Microphytobenthic biomass, species composition and nutrient availability in sublittoral sediments of the Gulf of Trieste (northern Adriatic Sea)

Tamara Cibic [a],[*], Oriana Blasutto [b], Claus Falconi [a], Serena Fonda Umani [c]

[a] *Biological Oceanography Department, National Institute of Oceanography and Experimental Geophysics, Via A. Piccard 54, 34014 Trieste, Italy*
[b] *Regional Environmental Protection Agency—FVG, Piazza Collalto 15, 33057 Palmanova, Udine, Italy*
[c] *Department of Biology, University of Trieste, Via A. Valerio 28/A, 34127 Trieste, Italy*

Abstract

Monthly sampling was carried out during a 2-year study (2003–2004) in order to analyse benthic microalgal composition, abundance (ABU) and biomass (BIOM). Temperature and photosynthetically available radiation (PAR) at the bottom were recorded during sampling. In the overlying water nitrite (NO_2^-), nitrate (NO_3^-), ammonium (NH_4^+), phosphate (PO_4^{3-}) and silicate (SiO_3^-) were analysed. The sediment consisted of 8.2% sand, 18.3% clay and 73.5% silt. BIOM showed its maximum in August 2004, while the minimum was recorded in October 2003. The microphytobenthic community was mainly composed of diatoms. Among all the 103 Bacillariophyceae taxa identified in the sediment, we distinguished 67 benthic, 8 epiphytic and 8 planktonic species. Among diatoms *Nitzschia* and *Navicula* were the most abundant genera (30.8 and 26.2%, respectively). The linear regression between total diatom ABU and PAR was statistically significant ($r = 0.66$, $p < 0.001$). A seasonal pattern of monthly samplings was highlighted both from cluster analysis and principal component analysis (PCA). The latter revealed two diatom assemblages: a winter assemblage and a spring–summer one. The genera *Diploneis* and *Pinnularia* showed an inverse relation against temperature. Considering the nutrient ratios the development of microphytobenthos (MPB) appeared to be potentially co-limited by Si and P almost throughout the study period. A clear inverse relation between NO_2^-, NO_3^- and BIOM was emphasised by the moving average and the PCA. The inverse relation among nutrients and light availability suggested that the photosynthetic activity of benthic diatoms in spring and summer may be one of the processes controlling sediment–water nutrient fluxes.

1. Introduction

The importance of diatoms and other microalgae in marine intertidal and shallow water benthic habitats has long been recognised. Comprehensive reviews of the biology and ecology of these organisms have been published by Round (1971), McIntire and Moore (1977), MacIntyre et al. (1996) and Cahoon (1999). A recent compilation, based on 85 worldwide studies, underlined the relevance of microalgae for ecosystem primary production in a number of estuarine, intertidal and shallow-water littoral environments (Cahoon, 1999). Considering the primary production both in the water column and in the sediment, the microphytobenthos (MPB) may contribute up to 50% of the total primary production (Perissinotto et al., 2002; Montani et al., 2003). MPB play an important role in the benthic and pelagic trophic web, constituting a substantial food source for sediment feeders (macro-meiobenthos) (Montagna et al., 1995; Buffan-Dubau and Carman, 2000). Moreover, MPB may influence the flux of inorganic nutrients

* Corresponding author.
E-mail addresses: tcibic@ogs.trieste.it (T. Cibic), blasuttoo@arpa.fvg.it (O. Blasutto), cfalconi@ogs.trieste.it (C. Falconi), labbioma@univ.trieste.it (S. Fonda Umani).

between sediment and water, acting as a filter (Welker et al., 2002; Sundbäck et al., 2004). The function of this filter is directly linked to nutrient uptake by microalgae both from the overlying water and sediment porewater (Sundbäck et al., 2000). The photosynthetically active microphytobenthic community has been shown to influence the chemical and physical properties of coastal sediments, influencing the nitrogen turnover process (Fredericks et al., 1999; Risgaard-Petersen, 2003) and the exchange rates for PO_4^{3-} at the sediment−water interface (Rizzo, 1990), and finally allowing the aerobic degradation of autochthonous and settled organic matter in sediments (Barranguet, 1997). While P has been most frequently inferred as the first limiting nutrient in coastal water, co-limitation have also been reported. N and Si can limit algal growth potential in seawater when light and temperature are adequate (Maestrini et al., 1997). Changes in the relative availability of N and Si may influence the relative as well as absolute abundance of various diatom species. A switch from N to Si limitation of diatom growth can influence both the quantity (cell numbers and biomass) and quality (composition of this biomass) of a diatom population (Gilpin et al., 2004 and references therein).

While the importance of inter-tidal benthic microalgae in near-shore foodwebs has been recognised and studied extensively over the past twenty years, the role of sub-tidal benthic microalgae has received vastly less attention. Similarly, in the northern Adriatic Sea, and particularly in the Gulf of Trieste, MPB and benthic nutrient fluxes have been investigated over the past two decades (Herndl et al., 1989; Bertuzzi et al., 1997; Sdrigotti and Welker, 2002; Welker et al., 2002). The

present work aimed to examine the abundance, distribution and species composition of the microphytobenthic community associated with the sediment in a sublittoral coastal area of the Gulf of Trieste (northern Adriatic Sea) during a 2-year period (2003−2004). Special emphasis was placed on the relation among benthic diatoms and PAR, temperature and inorganic nutrient availability.

2. Materials and methods

2.1. Study site

The Gulf of Trieste, located at the north-western end of the Adriatic Sea, is a shallow basin of about 500 km² and a coastline of about 100 km. It is almost completely surrounded by land except to the southwest and it is connected to the rest of the Adriatic Sea by a sill located in the southern part of the basin (Ogorelec et al., 1991); 10% of its area is <10 m and the maximum depth is about 25 m. The study was carried out at a coastal station (C1), 200 m offshore (45°42.05′ N, 13°42.60′ E) at a depth of about 17 m, within the Marine Reserve of Miramare, sheltered from boats, fishing and swimmers, representing a relatively undisturbed area (Fig. 1). The sediment texture is mud, with <10% of sand and a detrital component.

2.2. Sampling

Monthly sampling was performed from January 2003 to December 2004, but not in October 2004. Virtually
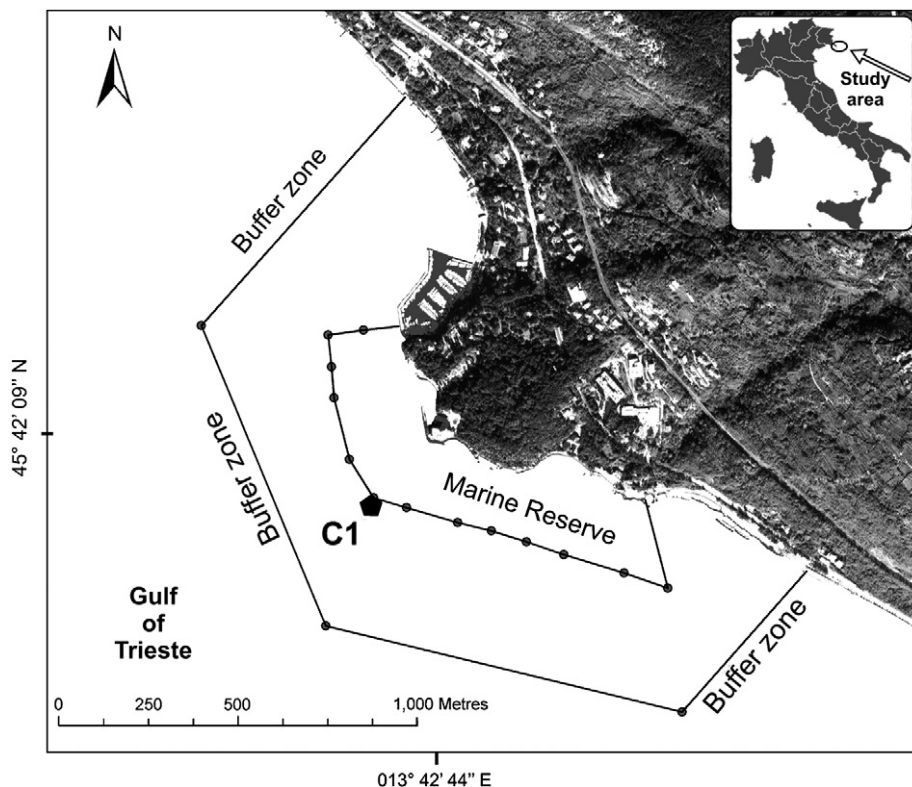

Fig. 1. Location of the study site in the Gulf of Trieste.

undisturbed sediment cores were taken by a KC Haps bottom corer (KC-Denmark, Silkeborg, Denmark) using a polycarbonate sample tube (13.3 cm i.d. with a sample area of 127 cm^2). At the moment of sampling PAR and sea water temperature were recorded *in situ* by a Profiling Natural Fluorometer PNF-300A (Biospherical Instruments Inc., San Diego, CA, USA), 50 cm above bottom. Both samplings and PAR measurements were performed at the same time of the day (ca. noon) each month.

2.3. Granulometry

Since there are no direct riverine or other inputs near the sampling station, the sediment granulometry did not change significantly during the sampling period (Cibic, pers. commun.). For this reason the grain-size analysis was performed only once. Sediment sample of about 10–15 g was taken for grain-size analysis. Small pebbles and shell fragments (>2 mm) were separated from the sand and mud fractions by sieving at 2 mm. Since a few grams of sediment are not representative of the gravel grain size, the gravel percentage is to be considered only a rough estimate. Each sample was disaggregated and the organic matter oxidised using 10% hydrogen peroxide (Carlo Erba, Rodano, Italy) at 60 °C for 24 h, then wet-sieved at 62.5 μm in order to separate sand from mud. The sand fraction (2000–62.5 μm) was analysed in a settling tube (Macrogranometer with tube length of 180 cm and internal diameter 20 cm) and the mud fraction on a Micromeritics ET5000 Sedigraph (Micromeritics Instrument Corporation, Norcross, GA, USA), using a 0.5% sodium hexametaphosphate dispersing solution (Carlo Erba, Rodano, Italy). Grain-size data were represented at $^1/_2$ phi intervals.

2.4. Nutrient analyses

Overlying water for the determination of dissolved inorganic nutrient concentrations (NO_2^-, NO_3^-, NH_4^+, PO_4^{3-} and SiO_3^-) was sampled and filtered through Millipore HA filters 0.45 μm pore size (Millipore Corp., Billerica, MA, USA). Analyses were conducted at room temperature on a five-channel Bran+Luebbe Autoanalyzer 3 Continuous Flow Analyzer (Bran+Luebbe, Norderstedt, Germany), using standard procedures (Bran+Luebbe, 2003a,b,c,d,e and references therein). The efficiency of the system was checked before and after sample analyses by doing replicates of internal standards. Dissolved inorganic nitrogen (the sum of NO_2^-, NO_3^- and NH_4^+) is referred to as N.

2.5. Microphytobenthic abundance

Subsamples for ABU and species composition were taken with cut-off plastic syringes (i.d. 10 mm) and frozen at −20 °C. After thawing ABU was determined in the uppermost sediment layer (0–1 cm). Two sediment layer replicates were fixed with 5 ml of 4% formaldehyde (Carlo Erba, Rodano, Italy) buffered solution ($CaMg(CO_3)_2$) in pre-filtered seawater (0.2 μm filter) (Schleicher & Schuell, Dassel, Germany). After

manual stirring, 20 μl aliquots of the sediment suspension were drawn off from the slurries and placed in a counting chamber (Thalassia, Trieste, Italy). Only cells containing pigments and not empty frustules were taken into consideration for ABU and BIOM estimates; their number varied from a minimum of 100 to a maximum of 400. For each sampling 3 replicates were counted under a Leitz inverted light microscope (Leica Microsystems AG, Wetzlar, Germany) (Utermöhl, 1958) using a 32× objective. The qualitative identification of microphytobenthic assemblages was carried out to the genus and, when possible, to the species level using floras of Van Heurck (1899), Hendey (1976), Germain (1981), Dexing et al. (1985), Ricard (1987), Round et al. (1992) and Tomas (1997).

2.6. Biomass estimation

Microalgal biomass was calculated to assess the carbon content of occurring algae varying in shape and size. We used a set of geometric shapes and mathematical equations for biovolume calculations from microscopically measured linear dimensions that included the entire range of microalgal shapes (Hillebrand et al., 1999). The carbon content (μg C) of microalgae was calculated by means of the transformation of cell volume to plasma volume including an estimate of the vacuole volume, and the calculation of cell carbon was in turn based on the plasma volume multiplied by a factor of 0.11 for diatom cells and Phytoflagellates without *theca* (Strathmann, 1967). BIOM expressed as μg C cm^{-3} was obtained multiplying ABU (cells cm^{-3}) by the carbon content of each counted cell.

2.7. Statistical analyses

In order to reduce the environmental noise of BIOM, NO_2^-, NO_3^-, NH_4^+, a weighted three point moving average filter was applied to biotic and abiotic data as follows:

$$y_n = \sum_{k=-1}^{1} c_k x_{n-k}$$

where x_n is the original data, y_n is the smoothed data and $c_k = {}^1/_2$, 1, $^1/_2$ for $k = -1, 0, 1$, respectively (Hamming, 1998). The inverse relation between ABU and temperature was approximated by means of least square regression according to the following formula: $y = a/x + b$. The goodness of fits was assessed applying an *F*-test (Altman, 1999).

Cluster analysis was carried out using PRIMER-5 software. Monthly samplings were gathered in four seasonal groups based on bottom temperature: January, February and March as winter; April, May and June as spring; July, August and September as summer; October, November and December as autumn. Square root was employed to transform the data matrix and Bray–Curtis similarity index with a complete linkage was calculated (Clarke and Warwick, 2001).

Principal component analysis (PCA) based on *r* algorithm (correlation coefficient) was performed using MATEDIT

software (Burba et al., 1992). The number of species was reduced performing a ranking of the variables which were ordered following their decreasing specific variance. The first 15 species, which corresponded to a cumulative variance of 99.1%, were used (Orloci, 1978). Simultaneous ordination of species, abiotic variables and sampling months was obtained using first and second autovectors and first and second principal components. The data matrix for PCA was constructed as follows: biotic (15 species) and abiotic variables in rows and samplings in columns. From this data matrix a correlation coefficient matrix among rows was calculated. We picked a 5% significance level for the correlation coefficients.

3. Results

3.1. Abiotic parameters

PAR at the bottom showed increasing values from January–February to June–July and decreasing ones from August to December, following a seasonal trend. A similar seasonality was also recognised for temperature with minima measured in February and maxima in late summer (Fig. 2a).

Nutrient concentrations, analysed from the overlying water, and Si:N, N:P and Si:P ratios are shown in Table 1. Applying the ratios proposed by Hillebrand and Sommer (1999) to our data, Si limitation was noticed all over the study period except for July 2003, and from July to December 2004. On the contrary, P limitation was observed all over the study period. The average Si:N ratio was 0.55, with the highest ratio in August 2004 (1.41) and the lowest one in May 2004 (0.09). An average N:P ratio of 92.9 was obtained, with the highest ratio in

January 2004 (551.6) and the lowest one in September 2004 (22.3).

Grain-size analysis was performed in April 2004. The sediment consisted of 8.2% sand (2000–62.5 µm), 73.5% silt (62.5–3.9 µm) and 18.3% clay (<3.9 µm).

3.2. Biotic parameters

In 2003 minimum ABU and BIOM were recorded in October (2.22×10^4 cells cm^{-3} and 6.27 µg C cm^{-3}, respectively), while the highest values were reached in May 2003 (9.74×10^4 cells cm^{-3} and 22.56 µg C cm^{-3}, respectively). In 2004 minimum ABU was observed in February (1.96×10^4 cells cm^{-3}) and minimum BIOM in December (7.16 µg C cm^{-3}). During 2004, maxima for both parameters were observed in August (10.64×10^4 cells cm^{-3} and 29.14 µg C cm^{-3}, respectively) (Fig. 2b).

Bacillariophyceae accounted for 96.68% of the total ABU, while phytoflagellates represented 2.85%. Resting cells, Dinophyceae and Cryptophyceae were minor components accounting for 0.38%, 0.07% and 0.02%, respectively. Diatoms represented 96.74% of the total BIOM, while resting cells, phytoflagellates and Dinophyceae accounted for 2.12%, 0.76% and 0.38%, respectively (Table 2). Resting cells which are rich in storage products and especially oil (Round et al., 1992), have a high carbon content, therefore their relative biomass (RB) is high, also with a low relative abundance (RA). On the contrary, phytoflagellates were more numerous than resting cells and their low carbon content led to a minor RB.

Among all the 103 Bacillariophyceae taxa identified in the sediment we distinguished 67 benthic, 8 epiphytic and 8
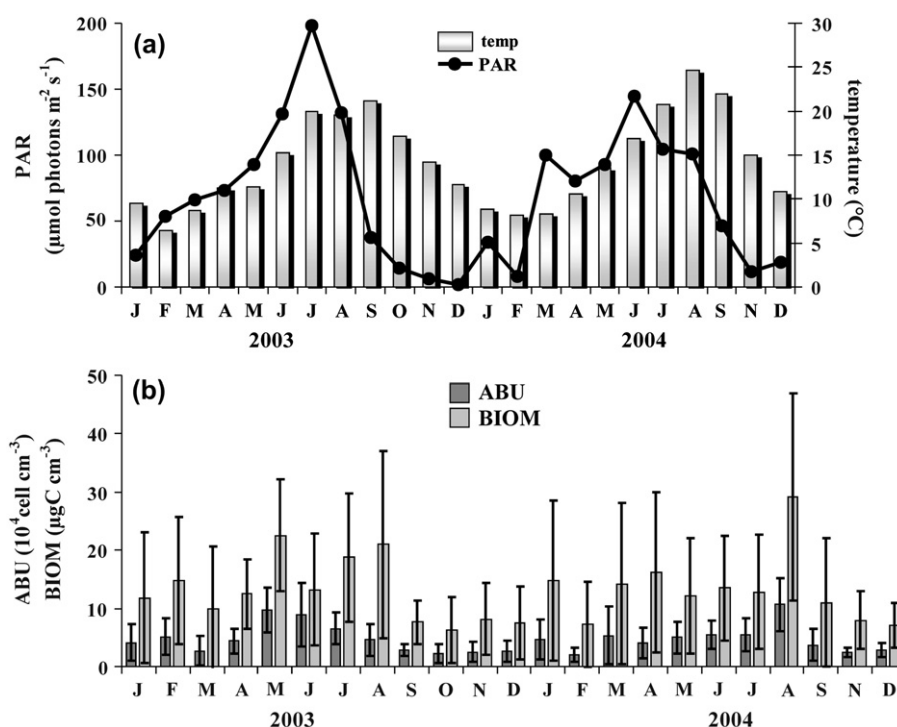

Fig. 2. Variations of temperature and PAR (a), ABU and BIOM (b) during the 2-year study.

Table 1

Nutrient concentrations (μM) in the overlying water and their ratios. N = dissolved inorganic nitrogen. Si/N ≤ 0.79 indicates Si limitation (Brzezinski, 1985); N/P > 22 indicates P limitation (Hillebrand and Sommer, 1999)

Sampling	PO_4^{3-}	SiO_3^-	N	Si/N	N/P	Limitation
Jan-03	0.22	5.50	13.51	0.41	61.7	Si and P
Feb-03	0.19	6.93	16.31	0.42	85.4	Si and P
Mar-03	0.23	7.05	18.93	0.37	82.7	Si and P
Apr-03	0.18	3.00	13.06	0.23	74.2	Si and P
May-03	0.33	9.29	18.36	0.51	55.8	Si and P
Jun-03	0.30	6.31	29.52	0.21	98.4	Si and P
Jul-03	0.19	9.32	6.70	1.39	34.9	P
Aug-03	0.32	20.80	25.11	0.83	78.0	Si and P
Sep-03	0.41	8.05	22.14	0.36	53.6	Si and P
Oct-03	0.25	11.12	55.53	0.20	225.7	Si and P
Nov-03	0.52	10.42	26.41	0.39	51.3	Si and P
Dec-03	0.51	10.20	23.83	0.43	46.5	Si and P
Jan-04	0.04	5.41	20.96	0.26	551.6	Si and P
Feb-04	0.52	3.64	22.11	0.16	42.8	Si and P
Mar-04	0.27	4.06	19.59	0.21	73.1	Si and P
Apr-04	0.18	4.44	8.80	0.50	48.9	Si and P
May-04	0.24	4.50	52.64	0.09	218.4	Si and P
Jun-04	0.25	5.07	12.89	0.39	52.6	Si and P
Jul-04	0.26	16.42	17.60	0.93	67.9	P
Aug-04	0.36	14.24	10.10	1.41	28.1	P
Sep-04	0.81	23.61	18.03	1.31	22.3	Light P
Nov-04	0.46	13.32	21.69	0.61	46.8	P
Dec-04	0.66	25.13	24.25	1.04	36.7	P

planktonic species (for a total of 33 genera and 83 species) (Round, 1971; Cibic et al., 2007) (Appendix A). *Nitzschia* and *Navicula* were the most abundant genera (30.8% and 26.2%, respectively), followed by the genera *Diploneis*, *Pleurosigma* and *Gyrosigma* (7.6%, 4.4% and 3.5%, respectively) (Table 2). Considering the Bacillariophyceae BIOM the genera *Nitzschia* and *Navicula* accounted for 25.0% and 15.7%, respectively, followed by *Gyrosigma*, *Pinnularia* and *Diploneis*. (9.0%, 7.8% and 7.6%, respectively) (Table 2). A significant difference in the genus *Navicula*, when expressed as RA or RB was observed, due to the presence of small sized species (*Navicula* spp. 1, *Navicula mollis* and *Navicula corymbosa*). The most abundant species of diatoms found over the 2-year study were: *Paralia sulcata*, which accounted for 11.1%, *Navicula directa* (5.7%) and *Cylindrotheca closterium* (4.9%) while as BIOM *N. directa* accounted for 7.7%, followed by *Paralia sulcata*, *Nitzschia sigma* and *Nitzschia lorenziana* with 6.2%, 5.1% and 5.1%, respectively (Fig. 3a,b).

Table 2

ABU and BIOM percentages of the main taxa constituting the microphytobenthic community

Taxa	ABU%	BIOM%	Diatom genera	ABU%	BIOM%
Bacillariophyceae	96.68	96.74	*Nitzschia*	30.8	25.0
Undet. Phytoflagellates	2.85	0.76	*Navicula*	26.2	15.7
Spores and cysts	0.38	2.12	*Diploneis*	7.6	7.6
Dinophyceae	0.07	0.38	*Pleurosigma*	4.4	5.3
Cryptophyceae	0.02		*Gyrosigma*	3.5	9.0
Total MPB	100	100	*Amphora*	2.7	5.3
			Pinnularia	0.6	7.8
			Other	24.2	24.3
			Total diatoms	100	100

3.3. Statistical analyses

The correlation between total diatom ABU against PAR was statistically significant, with $r = 0.66$, $p < 0.001$, df = 21 (Fig. 4). Least square inverse power regressions of the genera *Diploneis* and *Pinnularia* against temperature were significantly better ($F_{1,21} = 18.06$, $p = 0.0004$ and $F_{1,21} = 14.11$, $p = 0.001$, respectively) than the regression assumed by the null hypothesis (all regression coefficients equal to zero) (Fig. 5a,b).

The result of the hierarchical clustering is represented in Fig. 6. The dendrogram revealed two major clusters (A, B) at a Bray−Curtis similarity level of 48%. Within group A two subgroups (E, D) were recognised, in which a clear distinction between the year 2003 and 2004 was evident. This interannual distinction was not observed within group B.

Diatom BIOM and N-compound dynamics were better noticed applying the moving average, which highlighted the different pattern of these parameters. A clearly inverse dynamic was observed between BIOM and NO_2^- (Fig. 7a), which was also confirmed by the inverse relation between these two parameters ($r = -0.54$, $p < 0.01$). Overall, a similar dynamic, although with some differences, was detected between BIOM and NO_3^- ($r = -0.44$, $p < 0.05$) (Fig. 7b). The correlation between BIOM and NH_4^+ was not statistically significant. However, an inverse pattern between these two parameters was still recognisable (Fig. 7c).

The data matrix of 23 sampling months, all abiotic parameters and 15 species was used to construct an ordination plot to examine the distribution of the considered variables. Fig. 8 shows an ordination plot that accounts for 41.36% of the total variance. The principal component 1 (PC1) axis explained 24.69% of the total variance and was correlated with the following abiotic parameters (correlation coefficient are indicated in parentheses): NO_2^- (-0.25) and temperature (0.24). The PC2 axis explained 16.67% of the remaining variance and was correlated with PO_4^{3-} (-0.40), SiO_3^- (-0.40), NO_3^- (-0.34) and PAR (0.25). Sampling months could be seasonally gathered in three groups: quadrant I with winter-early spring months, quadrant II and III with late spring−summer months and quadrant IV with autumn months. Going from quadrant I to IV we observed a seasonal pattern of sampling months from winter towards autumn. The position of winter-early spring months in quadrant I was determined by low temperature; only 5 species were placed in this quadrant. Late spring−summer months were located in quadrant II and III due to high PAR and temperature. All the remaining species were situated in these 2 quadrants. No species were placed in quadrant IV due to low PAR. In this quadrant the majority of nutrients were positioned together with autumn samplings.

4. Discussion

PAR, temperature and nutrient availability at the bottom allowed the development of MPB in the investigated area throughout the study period. Miles and Sundbäck (2000) affirm that PAR is the most important factor influencing the development of the microalgal community. The statistically
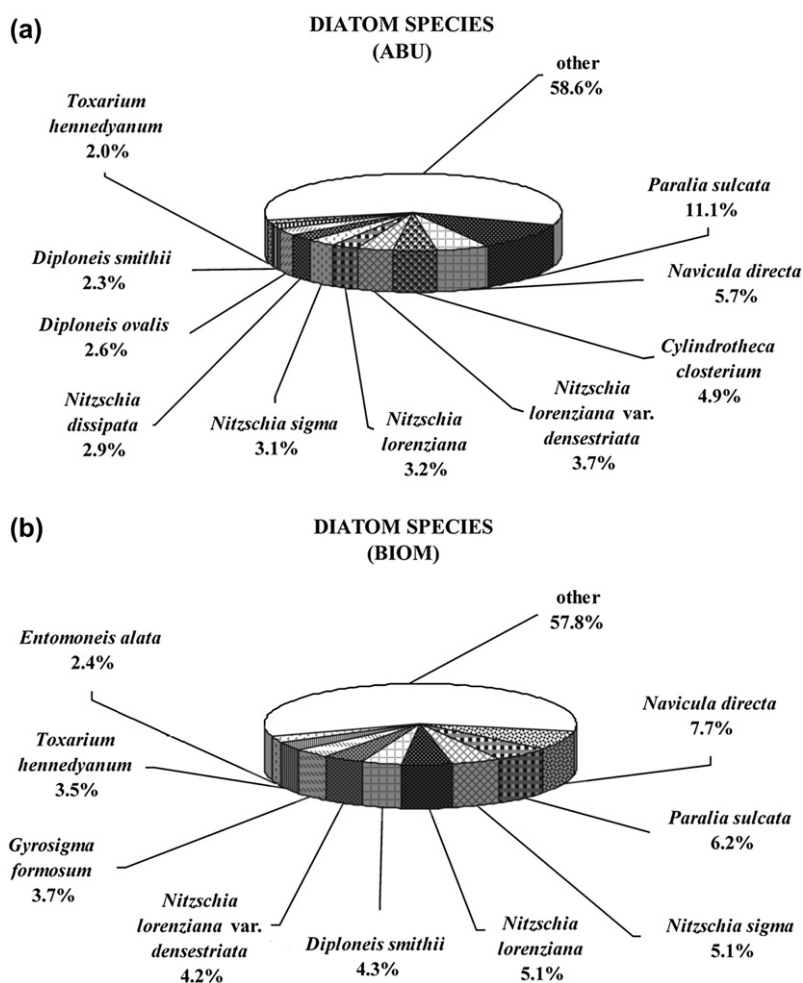
Fig. 3. Dominant diatom species expressed as ABU (a) and BIOM (b).

significant relation between total ABU of benthic diatoms and PAR found in our study confirmed the authors' statement. Besides PAR, high temperature can influence the microphytobenthic community (Montagnes and Franklin, 2001). At our study site, when diatoms were exposed to high temperature (21 °C in September 2003 and 22 °C in September 2004, respectively) for a prolonged period, a decrease of BIOM and ABU was observed.

Throughout the 2-year study the microphytobenthic community was mainly composed of Bacillariophyceae. We discriminated among benthic, epiphytic and planktonic species.
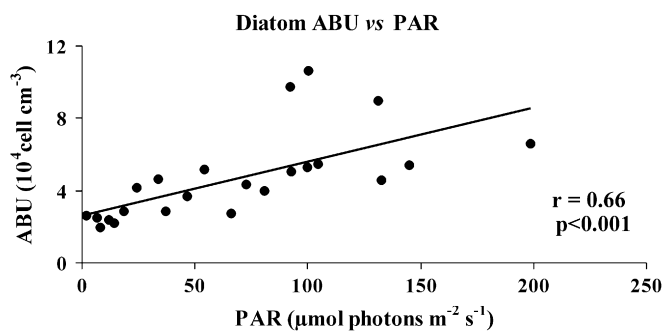
Among benthic species we further distinguished epipsammic and epipelic ones (Cibic et al., 2007). Most of the species was epipelic. Their presence was due to the high percentage of clay and silt in the sediment. Cohesive sediments enable the development of epipelic forms capable of moving through the sediment while sand supports the growth of epipsammic species, which adhere to the grains by means of apical pads, stalks and tubes (Miles and Sundbäck, 2000). In fact, the occurrence of a few epipsammic species among the genera *Amphora* spp., *Cocconeis* spp. and *Cymbella* spp., was due to the low percentage of sand in the sediment (Blasutto et al., 2005). In a previous study performed at the same sampling station we observed that only a few planktonic species became part of the microphytobenthic community. In the same study we inferred that the occurrence of epiphytic species in the sediment is related with the closeness to the coast, where the cells live attached to macroalgae and rocky substrata (Cibic et al., 2007).

ABU and BIOM showed a similar pattern in 2004, while in 2003 a different pattern of the two parameters was observed. In June 2003 ABU was higher than BIOM due to the presence of small sized species with a lower carbon content, i.e. *Cylindrotheca closterium*, very abundant in this month (RA = 26.0%, RB = 4.6%). On the contrary, in August 2003 BIOM was higher than ABU for the presence of a few large sized
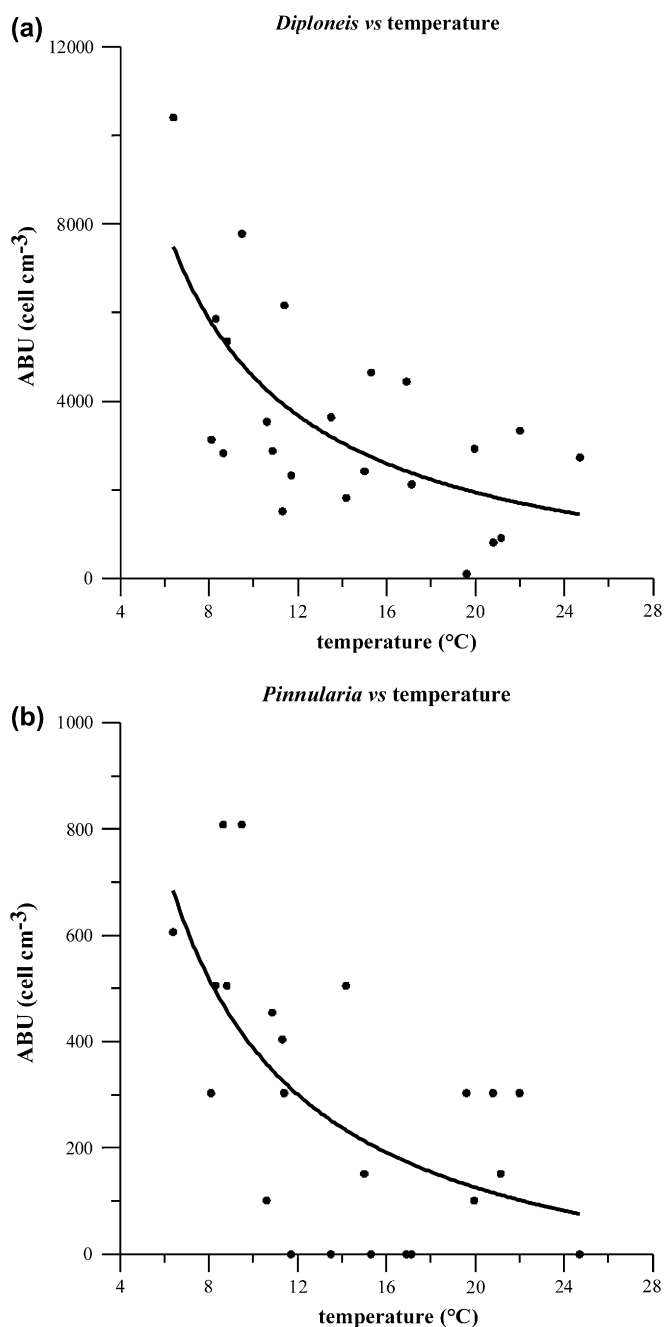

Fig. 4. Linear regression of total diatom ABU against PAR.

(a)

Diploneis vs temperature

(b)

Pinnularia vs temperature

Fig. 5. Least square regressions of the genera *Diploneis* (a) and *Pinnularia* (b) against temperature: interpolating function $y = a/x + b$.

Fig. 6. Cluster analysis, based on Bray–Curtis similarity. Monthly samplings were gathered in four seasonal groups based on bottom temperature.

Gyrosigma spencerii and *Nitzschia lorenziana* var. *densestriata*. In detail, group F was distinguished from AUT-04 and WIN-03 by the presence of *Bacillaria paxillifera* and the absence of several species of the genera *Pleurosigma*, *Gyrosigma* and *Nitzschia*. Group E differed from group D by the presence of *Amphora coffaeformis* and *Pleurosigma aestuarii*,
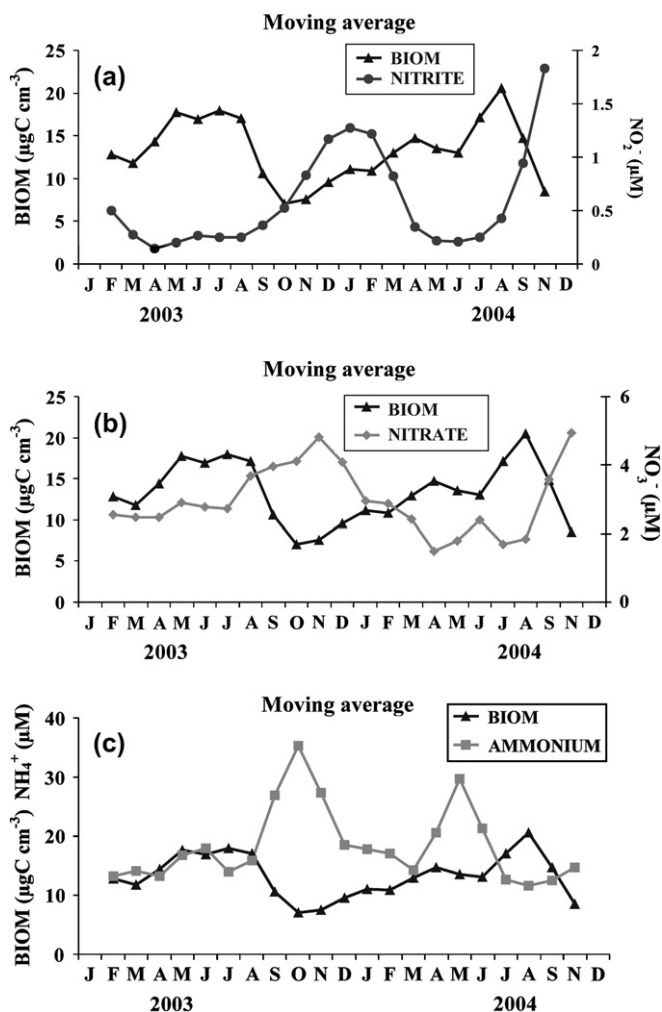
Fig. 7. Three point moving average applied to BIOM and N-compounds: NO_2^- (a), NO_3^- (b) and NH_4^+ (c).

genera, *Synedra* (RA = 3.7%, RB = 16.7%) and *Gyrosigma* (RA = 9.2%, RB = 19.7%).

The cluster analysis pointed out a clear distinction among seasons. Two main groups were identified; one large group gathered all winters and autumns, the other one all springs and summers. This difference between groups A and B was based on the presence or absence of several species. Group B differentiated from group A for the percentage of *Paralia sulcata* (11.0% and 20.0% in groups A and B, respectively), *Navicula* spp.1 (18.8% and 7.8% in groups A and B, respectively) and *Navicula directa* (3.8% and 7.8% in groups A and B, respectively) and the absence of *Gyrosigma macrum*,
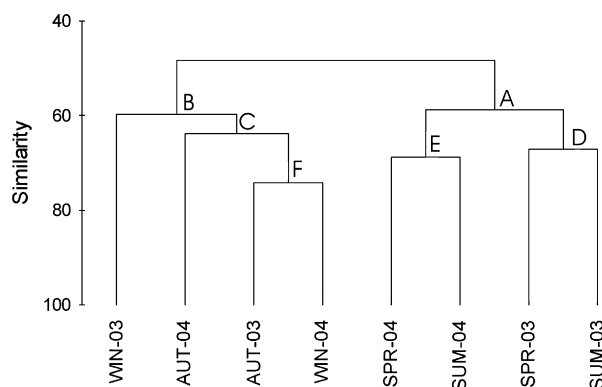
Fig. 8. Principal component analysis, based on *r* algorithm, of species (▲), samplings (●) and abiotic parameters (■) obtained using the first and the second autovectors. *Cylindrotheca closterium* = Cyc, *Diploneis ovalis* = Dio, *Diploneis smithii* = Dis, *Gyr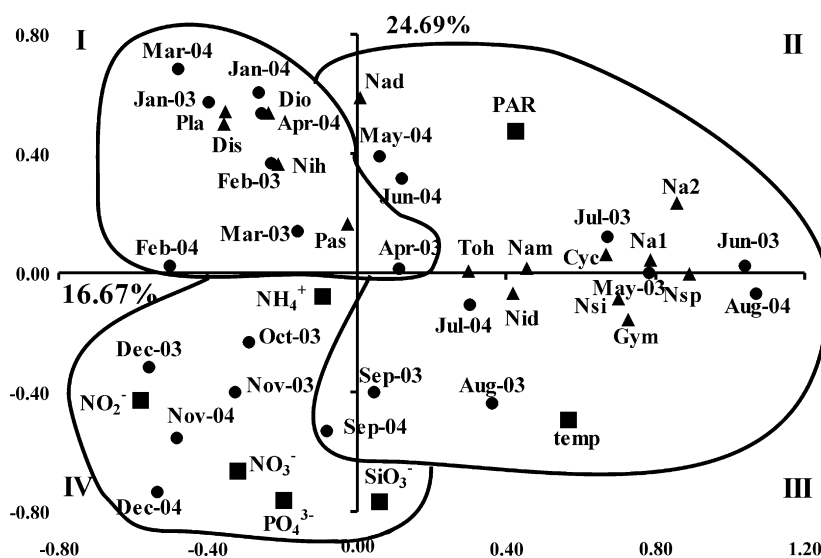osigma macrum* = Gym, *Navicula* spp. 1 = Na1, *Navicula* spp. 2 = Na2, *Navicula directa* = Nad, *Navicula mollis* = Nam, *Nitzschia dissipata* = Nid, *Nitzschia hantzschiana* = Nih, *Nitzschia sigma* = Nsi, *Nitzschia* spp. = Nsp, *Paralia sulcata* = Pas, *Pleurosigma aestuarii* = Pla, *Toxarium hennedyanum* = Toh.

while *Pleurosigma angulatum* and *G. macrum* were missing. Also the percentages of *Paralia sulcata* and *N. directa* were dissimilar between these two groups.

In literature BIOM is mostly estimated as chlorophyll *a* (MacIntyre et al., 1996; Cahoon, 1999; Urban-Malinga and Wiktor, 2003). de Jonge (1980) affirms that chlorophyll *a* is not necessarily a good and conservative unit to describe the quantity of algal biomass. Every biomass value produced is an estimate because none of the available parameters that can be measured (cell counts, organic carbon, chlorophyll *a*, intact diatom frustules, etc.) unambiguously reflects algal biomass expressed as organic carbon (de Jonge and Colijn, 1994 and references therein). These authors reported a carbon: chlorophyll *a* ratio varying from 1 to 40, determined for well-growing natural algae populations. Applying this ratio to our data (where BIOM represents the carbon content), we obtained a range of chlorophyll *a* varying from 0.16 to 29.14 μg cm^{-3}. These chlorophyll *a* values are comparable to those previously reported by Herndl et al. (1989) in the Gulf of Trieste. We

found only a few reports of the specific composition of the microphytobenthic community and cell abundances. There is a lack of a standard unit for ABU, which is often expressed either per area or volume. Moreover, different sampling methodologies and sampling depths are reported in literature making difficult the data comparison (Table 3).

Besides PAR and temperature other factors might control spatial and temporal patterns of the composition and microphytobenthic ABU, like grazing pressure and nutrient availability (Welker et al., 2002). Little work has been done on nutrient limitation in MPB, because it was assumed that this community has access to an inexhaustible nutrient supply from the sediment porewater. Although porewater often contains inorganic nutrients in concentrations several orders of magnitude higher than in the overlying water, MPB appeared to be nutrient-limited in the overlying water (Welker et al., 2002 and references therein). When a specific nutrient becomes limiting or ratios among nutrients are unbalanced, microphytobenthic ABU is determined by the limiting nutrient. Diatoms have an absolute

Table 3
Summary of microphytobenthic abundances from different studies

ABU	Unit	Thickness (mm)	Region	Depth (m)	Reference
$4.0 \times 10^4 - 1.5 \times 10^5$	cells cm^{-3}	5	Ems-Dollard Estuary, Netherlands	Intertidal site	Admiraal et al., 1982
$2.7 - 6.3 \times 10^{4b}$	cells cm^{-3}	10	Grado and Marano Lagoon, Italy	1.5	Blasutto et al., 2005
$1 - 17 \times 10^{6a}$	cells cm^{-2}	10	Long Island, New York, USA	Intertidal site	Cheng et al., 1993
$1.3 - 4.3 \times 10^6$	cells cm^{-3}	3–4	Venice Lagoon, Italy	0.8–0.9	Facca et al., 2002
$4.7 - 7.5 \times 10^6$	cells cm^{-2}	5	Gulf of Finland	1.5	Snoeijs et al., 1990
$0.25 - 2.1 \times 10^6$	cells cm^{-2}	5	Laholm Bay, Sweden	14–16	Sundbäck and Jönsson, 1988
750.30	cells mm^{-3}	5	Lindåspollene, Norway	10	Taasen and Høisæter, 1981
720–9278	cells cm^{-3}	10	Adriatic Sea, Italy	13.5–66	Totti, 2003
$0.1 - 5.7 \times 10^4$	cells g^{-1} sed d.wt.	10	Gulf of Trieste, Italy	20	Welker et al., 2002
$1.9 - 10.6 \times 10^4$	cells cm^{-3}	10	Gulf of Trieste, Italy	17	This study

[a] Value estimated from a figure.
[b] Cells g^{-1} sed d.wt. converted to cells cm^{-3} for data comparison.

requirement for Si (Egge and Aksnes, 1992; Rousseau et al., 2002). Silicic acid is essential for the skeletons of diatoms and its depletion inhibits the cell division and eventually suppresses the metabolic activity of the cell (Levinton, 1995). Si is required in broadly equimolar amounts to N at saturating nutrient concentrations (Dugdale and Wilkerson, 1998; Flynn and Martin-Jézéquel, 2000). Brzezinski (1985) found for 27 diatom species mean Si:N ratios of 1.12 ± 0.33. Thus, Si:N ratios ≤ 0.79 could potentially indicate Si limitation. On the other hand, N:P ratios >22 indicate P limitation for benthic microalgae growth (Hillebrand and Sommer, 1999). Applying these inequalities to our ratios we found Si limitation from January 2003 to June 2004, except for July 2003, and in November 2004 and P limitation all over the study period, except for September 2004. We inferred that all the available PO_4^{3-} was consumed by the benthic microalgae, becoming a growth limiting factor. In June 2004, despite the highest PAR of the year and Si availability, the community appeared to be limited by other factors, probably by P, which was below the potential phytoplankton-growth limiting threshold of 0.2 μM (Fisher et al., 1988).

A clear inverse relation between BIOM and N-compounds resulted from the observation of the three filtered data series. This is in accordance with Sundbäck et al. (2000) who found that $NO_3^- + NO_2^-$ concentrations in the overlying water showed a significant seasonality, with highest values being found during winter (November to February). Increased nutrient concentrations in the overlying water are probably controlled to a higher degree by stochastic events, mainly sediment resuspension. On the contrary, net uptake of inorganic nitrogen, mainly in spring and summer, reflects microalgal growth (Sundbäck et al., 2000 and references therein).

A pronounced seasonality is a typical feature of the sublittoral zone of the Gulf of Trieste, with a temperature range varying from 6.4 to 25.5 °C at the bottom (Bussani et al., 2003). Thus, temperature can be expected to be one of the main controlling factors in the shallow-water zone (Sundbäck et al., 2000). The development of large populations of given species seemed to be quite correlated with temperature; some species growing best under cool water conditions and others under warm conditions. A considerable shift in diatom species occurs with decrease of temperature (Patrick, 1977). In quadrant I of the PCA the position of *Pleurosigma aestuarii, Diploneis smithii, Paralia sulcata* and *Diploneis ovalis* was mainly due to low temperature. An inverse relation between ABU of each species and temperature was found (−0.40, −0.39, −0.35 and −0.34, respectively) corroborating the results shown in Fig. 4a,b. A similar outcome was found in a previous study carried out in the same area (Cibic et al., 2007). Hobson and McQuoid (1997) found that *P. sulcata* was most abundant in cool waters at times of short daylengths. McQuoid and Nordberg (2003) suggested that *P. sulcata* can be found in the water column during winter, due to a competitive advantage for this species in low light or as a result of increased vertical mixing. Moreover, *P. sulcata* and *Diploneis* spp. were reported by the same authors to be part of the same diatom assemblage.

In the second group (quadrant II−III) the position of species was mainly determined by high temperature and PAR. *Navicula* spp. 1, *Nitzschia dissipata, Gyrosigma macrum, Nitzschia* spp. and *Nitzschia sigma* showed positive significant correlation with temperature (0.59, 0.51, 0.49, 0.44, 0.43, respectively). *Navicula* spp. 1 and spp. 2 showed also a positive significant correlation with PAR (0.53 and 0.47, respectively). *Cylindrotheca closterium*, also belonging to this group, has been previously reported to be a summer species (Hobson and McQuoid, 1997).

Autumn samplings were all placed in quadrant IV due to low PAR. The absence of species observed in this quadrant confirmed the inverse relation between BIOM and N-compounds, which were all positioned here. Among N-compounds both NO_2^- and NO_3^- showed inverse relation with many species, corroborating the moving average pattern shown in Fig. 7a,b. The correlation coefficients between NO_2^- and *Nitzschia sigma, Navicula* spp. 1, *Navicula* spp.2 and *Nitzschia* spp. were statistically significant (−0.56, −0.44, −0.43 and −0.42, respectively). Similarly, NO_3^- showed negative significant correlation with *Nitzschia dissipata* (−0.42); other species showed an inverse relation, although not statistically significant, with NO_3^-: *Navicula directa* (−0.37), *Pleurosigma aestuarii* (−0.33) and *Navicula* spp. 2 (−0.32).

The PCA pointed out an inverse relation among PAR and nutrients. In particular, the correlation coefficient between PAR and NO_3^- was statistically significant (−0.42). This is in consistent with Sundbäck and Graneli (1988) who found an inverse relation between the amount of light and the release of NH_4^+ from the sediments. Moreover, according to Sundbäck et al. (1991) decreased outfluxes of NH_4^+, NO_3^- and PO_4^{3-} during the light period were shown to be mediated by photosynthetic benthic organisms. Nutrient exchange between sediment and water has traditionally been studied in darkened cores or chambers, also for sediments in shallow waters, thus excluding the influence of photosynthetic organisms. Photosynthesis of benthic microalgae has been suggested as one of the mechanisms explaining why porewater nutrient profiles cannot be used to predict sediment−water flux rates during conditions when light reaches the sediment surface (Sundbäck et al., 1991 and references therein). According to these authors MPB appear to regulate nutrient flux even at sublittoral depths; however, this effect is probably limited to the growth season, when enough light penetrates to the sediment. The indirect influence of MPB on the nutrient flux, i.e. by changes in the oxygen concentration, seem more important than the direct effect by uptake by the algae (Sundbäck and Graneli, 1988). Yet, other factors can interfere. Animal activity can also be expected to contribute to the temporal variations of MPB and sediment/water nitrogen fluxes through grazing, bioturbation and nutrient excretion (Sundbäck et al., 2000). Furthermore, chemical forms of nutrients and their concentration in the overlying water depend also on respiration processes of the whole benthic community, bacterial activity and chemical equilibria in the sediment (Welker et al., 2002).

5. Conclusions

Throughout the 2-year study the microphytobenthic community was mainly composed of Bacillariophyceae. The

statistically significant relation between diatom ABU and PAR confirmed that benthic microalgal ABU was mainly controlled by light availability at the bottom. Besides PAR, also temperature affected the composition of the benthic microalgal community. A seasonal pattern of monthly samplings was highlighted both from the cluster analysis and the PCA. The latter revealed two diatom assemblages: a winter assemblage and a spring—summer one. While the abundance of the majority of the diatom genera increased with increasing temperature, the genera *Pinnularia* and *Diploneis* appeared to prefer low temperature. Considering the nutrient ratios the development of MPB appeared to be potentially co-limited by Si and P almost throughout the study period. The clear inverse relation between NO_2^-, NO_3^- and BIOM was emphasised both by the moving average and the PCA. The inverse relation among nutrients and light availability suggested that the photosynthetic activity of benthic diatoms in spring and summer may be one of the processes controlling sediment—water nutrient fluxes. Our results corroborate the existing hypothesis that nutrients have a key role for the microphytobenthic development and *vice versa* that the metabolism of

benthic diatoms is important in regulating nutrient concentrations in the overlying water. However, these results are inferred from 23 monthly observations *in situ* rather than from experiments under controlled conditions.

Acknowledgements

Appendix A

Floristic list with mean abundance (+standard deviation) and abundance range (minimum to maximum) of the 2-year study.

St. C1 Bacillariophyceae		Mean ABU (cells cm^{-3})	SD (cells cm^{-3})	Range min—max ABU (cells cm^{-3})
Amphora arenaria Donkin	Benthic, epipsammic	26	55	0—202
Amphora coffeaeformis (Agardh) Kützing	Benthic	784	839	0—3232
Amphora ostrearia de Brébisson	Benthic, epipsammic	4	21	0—101
Amphora ovalis (Kützing) Kützing	Benthic, epipsammic	209	283	0—1010
Amphora ovalis var. *pediculus* (Kützing) Van Heurck	Benthic, epipsammic	13	46	0—202
Amphora proteus Gregory	Benthic, epipsammic	4	21	0—101
Amphora veneta Kützing	Benthic, epipsammic	13	46	0—202
Amphora spp.	Benthic, mostly epipsammic	226	254	0—1010
Auricula insecta (Grunow) Cleve	Benthic, epipelic	22	105	0—505
Auricula spp.	Benthic, epipelic	29	73	0—303
Bacillaria paxillifera (Müller) Hendey	Tycopelagic	268	571	0—2424
Campylodiscus spp.	Benthic, epipelic	11	37	0—152
Climacosphenia moniligera Ehrenberg	Benthic, epiphytic	9	42	0—202
Cocconeis spp.	Benthic, epipsammic	29	73	0—303
Coscinodiscus spp.	Planktonic	55	232	0—1111
Cylindrotheca closterium (Ehrenberg) Lewin et Reimann	Tycopelagic	2308	5232	0—23230
Cymbella cfr. *ventricosa* Kützing	Benthic	9	29	0—101
Cymbella spp.	Benthic, epipelic	373	592	0—2525
Diploneis bombus (Ehrenberg) Cleve	Benthic, epipelic	358	504	0—1818
Diploneis crabro (Ehrenberg) Ehrenberg	Benthic, epipelic	4	21	0—101
Diploneis marginestriata Hustedt	Benthic, epipelic	46	111	0—404
Diploneis ovalis (Hilse) Cleve	Benthic, epipelic	1221	1188	0—4545
Diploneis smithii (de Brébisson) Cleve	Benthic, epipelic	1069	965	0—4040
Diploneis spp.	Benthic, epipelic	852	1218	0—4646
Entomoneis alata (Ehrenberg) Ehrenberg	Benthic, epipelic	145	304	0—1111
Entomoneis spp.	Benthic, epipelic	75	248	0—909
Eunotia spp.	Mostly freshwater, benthic	57	110	0—455
Fragilaria spp.	Mostly freshwater, planktonic	18	50	0—202
Grammatophora marina (Lyngbye) Kützing	Benthic, epipelic	193	349	0—1515
Gyrosigma acuminatum (Kützing) Rabenhorst	Benthic, epipelic	149	228	0—707
Gyrosigma attenuatum (Kützing) Rabenhorst	Benthic, epipelic	13	46	0—202
Gyrosigma balticum (Ehrenberg) Rabenhorst	Benthic, epipelic	75	299	0—1414
Gyrosigma fasciola (Ehrenberg) Griffith et Henfrey	Benthic, epipelic	132	219	0—707
Gyrosigma formosum (W. Smith) Griffith et Henfrey	Benthic, epipelic	97	148	0—606
Gyrosigma macrum (W. Smith) Griffith et Henfrey	Benthic, epipelic	525	1019	0—4444

Appendix (*continued*)

St. C1 Bacillariophyceae		Mean ABU (cells cm^{-3})	SD (cells cm^{-3})	Range min–max ABU (cells cm^{-3})
Gyrosigma spencerii (W. Smith) Griffith et Henfrey	Benthic, epipelic	277	382	0–1313
Gyrosigma spp.	Benthic, epipelic	391	340	0–1212
Leptocylindrus danicus Cleve	Planktonic	9	42	0–202
Licmophora gracilis (Ehrenberg) Grunow	Benthic, epiphytic	13	46	0–202
Licmophora spp.	Benthic, epiphytic	29	66	0–202
Melosira spp.	Mostly freshwater	53	113	0–404
Navicula complanata Hustedt	Benthic, epipelic	4	21	0–101
Navicula directa (W. Smith) Ralfs	Benthic, epipelic	2677	2159	0–7070
Navicula liber (W. Smith) Peragallo	Benthic, epipelic	13	63	0–303
Navicula mollis (W. Smith) Cleve	Epiphytic	18	84	0–404
Navicula rhombica Gregory	Benthic, epipelic	4	21	0–101
Navicula spp. 1[a]	Benthic	4446	3900	202–15756
Navicula spp. 2	Benthic	4982	3127	1111–12827
Navicula spp. 3	Benthic	147	245	0–909
Nitzschia acicularis (Kützing) W. Smith	Benthic, epipelic	509	433	0–1616
Nitzschia acuminata (W. Smith) Grunow	Benthic, epipelic	35	131	0–606
Nitzschia angularis W. Smith	Benthic, epipelic	26	92	0–404
Nitzschia dissipata (Kützing) Grunow	Epiphytic	1348	4478	0–16564
Nitzschia fasciculata Grunow	Benthic, epipelic	742	659	0–2828
Nitzschia fruticosa Hustedt	Epiphytic	13	63	0–303
Nitzschia gracilis Hantzsch	Benthic, epipelic	9	42	0–202
Nitzschia hantzschiana Rabenhorst	Benthic, epipelic	4	21	0–101
Nitzschia intermedia Hantzsch	Benthic, epipelic	4	21	0–101
Nitzschia longissima (de Brébisson) Ralfs	Benthic, epipelic	53	149	0–606
Nitzschia lorenziana Grunow	Benthic, epipelic	1478	1009	101–4040
Nitzschia lorenziana var. *densestriata* (Peragallo) A. Schmidt	Benthic, epipelic	1735	2084	0–8484
Nitzschia obtusa W. Smith	Benthic, epipelic	9	42	0–202
Nitzschia panduriformis Gregory	Benthic, epipelic	119	235	0–909
Nitzschia panduriformis var. *minor* (Cleve et Grunow) Grunow	Benthic, epipelic	57	169	0–707
Nitzschia punctata (W. Smith) Grunow	Benthic, epipelic	141	179	0–606
Nitzschia recta Hantzsch	Benthic, epipelic	31	94	0–404
Nitzschia sigma (Kützing) W. Smith	Benthic, epipelic	1458	1097	202–3434
Nitzschia sigma var. *intercedens* Grunow	Benthic, epipelic	94	118	0–404
Nitzschia sigma var. *sigmatella* (Gregory) Grunow	Benthic, epipelic	53	149	0–606
Nitzschia sigmoidea (Ehrenberg) Smith	Benthic, epipelic	77	160	0–707
Nitzschia subtubicola Germain	Epiphytic	790	3577	0–17170
Nitzschia tryblionella Hantzsch	Benthic, epipelic	970	889	0–4343
Nitzschia vermicularis (Kützing) Hantzsch	Benthic, epipelic	332	340	0–1111
Nitzschia spp.	Benthic	4297	3574	808–13938
Paralia sulcata (Ehrenberg) Cleve	Benthic, epipelic	5210	2244	1717–11514
Pinnularia cardinalis (Ehrenberg) W. Smith	Benthic, epipelic	57	113	0–404
Pinnularia viridis (Nitzsch) Ehrenberg	Freshwater, benthic	48	96	0–303
Pinnularia spp.	Benthic, epipelic	182	151	0–404
Pleurosigma acutum Norman	Benthic, epipelic	57	149	0–606
Pleurosigma cfr. *aestuarii* (de Brébisson) W. Smith	Benthic, epipelic	77	180	0–606
Pleurosigma aestuarii (de Brébisson) W. Smith	Benthic, epipelic	876	988	0–4141
Pleurosigma angulatum (Queckett) W. Smith	Benthic, epipelic	382	321	0–1010
Pleurosigma elongatum W. Smith	Benthic, epipelic	13	46	0–202
Pleurosigma falx Mann	Benthic, epipelic	22	68	0–303
Pleurosigma minutum Grunow	Benthic, epipelic	215	297	0–1010
Pleurosigma spp.	Benthic, epipelic	430	362	0–1212
Podosira moniliformis Ehrenberg	Benthic	22	52	0–202
Podosira spp.	Benthic	15	51	0–202
Proboscia alata (Brightwell) Sundström	Planktonic	4	21	0–101
Pseudo-nitzschia cfr. *pungens* (Grunow ex Cleve) Hasle	Planktonic	40	151	0–707
Pseudo-nitzschia seriata (Cleve) Peragallo	Planktonic	40	190	0–909
Rhopalodia gibba (Ehrenberg) Müller	Benthic	156	203	0–606
Rhopalodia spp.	Mostly freshwater, benthic	66	116	0–404
Surirella spp.	Benthic	13	63	0–303
Synedra rumpens Kützing	Benthic, epiphytic	9	42	0–202
Synedra spp.	Benthic, epiphytic	143	359	0–1717

Appendix (*continued*)

St. C1 Bacillariophyceae		Mean ABU (cells cm^{-3})	SD (cells cm^{-3})	Range min−max ABU (cells cm^{-3})
Thalassionema nitzschioides (Grunow) Mereschkowsky	Planktonic	18	84	0−404
Thalassionema spp.	Planktonic	154	234	0−909
Thalassiosira eccentrica (Ehrenberg) Cleve	Planktonic	400	448	0−1818
Thalassiosira spp.	Planktonic	226	212	0−606
Toxarium hennedyanum (Gregory) Grunow	Benthic, epiphytic	927	2889	0−11110
Tropidoneis longa (Cleve) Cleve	Benthic, epipelic	11	37	0−152
Tropidoneis spp.	Benthic, epipelic	40	104	0−303
Undet. Centrales		9	29	0−101
Undet. Pennales		169	518	0−1919

[a] A distinction was made considering the dimensions of different species belonging to the genus *Navicula*: spp. 1 < 20 µm, 20 µm < spp. 2 < 40 µm, spp. 3 > 40 µm.

References

Admiraal, W., Peletier, H., Zomer, H., 1982. Observations and experiments on the population dynamics of epipelic diatoms of an estuarine mudflat. Estuarine, Coastal and Shelf Science 14, 471−487.

Altman, D.G., 1999. Practical Statistics for Medical Research. Chapman & Hall/CRC, London, 611 pp.

Barranguet, C., 1997. The role of microphytobenthic primary production in a Mediterranean mussel culture area. Estuarine, Coastal and Shelf Science 44, 753−765.

Bertuzzi, A., Faganeli, J., Welker, C., Brambati, A., 1997. Benthic fluxes of dissolved inorganic carbon, nutrients and oxygen in the Gulf of Trieste (northern Adriatic). Water, Air and Soil Pollution 99, 305−314.

Blasutto, O., Cibic, T., De Vittor, C., Fonda Umani, S., 2005. Microphytobenthic primary production and sedimentary carbohydrates along salinity gradients in the lagoons of Grado and Marano (Northern Adriatic Sea). Hydrobiologia 550, 47−55.

Bran+Luebbe, 2003a. Ammonia in Water and Seawater. Method No. G-171−96 Rev. 8 (Multitest MT19), Bran+Luebbe, Noderstedt, Germany.

Bran+Luebbe, 2003b. Nitrate and Nitrite in Water and Seawater. Method No. G-172−96 Rev. 9. (Multitest MT19), Bran+Luebbe, Noderstedt, Germany.

Bran+Luebbe, 2003c. Nitrite in Water and Seawater. Method No. G-173−96 Rev. 7 (Multitest MT18), Bran+Luebbe, Noderstedt, Germany.

Bran+Luebbe, 2003d. Silicate in Water and Seawater. Method No. G-177−96 Rev. 7 (Multitest MT19), Bran+Luebbe, Noderstedt, Germany.

Bran+Luebbe, 2003e. Phosphate in Water and Seawater. Method No. G-175−96 Rev. 10 (Multitest MT18), Bran+Luebbe, Noderstedt, Germany.

Brzezinski, M.A., 1985. The Si:C:N ratio of marine diatoms: interspecific variability and the effect of some environmental variables. Journal of Phycology 21, 347−357.

Buffan-Dubau, E., Carman, K.R., 2000. Diel feeding behaviour of meiofaunal and their relationships with microalgal resources. Limnology and Oceanography 45, 381−395.

Burba, N., Feoli, E., Malaroda, M., Zuccarello, V., 1992. Un sistema informativo per la vegetazione. Software per l'archiviazione della vegetazione italiana e per l'elaborazione di tabelle. Manuale di utilizzo dei programmi CETA Book 2, Gorizia, 78 pp.

Bussani, A., Celio, M., Comici, C., 2003. Climatological analysis (1991−2002) of the thermohaline characteristics in the Marine Reserve of Miramare (Gulf of Trieste). Bollettino di Geofisica teorica ed applicata 44, 11−17.

Cahoon, L.B., 1999. The role of benthic microalgae in neritic ecosystems. Oceanography and Marine Biology. An Annual Review 37, 47−86.

Cheng, I.J., Levinton, J.S., McCartney, M., Martinez, D., Weissburg, M.J., 1993. A bioassay approach to seasonal variation in the nutritional value of sediment. Marine Ecology Progress Series 94, 275−285.

Cibic, T., Blasutto, O., Fonda Umani, S., 2007. Biodiversity of settled material in a sediment trap in the Gulf of Trieste (northern Adriatic Sea). Hydrobiologia 580, 57−75.

Clarke, K.R., Warwick, R.M., 2001. Change in Marine Communities: an Approach to Statistical Analysis and Interpretation. PRIMER-E, Plymouth, 176 pp.

Dexing, J., Junmin, L., Zhaodi, C., Shicheng, L., 1985. Marine Benthic Diatoms in China. China Ocean Press, Beijing, 313 pp.

Dugdale, R.C., Wilkerson, F.P., 1998. Silicate regulation of new production in the equatorial Pacific upwelling. Nature 391, 270−273.

Egge, J.K., Aksnes, D.L., 1992. Silicate as regulating nutrient in phytoplankton competition. Marine Ecology Progress Series 83, 281−289.

Facca, C., Sfriso, A., Socal, G., 2002. Changes in abundance and composition of phytoplankton and microphytobenthos due to increased sediment fluxes in the Venice lagoon, Italy. Estuarine, Coastal and Shelf Science 54, 773−792.

Fisher, T.R., Harding, L.W., Stanley, D.W., Ward, L.G., 1988. Phytoplankton, nutrients, and turbidity in the Chesapeake, Delaware and Hudson estuaries. Estuarine, Coastal and Shelf Science 27, 61−93.

Flynn, K.J., Martin-Jézéquel, V., 2000. Modelling Si-N-limited growth of diatoms. Journal of Plankton Research 22, 447−472.

Fredericks, D.J., Heggie, D.T., Longmore, A., Palmer, D., Smith, C., Skyring, G.W.S., 1999. Nutrient recycling and benthic activity in a shallow coastal lagoon in Western Australia. AGSO Research Newsletter 31, 4−6.

Germain, H., 1981. Flore des Diatomées Eaux Douces et Saumâtres. Société Nouvelle des Éditions Boubée, Paris, 444 pp.

Gilpin, L.C., Davidson, K., Roberts, E., 2004. The influence of changes in nitrogen: silicon ratios on diatom growth dynamics. Journal of Sea Research 51, 21−35.

Hamming, R.W., 1998. Digital Filters. Dover Publications, New York, 284 pp.

Hendey, N.I., 1976. An Introductory Account of the Smaller Algae of British Coastal Waters. Otto Koeltz Science Publishers, West Germany, 317 pp.

Herndl, G.J., Peduzzi, P., Fanuko, N., 1989. Benthic community metabolism and microbial dynamics in the Gulf of Trieste (Northern Adriatic Sea). Marine Ecology Progress Series 53, 169−178.

Hillebrand, H., Sommer, U., 1999. The nutrient stoichiometry of benthic microalgal growth: Redfield proportions are optimal. Limnology and Oceanography 44, 440−446.

Hillebrand, H., Durselen, C.D., Kirschtel, D., Pollingher, U., Zohary, T., 1999. Biovolume calculation for pelagic and benthic microalgae. Journal of Phycology 35, 403−424.

Hobson, L.A., McQuoid, M.R., 1997. Temporal variations among planktonic diatom assemblages in a turbulent environment of the southern Strait of Georgia, British Columbia, Canada. Marine Ecology Progress Series 150, 263−274.

de Jonge, V.N., 1980. Fluctuations in the organic carbon to chlorophyll *a* ratios for estuarine benthic diatom populations. Marine Ecology Progress Series 2, 345−353.

de Jonge, V.N., Colijn, F., 1994. Dynamics of microphytobenthos biomass in the Ems estuary. Marine Ecology Progress Series 104, 185−196.

Levinton, J.S., 1995. Marine Biology. Function, Biodiversity, Ecology. Oxford University Press, New York, 420 pp.

MacIntyre, H.L., Geider, R.J., Miller, D.C., 1996. Microphytobenthos: the ecological role of the ''secret garden'' of unvegetated, shallow-water marine habitats. I. distribution, abundance and primary production. Estuaries 19, 186−201.

Maestrini, S.Y., Berland, B.R., Bréret, M., Béchemin, C., Poletti, R., Rinaldi, A., 1997. Nutrient limiting the algal growth potential (AGP) in the Po river plume and an adjacent area, Northwest Adriatic Sea: enrichment bioassays with the test algae *Nitzschia closterium* and *Thalassiosira pseudonana*. Estuaries 20, 416–429.

McIntire, C.D., Moore, W.W., 1977. Marine littoral diatoms: ecological considerations. In: Werner, D. (Ed.), The Biology of Diatoms. Botanical Monographs, vol. 13. Blackwell, Oxford, pp. 333–371.

McQuoid, M.R., Nordberg, K., 2003. The diatom *Paralia sulcata* as an environmental indicator species in coastal sediments. Estuarine, Coastal and Shelf Science 56, 339–354.

Miles, A., Sundbäck, K., 2000. Diel variation in microphytobenthic productivity in areas of different tidal amplitude. Marine Ecology Progress Series 205, 11–22.

Montagna, P.A., Blanchard, G.F., Dinet, A., 1995. Effect of production and biomass of intertidal microphytobenthos on meiofaunal grazing rates. Journal of Experimental Marine Biology and Ecology 185, 149–165.

Montagnes, D.J.S., Franklin, D.J., 2001. Effect of temperature on diatom volume, growth rate, and carbon and nitrogen content: reconsidering some paradigms. Limnology and Oceanography 46, 2008–2018.

Montani, S., Magni, P., Abe, N., 2003. Seasonal and interannual patterns of intertidal microphytobenthos in combination with laboratory and areal production estimates. Marine Ecology Progress Series 249, 79–91.

Ogorelec, B., Misic, M., Faganeli, J., 1991. Marine geology of the Gulf of Trieste (northern Adriatic): sedimentological aspects. Marine Geology 99, 79–92.

Orloci, L., 1978. Multivariate Analysis in Vegetation Research. Junk Publishers, Boston, 451 pp.

Patrick, R., 1977. Ecology of freshwater diatoms and diatom communities. In: Werner, D. (Ed.), The Biology of Diatoms. Botanical Monographs, vol. 13. Blackwell, Oxford, pp. 333–371.

Perissinotto, R., Nozais, C., Kibirige, I., 2002. Spatio-temporal dynamics of phytoplankton and microphytobenthos in a South African temporarily-open estuary. Estuarine, Coastal and Shelf Science 55, 47–58.

Ricard, M., 1987. Atlas du Phytoplancton Marin Volume II: Diatomophycées. Éditions du Centre National de la Recherché Scientifique, Paris, 297 pp.

Risgaard-Petersen, N., 2003. Coupled nitrification-denitrification in autotrophic and heterotrophic estuarine sediments: on the influence of the benthic microalgae. Limnology and Oceanography 48, 93–105.

Rizzo, W.M., 1990. Nutrient exchanges between the water column and a subtidal benthic microalgal community. Estuaries 13, 219–226.

Round, F.E., 1971. Benthic marine diatoms. Oceanography and Marine Biology. An Annual Review 9, 83–139.

Round, F.E., Crawford, R.M., Mann, D.G., 1992. The Diatoms. Cambridge University Press, Avon, 747 pp.

Rousseau, V., Leynaert, A., Daoud, N., Lancelot, C., 2002. Diatom succession, silicification and silicic acid availability in Belgian coastal waters (Southern North Sea). Marine Ecology Progress Series 236, 61–73.

Sdrigotti, E., Welker, C., 2002. The role of a macroalgal covering event on the microphytobenthos community in intertidal sediment (Marano Lagoon, northern Adriatic Sea). Marine Ecology 23, 370–383.

Snoeijs, P., Leskinen, E., Sundbäck, K., Kuylenstierna, M., Witkowsky, A., Hällfors, G., 1990. Microphytobenthic cell density and species composition in the surface sediment in a shallow brackish-water bay (Gulf of Finland). Aqua Fennica 20, 103–114.

Strathmann, R.R., 1967. Estimating the organic carbon content of phytoplankton from cell volume or plasma volume. Limnology and Oceanography 12, 411–418.

Sundbäck, K., Graneli, W., 1988. Influence of microphytobenthos on the nutrient flux between sediment and water: a laboratory study. Marine Ecology Progress Series 43, 63–69.

Sundbäck, K., Jönsson, B., 1988. Microphytobenthic productivity and biomass in sublittoral sediments of a stratified bay, southeastern Kattegat. Journal of Experimental Marine Biology and Ecology 122, 63–81.

Sundbäck, K., Enoksson, V., Graneli, W., Pettersson, K., 1991. Influence of sublittoral microphytobenthos on the oxygen and nutrient flux between sediment and water: a laboratory continuous-flow study. Marine Ecology Progress Series 74, 263–279.

Sundbäck, K., Miles, A., Gőrnasson, E., 2000. Nitrogen fluxes, denitrification and the role of microphytobenthos in microtidal shallow-water sediments: an annual study. Marine Ecology Progress Series 200, 59–76.

Sundbäck, K., Linares, F., Larson, F., Wulff, A., Engelsen, A., 2004. Benthic nitrogen fluxes along a depth gradient in a microtidal fjord: the role of denitrification and microphytobenthos. Limnology and Oceanography 49, 1095–1107.

Taasen, J.P., Høisæter, T., 1981. The shallow-water soft-bottom benthos in Lindåspolenne, western Norway. 4. Benthic marine diatoms, seasonal density fluctuations. Sarsia 66, 293–316.

Tomas, C.R., 1997. Identifying Marine Phytoplankton. Academic Press, San Diego, 858 pp.

Totti, C., 2003. Influence of the plume of the river Po on the distribution of subtidal microphytobenthos in the Northern Adriatic Sea. Botanica Marina 46, 161–178.

Urban-Malinga, B., Wiktor, J., 2003. Microphytobenthic production along a non-tidal sandy beach gradient: an annual study from the Baltic Sea. Oceanologia 45, 705–720.

Utermöhl, H., 1958. Zur Vervollkommnung der quantitativen Phytoplankton-Methodik. Mitteilungen. Internationale Vereiningung fuer Theoretische und Angewandte Limnologie 9, 1–38.

Van Heurck, H., 1899. Traité des Diatomées. Édité aux Frais de L'Auteur, Anvers, 572 pp.

Welker, C., Sdrigotti, E., Covelli, S., Faganeli, J., 2002. Microphytobenthos in the Gulf of Trieste (Northern Adriatic Sea): relationship with labile sedimentary organic matter and nutrients. Estuarine, Coastal and Shelf Science 55, 259–273.