

Impacts of high-nitrate freshwater inputs on macrotidal ecosystems. I. Seasonal evolution of nutrient limitation for the diatom-dominated phytoplankton of the Bay of Brest (France)

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ABSTRACT: The chemical factors (inorganic nitrogen, phosphate, silicic acid) that potentially or actually control primary production were determined for the Bay of Brest, France, a macrotidal ecosystem submitted to high-nitrate-loaded freshwater inputs (winter nitrate freshwater concentrations >700 μM , Si:N molar ratio as low as 0.2, i.e. among the lowest ever published). Intensive data collection and observations were carried out from February 1993 to March 1994 to determine the variations of physical [salinity, temperature, photosynthetically active radiation (PAR), freshwater discharges] and chemical (oxygen and nutrients) parameters and their impacts on the phytoplankton cycle (fluorescence, pigments, primary production). With insufficient PAR the winter stocks of nutrients were almost non-utilized and the nitrate excess was exported to the adjacent ocean, due to rapid tidal exchange. By early April, a diatom-dominated spring bloom developed (chlorophyll *a* maximum = 7.7 $\mu\text{g l}^{-1}$; primary production maximum = 2.34 $\text{g C m}^{-2} \text{d}^{-1}$) under high initial nutrient concentrations. Silicic acid was rapidly exhausted over the whole water column; it is inferred to be the primary limiting factor responsible for the collapse of the spring bloom by mid-May. Successive phytoplankton developments characterized the period of secondary blooms during summer and fall (successive surface chlorophyll *a* maxima = 3.5, 1.6, 1.8 and 1.0 $\mu\text{g l}^{-1}$; primary production = 1.24, 1.18 and 0.35 $\text{g C m}^{-2} \text{d}^{-1}$). Those secondary blooms developed under lower nutrient concentrations, mostly originating from nutrient recycling. Until August, Si and P most likely limited primary production, whereas the last stage of the productive period in September seemed to be N limited instead, this being a period of total nitrate depletion in almost the whole water column. Si limitation of spring blooms has become a common feature in coastal ecosystems that receive freshwater inputs with Si:N molar ratios <1. The peculiarity of Si limitation in the Bay of Brest is its extension through the summer period.

KEY WORDS: Coastal ecosystem · Phytoplankton dynamics · Macrotidal · Nutrient limitation · Silicon · Eutrophication

INTRODUCTION

The long-term increase of anthropogenic inputs of nitrogen and phosphorus into the coastal waters of most developed countries (Howarth et al. 1996) has 3

potential consequences. First, it can induce a spectacular enhancement of the productivity of phytoplankton populations that were previously N and/or P limited, leading to severe eutrophication problems (Meybeck & Helmer 1989, Smayda 1990). Second, declines in dissolved Si:N ratios in coastal waters can induce subsequent silicic acid (= 'silicates') limitation of diatom-dominated populations (Officer & Ryther 1980, Smayda 1990, Conley & Malone 1992, Conley et al. 1993). Third, this Si limitation can disturb the typical suc-

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sion of phytoplankton in temperate waters and provoke a shift of dominance from siliceous (diatoms) to non-siliceous (flagellates) species (Billen et al. 1991).

Numerous studies have already focused on the eutrophication of ecosystems highly loaded with N and P (review in Howarth et al. 1996) but the importance of Si as a limiting factor of phytoplankton growth has long been ignored (e.g. Tréguer et al. 1995) and only a few studies (Smayda 1990, Egge & Aksnes 1992, Bodeanu 1993) have envisaged the impact of the decline of the Si:N ratios in coastal waters on the composition of phytoplankton populations. The aim of this paper is to address the question of how the chemical factors (nitrogen, phosphorus, silicon) potentially or actually control pelagic primary production during the different seasons of a complete annual cycle in a macrotidal ecosystem of western Europe (the Bay of Brest, France), which receives freshwaters that have been dramatically enriched in nitrate in recent decades (Del Amo 1996, Le Pape et al. 1996). The response of the phytoplankton community structure to this nitrate perturbation is presented in a companion paper (Del Amo et al. 1997).

Among world ocean coastal waters, those of western Europe are characterized by an intensive tidal exchange with the ocean. The Bay of Brest is a typical macrotidal ecosystem that is strongly influenced by freshwater inputs originating from 2 main rivers: Elorn River, discharging in the northern basin, and Aulne River, in the south, whose catchment areas are submitted to intensive agriculture runoff (Daniel 1995). This semi-enclosed basin of 180 km² is connected to the Atlantic Ocean (Iroise Sea) by the Brest Channel, which is 1.8 km wide and 50 m deep. Except for the fairways of the main rivers and the Brest Channel, the Bay is generally shallow: more than 50% of the total area is <5 m deep, and only 13% is more than 20 m deep. Between 1/35 and 1/25 of the Bay volume is

renewed at each tide (maximum tidal amplitude: 8 m; tide periodicity: 12 h 15 min) by the ocean (Delmas & Tréguer 1983). Strong tidal currents (maximum tidal currents: 2.6 m s⁻¹; SHOM 1968) coupled with surface wind stress induce rapid vertical mixing of the Bay water, hindering the typical summer stratification observed in other coastal ecosystems (D'Elia et al. 1992, Turner & Rabalais 1994, Malej et al. 1995).

Within the framework of a multidisciplinary study (Contrat de Baie), an intensive data collection was carried out in the Bay of Brest during the 1993–1994 annual cycle, to describe the seasonal pattern of phytoplankton primary production and community structure in relation to nutrient availability and the physical environment. Although during the last 20 yr the riverine discharge of nitrate into the Bay has doubled, neither did the annual amount of phytoplankton biomass increase nor the oxygen concentrations above the sediments significantly decrease (Le Pape et al. 1996). The resistance of this ecosystem to eutrophication has been explained by the fast renewal rate of the Bay water (Delmas 1981). Although non-eutrophicated, the Bay is definitely chemically perturbed, as evidenced by the long-term decrease of the Si:N ratio (silicic acid : nitrate + nitrite + ammonium) during summer from 2.00 in 1975 to 0.33 in 1993 (Del Amo 1996, Le Pape et al. 1996). Consequences of such a drastic change in the chemical composition of the Bay of Brest waters on the spring phytoplankton dynamics have been partly described by Ragueneau et al. (1994), who suggested that the 1992 diatom-dominated bloom collapsed due to Si limitation. Because the dominance of diatoms was known to occur not only during spring but during the entire productive period (Quéguiner & Tréguer 1984), this study is concerned with the investigation of the chemical control of phytoplankton during the complete annual cycle of 1993–1994.

MATERIALS AND METHODS

Sampling strategy. Thirty cruises were conducted in the Bay of Brest (Fig. 1) from 15 February 1993 to 21 March 1994; sampling frequency ranged from twice a week during spring to once a month during winter. As phytoplankton developments have already been shown to respond to decreasing tidal mixing (Ragueneau et al. 1996), sampling dates were chosen close to neap tides. Water sampling was performed at Stn R₃ (48° 20' N, 4° 30' W), located in the mixing area between waters of the northern and southern basins of the Bay of Brest and the ocean (Delmas 1981, Quéguiner 1982, Delmas & Tré-

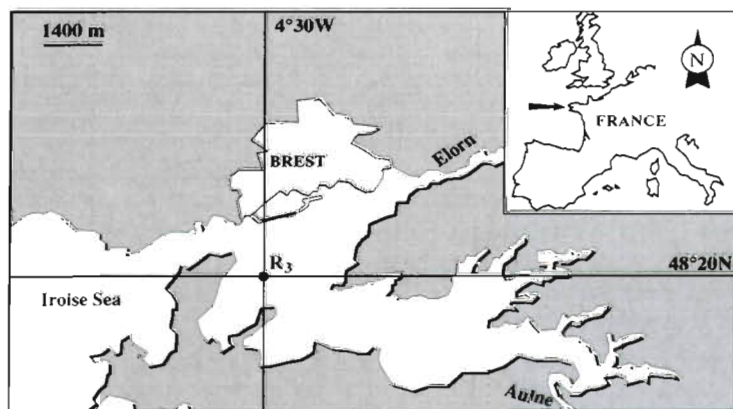


Fig. 1. Study area and location of station (R₃) sampled during 1993–1994 in the Bay of Brest, France

guer 1983). Stn R₃ is considered as typical of the major part of the Bay (see Ragueneau et al. 1994 for details), and has been extensively studied during the last 20 yr [R.N.O. (Réseau National d'Observation de la qualité du milieu marin, Ministère de l'Environnement, France) database, Quéguiner & Tréguer 1984, Jacq et al. 1985, Dauchez et al. 1991, Ragueneau et al. 1994, 1996, Daniel 1995].

Salinity, temperature, fluorescence and photosynthetically active radiation (PAR) profiles were made by using a Seabird SBE 19 Seacat CTD profiler, coupled with a LiCor LI-1000 PAR sensor. Discrete sampling was carried out by using 8 l or 30 l Niskin samplers, fitted with non-toxic silicone tubings, at depths corresponding respectively to 100 (surface), 25, 10, and 1 % of surface PAR, and 2 m above the bottom.

Ammonium and dissolved oxygen samples were immediately fixed by addition of the working reagents. Nutrient samples were either stored at 4°C for silicic acid analysis, or frozen (−20°C) for nitrate + nitrite and phosphate measurements. Filtrations for particulate organic carbon (POC) and nitrogen (PON), as well as for pigments [chlorophyll *a* (chl *a*) and phaeopigments], were also done immediately after sample collection. Dissolved oxygen, nutrients and carbon uptake measurements were performed at the 5 sampled depths, whereas measurements of pigments, POC, and PON were restricted to surface samples.

Daily river flow data for Elorn and Aulne Rivers were provided by Direction Régionale de l'Environnement - Service du bassin Loire Bretagne. Solar radiation data were provided by Météorologie Nationale (Brest-Guipavas weather station).

Analytical methods. Salinity values measured by the CTD were calibrated against discrete surface and bottom measurements which were made using a Guildline Autosol 8400 salinometer, precision: ± 0.001 psu. Dissolved oxygen was analyzed by the Winkler method according to Strickland & Parsons (1972), precision: ± 0.6 %. Ammonium was analyzed according to Koroleff (1969), precision: ± 3 %. Nitrate + nitrite and silicic acid concentrations were determined according to the spectrophotometric method of Gordon et al. (1993) using an Alpkem RFA-300 Analyzer, precision: ± 0.8 % for nitrate + nitrite and ± 0.5 % for silicic acid. Phosphate concentrations were measured on a Technicon Auto Analyzer II, according to Tréguer & Le Corre (1975), precision: ± 1 %.

For chl *a* and phaeopigment determinations, 0.5 to 1.0 l seawater was filtered through 0.6 μ m Nuclepore filters, which were immediately frozen (−20°C) until further analysis by the fluorometric method of Yentsch & Menzel (1963), using a calibrated Turner 112 fluorometer, precision: ± 1 %. For POC and PON measurements, 0.5 to 1.0 l seawater was filtered through pre-combusted (450°C for 4 h) Whatman GF/F filters,

which were immediately frozen (−20°C) until further analysis on a modified Carlo Erba analyzer model N 1500, following the method of Strickland & Parsons (1972), precision: ± 1 %.

For primary production measurements 24 h dawn-to-dawn incubations were performed under simulated *in situ* conditions by using nickel screens of various light transmissions. 1 ml (ca 147 kBq) of NaH¹⁴CO₃ solution (Carbon 14 Centralen®, Denmark) was added to 250 ml of unfiltered seawater into polycarbonate bottles. After incubation, samples were filtered onto 0.6 μ m Nuclepore filters, which were kept frozen until processing. At the laboratory, filters were dried at 30°C for 12 h and counted by liquid scintillation on a Packard Tri-Carb model 1600 TR liquid scintillation counter without previous acidification. Total CO₂ was estimated to be 2200 μ M for calculation of incorporated ¹⁴C. Specific activities of each serial batch of the ¹⁴C stock solutions were given by the supplier and were double-checked at the laboratory by drawing 20 μ l of the radioactive solution into a scintillation vial containing 50 μ l 6 N NaOH, followed by addition of liquid scintillation cocktail, which was then immediately measured (to avoid any CO₂ loss) by liquid scintillation. No attempt was made to measure dark or time-zero uptake of ¹⁴C. Water column integrated primary production ($\text{mmol C m}^{-2} \text{d}^{-1}$) was calculated by trapezoidal integration of the individual depth profiles of daily primary production. The rate nearest to the surface (1 m depth) was assumed to be constant up to 0 m, and the one nearest to the bottom (2 m above sediment) was assumed to be constant down to the bottom.

RESULTS

The physical environment

The highest river flows recorded during the study period mainly occurred during winter (Fig. 2a); these events followed the usual seasonal pattern, although they were much more pronounced in comparison with the 1970–1992 average evolution. Some smaller water discharges occurred during spring, while summer was characterized by very low river flows. Daily solar radiation (Fig. 2b) followed the seasonal evolution of temperate areas. The lowest values were observed in winter ($< 500 \text{ J cm}^{-2} \text{d}^{-1}$), while summer maximum values reached $3500 \text{ J cm}^{-2} \text{d}^{-1}$. However, the solar radiation was characterized by a high day-to-day variability, and values as low as $700 \text{ J cm}^{-2} \text{d}^{-1}$ (i.e. close to winter maxima) were frequently measured during summer. Surface PAR ranged from 40 (December 1994) to $1700 \mu\text{E m}^{-2} \text{s}^{-1}$ (May 1993) and the photic depth (1 % of surface PAR) ranged from 15 m (February 1994) to

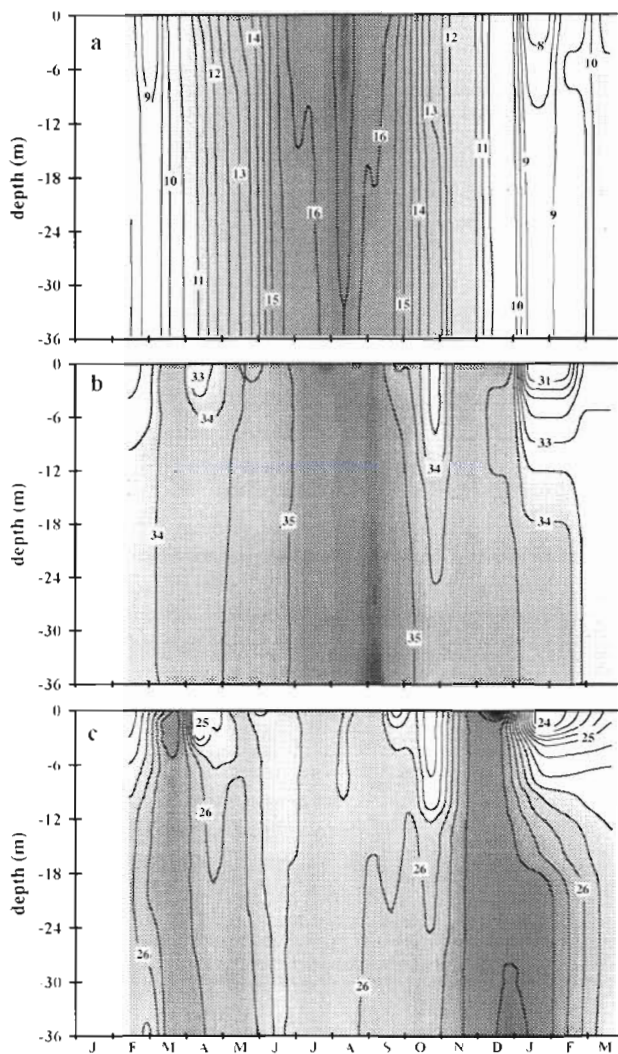


Fig. 3 Seasonal variations of physical parameters at Stn R₁ (Bay of Brest) during 1993–1994 based on CTD profiler measurements. (a) Temperature in °C, (b) salinity in psu, (c) density (σ_t)

processes (grazer excretion and bacterial remineralization), remained low ($\leq 1 \mu\text{M}$) during most of the year. The end of the phytoplankton uptake period in October was marked by a transitory ammonium increase (up to $2.2 \mu\text{M}$), afterwards, concentrations stabilized around $1 \mu\text{M}$ during winter. The seasonal variations of silicic acid and phosphate (Fig. 4c, d) were close to that of nitrate ($r_s = 0.62$ and 0.61 for silicic acid and phosphate, respectively, Table 1), with high winter concentrations during periods of freshwater inputs and low summer concentrations reflecting a net phytoplankton uptake. The most prominent feature in the silicic acid evolution (Fig. 4c) was the decrease to undetectable concentrations over the whole water column from 27 April to 3 May following uptake by diatoms. After the spring bloom, the enhancement of biogenic silica dissolution

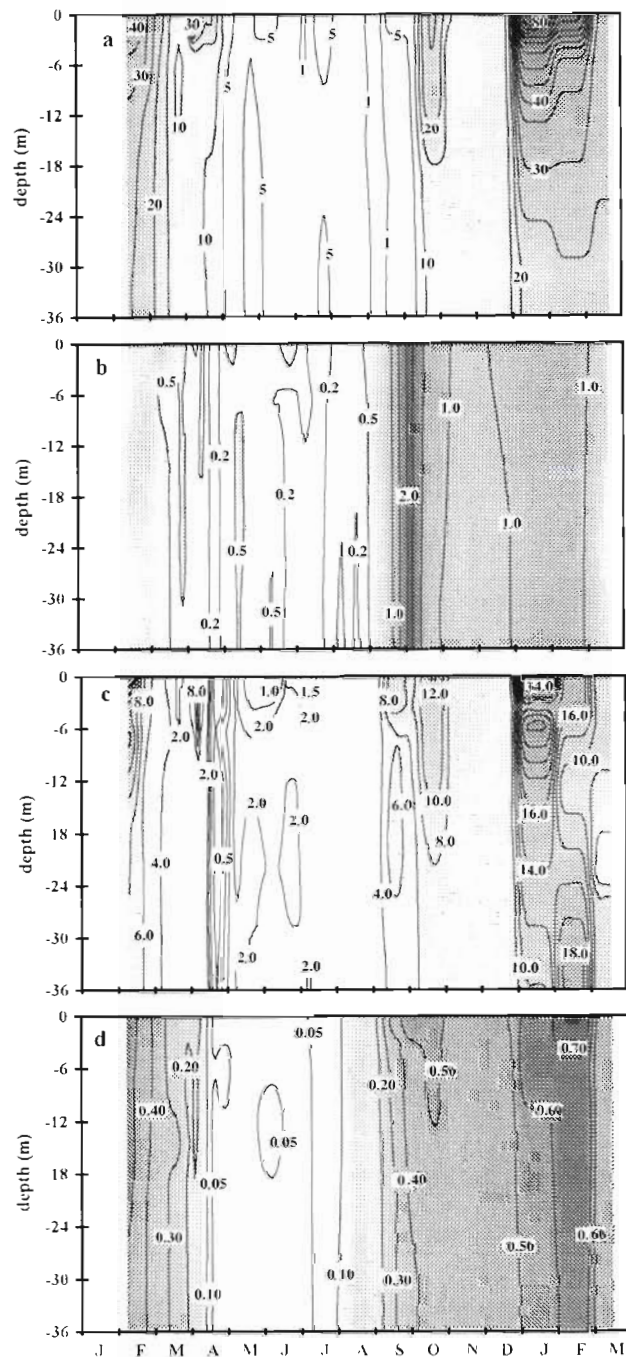


Fig. 4 Seasonal variations of nutrient concentrations (in μM) at Stn R₁ during 1993–1994. (a) Nitrate+nitrite, (b) ammonium, (c) silicic acid, (d) phosphate

in bottom waters (Ragueneau et al. 1994) caused the gradual increase of silicic acid concentrations (with faster increase in bottom waters; Fig. 4c). Concentrations remained below $4 \mu\text{M}$ throughout the water column until the resumption of freshwater inputs in September. Phosphate (Fig. 4d) remained $< 0.8 \mu\text{M}$ all year round. The lowest concentrations ($< 0.1 \mu\text{M}$) were ob-

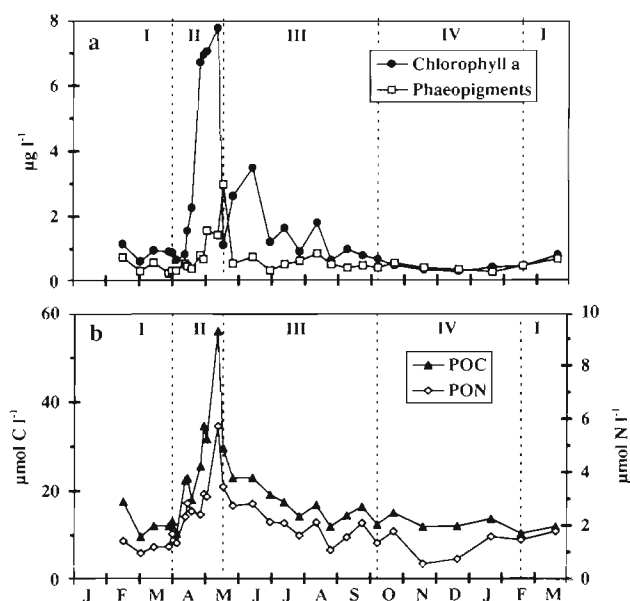


Fig. 5. Time-course variations of biomass in surface waters at Stn R₃ during 1993–1994. (a) Chlorophyll *a* and phaeopigments; (b) particulate organic carbon and nitrogen. Vertical dashed lines delimit the different periods distinguished during the annual plankton cycle: I, spring transition; II, spring bloom; III, secondary blooms; IV, fall/winter (see text for details)

served from late April to early August, reaching undetectable values (i.e. $<0.02 \mu\text{M}$) only on 1 occasion in surface waters (18 May).

The biological cycle

Based on the seasonal patterns of biomass parameters (Fig. 5) and primary production (Fig. 6), we divided the annual cycle into 4 periods characterized by distinct features: Phase I, the spring transition period (15 February to 1 April 1993), shows slight increases in biomass and primary production; Phase II, the spring bloom period (1 April to 18 May 1993), is marked by the rapid increase of the phytoplankton biomass and primary production; Phase III, successive phytoplankton developments of smaller amplitude characterize the secondary blooms period (18 May to 7 October 1993), and Phase IV, the fall/winter period (7 October 1993 to 17 February 1994), is the period of lowest phytoplankton biomass and activity.

Biomass parameters

Surface chl *a* concentrations (Fig. 5a) remained under $1 \mu\text{g l}^{-1}$ until 15 April. During the spring bloom, surface chl *a* reached its annual maximum ($7.7 \mu\text{g l}^{-1}$

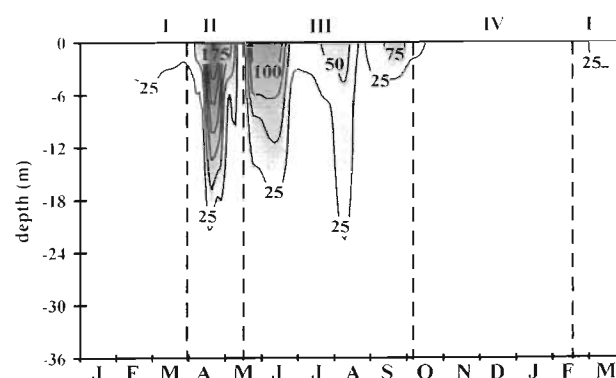


Fig. 6. Seasonal evolution of primary production rates (in $\text{mg C m}^{-3} \text{d}^{-1}$) at Stn R₃ during 1993–1994. Vertical dashed lines and numbers as in Fig. 5

on 13 May). The secondary blooms period was then characterized by successive peaks showing a somewhat gradual decreasing trend until the end of the period. Except during the secondary blooms period, the evolution of phaeopigments (Fig. 5a) was generally close to that of chl *a*, with a distinct maximum at the end of the spring bloom ($3.0 \mu\text{g l}^{-1}$ on 18 May).

The same observation was also made for POC and PON (Fig. 5b): only maximum concentrations ($55.8 \mu\text{mol C l}^{-1}$ and $5.8 \mu\text{mol N l}^{-1}$) were simultaneous with the chl *a* maximum of 13 May. Concentrations then decreased gradually during the secondary blooms period, and winter minima were reached by November ($<15.0 \mu\text{mol C l}^{-1}$ and $<2.0 \mu\text{mol N l}^{-1}$).

Primary production

Up to 86% of the integrated annual primary production occurred during the 6 mo of the productive period, from early April to early October (Fig. 6). Within that period (spring bloom and secondary blooms periods), several maxima in primary production (Fig. 6, see also Fig. 9) occurred. The maximum carbon production rate ($2.34 \text{ g C m}^{-2} \text{d}^{-1}$) corresponded to the spring bloom period and was reached on 19 April, 24 d before the maximum surface chl *a* concentration (Fig. 5a). During the secondary blooms period 3 peaks of primary production were observed. The first one ($1.24 \text{ g C m}^{-2} \text{d}^{-1}$ on 14 June) was reached simultaneously with the corresponding peak of chl *a*. In late July-early August, a peak of primary production ($1.18 \text{ g C m}^{-2} \text{d}^{-1}$) overlapped 2 small peaks of chl *a*. Finally, the productive period ended with an increase of the primary production restricted to the surface layer ($91.2 \text{ mg C m}^{-3} \text{d}^{-1}$ at the surface but only $0.35 \text{ g C m}^{-2} \text{d}^{-1}$ in the whole water column) and with low chl *a* content. It is noteworthy that from April to August 1993, when solar radiation was maximum (Fig. 2b), the major peaks of primary

production extended deep in the water column (Fig. 6). In contrast, during the spring transition periods (March 1993 and March 1994) as well as at the end of the secondary blooms period (September 1993), primary production was confined to the surface.

Due to the importance of vertical mixing and water renewal in the Bay of Brest, oxygen concentrations did not exhibit important seasonal variations; waters were close to saturation or super-saturated with respect to oxygen over the entire water column during most of the annual cycle (Fig. 7). Maximal super-saturation was observed during the fall bloom (up to 138% on 9 September), while transient under-saturation was only moderate and never went below 95% (minimum observed on 20 December).

DISCUSSION

As a common feature in many coastal ecosystems (e.g. Legendre et al. 1986, Peterson 1986, Tsuda et al. 1994, Malej et al. 1995) the onset of the productive period in the Bay of Brest is driven by physical parameters (Ragueneau et al. 1996). In spring 1993 the beginning of the diatom-dominated spring bloom coincided with the stabilization of the water column ($\Delta\sigma_t$ between surface and bottom waters: 2.78 g kg^{-1}) due to freshwater discharges, and increased light availability during stratification enabled the enhancement of primary production. However, light limitation was still liable to occur afterwards with regard to the high day-to-day variability of solar radiation (Fig. 2b), as also described in similar ecosystems (e.g. Wafar et al. 1983, Lohrenz et al. 1990, Pennock & Sharp 1994). In the course of the productive period, the fast tidal exchange between the Bay and the ocean waters (Delmas & Tréguer 1983, Souchu 1986) coupled with reduced freshwater inputs and increasing phytoplankton uptake during spring and summer led to progressive depletion of the nutrients of the Bay. Exhaustion occurred during this period (Fig. 4), suggesting that nutrients exert a close control on primary production.

The potential limitation of primary production by nutrient availability is first approached with reference to the composition of the biogenic matter. According to Redfield et al. (1963) and to Brzezinski (1985), the standard molar ratio for silicic acid, DIN (dissolved inorganic nitrogen = nitrate + nitrite + ammonium), and phosphate is Si:N:P = 16:16:1. This ratio corresponds to the composition of diatoms, which are the primary contributors of the Bay of Brest phytoplankton production (Del Amo et al. 1997), and it also gives estimates of the relative nutrient requirements by diatoms (e.g. Levasseur & Theriault 1987, Fisher et al. 1992). In Fig. 8a, b the Si:N = 1, N:P = 16 and Si:P = 16 lines delimit 6

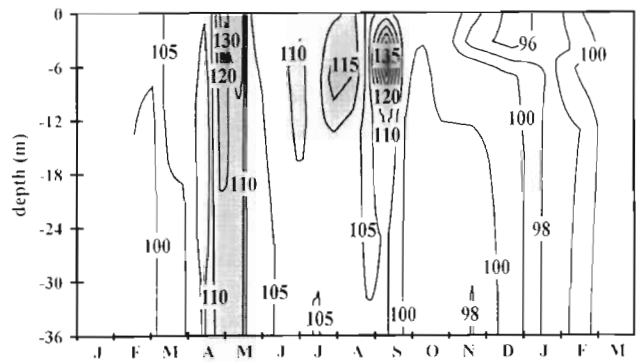


Fig. 7. Seasonal variations of dissolved oxygen saturation values (in %) at Stn R₃ during 1993–1994

areas, each of them being characterized by the potentially limiting nutrients in order of priority (see Fig. 8 legend). The data points of the 1993–1994 annual cycle are plotted in Fig. 8a, b.

During the fall/winter and the spring transition periods (Fig. 8a) nutrient concentrations were high (Fig. 4) and therefore nutrient limitation was not likely to occur. The Si:P ratios were close to 16 (18.5 ± 2.5), i.e. silicic acid and phosphate were balanced with regard to the potential diatom demand. In contrast, low Si:N (0.37 ± 0.04) and high N:P ratios (53.7 ± 6.3) indicated excess DIN originating from river inputs. During this period the 2 major processes that controlled nutrient concentrations in the Bay were the freshwater inputs and the tidal exchange with the adjacent Iroise Sea.

During the productive period (spring bloom and secondary blooms periods), these processes were predominated by phytoplankton uptake and nutrient recycling (N and P regeneration, biogenic silica dissolution) and the nutrient concentrations decreased to very low values (Fig. 4), with residual nutrient ratios clearly different from the standards (Fig. 8b). The locations of the data points in Fig. 8b clearly suggest that P and Si were the most common potentially limiting major nutrients, but some potentially N-limited events were also evidenced. A comprehensive view of the potential nutrient limitation of phytoplankton production during the productive period, referring to standard Si:N:P ratios, is illustrated in the 2-dimensional (depth and time) diagram (Fig. 8c).

Of course, examination of residual nutrient ratios alone does not give definitive conclusions about the actual limiting nutrient for algal growth, because real limitation depends on the species composition and the specific kinetic curves, as well as on the regeneration rates of each nutrient. An additional approach to determine nutrient limitation is therefore to compare the *in situ* nutrient concentrations to half-saturation constants for nutrient uptake (K_m). *In situ* nutrient concen-

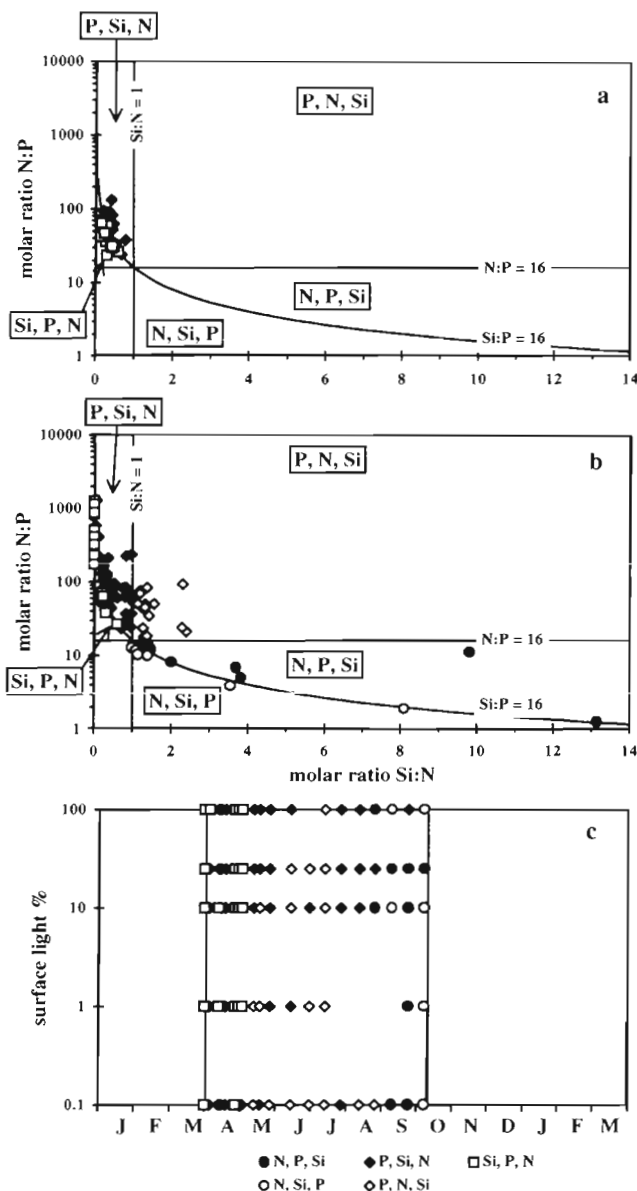


Fig. 8. Synthetic graph of Si:N:P molar ratios in the water column of the Bay of Brest at Stn R₃ during 1993–1994. In each area delimited by the Brzezinski (1985) ratio and by the Redfield et al. (1963) ratio (Si:N:P = 16:16:1), the potential limiting nutrients (PLN) are reported in order of priority. (a) Winter period: from 15 February to 1 April 1993 and from 7 October 1993 to 21 March 1994; (b) productive period: from 1 April to 7 October 1993 (see text for details); (c) 2-dimensional diagram (over depth and time) showing PLNs in order of priority within the productive period. Circles: N first PLN (●: P second PLN; ○: Si second PLN); diamonds: P first PLN (◆: Si second PLN; ◇: N second PLN); (□): Si first PLN and P second PLN

trations below the corresponding K_m corresponded to nutritional conditions that greatly reduced nutrient uptake rates and potentially limited algal growth rates. K_m values reported in the literature for natural popula-

tions present a large range of variations (Table 2). For the Bay of Brest, Dauchez et al. (1991) established uptake limitation when nitrate was below 1.6 μM , which is in agreement with mean K_m values for DIN uptake of 2.0 and 1.0 μM respectively proposed by Fisher et al. (1988) and Dortch & Whitledge (1992). The authors of both studies considered mean K_m values of 0.2 and 2.0 μM for phosphate and silicic acid uptake, respectively. Indeed, Egge & Aksnes (1992) reported that diatom dominance occurred when silicic acid concentrations exceeded a threshold of 2.0 μM . Natural diatom assemblages collected in the Bay of Brest during 1994 yielded K_m values for silicic acid uptake ranging from 1.6 μM for summer diatoms to 3.3 μM for spring diatoms (Del Amo 1996). Therefore, for the conditions prevailing in the Bay of Brest we herein considered that concentrations ≤ 0.2 , 2.0 and 2.0 μM , for phosphate, silicic acid and DIN, respectively, were limiting for nutrient uptake by phytoplankton during our study period. If we compare the *in situ* nutrient concentrations to these reference values, it is clear that Si, N or P can be in turn limiting during the productive period (Table 3).

Fig. 9 gives a synthetic view of the most probable chemical limiting factor(s) of phytoplankton growth in the Bay of Brest for each period, taking into account the 2 different approaches considered (potential limitation in reference to standard composition ratios, and comparison between the residual nutrient concentrations and K_m). The first diatom spring bloom (April to early May) is obviously Si limited (Fig. 9). From 27 April to 3 May 1993 this limitation was especially severe when silicic acid concentrations dropped to $<0.05 \mu\text{M}$, far below the 3.3 μM K_m value for spring diatom populations (Del Amo 1996). Such a severe limitation of silicic acid uptake rates provides indirect evidence for Si limitation of diatom growth rates. The silicic acid exhaustion period was indeed followed by the decline of the first primary production peak (diatom dominance $>97\%$; Del Amo et al. 1997) as in spring 1992 (Ragueneau et al. 1994).

During the first half of the secondary blooms period (until mid-July), most of the silicic acid concentrations stayed below or close to 2 μM throughout the water column (Fig. 4). Compared with the K_m value of 1.6 μM measured by Del Amo (1996) for summer diatoms, it is clear that, although those concentrations were still sufficient to sustain diatom production, the latter was probably not occurring at its maximal rate. During the same period and even later, phosphate concentrations remained $\leq K_m$ (0.2 μM) until early September, and with regard to residual nutrient ratios (Fig. 8c), P appeared as the primarily limiting nutrient until mid-August. However, despite these permanently low phosphate concentrations ($<0.1 \mu\text{M}$ from 27 April to 12 August),

Table 2. Ranges of half-saturation constants (K_m) for nutrient uptake, and optimal algal composition or uptake ratios reported in the literature. DIN: dissolved inorganic nitrogen

	Silicic acid	Phosphate	DIN	Source
K_m (μM)	1–5 0.03–3.37	0.1–0.5 0.0001–2	1–2 0.01–10.3	Fisher et al. (1988) Dortch & Whitledge (1992)
Composition ratios		N:P = 16; Si:P = 16; Si:N = 1 N:P = 7–87; Si:P = 1–96 N:P = 10–30; Si:P > 3; Si:N = 1		Redfield et al. (1963), Brzezinski (1985) Hecky & Kilham (1988) Dortch & Whitledge (1992)

Table 3. Number of data points which suggest nutrient limitation (total data points in parentheses) according to mean values of K_m (see text for details) during each period

	Fall/winter	Spring transition	Spring bloom	Secondary blooms
DIN $\leq 2 \mu\text{M}$	0 (25)	0 (27)	4 (49)	18 (41)
Silicic acid $\leq 2 \mu\text{M}$	0 (25)	1 (27)	19 (49)	11 (41)
Phosphate $\leq 0.2 \mu\text{M}$	0 (25)	0 (27)	45 (49)	35 (40)

phytoplankton production did occur during the productive period (Fig. 6). Such a feature has already been described for other coastal systems (e.g. Wafar et al. 1983, Cadée & Hegeman 1993). Among the 3 major nutrients (N, P and Si), phosphate is probably the more difficult nutrient to assess for primary production limitation due to its fast biological (Admiraal & Werner 1983) and geochemical (Lean et al. 1983, Conley et al. 1988) turnover. Furthermore, due to the high reactivity of phosphorus with the suspended matter, processes of adsorption/desorption of phosphate from the particulate matter are liable to control concentrations of dissolved phosphorus in the water column (e.g. Carrit & Goodgal 1954). A specific study of interactions between the suspended matter and the water column in the Bay of Brest (Delmas 1981) has shown that a single measurement of phosphate concentration in the water column does not allow the correct assessment of actual phosphate availability for phytoplankton growth. Furthermore, optimum composition ratios for phosphate, in comparison with those for the other major nutrients, vary over a large range among phytoplankton species (Table 2). The ranges of these ratios are greater than those of Redfield et al. (1963) and Brzezinski (1985). During the whole productive period, Si:P ratios were seldom >96 and high N:P values (≥ 87) were found from mid-April to mid-June. Therefore, it seems difficult to discard P as a limiting nutrient during this period, and both P and Si limitation could be inferred for the Bay of Brest.

Interestingly, within this secondary blooms period (from May to July), the P, Si limitation order of priority is gradually replaced by a P, N limitation order of priority, and this occurred from the bottom to the surface

layers (Fig. 8c). This suggests silicic acid regeneration from biogenic silica dissolution at the sediment-water interface, progressively influencing the upper layers during summer (Ragueneau et al. 1994). At that time, when nutrient inputs by riverine discharge are at their lowest (Fig. 2a), silicic acid regeneration should be the main process allowing diatoms to keep growing, the diatoms thus remaining the dominant phytoplankton group throughout the productive period (Del Amo et al. 1997).

Referring to nutrient ratios, N appears to be limiting during the last stage of the secondary blooms period, from late August to early October (Fig. 8). Referring to half-saturation constants for DIN uptake (Table 2), very low DIN concentrations likely limited phytoplankton uptake in early July (from 0.8 to 2.0 μM in the

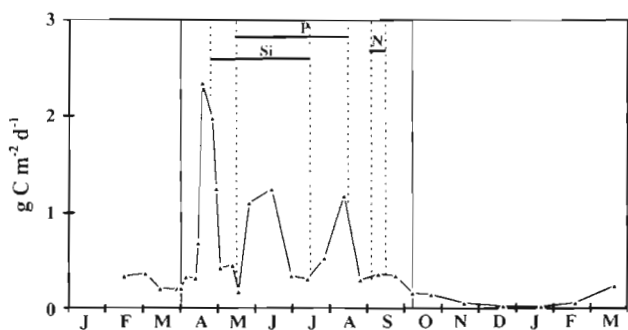


Fig. 9. Seasonal variation of depth-integrated primary production rates at Stn R₃ during 1993–1994 and periods of nutrient limitation: Si, silicic acid limitation; P, phosphate limitation; N, dissolved nitrogen limitation (see text). Solid vertical lines delineate the productive period

Table 4. Nutrients inferred for primary production limitation in some coastal ecosystems, and corresponding Si:N ratios in fresh waters from the respective main rivers. 'Pristine' conditions: natural conditions in the world's major unpolluted rivers. nd: not determined

River (coastal ecosystem)	Riverine Si:N ratio	Limiting nutrient	
		Spring	Summer
'Pristine' conditions	17.4 ¹	–	–
Sacramento-San Joaquin (San Francisco Bay)	1.4 ²	Si or N ²	Not limited ¹¹
Mississippi (Gulf of Mexico)	0.9 ³	Si and P ⁷	N ⁷
Po (Northern Adriatic Sea)	0.7 ³	nd	P ¹²
Susquehanna (Chesapeake Bay)	0.7 ⁴	Si ⁸	N ¹³
Morlaix River (English Channel)	0.5 ⁵	Si ⁹	N ⁹
Aulne/Elorn (Bay of Brest)	0.2 ⁶	Si ¹⁰	Si and P, then N ¹⁰

¹Meybeck (1982); ²Cloern (1996); ³Justic et al. (1995); ⁴Fisher et al. (1988); ⁵Wafar (1981); ⁶P. Tréguer unpubl. data; ⁷Nelson & Dortch (1996) and Dortch & Whittedge (1992); ⁸Conley & Malone (1992); ⁹Wafar et al. (1983); ¹⁰present study; ¹¹Alpine & Cloern (1988) and Cloern (1991); ¹²Revelante & Gilmartin (1976); ¹³Fisher et al. (1992) and D'Elia et al. (1992)

water column), and especially in early September (DIN ranged from 0.2 to 0.7 μM). During this last period nitrate was below the analytical detection limit ($<0.05 \mu\text{M}$) throughout most of the water column (Fig. 4). Consideration of N limitation could be biased by not taking into account of dissolved organic nitrogen, e.g. urea. Urea concentrations in surface samples were low in July and September (0.4 and 0.5 μM , respectively; Daniel 1995), and remained 0.6 μM until early October (Daniel 1995), i.e. total dissolved nitrogen concentrations were still $<2.0 \mu\text{M}$ during this period. Taking urea into account in Si:N:P ratio calculations for the surface water leads to the conclusion that potential N limitation is likely to occur only by early September. Thus, both approaches support the idea that N limitation seemed to occur only during the last stage of the productive period (Fig. 9).

Therefore, during the 1993–1994 annual cycle, the diatom-dominated phytoplankton in the Bay of Brest seemed to have been strongly Si limited in the earlier stage of the spring bloom, followed by a potential Si and P limitation period; late summer growth of phytoplankton populations being N limited. Fifteen years ago (1981–1982) the first nutrient depleted in the Bay during the summer period was nitrate and not silicic acid (Quéguiner 1982, Quéguiner & Tréguer 1984). Le Pape et al. (1996) reported that summer DIN depleted periods are now less frequent and of shorter duration than 15 yr ago.

Following Officer & Ryther (1980), Smayda (1990), and Conley & Malone (1992), this study supports the idea that Si limitation of siliceous-phytoplankton-dominated spring blooms represents the response of the Bay of Brest ecosystem to high-nitrate-loaded freshwater inputs. In the 1990s, winter nitrate freshwater concentrations have increased to $>700 \mu\text{M}$ and the Si:N molar ratio in the Bay of Brest tributaries has dropped to 0.2 (Table 4), i.e. far below the pristine conditions given

by Meybeck (1982). Such a Si:N ratio is among the lowest measured in freshwaters (Table 4); for instance, it is 7 times lower than that for the Sacramento and San Joaquin Rivers (San Francisco Bay; Cloern 1996). Table 4 shows that Si is the limiting factor for the spring diatom blooms in various marine ecosystems, at least when Si:N ratios in freshwater inputs drop below 1. But coastal ecosystems that are less perturbed by nitrate than the Bay of Brest did not show Si limitation for summer blooms (Table 4). For instance the Gulf of Mexico which receives Mississippi River inputs (Si:N = 0.9) supports summer phytoplankton blooms that are N limited (Nelson & Dortch 1996). Comparable situations have been described for the Northern Adriatic Sea (Po River; Revelante & Gilmartin 1976), the Chesapeake Bay (Susquehanna River; Fisher et al. 1992, D'Elia et al. 1992) and the western English Channel (Morlaix River; Wafar et al. 1983). In the Bay of Brest, the dominance of phytoplankton by diatoms during spring and summer in spite of the severe Si limitation is explained by an original 'silicate pump' of well-mixed ecosystems (Del Amo et al. 1997).

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