

Pigment Signatures of Phytoplankton Dynamics in the Northern Adriatic*

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Seasonal and spatial distribution of chlorophyll and carotenoid pigments were determined over an annual cycle in the central part of the northern Adriatic (transect Po River mouth – Rovinj) by the reversed-phase HPLC technique. The phytoplankton dynamics as reflected in chlorophyll *a* was affected by two distinct mechanisms: the new production fostered by an intensive nutrient supply to the surface layer by north Italian rivers and regenerated production prevailing in the bottom layer. The most prominent individual accessory pigments were fucoxanthin, 19'-hexanoyloxyfucoxanthin, chlorophyll *b*, peridinin, 19'-butanoyloxyfucoxanthin and zeaxanthin, which indicated the presence of diatoms, prymnesiophytes, green algae, dinoflagellates, chrysophytes and cyanobacteria, respectively, as the major phytoplankton groups. Phytoplankton blooms, which occur regularly after major freshets in spring and autumn, were dominated by diatoms (fucoxanthin) while, at the beginning of the stratification period, the diatom-dominated population was replaced by a more complex flora dominated by prymnesiophytes (19'-hexanoyloxyfucoxanthin). Intensive phytoplankton blooms, triggered by strong freshwater pulses from the Po River, can reach even the oligotrophic coastal waters of western Istria, but are mostly confined to the upper 5 m of the water column.

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INTRODUCTION

Phytoplankton pigments are a numerous class of organic compounds which play the key role in carbon cycling in the marine environment, providing the basis for photosynthetic assimilation of carbon dioxide, *i.e.* for primary production. Determination of chlorophyll *a* as the main photosynthetic pigment is still the most common method for estimating the crop of primary producers in the marine environment.¹ Moreover, it was shown that some phytoplankton pigments, often referred to as biomarker pigments, can be regarded as selective biomarkers of certain microalgal classes (see Refs. 2 and 3 for review and references), which led to an increasing use of chemotaxonomic approach in marine phytoplankton studies. An important prerequisite for the application of the biomarker approach in extensive field studies was the development of analytical methods for the determination of photosynthetic pigments using HPLC techniques,⁴ which enabled a routine and rapid simultaneous determination of various chlorophyll and carotenoid pigments along with their degradation products.

The HPLC-based methodology was successfully applied in a number of studies, carried out in open oceans, aimed at determining the taxonomic composition of phytoplankton as well as the group-specific distribution and degradation pattern of phytoplankton biomass.⁵⁻⁹ In contrast, systematic studies of that kind are still relatively rare in the more dynamic estuarine and coastal environments.¹⁰⁻¹⁵ Most of the published studies deal with short-living phytoplankton bloom events^{10,15} and/or cover only a few seasons of the annual cycle,^{11,12,14} which is not an adequate frequency with respect to the high temporal variability characteristic of coastal waters. Investigations of detailed spatial and temporal dynamics along the strong chemical and physical gradients present in the areas characterized by large freshwater inputs are still missing. A recent work by Barlow *et al.*¹⁶ revealed marked differences in the phytoplankton composition along the trophic gradients in the Mediterranean Sea, as reflected in biomarker pigments. The percentage of nano- and picoplankton clearly increased following the trophic gradients from the coastal to open oligotrophic areas. Such gradients can be also expected on smaller geographic scales, *e.g.* in the northern Adriatic.

Previous studies in this shallow temperate ecosystem, based on chlorophyll *a* and primary production measurements as well as on conventional identification and enumeration of phytoplankton using inverted microscopy,¹⁷⁻¹⁹ revealed large differences in the trophic status between the western and eastern parts of the basin as well as between the surface and deeper layers of the water column. It was shown that, in addition to diatoms, nanoplankton play an important role in phytoplankton dynamics. Since this phytoplankton size fraction is not accessible for detailed studies by inverted microscopy, the present knowledge of the composition of nano- and picoplankton of the northern Adriatic is very poor.

The aim of the present work was to investigate the distribution of pigment biomarkers in the northern Adriatic in order to achieve a more detailed insight into the phytoplankton community structure using the chemotaxonomic approach and to study phytoplankton dynamics in relation to hydrographical features of the basin by covering the whole annual cycle at monthly frequency.

EXPERIMENTAL

Study Area

The northern Adriatic is a shallow basin, with a maximum depth of 35 m, which exhibits a strong trophic gradient along the transect from the west to the east coast.¹⁹ The basin represents a very dynamic ecosystem with pronounced seasonal and interannual variability in chemical and biological properties, including the primary production rate and phytoplankton crop. However, recent analyses of the data collected over a 23-year period (1969–1992) revealed the existence of long-term trends which are primarily related to fluctuations in the Po River discharge and the prevailing meteorological conditions, which determine the water column stability and circulation pattern in the basin.²⁰ The western part of the basin is highly eutrophic due to river inputs from the Italian rivers, particularly the Po river with a long-term average water discharge of 1500 m³/s. The eastern part is generally oligotrophic as a result of the predominant cyclonic current system which brings oligotrophic waters from the central Adriatic along the coast of western Istria whereas nutrient rich Northern Adriatic water leave the area along the Italian coast (see Ref. 21 for review and references). It should be mentioned that in some seasons, particularly during the stratification period, the general current system could be significantly altered by formation of a cyclonic gyre which does not allow an efficient export of eutrophic waters from the basin. In such situations, even the oligotrophic coastal waters of western Istria can be affected by riverine inputs from the other coast.²²

Sampling

Seasonal and spatial distribution of pigment biomarkers were determined over an annual cycle in the middle part of the northern Adriatic on the transect Po River mouth – Rovinj (Figure 1). The stations along this transect were generally accepted as representative of the northern Adriatic¹⁹ and, since 1976, these stations have been regularly surveyed for basic hydrographic properties, nutrient dynamics and phytoplankton dynamics at frequencies ranging from monthly to seasonal (see Ref. 20 for review). During this study sampling was performed in the period January-December 1994 from the research vessel »Vila Velebica« at 6 stations and at 5–6 depths (0, 5, 10, 20, 30 m and near the bottom) using 5 l Niskin bottles.

Determination of Photosynthetic Pigments

Samples for the analyses of photosynthetic pigments (0.5 l) were filtered onto 25 mm GF/F filters. Filters were immediately stored at –20 °C until analysis. The filtrates were extracted in 3 ml of cold 90% acetone using sonication, centrifuged to clarify the extract, and the chlorophylls and carotenoids separated by reversed-phase

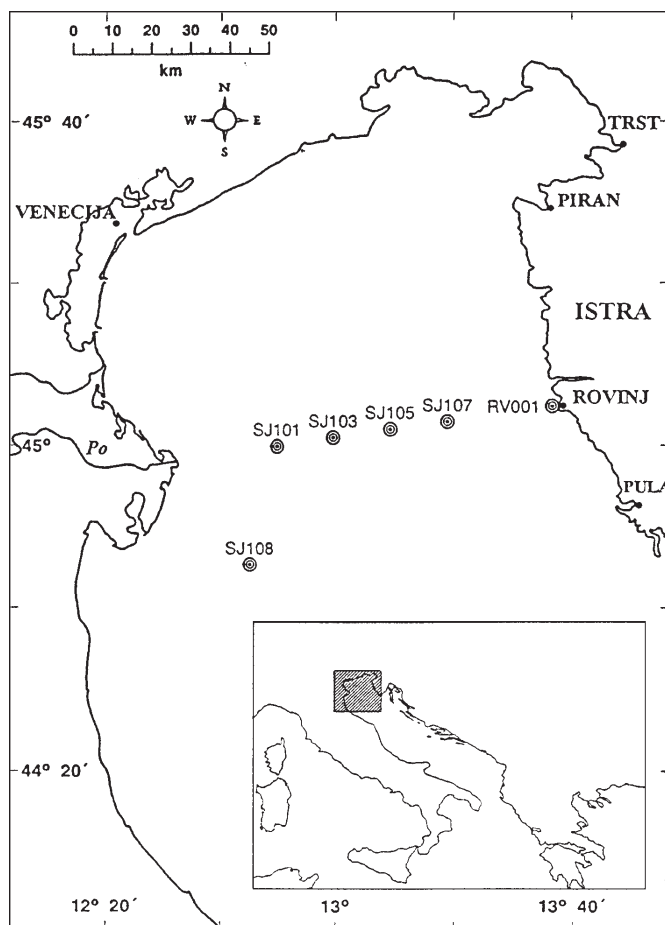


Figure 1. Map of the northern Adriatic with sampling stations on the transect: Po River mouth – Rovinj.

HPLC according to Barlow *et al.*⁷ Briefly, extracts were mixed (1:1; v/v) with 1 M ammonium acetate and injected into a HPLC system incorporating a gradient solvent delivery system (Varian Star 9010), injector (Rheodyne, Model 7125), C_{18} 3 μ m Pecosphere column (3.3 \times 0.45 cm, Perkin Elmer) and serially coupled spectrophotometric and spectrofluorimetric detectors. A binary linear gradient was used to separate the pigments. Solvent A consisted of methanol ($\phi = 0.80$) and 1 M ammonium acetate ($\phi = 0.20$), while solvent B contained methanol ($\phi = 0.60$) and acetone ($\phi = 0.40$). Chlorophylls and carotenoids were detected by absorbance at 440 nm (Spectra Physics UV 2000), while phaeopigments were detected by fluorescence (Spectra Physics F 2000) using excitation at 420 nm and emission at 672 nm. Data collection and re-processing involved use of Spectra Physics PC 1000 software. Qualitative identification and quantitative determination of individual pigments were performed according to Barlow *et al.*⁷ and Terzić.²³

RESULTS AND DISCUSSION

Pigment Signatures of Phytoplankton Composition

Chromatograms obtained by the UV-Vis and fluorimetric detection presented in Figure 2 illustrate typical patterns of phytoplankton pigments and their degradation products in the northern Adriatic. In order to indicate most of the characteristic pigments identified in the northern Adriatic, a sample showing high complexity of the pigment assemblage, collected in late spring, was chosen. However, it should be stressed that the composition was much simpler, in typical bloom periods, mostly exhibiting a strong predominance of one or two biomarker pigments. Apart from chlorophyll *a*, as a

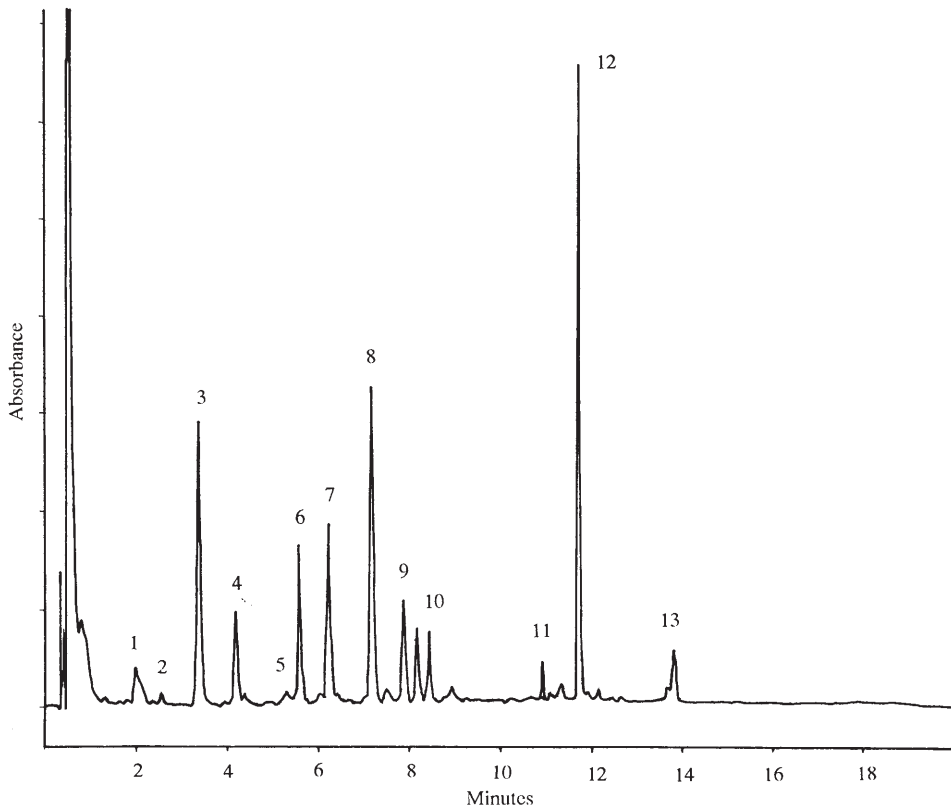


Figure 2. HPLC absorbance (440 nm) chromatogram of a sample obtained on May 17, 1994 from the surface layer in the northern Adriatic. Pigment identities are (1) chlorophyll *c*₃, (2) chlorophyllide *a*, (3) chlorophyll *c*₁ + *c*₂, (4) peridinin, (5) 19'-butanoyloxyfucoxanthin, (6) fucoxanthin, (7) 19'-hexanoyloxyfucoxanthin, (8) diadinoxanthin, (9) alloxanthin, (10) zeaxanthin/lutein, (11) chlorophyll *b*, (12) chlorophyll *a* and (13) β -carotene.

universal indicator of the total phytoplankton biomass, the most prominent pigments found in the northern Adriatic during the investigated period were chlorophyll c_3 , chlorophyll $c_1 + c_2$, peridinin, 19'-butanoyloxyfucoxanthin, fucoxanthin, 19'-hexanoyloxyfucoxanthin, diadinoxanthin, alloxanthin, zeaxanthin, lutein, chlorophyll b and β -carotene. Some of the most abundant pigments, such as chlorophyll $c_1 + c_2$ and diadinoxanthin, have a rather small diagnostic value for the analysis of phytoplankton dynamics,^{2,3} and are therefore not discussed in this paper. A special emphasis was put on those biomarker pigments which are suitable for the estimation of the group-specific phytoplankton biomass in the northern Adriatic, including diatoms (fucoxanthin), prymnesiophytes (19'-hexanoyloxyfucoxanthin), chrysophytes (19'-butanoyloxyfucoxanthin), dinoflagellates (peridinin), green algae (chlorophyll b) and cyanobacteria (zeaxanthin).²⁻³ It should be stressed that no separation between zeaxanthin and lutein, or between chlorophyll a , and divinyl chlorophyll a was achieved by the applied HPLC method, which precluded unambiguous identification of the procaryotic contribution to the total phytoplankton biomass.¹⁶ However, in some situations, when chlorophyll b was very low, suggesting virtual absence of green algae and prochlorophytes from the sample, the zeaxanthin/lutein peak was attributed entirely to zeaxanthin derived from cyanobacteria. Furthermore, 19'-butanoyloxyfucoxanthin was assumed to derive from chrysophytes since the alterantive group containing this biomarker pigment, pelagophytes, are more typical of oligotrophic open waters.^{16,24} For the same reason, occurrence of chlorophyll b was assumed to reflect the presence of green algae (prasinophytes and chlorophytes), since prochlorophytes, which also contain chlorophyll b , were shown to be only a minor component of phytoplankton populations in the Mediterranean coastal areas.¹⁶

Hydrographic and Nutrient Data

The two most important sources of nutrients in the northern Adriatic are freshwater discharges of the Po River to the surface layer and regeneration processes in the bottom layer.²⁶ Distribution of surface salinity during 1994 exhibited a strong seasonal and spatial variability (Figure 3), reflecting primarily strong seasonal fluctuations of the Po River runoff. The impact of freshwater discharges on the surface layer salinity was particularly strong during the major freshets of the Po River, which occurred in March, May and November. This was especially pronounced at the stations situated in the western part of the basin (SJ108 and SJ101), with salinity decreasing below 30. In contrast, the salinity decrease at station RV001 was very modest, with slightly lower values during the stratification period (June–September). It should be stressed that the influence of freshwater runoff was mostly confined to the top 5 meters of the water column. Such a distribution pattern of salinity was generally followed by similar gradients of riverborne nutrients, notably orthosilicate and nitrate in the surface layer (Figure 3).

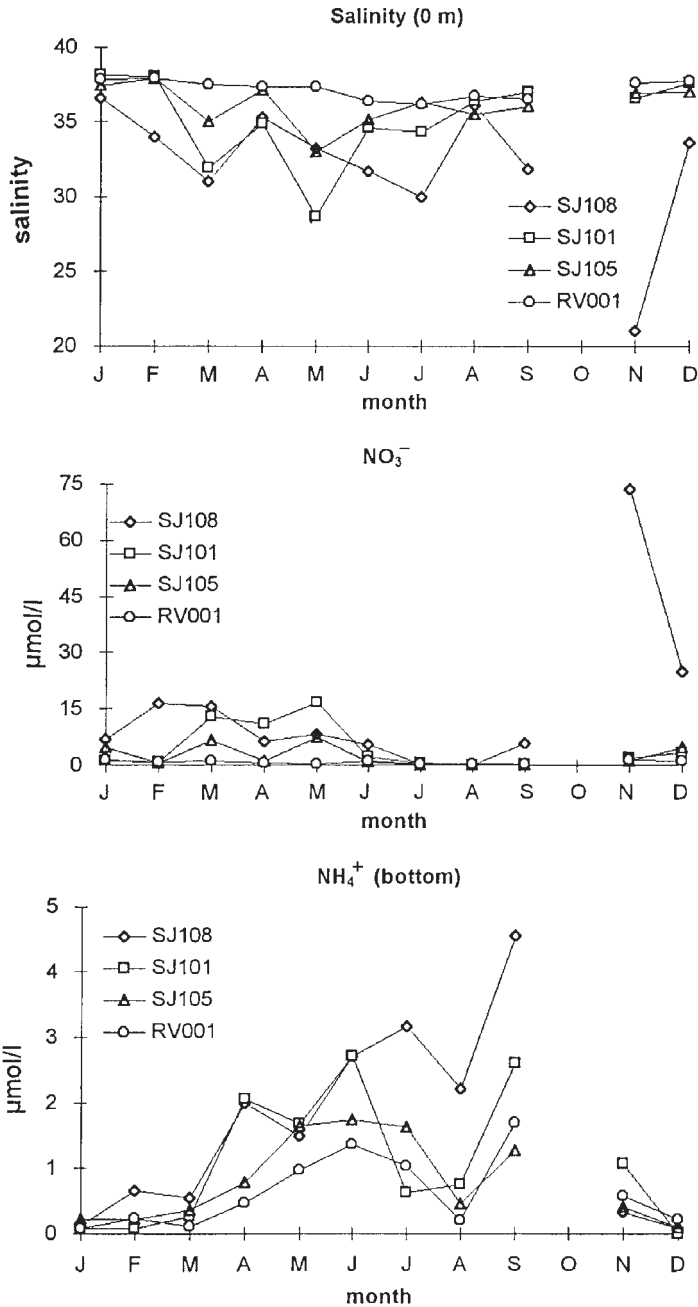


Figure 3. Seasonal fluctuations of salinity and nitrate in the surface layer and ammonia in the bottom layer of the northern Adriatic during 1994 at four selected stations along the transect Po River mouth – Rovinj.

The concentration of nitrate, the predominant inorganic nitrogen species in the surface layer, varied from 0.3 to 74 $\mu\text{mol/l}$, but the concentrations were below 20 $\mu\text{mol/l}$ in most seasons. As opposed to the surface layer, the predominant inorganic nitrogen species was ammonia, especially during the stratification period. The distribution of ammonia in the bottom layer exhibited a typical seasonal pattern, with the highest values in the period from April to September. A very similar seasonal distribution in the bottom layer was obtained for orthosilicate,²⁷ indicating the impact of regeneration processes. It should be pointed out that the concentrations of regenerated orthosilicate and ammonia were much higher in the more productive western part of the basin than in the rather oligotrophic eastern part.

Seasonal and Spatial Distribution of Chlorophyll a

Concentration levels of chlorophyll *a* as well as the composition of pigment biomarkers in the northern Adriatic were rather variable, depending strongly on the hydrological situation in the Po River watershed, distance of the given location from its mouth, seasonally-dependent type of circulation in the basin, water column stability and depth. The seasonal distribution of chlorophyll *a* presented in Figure 4 assumes a concept which regards the northern Adriatic as a quasi-three-layer system consisting of a) the surface layer (< 5 m) which is under direct impact of freshwater inputs, b) the bottom layer which is influenced by water sediment processes such as regeneration, and c) a large intermediate layer which couples the former two layers.²⁵ Moreover, characteristic stations which cover the entire eutrophic gradient along the transect Po River mouth – Rovinj were selected for the presentation: stations SJ108 and SJ101 are situated close to the Po River mouth and are under direct influence of freshwater discharges most of the year, station SJ105 is situated in the middle part of the transect, while station RV001 represents the oligotrophic coastal waters of western Istria.

Seasonal dynamics of chlorophyll *a* during 1994 was quite variable along the investigated transect, and the variability is especially pronounced when comparing the two extreme stations, S108 and RV001. Furthermore, the differences between these stations are more expressed in the surface layer (0 m) than in the bottom layer. The data show that the surface layer is the most variable part of the ecosystem with respect to phytoplankton dynamics, which is clearly related to freshwater pulses of the Po River as the dominant source of nutrients in the area.²⁶ The strongest impact of river-borne nutrients on the phytoplankton was observed at stations SJ108 and SJ101, which were, in most seasons, under direct influence of river discharges,²⁸ as reflected in a significant decrease of salinity in the upper 5 m of the water column, accompanied by an enhanced nitrate concentration (Figure 3). The chlorophyll *a* concentration at these stations reaches up to 8000 ng/L and coincides with major freshets from the Po River, which occurred in March, May, September and November. However, it should be noted

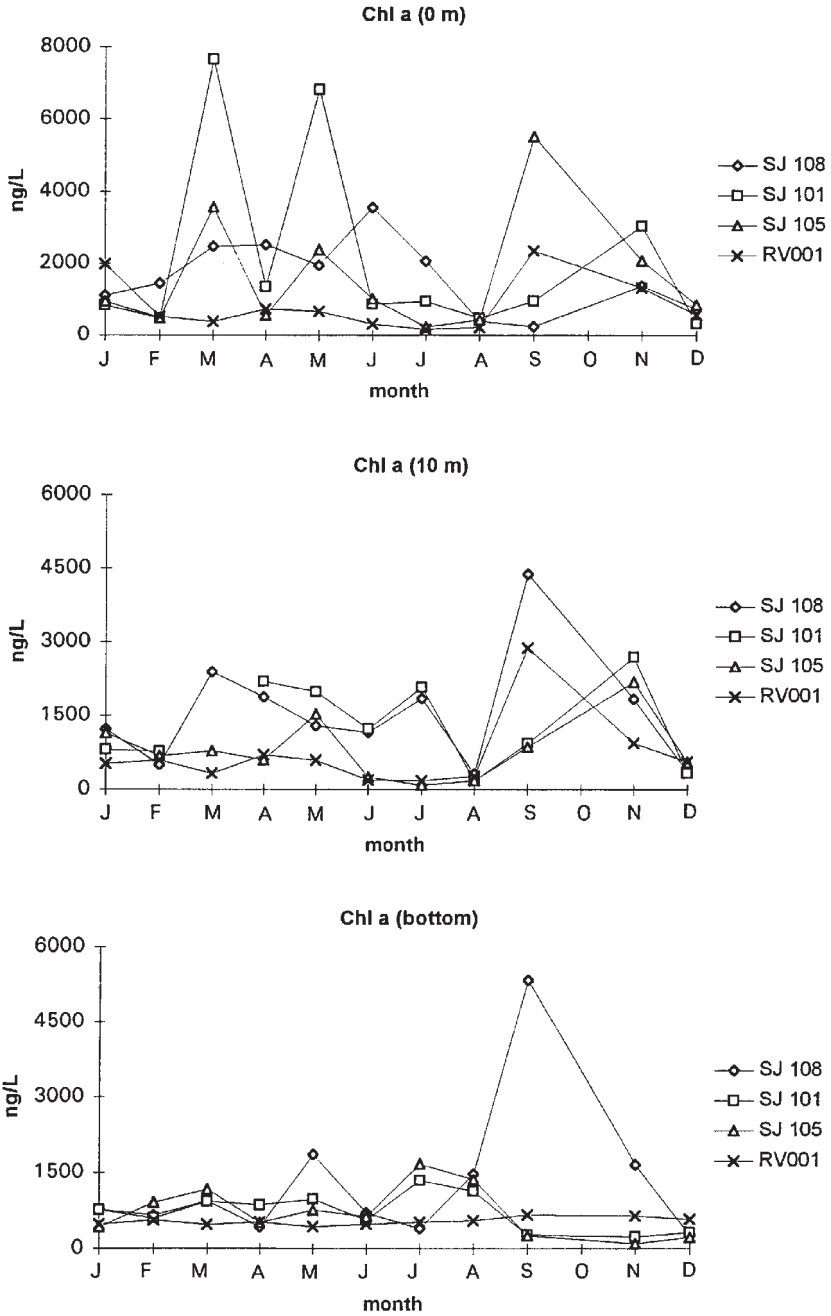


Figure 4. Seasonal variability of chlorophyll *a* in the surface (0 m), intermediate (10 m) and bottom (27–35 m) layers of the northern Adriatic during 1994 at four selected stations of the transect Po River mouth – Rovinj.

that the chlorophyll *a* maxima for stations SJ108 and SJ101 do not always coincide. This is a consequence of different hydrodynamics of water masses in the western part of the basin in various seasons.²¹ In a typical winter situation, the river water plume from the Po mouth spreads southwards in a relatively narrow (10–20 km) strip along the Italian coast and builds a well defined frontal system that prevents the spreading of freshwater in to the basin interior.²⁹ In such situations, the station most affected by freshwater inputs is SJ108, while the biomass remains low to moderate (below 2000 ng/l of chlorophyll *a*) at other stations. However, stronger pulses from the Po River result in an intensive spreading of the freshwater into a wider area of the basin, but not reaching layers below 5 m. The strongest impact on the phytoplankton crop in such situations was observed at the western most station SJ101. However, the chlorophyll *a* distribution in September indicated that very complex hydrodynamic situations are also possible, with occurrence of larger buoyant brackish layers floating in the middle of the basin, characterized by enhanced chlorophyll *a* levels, while the concentration of chlorophyll *a* was lower at more western stations of the transect.

Influence of freshwater discharges does not usually reach layers below 5 m to any significant extent. However, in a rather exceptional situation in September 1994, fast cooling of the surface layer caused vertical convective mixing of the water column, bringing nutrients to deeper layers.²⁸ Because of that, chlorophyll *a* levels up to 4000 ng/l were found in the intermediate layer (10 m). In the bottom layer (Figure 4), the maximal concentration of chlorophyll *a* was significantly lower than that in the surface layer and remained below 2000 ng/l throughout the year, except for one occasion at station SJ108. At stations SJ101 and SJ105, there was a significant increase in chlorophyll *a* during summer, which is attributed to the impact of regenerated nutrients. A similar observation was made earlier in the Gulf of Trieste,^{23,30} which has similar characteristics to those of the northern Adriatic regarding increased eutrofication caused by riverborne nutrient inputs. It is interesting to note that such an increase was not evident at the oligotrophic station RV001, suggesting a comparatively lower flux of phytoplankton derived organic matter to the bottom layer and therefore less intensive nutrient regeneration in that part of the basin.

The levels as well as the seasonal and spatial pattern of chlorophyll *a* distribution during 1994 are in good agreement with the long-term average values reported by other authors.^{17,19,20}

Seasonal and Spatial Distribution of Biomarker Pigments

The dynamics of the group-specific phytoplankton biomass in the northern Adriatic can be successfully described by the 6 most prominent biomarker pigments: fucoxanthin, 19'-hexanoyloxyfucoxanthin, chlorophyll *b*, peridinin, 19'-butanoyloxyfucoxanthin and zeaxanthin because these pig-

ments explain more than 90% of the biomass associated with chlorophyll *a* in the northern Adriatic.²³ Large differences in seasonal distributions of biomarker pigments were observed between the western (station SJ101; Figure 5) and eastern (station RV001; Figure 6) parts of the basin.

The main feature of biomarker pigment distribution in the surface layer (0.5 m) of the western part of the basin are the strong maxima representing major phytoplankton blooms that follow major freshets from the Po River. These blooms are typically dominated by diatoms.³¹ However, in 1994, two major spring blooms (March, May) were dominated by green algae (chlorophyll *b*) and prymnesiophytes (hexanoyloxyfucoxanthin), respectively. Nevertheless, diatoms (fucoxanthin) were significant contributors to the total biomass in March and the most abundant species was *Skeletonema costatum* with 5.2×10^6 cell/l.²⁷ In contrast, microscopic examination of the samples in May showed a comparatively small number of diatoms ($< 10^5$ cells/l), suggesting that a significant part of fucoxanthin detected in the samples may have originated from nanoplanktonic non-diatom species, which could contain smaller but significant quantities of this pigment.² Such a strong bloom of green algae, like the one in March 1994, is quite unusual for the northern Adriatic and was attributed to a rather local event confined only to station SJ101. On the contrary, predominance of prymnesiophytes in late spring seems to be typical for the entire northern Adriatic. A similar situation was recorded in the northernmost part of the basin, in the Gulf of Trieste.^{23,30} This is also in agreement with some earlier studies,³¹ which reported regular predominance of microflagellates (as defined by counting after the Utermoehl method) in late spring-summer phytoplankton populations. Peridinin was also rather abundant in May 1994, indicating a significant presence of dinoflagellates in late spring (Figure 5). The most abundant species were *Prorocentrum micans* and *Prorocentrum balticum*.²⁷ In all other seasons throughout 1994, peridinin levels were low, suggesting that dinoflagellates were not important contributors to the phytoplankton biomass in the surface layer. Similar conclusions can be drawn for cyanobacteria based on continuously low zeaxanthin/lutein levels, except during the May bloom (Figure 5). It should be stressed that the phytoplankton composition during the bloom was very complex which was quite unique relative to the other bloom events during 1994, which showed the typical strong predominance of a few species. In non-bloom situations, the concentration of biomarker pigments in the surface layer was mostly below 200 ng/l, with characteristic features reflecting the general dynamics of chlorophyll *a*, *i.e.* showing lower concentrations during summer and enhanced concentrations in spring and autumn.

The seasonal dynamics of biomarker pigments in the intermediate layer (10 m; Figure 5) was significantly different from that in the overlying surface water. The two largest concentration maxima of individual biomarkers were observed for fucoxanthin. These two maxima reflected the impact of

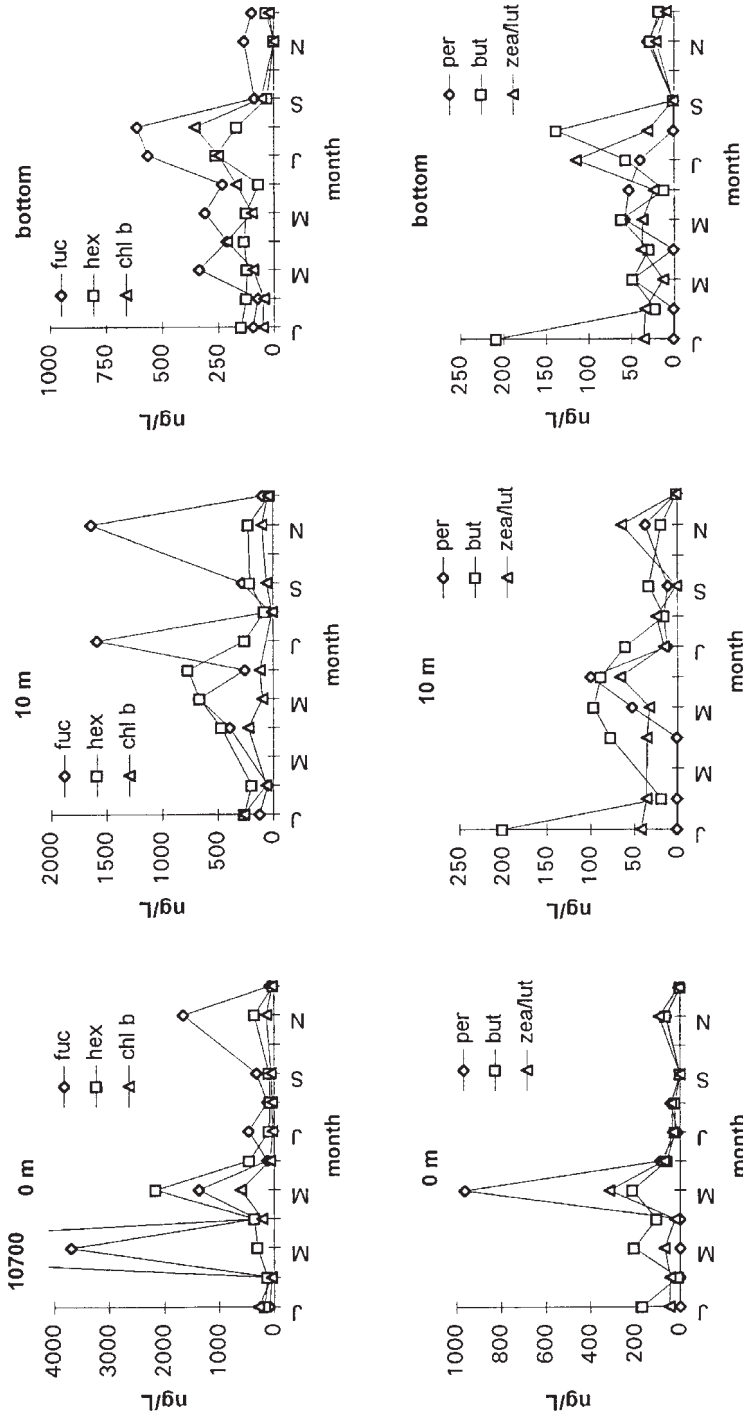


Figure 5. Seasonal variability of fucoxanthin (fuc), 19-hexanoyloxyfucoxanthin (hex), chlorophyll *b* (chl b), peridinin (per), 19-butanoxyfucoxanthin (but) and zeaxanthin/lutein (zea/lut) in the surface (0 m), intermediate (10 m) and bottom (27–35 m) layers of the western part of the northern Adriatic (station SJ101; 1994).

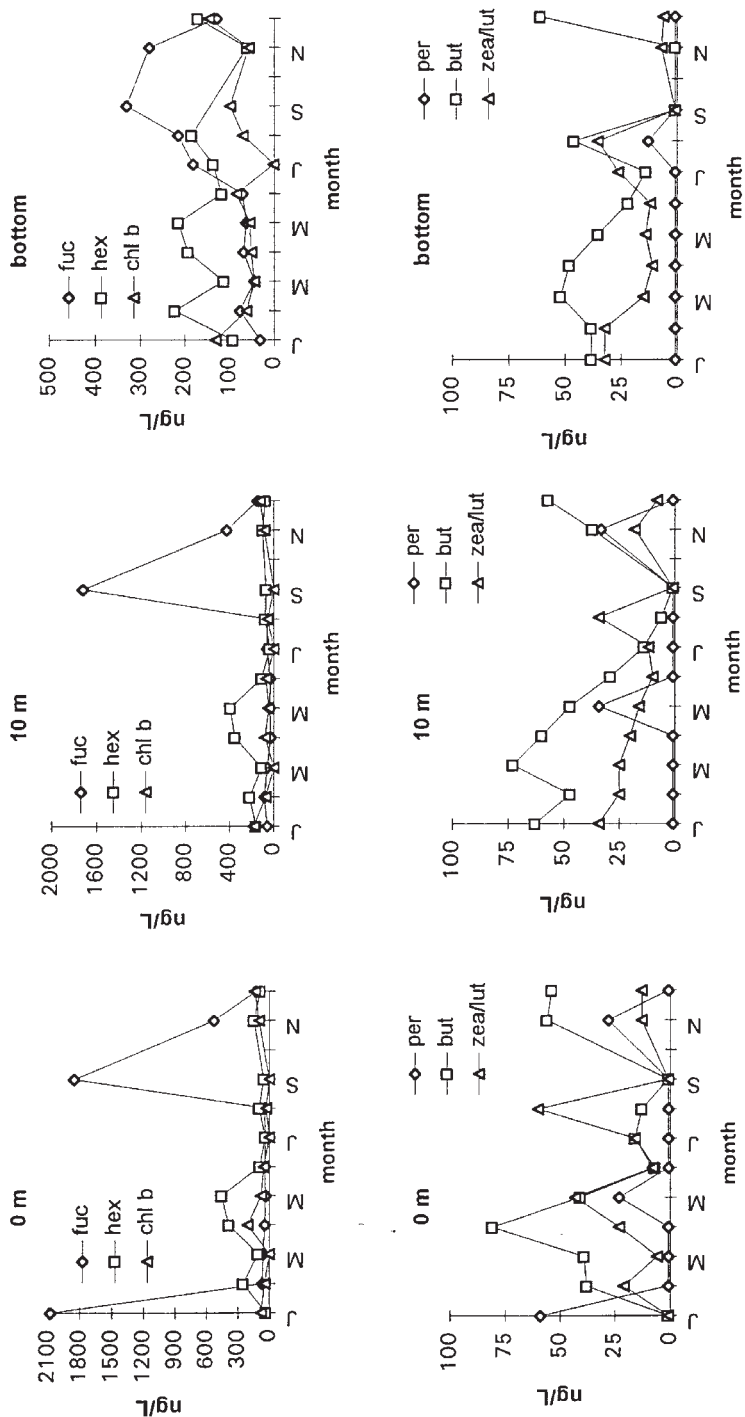


Figure 6. Seasonal variability of fucoxanthin (fuc), 19'-hexanoyloxyfucoxanthin (hex), chlorophyll *b* (chl *b*), peridinin (per), 19'-butanoyloxyfucoxanthin (but) and zeaxanthin/lutein (zea/lut) in the surface (0 m), intermediate (10 m), and bottom (27-35 m) layers of the eastern part of the northern Adriatic (station RV001; 1994).

diatom blooms that occurred in the surface layer in July and November. Apart from these two seasons, the prevailing pigment biomarker in the intermediate layer was hexanoyloxyfucoxanthin, with a maximum at the beginning of the stratification period (June), which indicated a clear predominance of prymnesiophytes in that layer. Other accessory pigments remained rather low (<100 ng/l) throughout the year with somewhat enhanced values in late spring.

Although the water column of the northern Adriatic is well mixed during the colder season, the pigment patterns in the surface and bottom layers do not seem to be well linked. It was especially in the warmer part of the year, when the water column was thermally stratified, that the phytoplankton dynamics in the bottom layer was rather different than in the overlying water. The phytoplankton in the bottom layer are dominated by diatoms (fucoxanthin) throughout the year and the maximum was observed during the stratification period (Figure 5). This is exactly the opposite of the situation found in the surface layer. This feature of the seasonal pigment distribution in the bottom layer is a consequence of intensive regeneration processes during summer, which supplied additional nutrients to the near bottom layer²⁶ while the surface remained depleted of nutrients. A very similar comparison of phytoplankton distributions in the surface and bottom layers was reported for the Gulf of Trieste.^{23,30} It is also interesting to point out a significant increase of chlorophyll *b* levels during July-August which was attributed to an enhanced biomass of green algae, very probably caused by the additional availability of nitrogen in the form of ammonia. This is in agreement with observations that green algae preferentially respond to the availability of ammonia as compared to other phytoplankton groups.³² All other accessory pigments detected in the bottom layer were present at much lower levels but they also showed a characteristic enhancement during the summer stratification period.

The seasonal distribution of various phytoplankton pigments in the surface layer of the eastern part of the basin (station RV001; Figure 6), which is rarely affected by freshwater pulses of the Po River, indicates rather different features both in terms of phytoplankton pigment composition and their seasonal maxima. The highest values of individual biomarkers were observed for fucoxanthin (up to 2000 ng/l in January and September). However, these two events were a result of rather unusual hydrographic conditions in the northern Adriatic.²⁸ The most abundant accessory pigment in most seasons during 1994 was 19'-hexanoyloxyfucoxanthin, suggesting that in these waters diatoms were outcompeted by hexanoyloxyfucoxanthin-containing microflagellates, most probably prymnesiophytes. Such conclusion was supported by microscopical analyses which showed a strong predominance of flagellated nanoplankters.²⁷ Low biomarker pigment concentrations accompanied by their rather complex composition underlined the basically oligotrophic character of the waters along the Istrian coast.¹⁹

The dynamics of phytoplankton in the intermediate layer at station RV001 (Figure 6) revealed a pattern that was very similar to that described for the surface layer. This can be explained by the fact that the surface layer at station RV001 was not influenced by any freshwater input from the coast and consequently the main characteristic of that layer, which makes it distinctive from the underlying water, is missing.

The pattern of biomarker pigments in the bottom layer (Figure 6) during winter and spring was similar to that in overlying waters while, during the summer stratification significant differences were observed that consisted of comparatively enhanced levels of several biomarker pigments, especially fucoxanthin. The observed difference was interpreted as a consequence of the response of bottom phytoplankton to regenerated nutrients. However, due to the rather oligotrophic character of waters along the west Istrian coast, the flux of detritus to the bottom and the subsequent regeneration processes are not so pronounced as in the eutrophic part of the basin.¹⁹ Moreover, the origin of waters in the bottom layer of the eastern part of the basin are mostly the nutrient-poor southern waters from the middle Adriatic.²¹ As a consequence of comparatively lower nutrient levels in the bottom layer at station RV001, the phytoplankton growth is very limited, resulting in a rather low biomass for all phytoplankton groups. In the first part of the year, the phytoplankton were dominated by prymnesiophytes (19'-hexanoyloxyfucoxanthin), while fucoxanthin prevailed in autumn. Low levels of chlorophyll *b* (green algae) seem to be consistent with very low ammonia levels throughout the year, as compared with station SJ101 (Figure 3). In the first part of the year, diatoms were outcompeted by microflagellates (prymnesiophytes and silicoflagellates), probably because low nutrients are more efficiently taken up by small flagellated cells.³³ Diatoms, however, became more successful in the second part of the year, which was stimulated by the additional input of nutrients *via* regeneration processes.

CONCLUSIONS

Photosynthetic pigments proved to be good chemotaxonomic markers of phytoplankton dynamics in the northern Adriatic and provided very useful information about the phytoplankton community structure which is complementary to the classical optical methods. The applied approach proved especially advantageous for the characterization of smaller phytoplankton fractions that cannot be properly assessed by inverted microscopy. It is well known that the phytoplankton dynamics in the northern Adriatic is dominated by pronounced diatom blooms and by, so far, poorly defined flagellated nanoplankton. Application of the biomarker approach in this paper revealed a great significance of prymnesiophytes as a major phytoplankton group and confirmed the ubiquity of this microalgal class. In addition to prymnsio-

phytes, the less prominent but significant contributors to the nanophytoplankton were also green algae and chrysophytes. The group specific characterization of the phytoplankton biomass is very important for a proper balance of carbon and consequently for a better understanding of ecological processes in the northern Adriatic, such as exudation of carbohydrates, mucilage formation and bottom anoxia.

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REFERENCES

1. J. D. H. Strickland and T. R. Parsons, *A practical handbook of seawater analysis*, 2nd edition, *Bull. Fish. Res. Board Can.* **167** (1972) 1–310.
2. S. K. Rowan, *Photosynthetic pigments of algae*, Cambridge University Press, Cambridge, 1990, p. 333.
3. D. F. Millie, H. W. Paerl, and J. P. Hurley, *Can. J. Fish. Aquat. Sci.* **50** (1993) 2513–2527.
4. S. W. Wright, S. W. Jeffrey, R. F. C. Mantoura, C. A. Llewellyn, T. Bjørnland, D. Repeta, and N. Welschmeyer, *Mar. Ecol. Prog. Ser.* **77** (1991) 183–196.
5. W. W. Gieskes and G. W. Kraay, *Mar. Biol.* **91** (1986) 567–576.
6. R. R. Bidigare, J. Marra, T. D. Dickey, R. Iturriaga, K. S. Baker, C. S. Raymond, and H. Pak, *Mar. Ecol. Prog. Ser.* **60** (1990) 113–122.
7. R. G. Barlow, R. F. C. Mantoura, M. A. Gough, and T. W. Fileman, *Deep-Sea Res. II* **40** (1993) 459–477.
8. R. M. Letelier, R. R. Bidigare, D. V. Hebel, M. Ondrusek, C. D. Winn, and D. M. Karl, *Limnol. Oceanogr.* **38** (1993) 1420–1437.
9. D. A. Everitt, S. W. Wright, J. K. Volkman, D. P. Thomas, and E. J. Lindstrom, *Deep-Sea Res.* **37** (1990) 975–977.
10. B. Klein and A. Sournia, *Mar. Ecol. Prog. Ser.* **37** (1987) 265–275.
11. V. Denant, A. Saliot, and R. F. C. Mantoura, *Mar. Chem.* **32** (1991) 285–297.
12. E. Jalliffier-Merlon, J.-C. Marty, V. Denant, and A. Saliot, *Estuarine Coastal Shelf Sci.* **32** (1991) 463–482.
13. T. S. Bianchi, S. Findlay, and R. Dawson, *Estuarine Coastal Shelf Sci.* **36** (1993) 359–376.
14. S. Terzić, M. Ahel, J.-J. Naudin, and G. Cauwet, *Rapp. Comm. Int. Mer. Medit.* **34** (1995) 72.
15. M. Ahel, R. G. Barlow, and R. F. C. Mantoura, *Mar. Ecol. Prog. Ser.* **143** (1996) 289–295.
16. R. G. Barlow, R. F. C. Mantoura, D. G. Cummings, and T. W. Fileman, *Deep-Sea Res. II* **44** (1997) 833–850.

17. N. Smodlaka, *Sci. Total Environ.* **56** (1986) 211–220.
18. N. Revelante and M. Gilmartin, *Mar. Biol.* **34** (1976) 259–271.
19. M. Gilmartin, D. Degobbi, N. Revelante, and N. Smodlaka, *Int. Rev. Gesamten Hydrobiol.* **75** (1990) 425–445.
20. R. Precali, PhD. Thesis, University of Zagreb, 1995 (in Croatian).
21. M. Orlić, M. Gačić, and P. A. la Violette, *Oceanol. Acta* **15** (1992) 109–124.
22. D. Degobbi, *Mar. Pollut. Bull.* **20** (1989) 452–457.
23. S. Terzić, PhD. Thesis, University of Zagreb, 1996 (in Croatian).
24. N. Simon, R. G. Barlow, D. Marie, F. Partensky, and D. Vaultot, *J. Phycol.* **30** (1994) 922–935.
25. P. Franco and A. Michelato, *Sci. Total Environ. Supplement* (1992) 35–62.
26. D. Degobbi, *Mar. Chem.* **29** (1990) 235–253.
27. Center for Marine Research, Rovinj, Croatia, unpublished data.
28. N. Supić, I. Ivančić, Ž. Stipić, and D. Degobbi, *Izvanr. meteorol. hidrol. prilike Hrvat.* **18** (1995) 1–5 (in Croatian).
29. A. Bergamasco, M. Gačić, R. Boscolo, and G. Umgiesser, *J. Mar. Syst.* **7** (1996) 67–94.
30. A. Malej, P. Mozetič, V. Malačić, S. Terzić, and M. Ahel, *Mar. Ecol. Prog. Ser.* **120** (1995) 111–121.
31. N. Revelante and M. Gilmartin, *Thalassia. Jugosl.* **19** (1983) 303–318.
32. C. Courties, A. Vaquer, M. Troussellier, M.-J. Chretiennot-Dinet, J. Neveux, C. Machado, and H. Claustre, *Nature* **370** (1994) 255.
33. M. J. Furnas, *J. Plankton Res.* **12** (1990) 1117–1157.

SAŽETAK

Praćenje dinamike fitoplanktona u sjevernom Jadranu određivanjem fotosintetskih pigmenta

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Upotrebom metode HPLC s obrnutom fazom istražena je sezonska i prostorna raspodjela klorofila i karotenoida u srednjem dijelu sjevernog Jadrana (profil ušće rijeke Po–Rovinj) tijekom 1994. godine. Dinamika biomase fitoplanktona (klorofila *a*) bila je određena dvama različitim mehanizmima: novom proizvodnjom potaknutom intenzivnim donosima hranjivih soli u površinski sloj posredstvom sjevernotalijanskih rijeka i regeneriranom proizvodnjom koja je dominirala u pridnenom sloju. Najzastupljeniji biomarkerski pigmenti bili su fukoksantin, 19'-heksanoiloksifukoksantin, klorofil *b*, peridinin, 19'-butanoiloksifukoksantin i zeaksantin koji su pokazali da su najvažnije skupine fitoplanktona bile dijatomeje, primnezioficeje, zelene alge, dinoflagelati, krizoficeje i cijanobakterije. U fitoplanktonskim cvatovima, koji se javljaju nakon jačih donosa slatkih voda u proljeće i jesen, prevladavale su dijatomeje (fukoksantin), dok je na početku ljetne stratifikacije dijatomejska populacija bila zamijenjena populacijom složenijeg sastava u kojoj su prevladavale primnezioficeje (19'-heksanoiloksifukoksantin). Intenzivni cvatovi koji nastaju nakon jačih pulseva riječnih donosa mogu doseći i oligotrofne vode zapadne Istre, ali su najčešće ograničeni na površinski sloj iznad 5 m dubine.