	714	33.33
<i>T. fusus</i> (Ehrenberg) F. Gómez	714	16.67	80	2.38	240	16.67	240	9.52	160	11.9
<i>T. horridus</i> (Cleve) F. Gómez			160	2.38						
<i>T. setaceus</i> (Jørgesen) Gómez	960	23.81	160	21.43	714	14.29	240	9.52	357	4.76
<i>T. macroceros</i> (Ehrenberg) Hallegraeff & Huisman			80	2.38	160	4.76				
<i>T. massiliensis</i> (Gourret) F. Gómez							80	2.38		
<i>T. muelleri</i> Bory de Saint-Vincent	240	14.29	240	14.29	240	11.9	240	16.67	160	7.14
<i>T. teres</i> (Kofoid) F. Gómez	80	2.38	80	7.14	80	2.38	80	2.38	80	2.38
<i>Tripas</i> spp.										
Coccolithophorides										
<i>Calciosolenia brasiliensis</i> (Lohmann) J. R. Young	714	16.67	320	9.52	320	7.14			714	9.52
<i>Calyptrosphaera oblonga</i> Lohmann	5355	66.67	3570	57.14	2499	66.67	3570	57.14	4998	59.52
<i>Helicosphaera walichii</i> (Lohmann) Okada & McIntyre	2142	9.52	240	2.38	160	4.76				
<i>Rhabdosphaera tignifer</i> J. Schiller	1428	33.33	2856	61.9	2142	45.24	2142	28.57	2142	38.09
<i>Syracosphaera pulchra</i> Lohmann	2142	66.67	15708	73.81	2856	73.81	2142	61.9	8925	64.29
Silicoflagellates										
<i>Dictyocha fibula</i> Ehrenberg	160	7.14	4998	11.76	160	7.14			80	2.38
<i>Octactis octonaria</i> (Ehrenberg) Hovasse	80	2.38	714	11.76						
Chlorophytes										
<i>Pediastrum duplex</i> Meyen	960	2.38					12138	14.29	42840	59.52
<i>Pediastrum</i> sp.									2160	11.9
<i>Scenedesmus quadricauda</i> (Turpin) Brébisson									2142	9.52

MAX-maximal value (cells/l); FR.-frequency of appearance

major nutrient sources for the region. The interplay between hydrological dynamics and biological processes in these areas sheds light on the complexities of the ecosystem, providing valuable insights for further understanding and management.

During research, regarding the difference in the phytoplankton community in two areas, the bay area and area outside of the bay, the reported values showed higher abundances of phytoplankton in the bay area, especially at locations in its inner part (Kotor Bay) (Figure 5). Notably, during the precipitation season, this area is under higher pressure from the land and under the influence of numerous streams and springs located there. The values of phytoplankton in the current research for the bay area were similar to values (abundances reached values of 10^5 and rarely 10^6 cells/l) previously found in Boka Kotorska Bay (Drakulović et al., 2012; 2017; Bosak et al., 2012) and for the northeastern (Burić et al., 2007; Bosak et al., 2009), middle (Skejić et al., 2014) and southern Adriatic Sea (Saracino & Rubino,

2006). The abundances recorded in this study were one order of magnitude lower than those from the northern Adriatic Sea, both from the western (Bernardi Aubry et al., 2004; Totti et al., 2019) and eastern (Cabrini et al., 2012; Cerino et al., 2019) sides.

Temporarily, the highest values of phytoplankton in these two investigated areas were recorded in winter and spring. These two areas during both investigated years showed slight differences in the occurrence of maximal values of phytoplankton. In Boka Kotorska Bay, the highest values were in the spring, while in the coastal open area, it was during the winter – late winter and spring period. In a previous study, Drakulović et al. (2012) recorded the highest value during late winter-early spring for the Boka Kotorska Bay area. When these higher values were noticed, the concentration of nutrients was generally lower, which could be due to their adoption by phytoplankton. Cerino et al. (2012) and Viličić (1983; 1989) recorded that phytoplankton abundances in the South Adriatic were low in winter,

Table 6. Spearman’s rank order correlation matrix for physico-chemical and biological parameters for Boka Kotorska Bay

	TP (logcells/l)	Diatoms (logcells/l)	Dinoflagellates (log cells/l)	Coccolithophores (logcells/l)
Tem. (°C)	-0.0767	-0.1764	0.2758	0.2948
Sal. (psu)	-0.1236	-0.1259	0.1294	0.1902
NO ₃ ⁻ (μmol/l)	0.0564	0.1082	-0.2058	-0.2625
PO ₄ ³⁻ (μmol/l)	0.0683	0.1462	-0.1824	-0.1321
SiO ₄ ⁻ (μmol/l)	0.0065	-0.0201	-0.0688	-0.1069

Table 7. Spearman’s rank order correlation matrix for physico-chemical and biological parameters for open coastal part

	TP (logcells/l)	Diatoms (logcells/l)	Dinoflagellates (log cells/l)	Coccolithophores (logcells/l)
Tem. (°C)	-0.2199	-0.3251	0.3740	-0.0537
Sal. (psu)	-0.3448	-0.3072	0.1601	-0.0324
NO ₃ ⁻ (μmol/l)	0.2697	0.2762	-0.1179	-0.0266
PO ₄ ³⁻ (μmol/l)	0.3193	0.3366	-0.1540	0.0057
SiO ₄ ⁻ (μmol/l)	0.3681	0.2743	-0.1039	0.1449

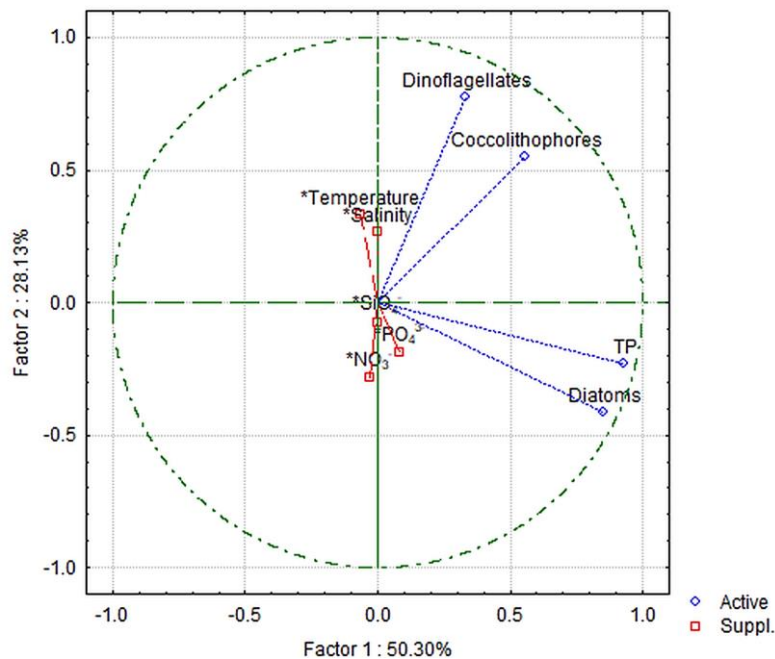


Figure 9. PCA of the relationship between environmental parameters and phytoplankton groups abundance in the Montenegrin waters from 2019-2020. Principal component analysis (PCA) of environmental variables (temperature, salinity, nitrates (NO₃⁻), phosphates (PO₄³⁻) and silicates (SiO₄⁻) and phytoplankton groups (diatoms, dinoflagellates and coccolithophores).

with higher values in late spring. During short spring blooms recorded in April (Cerino et al., 2012; Viličić, 1989, Ljubomir et al., 2017), the recorded phytoplankton abundance was typical of eutrophicated ecosystems (Viličić, 1989).

In the bay area, a recent study on the temporal distribution of dominant phytoplankton groups - diatoms - revealed a mostly uniform distribution with a slight increase in abundance during late summer. This contrasts with the findings of Drakulović et al. (2012), who observed maximal diatom abundance in late winter-early spring. Drakulović et al. (2017) also noticed differences in the occurrence of maximal abundance during the autumn period in the inner part of the bay. In

the open part of the coastal area, the situation was slightly different, with high values during summer and autumn in 2020, while in 2019, the highest values were during winter and spring. This pattern is generally consistent with findings from the northern Adriatic region (Cabrini et al., 2012; Kraus & Supić, 2011; Marić et al., 2012; Godrijan et al., 2013; Talaber et al., 2014; Totti et al., 2019; Cerino et al., 2019). Regarding phytoplankton succession, diatoms are an adaptable group that is present during almost all investigation periods, while dinoflagellates and coccolithophores were mostly present in warmer, less turbulent periods which was confirmed by PCA analysis.

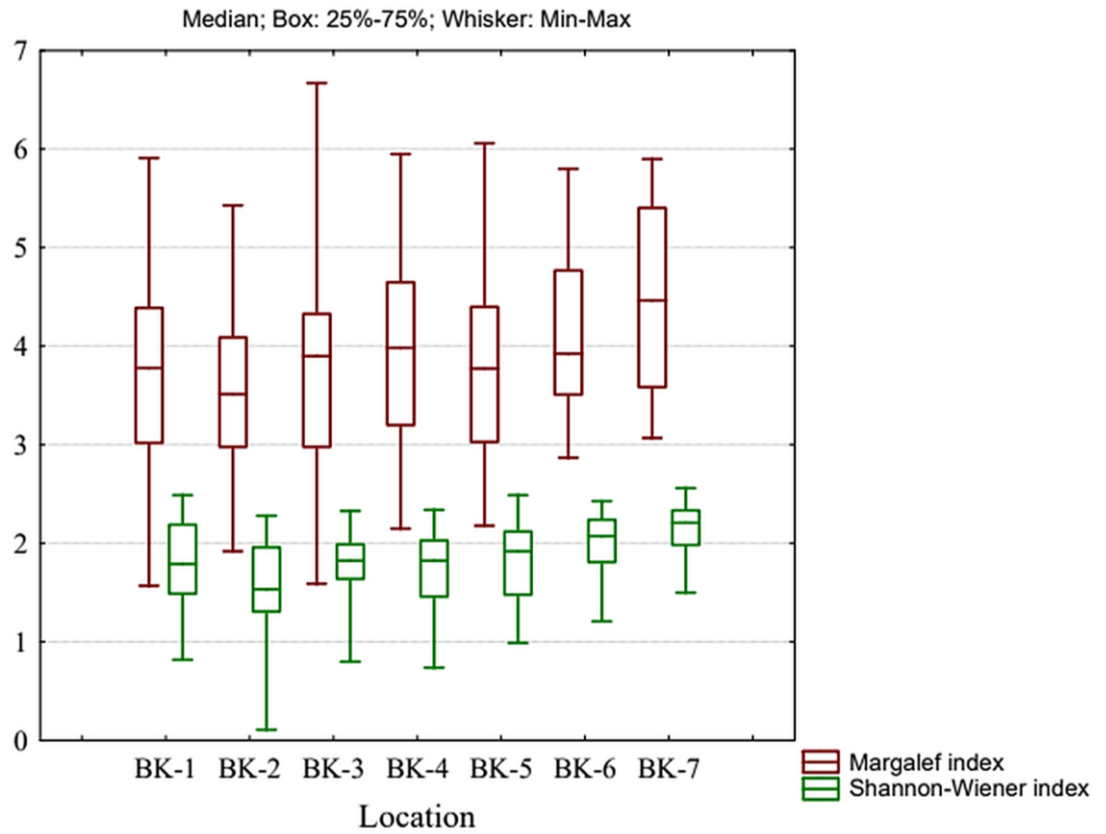


Figure 10. Box-Whisker presentation of the Margalef and Shannon-Wiener diversity indices for the phytoplankton community in the Bay area in the period 2019-2020

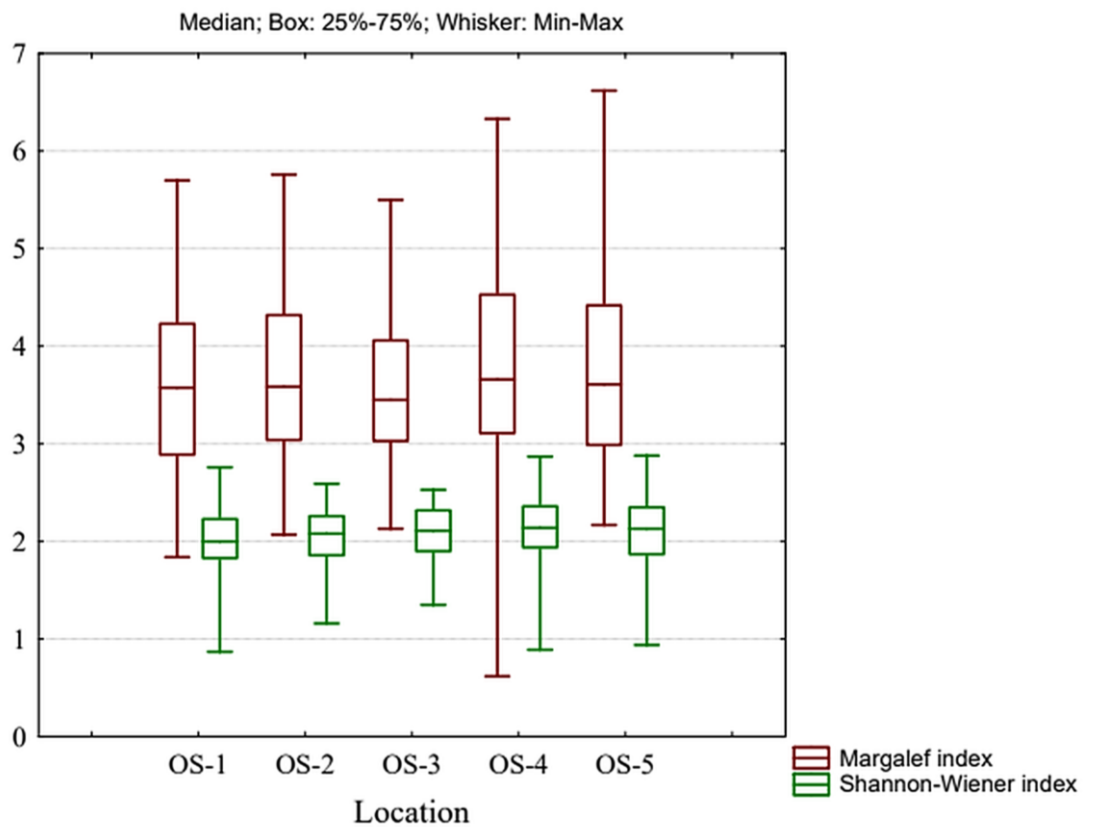


Figure 11. Box-Whisker presentation of the Margalef and Shannon-Wiener diversity indices for the phytoplankton community in the area outside of the bay for period 2019-2020

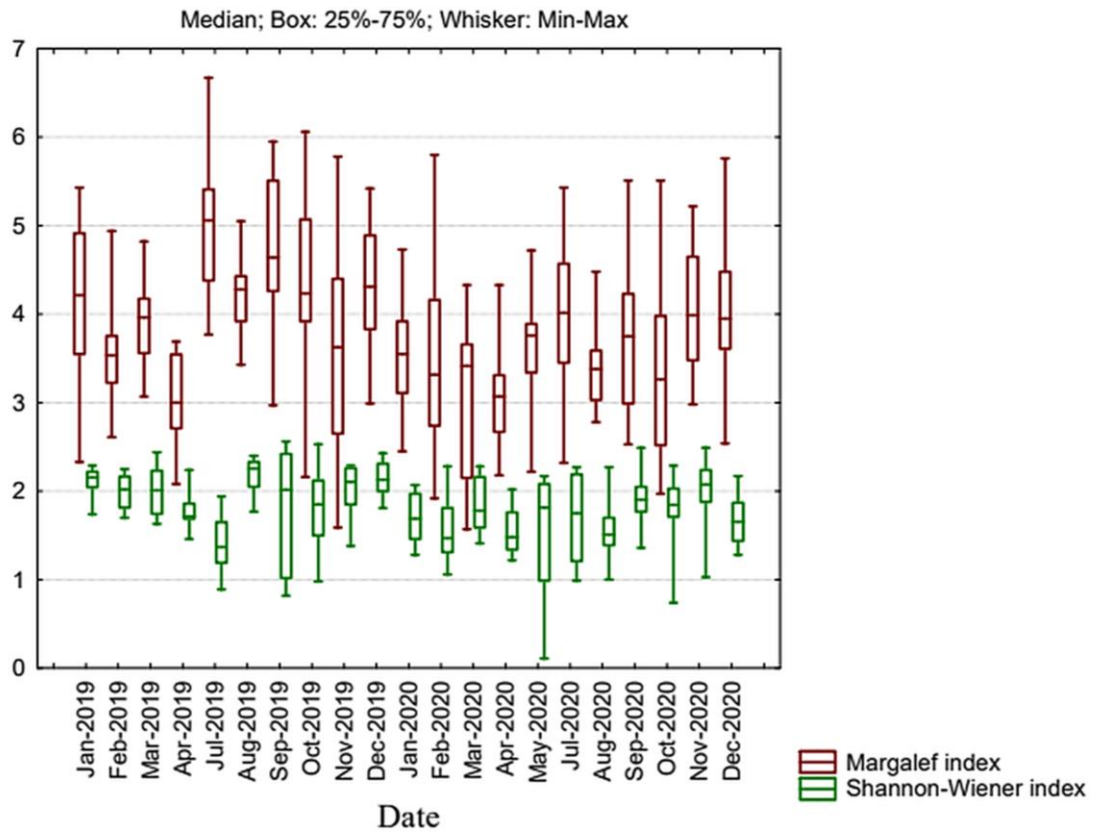


Figure 12. Box-Whisker presentation of the Margalef and Shannon-Wiener diversity indices for the phytoplankton community in the area outside of the bay for period 2019-2020

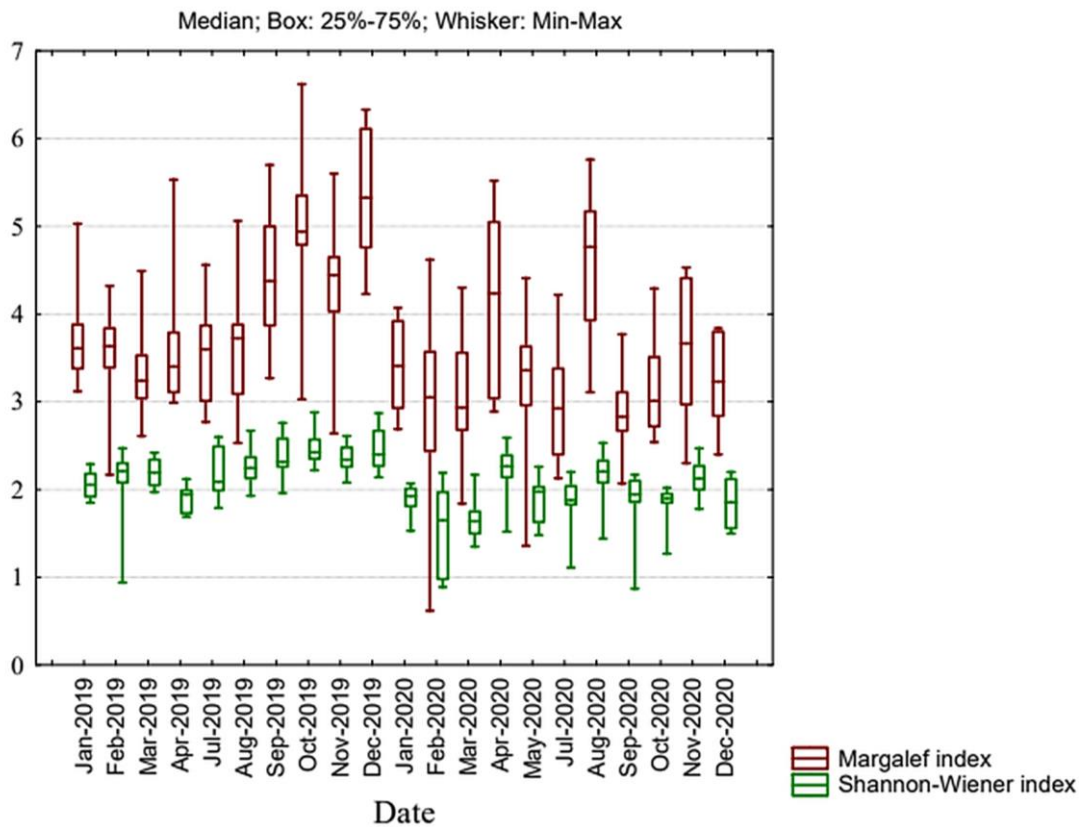


Figure 13. Box-Whisker presentation of the Margalef and Shannon-Wiener diversity indices for the phytoplankton community in the area outside of the bay for period 2019-2020

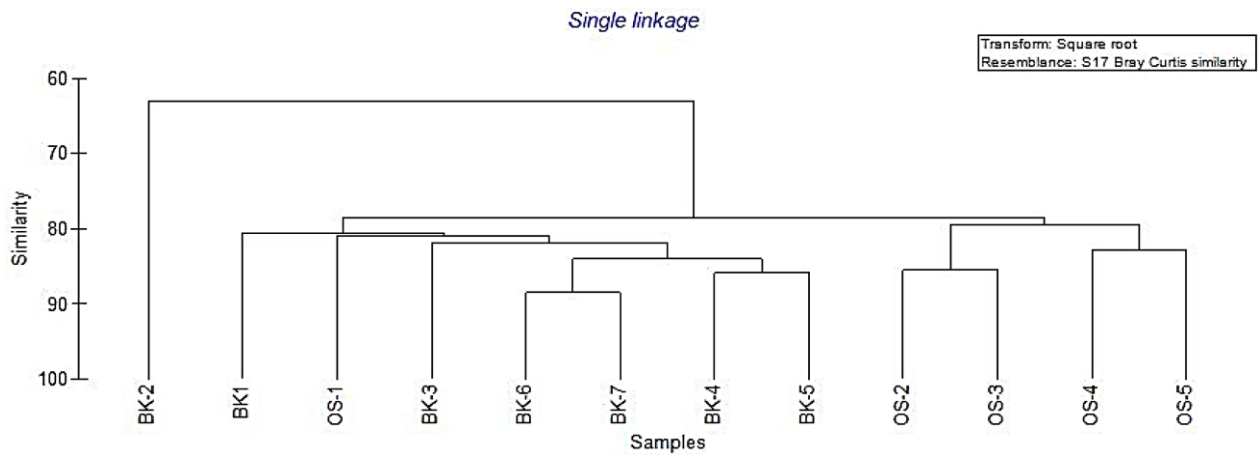


Figure 14. Hierarchical cluster dendrogram for the different locations (Boka Kotorska Bay- BK-1, BK-2, BK-3, BK-4, BK-5, BK-6 and BK-7 and area outside of the bay - OS1, OS-2, OS-3, OS-4, OS-5) versus the absence or presence of species

In our study, most of the dominant diatoms were species that had a preference for nutrient-enriched conditions. Additionally, in the northeastern Adriatic (Bosak et al., 2012), the majority of the dominant diatoms recorded in the study prefer nutrient-enriched conditions. In general, ecosystems rich in inorganic nutrients will support microphytoplankton, while organic nutrients are preferred by nano- and picophytoplankton (Thingstad & Sakshaug, 1990, Turchetto et al., 2000, Matek et al., 2023).

Regarding the presence of phytoplankton taxa in these two areas, the recorded values were slightly higher in the bay area (128 taxa) than in the open part (118 taxa). In recent research related to the area of Boka Kotorska Bay, diatoms were present with 63 taxa, and dinoflagellates were present with 55 taxa, showing slightly higher diversity compared to previous results (Drakulović et al., 2012, 2017; Bosak et al., 2012). Drakulović et al. (2017) identified a total of 100 taxa, which is comparable with the results of Drakulović et al. (2012) for the same bay area, where 109 taxa were identified. In the open coastal part, the ratio of phytoplankton groups was similar to that in the bay area, with 58 diatoms, 50 dinoflagellates, 5 coccolithophores, 2 silicoflagellates, and 3 chlorophytes.

The dominance of diatoms was previously observed in Boka Kotorska Bay (Drakulović et al., 2012, 2017) and the northern Adriatic Sea (Burić et al., 2007; Totti et al., 2019; Aubry et al., 2004; Aubry et al., 2021) on both sides. In the middle of the Adriatic Sea (Skejić et al., 2014) and the northern Adriatic Sea (Cabrini et al., 2012; Cerino et al., 2019), diatoms and flagellates were equally represented. On the other hand, in the southern Adriatic Sea (Saracino & Rubino, 2006), dinoflagellates were the most identified taxa (55% in autumn and 58% in spring), while diatoms constituted 32% of the total taxa in both sampling periods.

The phytoplankton community in both areas was predominantly composed of diatoms, with fewer dinoflagellates. *Pseudo-nitzschia* spp. stood out as the dominant diatom, present in 100% of samples from both locations. The dominance of *Pseudo-nitzschia* spp. was

also observed in the Bay area (Drakulović et al., 2012, 2017), with higher values reported in the current study compared to previous findings by Drakulović et al. (2017). This genus has been consistently present in the Mediterranean and Adriatic Seas (Socal et al., 1999; Orsini et al., 1992; Quiroga, 2006; Bosak et al., 2009; Totti et al., 2019). A total of 33 species (updated by current IOC-UNESCO Taxonomic Reference List of Harmful Micro Algae) were identified, of which mostly are capable of producing domoic acid (Quiroga, 2006; Bates et al., 1998). As a result, there has been increased interest in *Pseudo-nitzschia* spp. in the Adriatic Sea.

In the middle Adriatic area, particularly in the western coastal area, Totti et al., 2000 observed a remarkable maximal value of diatoms attributed to the growth of diatom *Pseudo-nitzschia* spp., with abundances reaching up to 10^6 cells/l. A similar finding was reported for the southern Adriatic Sea by Caroppo et al., 2005 with abundances reaching up to 10^5 cells/l.

During the study, several species were found to be more frequent and abundant, including *Proboscia alata*, *Thalassionema nitzschioides*, and some species from the genus *Chaetoceros*. These findings were consistent with a previous study by Drakulović et al. (2012), where the dominance of *Thalassionema nitzschioides* (abundance of 1.57×10^5 cells/l and frequency of 69%) was also recorded. Dominance of *Thalassionema nitzschioides* (with a frequency of 56%) and species from the genus *Chaetoceros* were similarly noted in the south Adriatic Sea by Saracino and Rubino (2006). In the northern Adriatic, both on the western (Totti et al., 2019) and eastern (Cerino et al., 2019) sides, the dominance of *Proboscia alata*, some *Chaetoceros* species, and *Pseudo-nitzschia* spp. were likewise recorded during the investigations.

In comparison to the investigation of Boka Kotorska Bay by Drakulović et al. (2017), the frequency of toxic dinoflagellates was lower, with *Dinophysis acuminata* having a frequency of 33.33%, *D. fortii* with a frequency of 23.19%, and *Prorocentrum cordatum* with a frequency of 21.74%.

Overall, these findings shed light on the diverse and dynamic phytoplankton communities in different areas of the Adriatic Sea and provide valuable insights into the distribution and abundance of various species, including those with potential ecological significance.

For the northwestern part of the Adriatic Sea, Totti et al. (2019) recorded the presence of certain species that have the potential to trigger harmful algal blooms (HABs) during the period from early spring to late summer. These species include dinoflagellates such as *Alexandrium minutum*, *Dinophysis caudata*, *D. fortii*, *D. sacculus*, *Gonyaulax polygramma*, *G. spinifera*, and *Prorocentrum rhathymum*, as well as diatoms such as *Halamphora coffeiformis*, *Pseudo-nitzschia delicatissima*, *P. pseudodelicatissima* complex, *P. fraudulenta*, *P. galaxiae*, and *P. pungens*.

The Spearman's and PCA results suggest that among the abiotic parameters, temperature, salinity and inorganic nitrogen and phosphates availability had the greatest influence on phytoplankton variability during our investigation. An even higher impact was recorded for biotic variables such as dinoflagellates and coccolithophores, which determined a great part of the overall variability in the summer phytoplankton community. These indicated that the nature of relationships within the plankton community was affected by the supply of nutrients.

Finally, the test shows a negative correlation with nitrate and silicate that are therefore consumed at the site.

Environmental pressures can have negative impacts on phytoplankton indices by favoring only the most stress-tolerant taxa. Eutrophication stress in phytoplankton communities often leads to massive blooms of a few species, resulting in strong dominance (Francé et al., 2021; Cozzoli et al., 2017). In our study, the results from the phytoplankton diversity indices exhibit similar fluctuations in the bay area, which is more influenced by human factors than the open sea. This finding is consistent with research by Skejić et al. (2014), where it was observed that sewage effluents in the Brač Channel did not negatively impact phytoplankton diversity. Instead, a mild increase in phytoplankton diversity was noted throughout the investigative period, supporting the hypothesis that moderate nutrient enrichment stimulates diversity (Spatharis et al., 2007). In our case, in the more closed area of the bay, under higher pressure from land-based activities, there were no records of higher values of indices compared to the open sea. On the other hand, France et al. (2021) conducted large-scale testing of phytoplankton diversity indices for environmental assessment in the Mediterranean subregions (Adriatic, Ionian, and Aegean Seas), and they noticed that a decrease in diversity and the predominance of a single taxon or a few taxa were only evident at locations with higher anthropogenic impacts.

The recent study highlighted variations in the temporal distribution of phytoplankton indices, with the

highest values being observed during the summer period, late summer-early autumn, and winter period. In the Lim Bay, Bosak et al. (2009) reported that both species richness (d) and the biodiversity index (H') exhibited high values during autumn, in contrast to lower values observed in summer, despite similar phytoplankton cell abundances. The high diversity recorded during autumn was mainly attributed to the presence of a variety of planktonic diatom species, including *Chaetoceros* and *Bacteriastrium*. During an investigation of phytoplankton composition and distribution along the Albanian coast, Saracino and Rubino (2006) recorded the highest value for the Shannon & Wiener index in April 2012 compared to October 2020. Interestingly, this recorded value of the Shannon & Wiener index aligns with our own findings. These observations highlight the dynamic nature of phytoplankton communities and underscore the importance of seasonal and regional factors in shaping phytoplankton community diversity and distribution.

Conclusion

In this study, the peak phytoplankton abundances were recorded in the bay area on the order of up to 10^6 cells/l, as this part is under higher human pressure than the open part; hence, phytoplankton growth will be higher in this part. Diatoms dominated throughout the entire research period, while dinoflagellates were the second most abundant group. The peak phytoplankton abundances recorded in the bay area and the high frequency of eutrophic species from the genera *Pseudo-nitzschia* and *Thalassionema nitzschioides* both suggest a slow increase in anthropogenic influences primarily in Boka Kotorska Bay (Drakulović et al., 2017; Bosak et al., 2012). The presence of diatom *Pseudo-nitzschia* spp. is important due to the possibility of producing domoic acid (Ujević et al., 2010). Thus, in the future, this region may become a eutrophic area, where toxicity events can be expected. Therefore, sustained monitoring is advised. The present results of phytoplankton assemblages and distribution provide valuable information for this part of the Montenegrin coast, especially as there is a lack of data for the area outside of the bay. However, hopefully, more research will be conducted in this area in the future, thus providing reliable data for comparison, especially for the area outside of the bay where data and information are lacking.

Ethical Statement

The study does not involve any live animals and human subjects and ethical statements are not applicable.

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Author Contribution

Conceptualization, D.D. and A.H.; methodology, D.D. and B.P.; software, B.P.; validation, D.D. and D.Š.; formal analysis, D.D. and D.Š.; investigation: D.D. and A.H.; resources, D.D. A.H., B.P and D.Š.; data curation, B.P.; writing—original draft preparation, D.D.; writing—review and editing, D.D., A.H., B.P and D.Š.; visualization, D.D.; supervision, A.H., B.P and D.Š.; All authors have read and agreed to the published version of the manuscript.

Conflict of Interest

The authors declare that they have no known competing financial or non-financial, professional, or personal conflicts that could have appeared to influence the work reported in this paper.

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