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# Biomass and composition of the phytoplankton in the Río de la Plata: large-scale distribution and relationship with environmental variables during a spring cruise

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## Abstract

The distributions of phytoplankton taxa and biomass were recorded to assess their association to environmental variables in the Río de la Plata, a shallow and highly turbid estuary. Forty-seven CTD stations covering the whole estuary were sampled from 5 to 16 November 2001. At 31 stations, dissolved oxygen concentration, pH, and light intensity at 50 cm depth intervals were recorded in situ; also, samples were taken for measurement of dissolved nutrient levels (nitrate, ammonium, phosphate, and silicate), suspended particulate matter and particulate organic matter loads, chlorophyll (*a*, *b*, and *c*) concentration, and phytoplankton identification. Stations ordination (multi-dimensional scaling, MDS, analysis) on environmental data suggested two main groups (upper–middle and lower estuary) but stressed the gradual change of conditions along the ecosystem. MDS on pigment data suggested a tendency for higher biomass levels in the lower estuary; MDS based on species indicated two main groups corresponding to lower and upper–middle estuarine stations. Spearman rank correlations of environmental and pigment similarity matrices suggested that salinity, the ratio of mixed to photic depth, and nitrates were the variables best explaining pigments distribution; in turn, the variables best explaining species distribution were salinity and seston. Results suggest an overriding importance of salinity–light gradients in modulating biomass levels and species distribution in the Río de la Plata. This result is consistent with processes known to modulate phytoplankton distribution and production in other highly turbid estuaries.

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**Keywords:** Phytoplankton; Chlorophyll; Estuaries; Uruguay; Argentina; Río de la Plata

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## 1. Introduction

The distribution of phytoplankton biomass is largely associated to nutrients availability at large

spatial scales; at medium and small scales biophysical processes like the light environment, water column stratification/turbulence, temperature, and grazing may also modulate productivity and biomass levels (Platt, 1972; Therriault et al., 1978; Joint and Pomroy, 1981; Pennock and Sharp, 1994).

In highly turbid coastal waters where nutrients are present at moderate to high concentrations, light availability is usually the limiting factor for primary production (Underwood and Kromkamp, 1999). Under such conditions the distribution of phytoplankton biomass can be expected to reflect horizontal gradients in those variables that modulate light availability (Pennock, 1985; Cole and Cloern, 1987; Cloern, 1999; Underwood and Kromkamp, 1999, and references therein).

Such horizontal gradients may result from the mixing of continental freshwaters with marine coastal waters. In many estuaries, the abrupt increase in ionic strength causes particles to flocculate, to sink and, after being carried landward by bottom saline waters, to accumulate at the tip of the salt-wedge (Postma, 1967; Mann and Lazier, 1996). This hydrodynamic trap results in an estuarine turbidity maximum followed seaward by a sharp reduction of the seston load and a consequent increase in water transparency. Phytoplankton production tends to peak seaward of the front as response to improved light environment; vertical haline stratification in this zone also contributes to enhance light availability for the phytoplankton (Pennock, 1985). Changes in the phytoplankton community representing an ecological succession also occur associated to such environmental gradients (Underwood and Kromkamp, 1999).

Because of their hydrodynamics and massive human settlements, estuaries are particularly susceptible to anthropogenic impact. Their integrity is currently under risk worldwide (Flindt et al., 1999; Kiddon et al., 2003), and the Río de la Plata estuary is no exception (Wells and Daborn, 1997; CARP-CTMFM, 1999). Understanding the modulation of biological processes by physical variability in estuaries is relevant to implementation of proper management strategies (Cloern, 2001). However, knowledge of the ecology of the Río

de la Plata is rather poor, particularly on basic aspects related to plankton distribution, dynamics, and regulating processes (Wells and Daborn, 1997; Calliari et al., 2003). Such lack of knowledge appears as critical in this case, considering the importance of the Río de la Plata as nursery area for several fish species (Macchi et al., 1996; Acuña et al., 1997; Vizziano, 2001; Martínez and Retta, 2001), some of them under heavy commercial exploitation by Uruguayan and Argentinean fleets (Rey and Arena, 2003).

Accordingly, the aim of this paper is to provide information on the phytoplankton biomass distribution (as chlorophyll (Chl) pigments) and taxonomic composition in the Río de la Plata, and assess their association to environmental variables. In particular, we explore potential regulating mechanisms of biomass levels within the estuary (i.e. light vs. nutrients limitation). A detailed taxonomic analysis of the phytoplankton community, the biology and adaptations of most representative species is presented elsewhere (Gómez et al., 2003; Gómez et al., submitted). Here, we explore how the spatial distribution of Chl and species reflects large-scale environmental variability. Considering the characteristics of the study area, we expected biomass and composition of the phytoplankton to follow main environmental gradients, namely salinity and turbidity.

## 2. Methods

### 2.1. Study area

The Río de la Plata is a coastal plain, microtidal estuary 250 km long and 230 km wide at the mouth (Fig. 1). Main tributaries are the Paraná and Uruguay rivers, characterized by flows with opposing patterns of seasonal variability. Resulting inflow (annual average ca.  $22,000 \text{ m}^3 \text{ s}^{-1}$ ) has moderated maximum and minimum in March–June (late summer–early winter) and January (summer), respectively (Nagy et al., 2002). Also, important variability can occur at larger time scales (i.e. inter annual) associated to ENSO events which induce floods (warm phase) or droughts (cold phase).

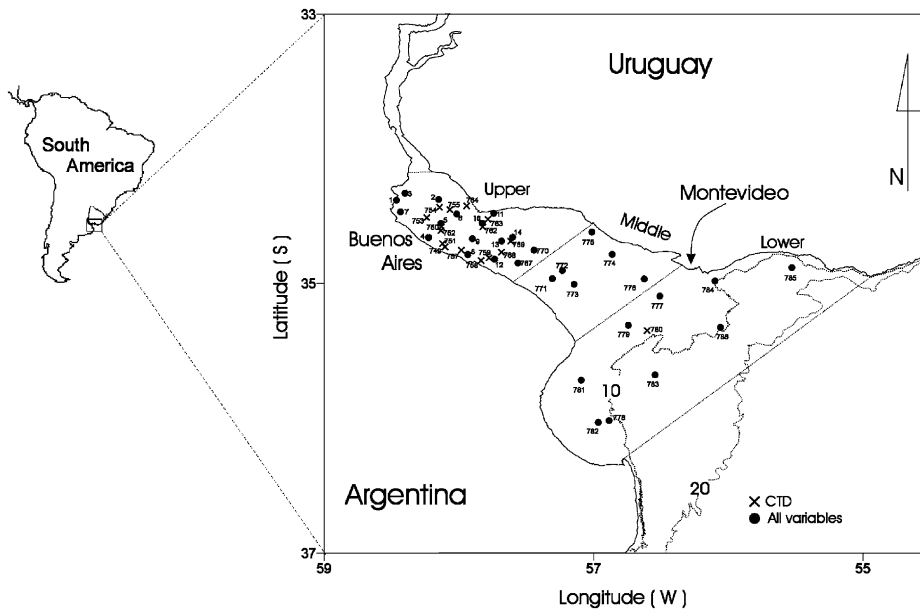


Fig. 1. Map of the Río de la Plata on a regional scale, showing 10 and 20 m isolines. Sampling stations depicted as crosses (only CTD) or full circles (all variables).

Understanding of the physical oceanography and basic chemical processes of the Río de la Plata is under development (Framiñan and Brown, 1996; Guerrero et al., 1997; Nagy et al., 2002). Mixing of fresh and marine waters results in the typical salt-wedge pattern with potentially strong vertical gradients, which can be eroded by wind stress due to prevailing shallow depths (<10 m). The position of the turbidity maximum is highly variable at short time scales according to tide, freshwater flow, and wind variability (Framiñan and Brown, 1996). Nutrient levels are high along the freshwater areas (salinity <1) but may drop seaward of the turbidity front due to dilution with marine waters (and presumably due to phytoplankton assimilation as well; Nagy et al., 2002).

The plankton of the Río de la Plata is to a large extent unknown, although several studies have advanced knowledge for given areas and taxa (i.e. Frenguelli, 1933; Müller-Melchers, 1949, 1959; Ferrando, 1962; Gómez and Bauer, 1998a, b). In particular, there is no published information on large-scale distribution of phytoplankton biomass and its relationship with environmental variables and processes. Reported biomass values (as Chl-*a*)

ranged between 0.1 and  $38 \mu\text{g l}^{-1}$ , and appear influenced by water transparency (Gómez et al., 2001; Nagy et al., 2002; Cervetto et al., 2002). Toxic algal blooms caused by dinoflagellates (*Alexandrium tamarense*, *Gymnodinium catenatum*, and *Dynophysis acuminata*) and cyanophyceas (*Microcystis aeruginosa*) have been recorded at inshore stations (Méndez et al., 1997; De León and Yunes, 2001).

## 2.2. Sampling and laboratory methods

A total of 47 stations were sampled from 5 to 16 November 2001 (Fig. 1) on board R./V. *Holmberg* at stations deeper than 5 m, and on the smaller A.R.A. *Cormorán* vessel over shallower waters (ca. 2.5–5 m). Vertical profiles of salinity and water temperature were recorded with a SEABIRD 19-01 CTD. At 31 stations, surface water samples were taken for pigment analysis and for taxonomic identification of phytoplankton, for dissolved nutrients analyses (nitrate, ammonium, phosphate, and silicate), and for the estimation of suspended particulate matter (SPM) and suspended particulate organic matter (POM) loads. Dissolved

oxygen concentration was recorded using an electronic sensor (YSI). Vertical profiles of light intensity in the water column were obtained with a radiometer (Li-Cor, fitted with a  $2\pi$  quantum sensor). Light attenuation was estimated according to the Lambert–Beer equation  $I_z = I_0 e^{-kz}$ , where  $k$  is the attenuation coefficient,  $I_z$  is the irradiance at depth  $z$ ,  $I_0$  is the irradiance at 0 depth, and  $z$  is the depth (m). Depth of the photic layer ( $Z_{\text{pho}}$ ) was assumed to extend to 1% of surface light intensity, as generally accepted (i.e. Cole and Cloern, 1987; Cloern, 1999; Jassby et al., 2002). Stratification (surface–bottom salinity difference, divided by water column depth) and depth of the mixed layer ( $Z_{\text{mix}}$ ) were determined from CTD data.

Chl-*a*, Chl-*b*, and Chl-*c*, and phaeopigments were determined spectrophotometrically according to Parsons et al. (1985) from 250 to 1000 ml aliquots filtered through Whatman GF/F filters (pore size ca. 0.7  $\mu\text{m}$ ), which were stored frozen at  $-80^\circ\text{C}$  for ca. 15 days. A second aliquot was taken and pre-filtered through a 20  $\mu\text{m}$  sieve in order to estimate pigments in the  $<20\ \mu\text{m}$  size fraction. At the laboratory, filters were extracted in 90% acetone and the absorbance of the extract read on a Beckman UV-26 spectrophotometer. Pigment concentrations were calculated employing the equations of Jeffrey and Humphrey (1975). Water samples for nutrients were kept frozen until laboratory analyses, which were performed following Parsons et al. (1985) using a DU-650 spectrophotometer. For SPM and POM estimation, 500–1500 ml of water was filtered on pre-combusted and weighed GF/F filters, which were kept frozen ( $-80^\circ\text{C}$ ) for ca. 15 days. At the laboratory filters were dried at  $60^\circ\text{C}$  for 24 h and weighed (SPM estimation), thereafter combusted at  $500^\circ\text{C}$  for 30 min and weighed again (POM estimation).

Phytoplankton species were identified using an optical microscope (Olympus BX 50) with phase and interferential contrast. Diatoms were cleaned with  $\text{H}_2\text{O}_2$ , washed thoroughly using distilled water and mounted on microscope slides with Naphrax<sup>®</sup>. Phytoplankton were counted according to Lund et al. (1958) with an inverted microscope (Olympus CK2) at  $400\times$  using 5 ml sedimentation chambers, a suitable size according

to the amount of suspended solids. The entire chamber was examined and each cell was counted as a unit.

### 2.3. Data analysis

Patterns in the ordination of samples were explored by non-metric multi-dimensional scaling (MDS) (Field et al., 1982) for both environmental and biological (pigments and species, separately) data. MDS for environmental data was based on an Euclidean Distance similarity matrix calculated from fourth root transformed data (Clarke and Warwick, 1994); similarity matrix for pigments and species data was constructed employing the Bray–Curtis index. A complementary cluster analysis with group average sorting, and based on the same distance matrix aided in the identification of groups. Groups were defined using a dissimilarity threshold value equal to the average of the distance matrix (Arancibia, 1988; Rodríguez and Castro, 2003). The relationship between environmental and biological data was tested by weighed Spearman rank correlation of the corresponding similarity matrices (Clarke and Ainsworth, 1993). This approach quantifies how well the community structure is explained by a given set of environmental variables. The combination of environmental variables resulting in the maximum correlation was considered the best ‘explanation’ for the observed distribution. We present a slight modification of such correlation method where, instead of species abundances (a matrix of species  $\times$  sites), we used pigments concentration (biomass) data (a matrix of pigments  $\times$  sites). The significance of Spearman rank correlations was tested by a random permutation algorithm under the hypothesis of no relation between multivariate patterns from the two sets of samples. Environmental variables included in the analysis were salinity, temperature, pH, oxygen saturation, SPM, POM, nitrate, and  $Z_{\text{mix}}:Z_{\text{pho}}$  ratio. A pre-screening excluded the variables ammonium, phosphates, and silicates due to their availability well above limiting conditions for phytoplankton over all of the study area; to avoid co-linearity we also removed the light attenuation coefficient and photic depth, and stratification (correlated with

$Z_{\text{mix}}:Z_{\text{pho}}$  ratio, and salinity, respectively). Variables in our pigment matrix were Chl-*a*, Chl-*b*, Chl-*c*, and Chl-*a* < 20  $\mu\text{m}$ . Species considered for MDS analysis were those whose numerical abundance was equal to (or higher than) 1% summed over all samples, and with an incidence of at least 15%. Species with abundance equal or higher than 50% at any given sample were included, even if they did not fulfil the former criteria. Statistical analyses were performed using PRIMER 5.0.1 (multivariate analyses) and Statistica 6.0 packages (Spearman correlations and Mann–Whitney *U*-test).

### 3. Results

#### 3.1. Environmental variability

Maps of selected environmental and biological variables are shown in Figs. 2 and 3. Surface and bottom salinity ranged from 0 to 18.4 and 0 to 28.8, respectively (Fig. 2), and a salinity front was present in the range ca. 1–18 at the surface and 1–22 at the bottom. The spatial scale of the front was ca. 70 km at the surface and 85 km at the bottom (maximum gradients 0.4 and 0.5  $\text{km}^{-1}$ , respectively). Moderate surface–bottom differences in salinity (up to 1.29  $\text{m}^{-1}$ ) occurred at the lower estuary. Surface temperature varied from 23.8 °C in the upper estuary to 18.7 °C at the mouth (Fig. 2); bottom temperature range was similar and no important vertical differences existed (max 0.15 °C  $\text{m}^{-1}$ ).

Highest nutrient levels were generally found in the upper estuary stations near Buenos Aires and in some cases near Montevideo (Table 1, Figs. 2 and 3). A general pattern for nutrients was to decrease from the river to the marine-influenced area. This pattern was particularly clear for silicates, and for nitrates and phosphates to a lesser extent, but not for ammonium (see Fig. 4 for plots and statistics). Seston load also tended to decrease from upper to lower estuary (Fig. 3) and POM comprised between 7% and 41% of SPM. The light attenuation coefficient was highest at upper and mid-estuary (Fig. 3); the ratio of the surface layer mixing depth to photic depth ( $Z_{\text{mix}}:Z_{\text{pho}}$  ratio) ranged between 0.4 and 13.7,

also with highest values at the upper and mid-estuary (Fig. 3).

MDS and cluster analyses on physical–chemical data evidenced two main groups representing upper–middle, and lower estuarine environments, plus a small transition group (St. 776, St. 777, and St. 779; Fig. 5). The upper–middle group appeared on the MDS plane as two clouds, one formed by most upper estuary stations and the second by the middle estuary stations plus a few upper ones. The low estuary group was rather spread, reflecting a gradual increase in the marine influence according to the order: St. 781, St. 784, St. 782, St. 783, and Sts. 785–786. St. 8 did not join any group, reflecting the very high nutrient loads and low oxygen levels recorded there.

#### 3.2. Pigments distribution and association to environmental variables

Chl-*a* levels were moderate to high (1–27.3  $\mu\text{g l}^{-1}$ , Fig. 3), while Chl-*b* and Chl-*c* were present at much lower concentrations: 0–0.7 and 0–3.1  $\mu\text{g l}^{-1}$ , respectively. No clear pattern of Chl-*a* distribution was evident for the upper and middle estuary. But Chl-*a* (particularly the < 20  $\mu\text{m}$  size fraction) and Chl-*c* levels clearly increased in the lower estuary at the salinity front (Mann–Whitney test,  $U = 41$ ,  $p < 0.05$ ,  $n = 31$ ; and  $U = 49$ ,  $p < 0.05$ ,  $n = 31$ , for Chl-*a* and Chl-*c*, respectively). A localized bloom was recorded on the Uruguayan coast dominated by *Protoperidinium* spp., *Skeletonema costatum* and *Prorocentrum* spp. together reaching a density over 10<sup>5</sup> cells  $\text{l}^{-1}$  (St. 784, Fig. 3). Phaeopigments ranged between 0.5 and 11.9  $\mu\text{g l}^{-1}$ , and at some station levels were higher than the sum of Chl-*a*, Chl-*b*, and Chl-*c*; Chl:phaeopigments ratio ranged between 0.5 and 27.6 (average 3.7), and highest values tended to occur associated to Chl maxima.

MDS ordination of stations based on pigment data did not reveal any clear pattern, and most clustered in a large group (Fig. 5). Exception was St. 784 (lower estuary), where highest biomass occurred; there was also a tendency for stations in the lower estuary to form a sub-group, reflecting the increase in Chl-*a*, Chl-*c*, and Chl-*a* < 20  $\mu\text{m}$  at that zone. Rank correlations between

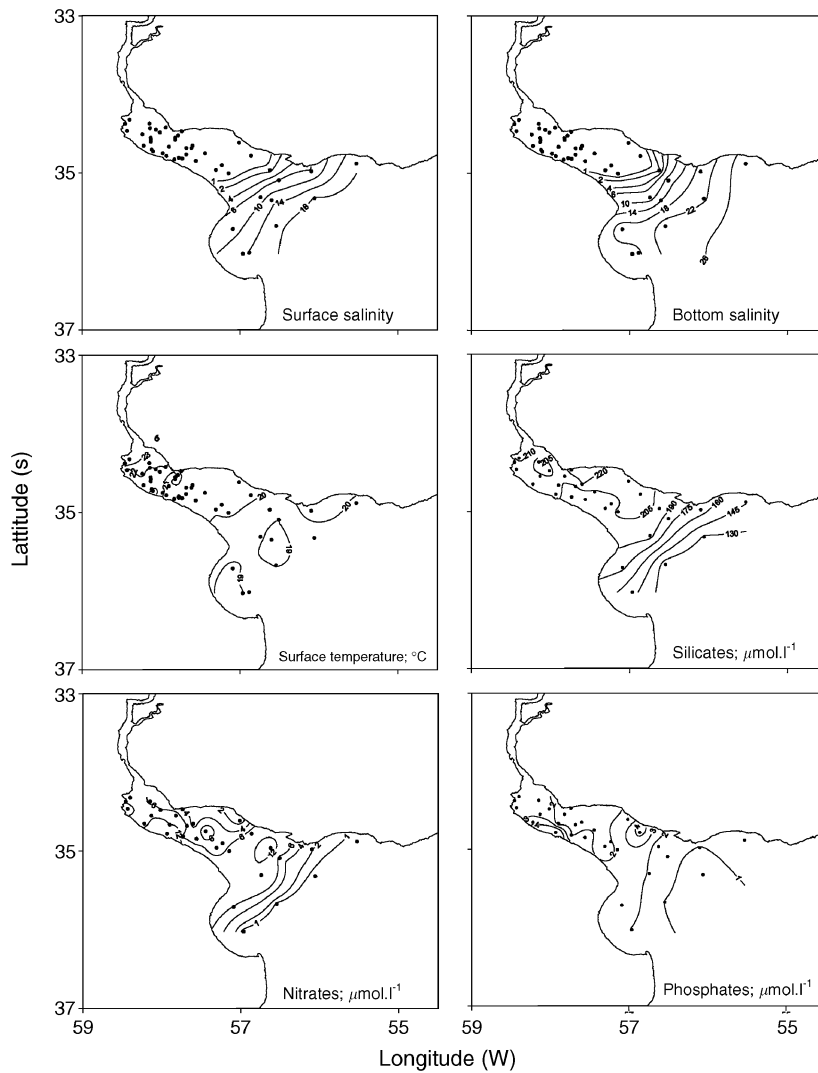


Fig. 2. Horizontal distribution of selected variables measured at the Río de la Plata during the period 5–16 November 2001. Sampling stations are indicated as full circles.

environmental and pigment similarity matrices (best correlation = 0.4,  $p < 0.01$ ) selected a combination of salinity,  $Z_{\text{mix}}:Z_{\text{pho}}$  ratio, and nitrate levels as the variables that best explained pigment distribution.

### 3.3. Phytoplankton assemblage composition

Main phytoplankton taxa are listed in Table 2. Ordination of stations from the species-based MDS analysis resulted in two main groups

corresponding to lower estuary on one side, and the upper and mid-estuarine stations on the other side (Fig. 5). A transition group formed by St. 776, St. 777, St. 779, and St. 781 was also suggested. This pattern was best explained by salinity and seston (Spearman rank correlation = 0.8,  $p < 0.01$ ). Most representative taxa at the upper estuary were filamentous chlorophytes, chlorococals, desmidiaceans, and diatoms. The lower estuary was dominated by diatoms (*S. costatum*) and dinoflagellates (*Protoperidinium* spp. and *Prorocentrum*



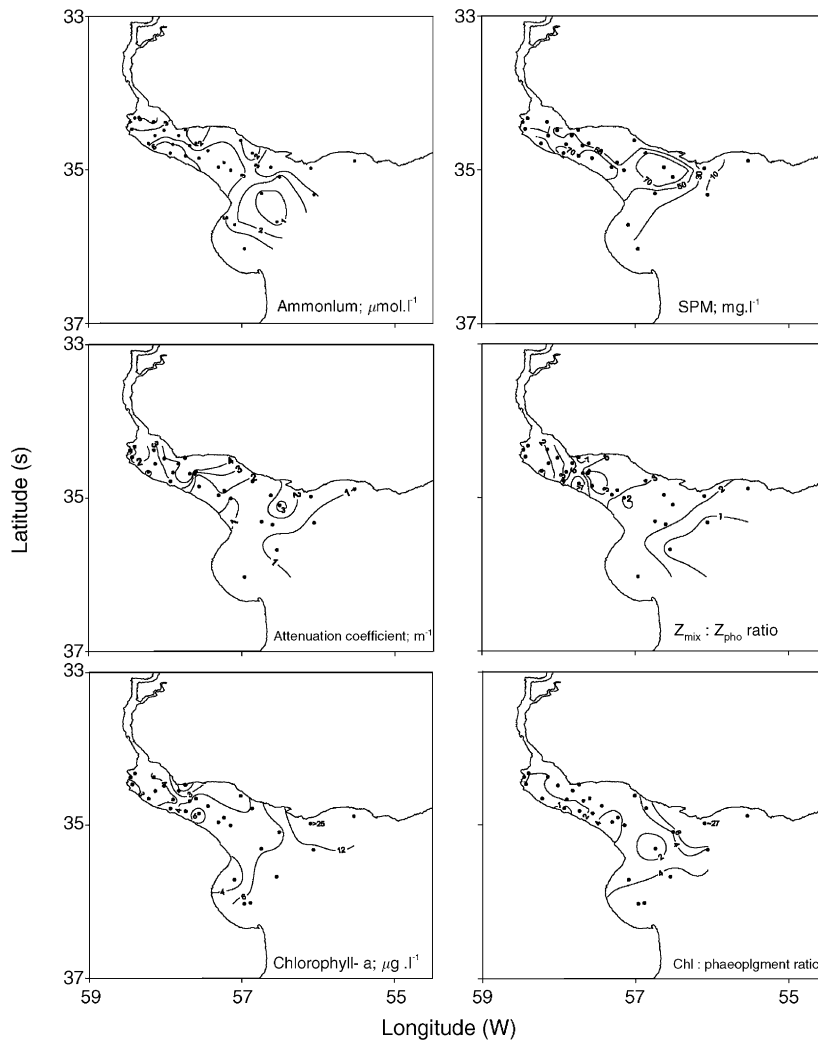


Fig. 3. Distribution of Chl-*a* Chl:phaeopigments ratio and selected variables measured at the Río de la Plata during the period 5–16 November 2001. Sampling stations are indicated as full circles.

spp.), while at the transition stations the same taxa dominated accompanied by few freshwater taxa present at lower densities.

#### 4. Discussion

##### 4.1. Environmental variability

Hydrographic conditions during sampling were normal for November according to inflow, tem-

perature, and salinity, as compared to time series data (Nagy et al., 1996; INAA, 2002): inflow was moderately above historic values during October, and slightly below average during November. Thus, results obtained during this survey should be broadly representative of mid-spring time conditions in the Río de la Plata.

Main environmental variability occurred along the longitudinal axis, most notably the salinity gradient at the lower estuary. Such gradients occurred also for almost every physical–chemical

Table 1

Mean value of environmental and biological variables recorded at the three regions of the Río de la Plata during the period 5–16 November 2001

	Upper	Middle	Lower
Salinity	0.02	0.32	11.52
Temperature (°C)	21.1	20.1	19.3
SPM ( $\text{mg l}^{-1}$ )	70.4	58.2	31.2
POM ( $\text{mg l}^{-1}$ )	11.2	9.8	5.3
$K$ ( $\text{m}^{-1}$ )	3.7	1.6	1.4
$Z_{\text{mix}}:Z_{\text{pho}}$ ratio	7.3	2	1.1
$\text{NO}_3^-$ ( $\mu\text{mol l}^{-1}$ )	8.41	8.28	3.34
$\text{NH}_4^+$ ( $\mu\text{mol l}^{-1}$ )	4.02	3.81	2.77
$\text{PO}_4^-$ ( $\mu\text{mol l}^{-1}$ )	2.50	2.15	1.54
$\text{SiO}_2$ ( $\mu\text{mol l}^{-1}$ )	208.5	203.6	158.4
Oxygen (%)	71.5	85.3	87.5
Chl- <i>a</i> ( $\mu\text{g l}^{-1}$ )	4.09	4.70	9.39
Chl- <i>b</i> ( $\mu\text{g l}^{-1}$ )	0.22	0.18	0.19
Chl- <i>c</i> ( $\mu\text{g l}^{-1}$ )	0.46	0.24	1.26

variable recorded (Figs. 2–4). In particular, silica levels closely followed the mixing of fresh and marine waters without evidence of assimilative losses to (or sources from decaying) planktonic biomass. And the reduction of SPM and POM along the estuary lead to improved light penetration in the lower estuary (Fig. 3, Table 1).

Overall nutrient concentrations were high and within the ranges earlier reported for this ecosystem more than a decade ago (Nagy et al., 2002); however, values were similar or even much lower than those reported for highly eutrophic estuaries (i.e. Gironde:  $\text{NO}_3^-$  range 53–202  $\mu\text{M l}^{-1}$ ;  $\text{PO}_4^-$  range 2.4–5.5  $\mu\text{M l}^{-1}$ , Irigoien & Castel, 1997; Colne:  $\text{NO}_3^-$  range 3–190  $\mu\text{M}$ , Kocum et al., 2002a). High amounts of ammonium, nitrate, and phosphate along with relatively low oxygen levels (i.e. St. 1 and St. 8) indicate punctual inputs in the upper estuary. This is emphasized by the close vicinity of such stations to the most heavily populated and industrial area of the region, and is consistent with earlier estimations that untreated sewage from that area accounts for ca. 25% of the total nutrient loads entering the upper estuary (Pizarro and Orlando, 1985).

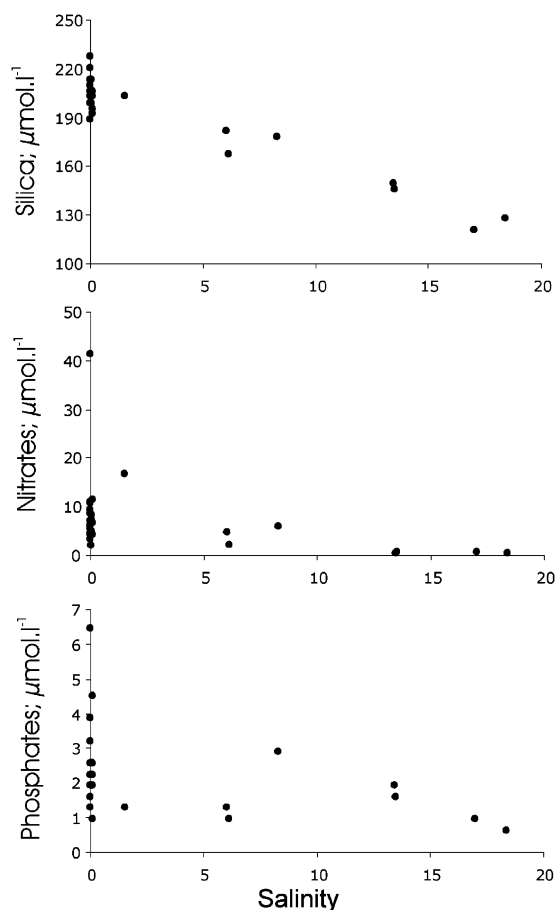


Fig. 4. Scatterplots of the concentration of selected nutrients vs. salinity (mixing diagrams). Spearman rank correlations were  $-0.78$ ,  $p < 0.01$  (silica);  $-0.47$ ,  $p < 0.01$  (nitrates);  $-0.41$ ,  $p < 0.05$  (phosphates);  $-0.1$ ,  $p > 0.05$  (ammonium, not shown);  $n = 30$  in all cases.

#### 4.2. Biological variability

Chl-*a* levels were consistent with antecedents available (Nagy et al., 2002). Phytoplankton biomass increased along the salinity front and peaked at salinities above ca. 10, close to the Uruguayan coast. This result suggests that conditions for phytoplankton improved at increasing salinities. Rank correlation indicated that such pattern was best explained by a combination of salinity,  $Z_{\text{mix}}:Z_{\text{pho}}$  ratio and nitrates, stressing the role of the light environment for modulating



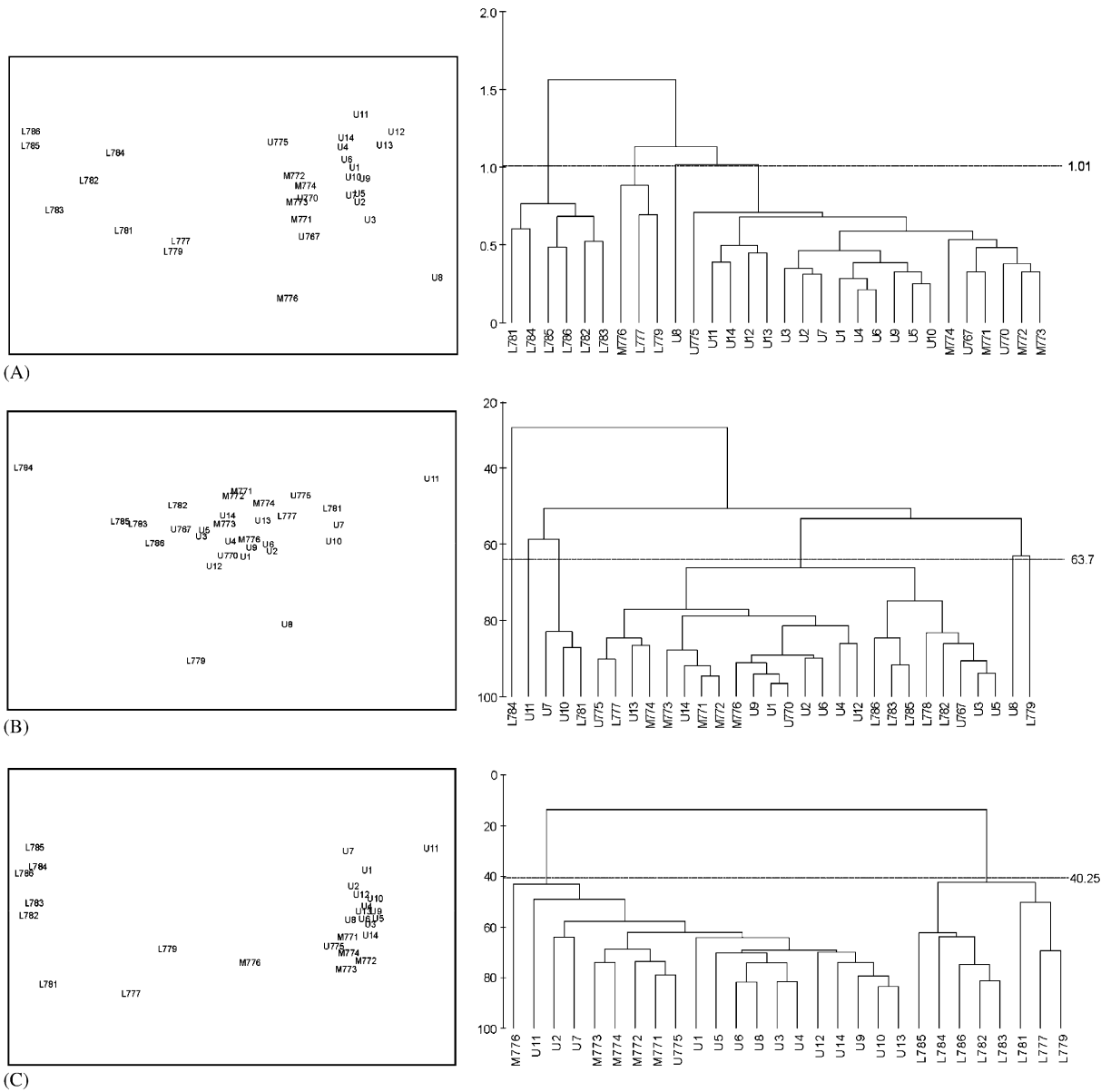


Fig. 5. Non-metric MDS analysis and corresponding cluster diagrams of sampling stations, according to (A) physical and chemical data; (B) pigments data; and (C) phytoplankton species abundance. Prefix U, M, or L before station number corresponds to upper, middle, or lower estuary, respectively. Dashed line in the cluster diagrams indicates the mean value of the corresponding similarity matrix.

phytoplankton biomass in the Río de la Plata. That is consistent with findings at other turbid estuaries, i.e. San Francisco (Alpine and Cloern, 1988), Delaware (Pennock, 1985), Schelde (Kromkamp and Peene, 1995), Gironde (Irigoien and

Castel, 1997), Colne (Kocum et al., 2002b), and in general with the overriding importance of light for the phytoplankton in turbid estuaries (Wofsy, 1983; Cloern, 1987; Grobbelaar, 1990; Cloern, 1996).

Table 2

Main phytoplankton taxa identified in samples from the Río de la Plata for the period 5–16 November 2001

Taxa	Upper		Middle		Lower	
	Avg.	Stdev	Avg.	Stdev	Avg.	Stdev
<i>Actinocyclus normanii</i>	58	164	525	718	240	321
<i>Aulacoseira ambigua</i>	3553	3840	300	424	0	0
<i>Aulacoseira distans</i>	1326	3562	400	616	0	0
<i>Aulacoseira granulata</i> (four forms)	14,632	15,097	2825	1725	460	654
<i>Chaetoceros</i> sp.	0	0	0	0	1840	820
<i>Cyclotella meneghiniana</i>	1205	1101	75	96	20	45
<i>Skeletonema costatum</i>	0	0	32,825	28,907	218,440	127,382
<i>Eunotia monodon</i>	84	90	0	0	0	0
<i>Fragilaria heideni</i>	242	516	0	0	0	0
<i>Nitzschia hungarica</i>	32	58	0	0	0	0
<i>Actinastrum hantzschii</i>	1832	2074	0	0	0	0
<i>Closterium cynthia</i>	84	90	0	0	0	0
<i>Closterium jenneri</i>	258	353	75	96	0	0
<i>Eutetramorus fotti</i>	942	1866	0	0	0	0
<i>Monorafidium arcuatum</i>	1105	961	0	0	120	179
<i>Monorafidium mirabile</i>	847	678	25	50	0	0
<i>Monorafidium tortile</i>	142	198	25	50	20	45
<i>Planctonema lauterbornii</i>	20,837	18,646	500	1000	0	0
<i>Scenedesmus acuminatus</i>	358	460	0	0	0	0
<i>Scenedesmus intermedius</i>	105	181	0	0	0	0
<i>Scenedesmus quadricauda</i>	1226	696	0	0	0	0
<i>Schoederia antillanum</i>	47	90	0	0	6760	10,347
<i>Schoederia setigera</i>	11	32	0	0	2620	2463
<i>Ulothrix</i> cf. <i>subconstricta</i>	14,471	14,190	475	950	0	0
<i>Pseudoanabaena constricta</i>	7626	31,796	0	0	0	0
<i>Ceratium tripos</i>	0	0	0	0	60	55
<i>Protoperidinium</i> spp. (two spp.)	5	23	6125	7497	28,220	38,255
<i>Prorocentrum</i> spp. (two spp.)	0	0	125	96	23,740	19,928

Numbers are expressed in cells l<sup>-1</sup>; Avg. = average, Stdev = standard deviation.

The formation of a halocline is one of the processes triggering the development of blooms in turbid estuaries (Pennock, 1985). A  $Z_{\text{mix}}:Z_{\text{pho}}$  ratio ca. 5–6 is the threshold to allow for net primary production under light limiting conditions (Wofsy, 1983; Cloern, 1987, 1996; Grobbelaar, 1990). Our results show that light attenuation was higher in the upper and mid-estuary, but due to a rather shallow water column (2–4 m) that ratio did not reach very high values (mean  $Z_{\text{mix}}:Z_{\text{pho}} = 7.3$ ; only in four out of 17 cases was such ratio > 5). At the lower estuary, stratification and lower light attenuation effectively reduced the  $Z_{\text{mix}}:Z_{\text{pho}}$  index (mean  $Z_{\text{mix}}:Z_{\text{pho}} = 1.4$ ; such ratio was never > 5 at this zone).

This mechanism can explain the higher biomass levels found at the lower estuary. This interpreta-

tion is also consistent with preliminary primary production estimates in the Río de la Plata that suggested light limitation at mid-estuarine stations (Gómez et al., 2001). The alternative explanation stating that higher Chl-*a* levels at the outer estuary reflected accumulation at the frontal area rather than active growth does not seem very likely: highest Chl:phaeopigments ratios were associated to Chl maxima, indicating that they resulted from active growth. If Chl maxima had resulted from accumulative processes, then a large fraction of detrital matter (indicated by high phaeopigments concentration) would be expected to occur along with high Chl-*a* levels.

Nutrients could be ruled out as playing a primary modulating role given their availability

at saturating levels. Over most of the estuary nutrients were present at concentrations well above—by a factor of 5–10, or more—the half saturating levels for nutrient-limited phytoplankton (Chapra, 1997; *vide*, Jassby et al., 2002; Irigoien and Castel, 1997); only at the outer region their concentration fell to comparatively lower values, and at a single station was nitrogen close to limiting levels (St. 783, nitrates + ammonium =  $1.42 \mu\text{mol l}^{-1}$ ). This is consistent with findings at the hyper eutrophic Colne where nutrient limitation was only bound to happen in waters outside the estuary (Kocum et al., 2002a).

Taxonomic data divided stations into two main clusters, largely reflecting salinity and seston gradients. The upper estuary assemblage was an association of shallow and eutrophic environments, and waters rich in phosphates and ammonium (Reynolds, 1997); the presence of the cyanophyceae *Pseudoanabaena constricta* is indicative of high nutrient and low oxygen levels (Margalef, 1983). These cyanobacteria are frequent taxa where rivers with high organic matter content flow into the upper Río de la Plata (Luján River; Gómez and Bauer, 1998b). The lower estuary assemblage was a coastal marine association, although some freshwater species occurred along, probably advected from upper estuarine areas.

Time variability remains a key unknown in the Río de la Plata; i.e. seasonal patterns of primary production and phytoplankton biomass. We cannot state whether our sampling (mid-/late spring) took place before or after an eventual spring bloom. In the case blooms occur in the Río de la Plata during early spring as in other mid-latitude ecosystems (Mann and Lazier, 1996), then we probably observed post-bloom conditions. In addition, enhanced stratification could be expected from late summer through autumn due to increased freshwater run-off (Nagy et al., 1996, see Study area above); given the critical importance of the light environment (i.e.  $Z_{\text{mix}}:Z_{\text{pho}}$  ratio), we could also expect high biomass levels during end of summer/autumn driven by still favourable light conditions and the further stabilizing effect of solar radiation, as shown for other estuaries (Pennock, 1985).

#### 4.3. Implications for future work

This work presents a preliminary exploration of mechanisms regulating phytoplankton distribution in a poorly known ecosystem. The main suggestion emerging is that biomass increased at the lower estuary as a response to the improvement in the light environment, presumably mediated by a reduction in SPM and the onset of stratification. Temporal replicability seems in order to explore seasonal and inter annual patterns; also the investigation of other potentially relevant processes. For instance, grazing by microplankton (Gallegos et al., 1996; Jochem, 2003), mesoplankton (Dam and Peterson, 1993), and benthic filter-feeders (Alpine and Cloern, 1992) can also modulate phytoplankton distribution, and should be considered in next studies.

If light limitation is established, an important basis would be set to refine our understanding by recurring to theoretical frameworks derived from studies at comparable turbid estuaries. Under such scenario, models analogous to Cole and Cloern's (1987) relating production to light availability and biomass could be developed to obtain synoptic estimates of primary production over the whole estuary.

Light limitation also has implications for eutrophication processes and water quality management. We can no longer extrapolate to coastal waters some insights obtained from studies of lakes and rivers (see Cloern, 2001), where a clear relation between nutrient loading and biological response (increase in Chl-*a*) has been established. Current understanding of eutrophication processes identifies differences on how estuarine and coastal waters respond to nutrient enrichment, depending on the characteristics of the ecosystem (Cloern, 2001). For example, in turbid estuaries with excess nutrients and light limited phytoplankton, further increases in the amount of nutrients will not lead to increased phytoplankton biomass (Cloern, 1999); consequently, Chl-*a* levels would not necessarily reflect eutrophication, and complementary indicators should be looked upon instead, like community structure and dissolved oxygen levels (Kiddon et al., 2003).

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