Elemental Stoichiometry in Nutrient Pools in Oligotrophic Marine Ecosystems

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Abstract

The stoichiometric ratios are powerful tools to model basic biogeochemical patterns of the sea when the fluxes of a single element are known. It is, therefore essential to understand the full implications of variable stoichiometries to predict the effect of the living components of the ocean on biogeochemical processes. Here, the stoichiometry between C, N, P and Si in different chemical pools (particulate and dissolved organic and dissolved inorganic matter) were examined in contrasting oceanic and littoral ecosystems, and the changes in nutrient partitioning in response to nutrient inputs was tested through experimental research.

In the Central Atlantic Ocean, the average ratio between dissolved inorganic nitrogen and phosphorus in the estimated vertical diffusive fluxes was similar to the Redfield ratio, but tended to be above the Redfield ratio at the center of the South subtropical gyre. The N:P supply ratio and the N:P ratio of dissolved inorganic nutrients in the biogenic layer were strongly correlated, but were not positively correlated to that in the intermediate waters. The vertical nutrient conveyor belt of nutrients in the upper waters operates relatively independently of the underlying waters in the Central Atlantic, so that both the biogenic and the biolythic components should be able to adapt to local variation about the Redfield ratio.

In the stratified NW Mediterranean Sea, the stoichiometry between dissolved inorganic, organic and particulate organic matter pools indicated an excess nitrogen relative to phosphorus, particularly in the biolythic layer, as well as a general silicate deficiency relative to both N and P. Most (> 80 %) of the organic carbon was present as dissolved organic carbon, with POC representing a minor percent of total organic C throughout the water column. The increasing C/N ratio of DOM with depth indicates that N is recycled faster than C in the DOM. There exists a dynamic equilibrium between the biological transformations between these pools with depth, with a dominance of DON production in surface waters and remineralization in the underlying layers, from which dissolved inorganic nitrogen is re-supplied to the biogenic layer.

Alocthonous N inputs must be important in the region since the downward DON flux exceeded the diffusive DIN supply.

The coupling between anabolic and catabolic compartments of a littoral area in the NW Mediterranean Sea are characterized. The pelagic compartment was heterotrophic, supported by significant allochthonous inputs of land material, whereas the benthic compartment was autotrophic, with the excess net benthic community production balancing the deficit in pelagic community production, leading to a system in metabolic equilibrium. Sedimentary inputs of phosphorus and silicon were compensated by sediment release of phosphate and silicate, whereas nitrogen was lost or accumulated in the sediment compartment. Carbon inputs to the benthic compartment also exceeded requirements, due to the allocthonous subsidies to the system, so that the benthic compartment stored or exported organic carbon. Experimental nutrient additions lead to a parabolic change in C/N and C/P ratios in the dissolved organic matter with increasing nutrient inputs. The relative size of the dissolved inorganic nutrient pools did not vary, but there was a tendency towards an increase in the relative size of the particulate pool at the expense of a decrease in the relative importance of DOM as a reservoir of N, P and C, with increasing nutrient inputs.

The relative importance of different nitrogen pools was examined in relation to the total nutrient stoichiometry of the oligotrophic system. The ratio of dissolved inorganic nutrients reported in the research presented is indicative of a general deficiency in nitrogen compared to the global ratios reported in literature. The dissolved organic matter was highly depleted in P relative to N and C at all locations investigated and the resulting POC:PON ratio (11.7) of this study in the particulate pool deviates greatly from the literature values which approximates Redfield value (5.5-6). The shift of the dominance of DON towards PON at TOC/TN values higher than 20 on the oligothrophic areas of the study, gives evidence of increasing carbon export fluxes in a system dominated by particulate pool and points to the effect of the biota on the gradient-driven export to sinking carbon fluxes in the ocean.

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1.1. General Introduction.

Whenever we read of the control that living systems, including humans, exert on the composition of the oceans and atmosphere and on the rate of weathering of the Earth's crust, we build upon our recognition that biogeochemistry studies are the key for understanding the interactive physical, chemical and biological processes that regulate the global changes occurring in the Earth system.

The ocean, in particular, plays a fundamental role in the global biogeochemical cycles through the cycling of non-conservative elements (C, N, P and Si) in seawater, often limiting the rate of net primary production (NPP) on land and in the sea. Commonly present in the ocean as $NO3^-$, $NO2^-$ and $NH4^+$ species, nitrogen is a very reactive element undergoing a number of transformations, such as those derived from the nitrification, denitrification, nitrogen fixation and ammonification reactions in which organic matter is reduced or oxidized. All these biochemical transformations are possible since nitrogen is found at valence states ranging from –3 to +5 and the energy released by these changes in redox potential is used by various microbes to maintain their life processes (Rosswall, 1982). Phosphorus, mostly present in the ocean in combination with oxygen (i.e., as PO_4^{3-}), is an essential component of DNA, ATP, and the phospholipid molecules of cell membranes. Silicon is an important component of the marine biogenic matter that accumulates in coastal and abyssal sediments. Dissolved Si in seawater occurs mostly as the undissociated monomeric silicic acid [Si (OH) $_4$]. The balance between geological and biological cycles of Si determines the silicic acid content. The surface reservoir receives silicic acid inputs from the lithosphere via chemical weathering of the continental crust and through eolian transport. Marine organisms (diatoms, silicoflagellates, and radiolarians build up their skeletons by taking up silicic acid from seawater. After these organisms die the biogenic silica accumulated in their skeletons redissolves or settles downward to reach the sediments. The transformation of opal (amorphous biogenic silica) deposits in

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sediments trough diagenetic processes is a way for silica to re-enter the geological cycle.

Marine phosphorus and nitrogen cycles are inextricably linked to the carbon cycle in the ocean. NPP fixes CO_2 in a biochemical process called photosynthesis that acts to transfer carbon from its oxidized form in the atmosphere to reduced forms in the ocean. As much as half of the global NPP occurs in the surface mixed layer of the ocean (Whittaker and Likens, 1973) with the highest values in the coastal regions and in regions of upwelling. However, the open ocean accounts for about 80% of the total marine NPP as a result of its large area, with continental shelf areas accounting for the remainder (Martin et al., 1987). NPP is degraded to inorganic compounds in surface waters, and the remaining organic matter produced sinks below the euphotic zone to be ultimately remineralised in deep ocean (Eppley and Peterson, 1979). Oxidation of organic matter in the coastal zone is particularly important due to the increase on river loading of organic material which has altered the estimates of long-term oxidizing in the global ocean (Smith et al., 1993).

Changes in the availability of nitrogen or phosphorus through geologic time have controlled the size and activity of the biosphere. In 1934, Albert Redfield published a paper that has served as a focal point in marine biogeochemistry. Redfield postulated that organic mechanisms (biota) controlled the movement of nitrogen and phosphorus in the ocean according to a constant atomic stoichiometry of 106C: 16N: 1P (Redfield, 1958). He also noted that the biotic demand for nitrogen and phosphorus was closely matched to the availability of these elements in upwelling waters (Redfield, 1963). Broecker (1974) recalculated Redfield's ratio considering the downward organic carbon pool. His modified Redfield ratio in sinking particles was 120C: 16N: 1P. This fundamental relationship among the major bioelements has provided a theoretical framework in diverse studies of organic matter production, diagenesis and nutrient regeneration rates in marine environments.

Although the Redfield stoichiometry does not apply universally, as for instance the draw-down of inorganic nutrients during bloom conditions may deviate from

Redfield stoichiometry (Sakshang et al., 1977, Laws et al., 1983), the N: P ratio has been shown to be conservative and predictable in most oxygenated marine environments (Holm-Hansen et al., 1966; Copin-Montegut and Copin-Montegut, 1983; Smith, 1984; Martin, 1987; Fanning, 1989). For example, changes in nutrient levels along neutral density surfaces in the deep ocean retain ratios near those described by Redfield (Anderson and Sarmiento, 1994). Takashi et al., (1985), have confirmed the Redfield N: P stoichiometry (i.e, mean N: P = 16.3 SD = 1.1) for the Indian and Atlantic oceans using data collected during GEOSECS and TTO expeditions. For Pacific Ocean waters, the N: P ratio was determined to be 17.5 ± 2.45 (Stefansson and Richards, 1963) and 17.5 ± 4 (Chow and Mantyla, 1965).

The uptake of atmospheric CO_2 by the oceanic "biological pump" is driven by the export of carbon from the euphotic zone to deep waters (Sarmiento et al., 1992). However, much of the excess organic carbon produced by plankton back to the atmosphere before it reaches the thermocline (del Giorgio and Duarte, 2002). Examination of the consumption of carbon relative to the limiting nutrients from the surface of coastal and open-ocean showed considerably deviations from the Redfield ratio (Broecker et.al, 1985), and greatly exceeded that predicted from the Redfield C: N ratio (Sambrotto et al., 1993). Deviant C: N stoichiometry has also been reported in many areas of low oxygen concentration over the world ocean, due to the low nitrate: phosphate ratios associated with the anaerobic decomposition by denitrifying bacteria (Kamykowski and Zentara, 1990; Anderson and Sarmiento, 1994; Tyrrell and Law, 1997). Growing awareness of the importance of biological processes in determining elemental stocihiometry in the ocean has increased the attention to biological processes as significant controls in the regulation of the exchange of carbon between oceanic and atmospheric pools.

The transformations exerted by biological processes differ vertically in the ocean. Reduction, through NPP, dominates in the photic layer. The depth at which the photosynthetically active irradiance (400- 700m) is 1% of the surface value (Wilkenson and Dugdale, 1992) defines the base of this layer. The balance between biological (and

photochemical) reduction and oxidation processes together with the resuply of inorganic nutrients provided by physical factors driven the upward nutrient transport from the deep ocean occurring by diffusion, eddy diffusion and upwelling, determine the partitioning of nutrients among different inorganic and organic (dissolved and particulate) pools (Sverdrup, 1938; Redfield, 1963).

Nutrient gradients and vertical diffusion coefficients can be combined to estimate the nutrient transport across the top layers of the water column (Lewis et al., 1986; Chavez and Barber, 1987; Crawford and Dewey, 1989) and consequently the primary production fueled by newly introduced nitrate into the mixed layer (new production), (Dugdale and Goering, 1967; Eppley and Peterson, 1979). If the mixed layer were deeper than the euphotic layer, nitrate supply would be determined by the flux through the base of the mixed layer or throughout the thermocline. Bellow the thermocline, nutrient recycling has an important role in re-supplying approximately 90% of the nutrient requirement in the surface waters each year (Wollast, 1981). Patterns of vertical particle fluxes and distributions of elemental concentrations suggest that most biomatter is consumed and transformed back into inorganic forms within 300-400 m of the surface (biolythic layer). The phytoplankton production based on nutrients recycled within the photic layer is called "regenerated production" (Eppley et al., 1983). Since the biological demand and remineralization of nutrients in the upper ocean determine their vertical distribution, the diffusional fluxes of nutrients to the mixed layer are then governed by nutrient gradients that depend on the uptake rates and physical redistribution of water masses, characteristics of each different areas of the ocean.

Whereas the rate of transport of deep rich water in nitrate in the oligothrophic eastern Atlantic was no different from the integrated rate of nitrate uptake (Wilkenson and Dugdale, 1992; Dugdale et al., 1992), discrepancy was found between new production estimated from physical supply and that estimated from nitrate incorporation into particles in the equatorial Pacific Ocean (Roemmich, 1989; Fiedler et al., 1991; Eppley and Renger, 1992). The elevated nutrient influx into the euphotic layer lead to a

higher consumption relative to that expected from the particulate organic matter (POM) production measurements (Carr et al., 1995). Indeed, particulate organic production is comparable to sediment trap results when half of the new production due to upwelled nutrients goes into a pool of dissolved organic compounds (Toggweiler, 1989).

POM is exported across the thermocline into Deep Ocean. While the distribution of total dissolved and particulate nutrient pool is constant with depth, nutrient ratios in sinking particles increase higher in biolythic layers than in biogenic area (Martin et al., 1987; Karl et al., 1996). The study of total nutrient partitioning of the dissolved upward transport and that of the organic downwell flux provides insight into the role of nutrients as determining the variability of the nutrient stoichiometry within water column.

Whereas the study of elemental ratios in the ocean has traditionally emphasized that of inorganic nutrients, organic – particularly dissolved – nutrient pools may comprise much of the elemental content of the photic layer of the ocean (Jackson and Williams, 1988; Druffel et al., 1992; Carlson and Ducklow, 1995; Williams, 1995). There is, indeed, growing evidence that the differences in elemental ratios between the upward flux of inorganic nutrients from the biolythic zone to the upper biogenic zone are driven by a differential partitioning of these nutrients between organic and inorganic pools. Indeed, nutrients are not exchanged between a living and an inorganic pool, but are also trapped and recycled from a non-living organic pool of dissolved organic matter (DOM).

There has been considerable progress over the past 15 years in estimating the concentrations of elements within DOM dissolved organic (Sugimura et al., 1988, Suzuki et al., 1992, Hedges and Farrington, 1993). The nature of DOM is, however, still poorly understood, in part, because it has been difficult to isolate sufficient amounts of fulvic and humic acid compounds (Benner et al., 1992). Polysaccharides appear to be more abundant and reactive components of DOM when supporting heterotrophic activity (Pomeroy, 1974; Azam et al., 1983) in the euphotic layer. Higher concentrations of DOM in the photic layer have been useful in explaining the vertical

distributions of nutrients in models where expected particulate fluxes and regeneration have been inadequate (Toggweiler, 1989). Jackson and Williams (1995), stress the importance of DOM in controlling the elemental ratios in the upper layers of the water column, since total nutrient ratios estimated as the sum of dissolved organic and inorganic pools, approximated those described by Redfield. Because the sum of dissolved inorganic pool (DIM) and DOM is almost constant, the vertical fluxes distribution in the ocean are not controlled by settling particulate matter alone, but also by oxidation of the DOM carried in a parcel of water (Christian et al., 1997). On the other hand, the substantial concentrations of DOM found in the photic layer indicates that the new nitrogen introduced depends on the total dissolved nitrogen (TDN) nutrient gradient rather than just the DIM gradient. Indeed, consideration of DOM in transport models, where the diffusive flux of a substance being mixed is proportional to its gradient, have altered significantly our perception of the nature of these diffusive gradients, both in magnitude and direction. Available evidence indicates that the role of DOM is highest in oligotrophic environments, where it represents the dominant elemental pool, which contains a labile fraction flux of DON and dissolved organic phosphorus (DOP) being recycled by pelagic systems to maintain their biochemical processes in systems provided of low nutrient supply (Zweifel et al., 1993; Thingstad et al., 1995). Hence, the examination of elemental fluxes in the ocean requires, therefore, not only consideration of their stoichiometric relationship, but also their partitioning among the different pools of POM, DOM and DIM; as these differ in elemental stoichiometry and turnover.

The importance of the DOM pool as a reservoir of C, N and P in oligotrophic environments can be tested through the examination of the changes in nutrient concentrations in enclosure ecosystems exposed to controlled nutrient additions. Large-scale experiments help test hypothesis as to how the partitioning of elements between nutrient pools and their stoichiometric balance change across a gradient from oligotrophic to eutrophic conditions. The examination of the partition of nutrient elements among these pools is still scarce and poorly considered in marine

biogeochemistry studies. Deviations from Redfield stocihiometry are, therefore, likely to be largest in oligotrophic environments, both coastal and oceanic. Furthermore, a better knowledge of dissolved organic nutrient partitioning within the biogenic and biolythic layers is required to make reliable inferences on the recycling rate of nitrogen and phosphorus and also to determine the dynamic balance established among the downward fluxes of dissolved organic matter and the upward transport of dissolved inorganic matter (Cauvet et al., 1997).

The coherent study of organic, inorganic and particulate nutrient pools is particularly important in coastal areas, where the nutrient stoichiometry may depend on the seasonal variability of antropogenic discharges. The huge increment on reactive phosphorus and total inorganic nitrogen concentrations in river waters during the 20th Century has led to the development of new budgetary procedures to define the mechanisms that regulate nutrient partitioning within the coastal ocean (Smith and Mackenzie, 1987; Buscail et al., 1990; Monaco et al., 1990).

The elucidation of patterns in the stoichiometry of C, N, P in the ocean is a major focus of interdisciplinary research in oceanography, driving large-scale international studies (e.g. JGOFS; WOCE). Yet, this research has revealed that the consideration of a single, universal Redfield ratio fails to predict the behavior of the ocean under changing environmental conditions (Michaels, 2001). Yet, stoichiometric ratios are powerful tools to model the basic biogeochemical patterns of the sea (river flow, upward transport, and internal recycling related to the net primary production) when the fluxes of a single element are known. It is, therefore essential to understand the full implications of variable stoichiometries to predict the effect of the living components of the ocean on biogeochemical processes.

1.2. Goals of the thesis.

The general goal of this thesis is to test the universality of the Redfield's stoichiometric ratio and to search for consistent patterns in the deviations from the average ratio in oligotrophic environments. This is mainly achieved through the examination of the stoichiometry between C, N, P and Si in different chemical pools (particulate and dissolved organic and dissolved inorganic matter) in contrasting oceanic and littoral ecosystems, as well as the response of these ratios to experimental nutrient additions. The stoichiometry of nutrient elements in the open ocean is examined in the Mediterranean Sea and the subtropical Atlantic Ocean. Nutrient ratios are then examined in the northeast Mediterranean littoral area, subject to antrophogenic nutrient inputs. The predictive capacity of the patterns derived from these studies is then tested by examining how nutrient partitioning changes in response to experimental nutrient inputs in a mesocosm experiment. These objectives result in the four specific goals of this thesis:

I. Dissolved inorganic nutrients and diffusional supply to the biogenic layer of the Central Atlantic (Chapter 2).

The aim of the study is to test the hypothesis that contrasting biochemical processes occurring within the upper 1000 meters of the water column may induce variability in the onset of a global nutrient stoichiometry in the ocean.

The test is based on the description of meridional patterns in the upward diffusive flow of nitrogen and phosphorus into the biogenic layer of the Central Atlantic Ocean. The variability in the stoichiometry of dissolved inorganic nitrogen (DIN) and phosphate (DIP) is examined and dissolved inorganic nutrient ratios in the diffusive flux are compared to those found in the biogenic layer. This examination helps assess the role of nitrogen versus phosphorus as nutrient limiting new production, provided the

differences in the time scales of coupling of vertical nutrient cycling in the Central Atlantic Ocean.

II. Nutrient (N, P and Si) and carbon partitioning in the stratified NW Mediterranean (Chapter 3).

The second specific goal is to test the hypothesis that there are stoichiometric predictable relations in the stoichiometry of the dissolved organic pool, which are balanced with the particulate and dissolved inorganic pools.

This hypothesis is tested by examining the distribution of C, N, P and Si in their particulate, dissolved organic and inorganic forms in the stratified NW Mediterranean Sea. I first describe the nutrient partitioning within the stratified Mediterranean Sea, and then use a budgetary approach to describe the nutrient exchange (export-inputs) between dissolved organic and inorganic pools within the system.

III. Nutrient dynamics and ecosystem metabolism in the Bay of Blanes (NW Mediterranean) (Chapter 4).

The third goal is the characterization of the coupling between the pelagic and benthic components of a littoral area in the NW Mediterranean sea; an anabolic compartment which produces organic matter versus a catabolic one as oxidizing agent of the organic matter pool. The simultaneous quantification of annual sedimentary fluxes of C, N, P and Si nutrients and the sediment-water fluxes of dissolved organic and inorganic chemical species as well as the mineralization rates derived from a budgetary exercise, provides insight into the role of sediments as a source or as a sink of the organic matter. The impact of antrophogenic perturbations, derived from anomalous summer increases in nutrient inputs due to tourism, is also evaluated as a source of variability in the nutrient stoichiometric ratios.

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IV. The effect of nutrient additions on the partitioning of nutrients in an experimental coastal Mediterranean system (Chapter 5).

The main goal of this chapter is to test the hypothesis that the importance of dissolved organic matter (DOM) as a reservoir of C, N and P declines, relative to that of the particulate pool, with increasing nutrient inputs, thereby increasing the departure of nutrient rations from Redfield stoichiometry. This is achieved by means of a mesocosm experiment carried out in the NW Mediterranean littoral. The changes in the partitioning of nutrients among different pools (inorganic and dissolved and particulate organic) are analyzed under controlled nutrient additions. This exercise contributes to test the role of DOM as a major nutrient reservoir in oligotrophic systems.

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Dissolved inorganic nutrients and vertical diffusional supply to the biogenic layer of the Central Atlantic

2.1. Abstract.

The vertical distribution of nutrients and the eddy turbulent diffusivity were examined to describe latitudinal patterns in the upward diffusive flow of nitrogen and phosphorus into the biogenic layer along the Central Atlantic Ocean (27 ° 13' N to 36 ° 21' S). Water flow and vertical turbulent diffusivity were low ($< 10^{-5}$ m^{-2} s^{-1}) at the southern subtropical gyre, and the fastest flow and vertical turbulent diffusivity was greatest ($< 10^{-2}$ m^{-2} s^{-1}) at the Equator. The depth and strength of the nutricline were strongly correlated ($r^2 > 0.80$) to the depth and the strength of the thermocline, respectively. The calculated upward diffusive flux of nitrate plus nitrite and phosphate into the biogenic layer was highest at the Equator (6 and 105 mmol phosphate and nitrate m^2 d^{-1}), and remained high (> 0.1 and > 1 mmol phosphate and nitrate m^2 d^{-1}) throughout the extent of the dome present between 5°S and 20°N, and was very low (< 0.1 and < 1 µmol phosphate and nitrate m^2 d^{-1}, respectively) in gyre areas. The average ratio between dissolved inorganic nitrogen and phosphorus in the estimated vertical diffusive fluxes (14.4 ± 1.7) was similar to the Redfield ratio, but tended to be well below the Redfield ratio at the southern end of the transect and above the Redfield ratio at the center of the South subtropical gyre (10° S - 15°S). The N:P supply ratio and the N:P ratio of dissolved inorganic nutrients in the biogenic layer were strongly correlated ($r^2 = 0.87$, $P < 0.001$), but were not positively correlated to that in the intermediate waters. The results indicate that the vertical nutrient conveyor belt is able to operate at nitrate to phosphate ratios well away from the Redfield ratio, so that both the biogenic and the biolithic components should be able to adapt to local variation about the Redfield ratio.

2.2. Introduction.

Biological consumption and subsequent export and remineralization of nutrients in the upper ocean determine their vertical distribution (Redfield et al., 1963; Anderson and Sarmiento, 1994). These processes involve the biogenic layer, which is characterized by a high demand of dissolved inorganic nutrients to support planktonic production. This demand generates a vertical concentration gradient driving an upward flux of dissolved inorganic nutrients regenerated in deeper waters.

The resulting fluxes lead to a conveyor belt dynamics in which nitrogen and phosphorus are transported upwards in inorganic form and downwell in organic form, both fluxes being in approximate equilibria in a steady-state ocean (e.g. Eppley and Peterson, 1979; Jenkins, 1988). Because of the differential importance of nitrogen and phosphorus as limiting nutrients in different areas of the ocean, their turnover time within the biogenic layer may also differ, so that the conveyor belt may function at somewhat different time scales for nitrogen and phosphorus. There are, however, limits to these differences imposed by the uptake rates which generates gradients of nutrient elements and govern the nutrient fluxes in the ocean (Redfield et al., 1934). Yet, planktonic communities have differential requirements for nitrogen and phosphorus, so that planktonic production is believed to be mostly limited by nitrogen supply (Dugdale, 1967; Ryther and Dunstan, 1971; Falkowski, 1997), whereas bacterioplankton growth is often P limited (Cotner et al., 1997; Rivkin and Anderson, 1997; Thingstad et al., 1998). It is possible, therefore, that the stoichiometry of dissolved inorganic nitrogen and phosphorus may experience changes between the deep source waters and those in the biogenic layer, generating variability in nitrate: phosphate ratios in surface waters (cf. Fanning , 1992).

Nutrient supply to the biogenic zone of the ocean is driven, away from the limited areas where advective transport is important, by diffusive supply across the thermocline (Dugdale and Goering, 1967; Eppley and Peterson, 1979; Eppley et al., 1979). The nutrient supply due to vertical mixing to the biogenic zone is dependent on

the gradient of dissolved inorganic nutrients across the thermocline, controlled by the balance between nutrient consumption above the thermocline and regeneration within and below the thermocline, and the vertical turbulent diffusion coefficient at the nutricline, largely governed by the turbulence generated by internal waves at the thermocline and shear flow instability (Gregg, 1989). The upward diffusional supply of nutrients to the biogenic zone is particularly important in the tropical ocean, where it is believed to be the main driving force for new production (e.g. Platt and Harrison, 1985; Lewis et al., 1986; Chavez and Barber, 1987; Jenkins, 1988). This believe is supported by a general agreement between estimates of the diffusional flux and nitrate uptake rates in different regions of the ocean (e.g. Lewis et al., 1986; Planas et al., 1999). Our knowledge on the diffusion supply of nutrients to the biogenic zone of the tropical ocean is, however, still patchy, estimates being available only for a few regions of the oceans (e.g. King and Devol, 1979; Eppley et al., 1979; Lewis et al., 1986; Jenkins, 1988; Carr et al., 1995).

Our main goal here is to describe meridional patterns in the diffusive flow of dissolved inorganic nitrogen and phosphorus into the biogenic layer of the Central Atlantic Ocean. We do so based on a description of the vertical distribution of nutrients along a latitudinal gradient across the Central Atlantic Ocean (27 ° 13' N to 36 ° 21' S). We first examine the gradient of dissolved inorganic nutrients across the thermocline, and combine these gradients with estimates of the eddy turbulent diffusivity at the thermocline to calculate upward diffusive fluxes. We then examine the variability in the stoichiometry of dissolved inorganic nitrogen (DIN) and phosphate within the upper 1000 meters in the Central Atlantic Ocean, and compare the DIN:phosphate ratio of the diffusive flux with that in the biogenic layer. This comparison provides insight into the role of nitrogen versus phosphorus supply as bottlenecks for new production and on possible differences in the time scales of the nitrogen and phosphorus vertical conveyor belts in the Central Atlantic Ocean.
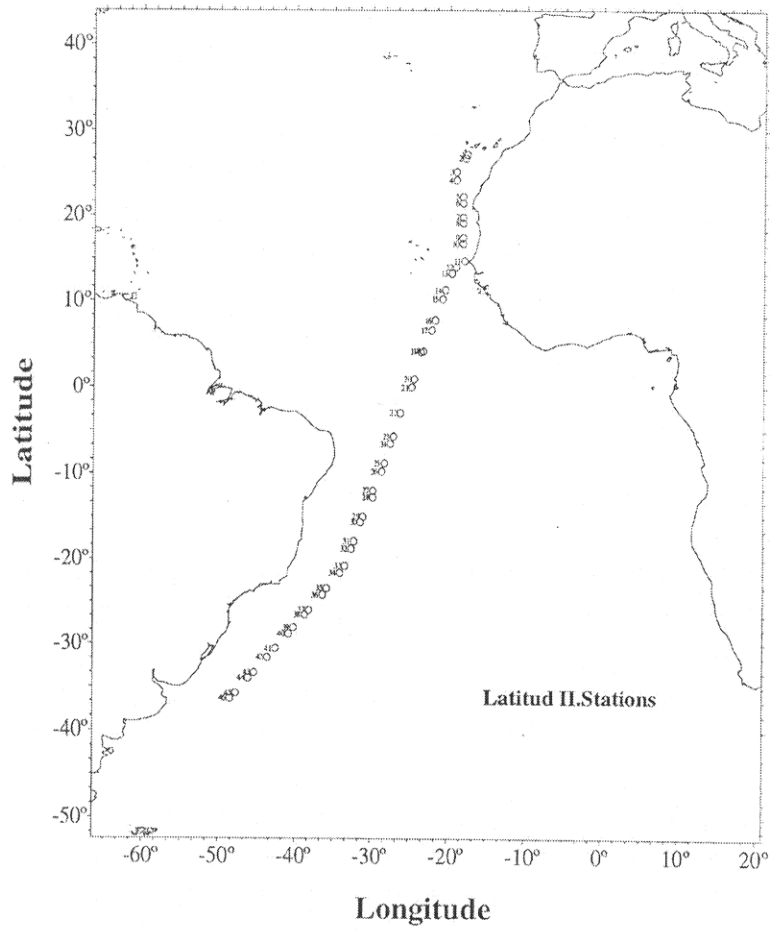
Fig. 1.   Stations sampled during the Latitude II cruise.

2.3. Methods.

The study was conducted in meridional transect across the Central Atlantic between the Canary Islands and Argentina (Fig.1). The cruise was carried on board the Spanish research vessel BIO Hespérides, occupying 46 stations between October 21 and November 16, 1995. At these stations, water samples were collected by means of a rosette sampler system fitted with 24, 12 L Niskin bottles attached to a CTD (Mark III or Mark V, Sea Bird). The CTD casts reached down to 1000 m depth, except for 3 stations where technical difficulties forced a somewhat shallower sampling. Water samples collected were from a total of 17-18 depths at each station with a higher density of samples at the thermocline, where changes in nutrient concentrations are greatest.

Subsamples were drawn from the Niskin bottles into pre-cleaned polyethylene bottles, and were immediately analyzed on board. Dissolved inorganic PO_4^{3-} was measured spectrophotometrically using a 10-cm cuvette cell according to a standard method (Koroleff, 1976; Grasshoff, 1983). $NO_3^- + NO_2^-$ concentrations within the mixed layer were analyzed spectrophotometrically, following the methods of Koroleff (1976) and Grasshoff (1983), using a 10-cm cuvette to increase the detection limit within these nutrient-depleted waters, whereas nitrate plus nitrite concentrations in water samples below the thermocline were determined using a Skalar autoanalyser. The detection limits of dissolved nutrient concentrations were 0.005, and 0.01 µmol L^{-1} for spectrophotometric determinations of $NO_3^- + NO_2^-$, and PO_4^3, respectively, and 0.02 µmol L^{-1} for $NO_3^- + NO_2^-$ measured with the autoanalizer.

Conductivity-temperature-pressure (CTD) data were obtained with a vertical resolution of 10 cm. Salinity and temperature estimates derived from CTD casts were calibrated along the cruise using direct salinity measurements from a Guildline Autosal salinometer calibrated with IAPSO standard seawater (34.993 ppt), and a highly
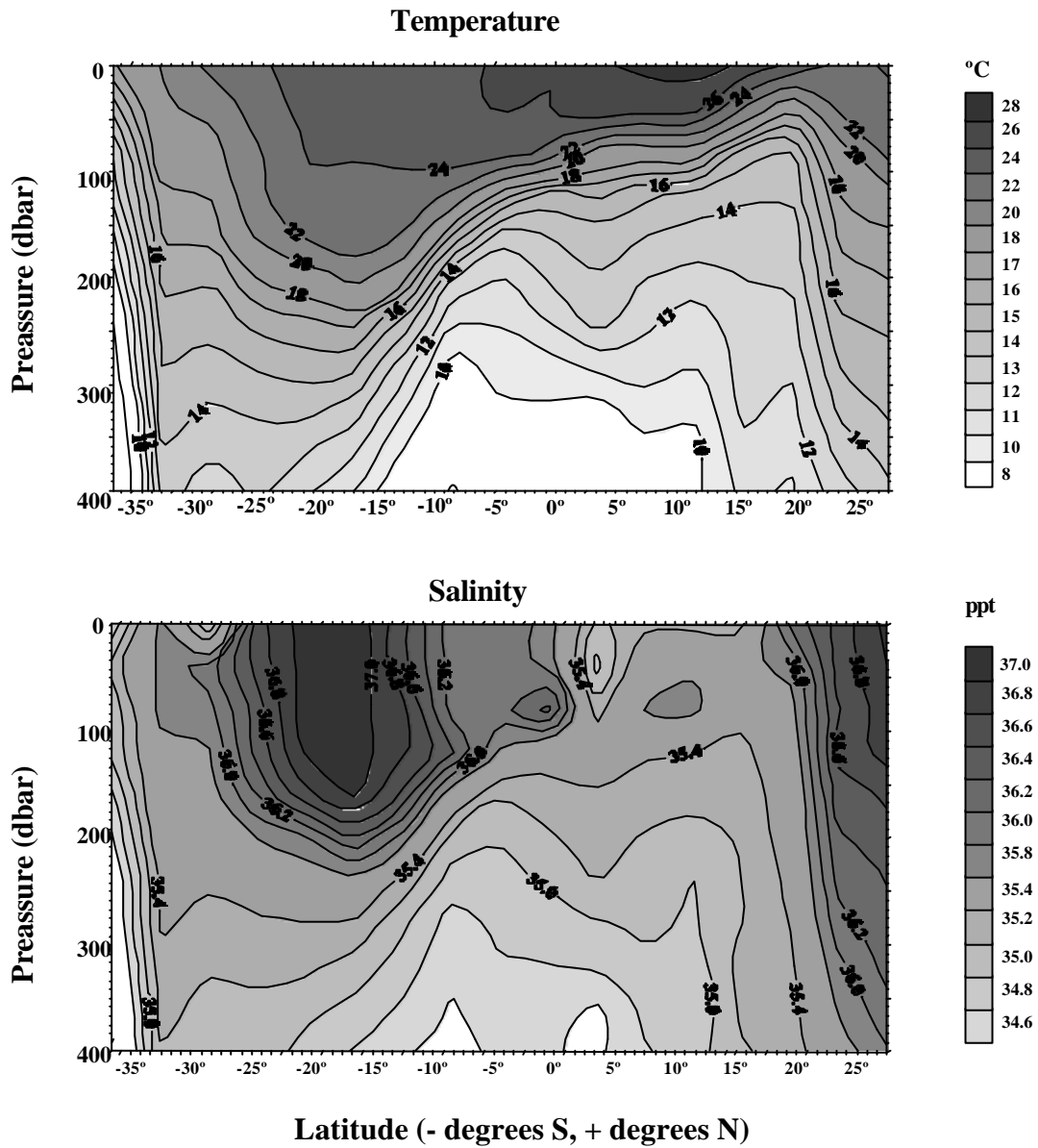
Fig. 2. Contour plots describing the distribution of seawater temperature and salinity along the Central Atlantic.

accurate reversible thermometer, respectively. Vertical profiles of salinity and temperature derived from the (corrected) CTD data were screened for errors. We then calculated the density anomaly, σt, from temperature and salinity following the algorithms of Fofonoff and Millard (1981), and then averaged the data at 1 m intervals.

The vertical gradient in σt ($\frac{\Delta\boldsymbol{r}}{\Delta z}$) was then used to calculate the Brunt-Väisälä buoyancy frequency (N^2), according to the equation:

$$N^2 = \frac{-g}{\boldsymbol{r}}\frac{\Delta\boldsymbol{r}}{\Delta z} \qquad\qquad\text{(Eq.1)}$$

where g is the gravitational acceleration, and ρ is the mean density averaged over the Δz (1 m) depth interval.

The horizontal velocity was measured continuously along the cruise using a VM150 narrowband acoustic Doppler current profiler (RD Instruments) mounted on the mid-hull of the ship. The two horizontal velocity components E-W (u, m s^{-1}) and N-S (v, m s^{-1}) were estimated every 1.2 seconds within 8 m bins from the subsurface (16 m) down to 400 m depth. The calibration of the average velocities was done as recommended by Joyce (1989). The estimates were averaged within 5 min intervals and scanned for errors due to changes in ship position and speed following King and Cooper (1993) and Saunders and King (1995). The estimates of velocity obtained during sampling at the stations were found to contain faulty data, so that only those obtained during navigation at a uniform speed, which were of much higher quality, were used. Hence, the average vertical velocity structure at each station was obtained by averaging the velocity profiles obtained in the 15 minutes preceding and subsequent to the occupation of the stations, while the ship was navigating at cruise speed (about 11-13 knots). The average vertical velocity profiles obtained for each station were used to calculate vertical shear (S) following the formula:

$$S = \sqrt{\left[\left(\frac{\Delta u}{\Delta z} \right)^2 + \left(\frac{\Delta v}{\Delta z} \right)^2 \right]} \qquad \text{(Eq.2)}$$

Buoyancy frequencies estimates were averaged at intervals corresponding to those used in the calculation of vertical shear, and both estimates were combined to derive an estimate of the vertical diffusivity across the thermocline, following the parametrization developed by Granata et al. (1995), after Gregg (1989):

$$K_r(z) = K_0 \left\langle \frac{S^4}{S_{GM}^4} \right\rangle \qquad \text{(Eq.3)}$$

where $K_r(z)$ is a parameterization of turbulent diffusivity at the thermocline, z is depth, K_0 is the background diffusivity, set to 5×10^{-6} m^2 s^{-1} (Gregg and Kunze, 1991), S_{GM} is the Garrett-Munk Shear which was set to $1.66 \times 10^{-10} < (N^2/N_0^2) >$, N_0 was 5.2×10^{-3} s^{-1} (Gregg, 1989) and S was the shear measured over the 8 m bins (broadband).

The thermocline depth was defined as the depth at which maximum stratification occurred (i.e. maximum N^2). This depth was found to correspond closely to the depth of the biogenic layer ($z_{biog.}$, where irradiance = 1 % of that at the surface and net primary production >0) across the transect (Agustí and Duarte, 1999). The depth of the base of the photic layer was closely correlated ($r^2 = 0.83$) with the depth of the thermocline used as the reference for flux calculations (Agustí and Duarte, 1999).

The turbulent flux of dissolved inorganic nutrients across the thermocline (F_N, mmol m^{-2} d^{-1}) was , therefore, calculated (assuming gradient transport) as the product between the coefficient of vertical diffusivity, ($Kr(z)$, m^2 s^{-1}) and the gradient in nutrient concentration across the thermocline, $\frac{\partial [N]}{\partial z}$, mmol m^{-4} (Lewis et al. 1986; Carr et al. 1995):

$$F_N = K_r(z)\frac{\partial[N]}{\partial z} \tag{Eq.4}$$

The turnover rate (T, d^{-1}) of dissolved inorganic nutrients within a steady-state mixed layer layer was calculated as follows:

$$T = \frac{F_N}{M_N} \tag{Eq.5}$$

where F_N is the vertical turbulent flux of dissolved inorganic nutrients in mmol m^{-2} d^{-1}, and M_N (mmol m^{-2}) is the integrated pool of dissolved inorganic nutrients within the upper layer (from the surface to the depth where the Brunt-Väisälä buoyancy frequency was maximal):

$$M_N = \int_{z_0}^{z_{bioh}}[Nutrient]dz \tag{Eq.6}$$

The residence time of dissolved inorganic nutrients within the biogenic layer is, therefore, the inverse of the turnover rate (T). In fact, the estimates of turnover rate and residence time derived here represent minimum and maximum values, respectively, for these calculations do not consider other possible inputs of dissolved inorganic nutrients (e.g. atmospheric deposition and lateral inputs). Upwelling motion occurring at 0º and near the NW African coast (Guinea Dome) has not been either considered as a loss or a source term to the biogenic layer. The estimated nitrate supply to the biogenic layer was subsequently used to derive an estimate of the new production (N_{pp}, mmol C m^{-2} d^{-1}) possible as:

$$N_{pp} = 6.6 \times F_N \tag{Eq.7}$$
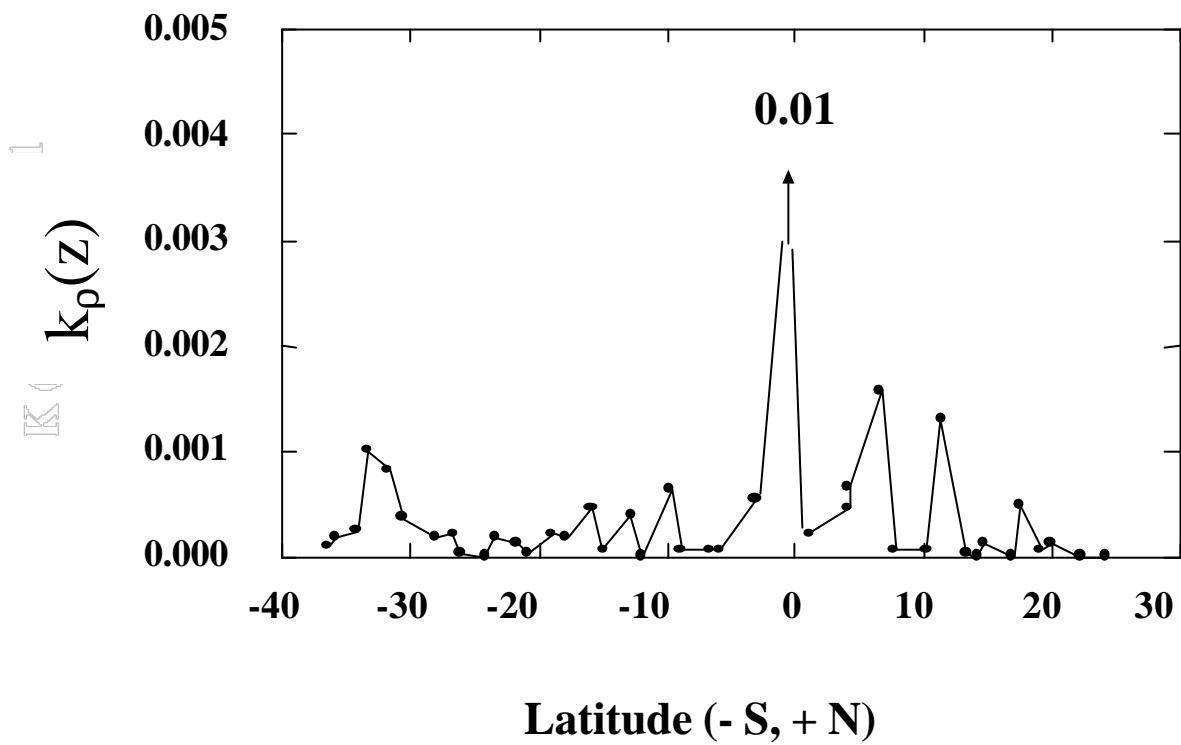
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where 6.6 is the Redfield C:N ratio (Redfield et al., 1963).

Fig. 3. The distribution of the estimated vertical turbulent diffusivity at the thermocline ($K_r(z)$) along the Central Atlantic.
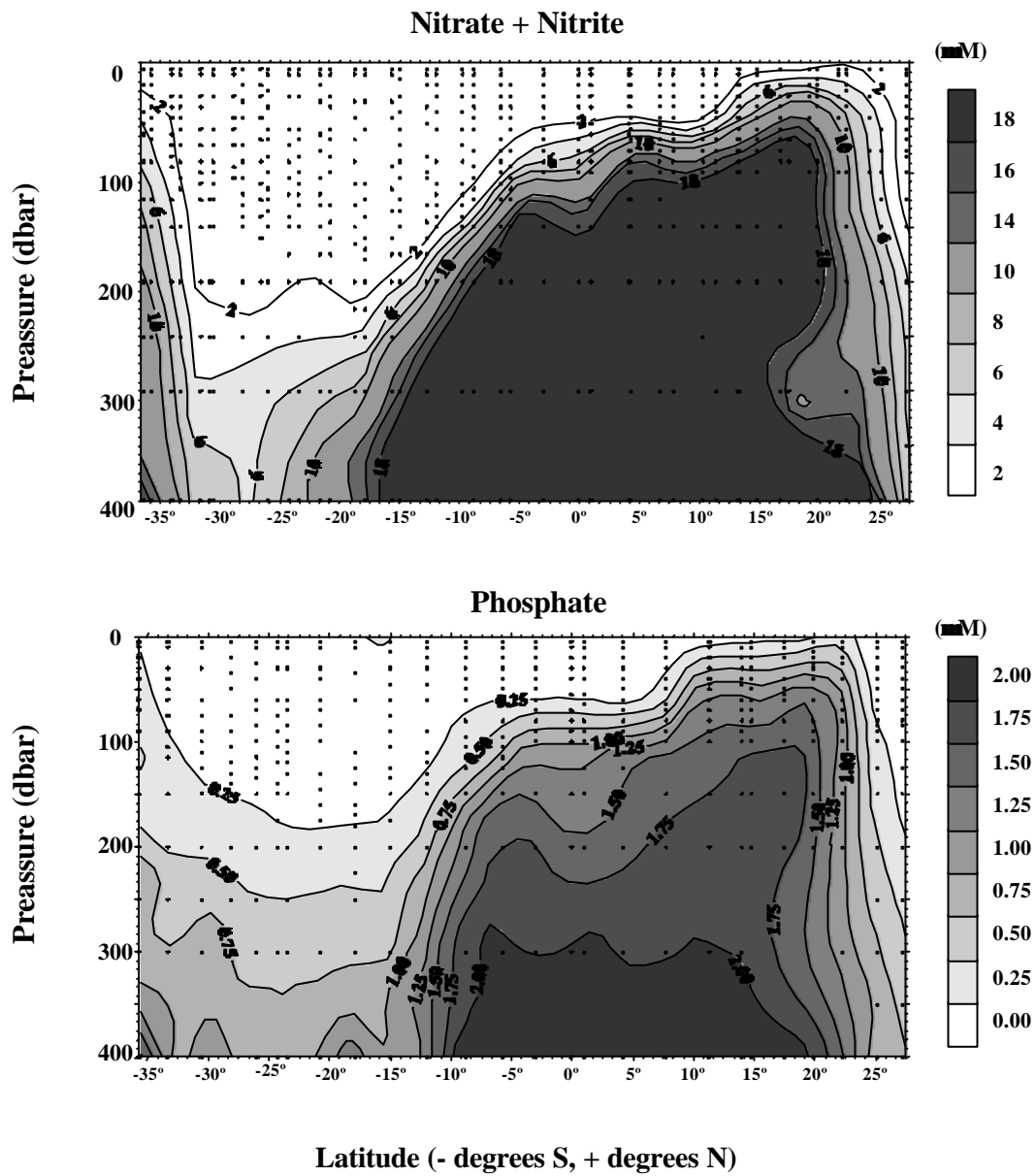
Nitrate + Nitrite

Phosphate

Latitude (- degrees S, + degrees N)

Fig. 4.   Contour plots describing the distribution of nitrate + nitrite and phosphate

along the Central Atlantic.

2. 4. Results.

The distribution of temperature and salinity along the transect reflects the flow patterns in the Central Atlantic (Fig. 2). The deep, cold waters domed between 15°S and 10°S, as a consequence of the equatorial divergence (Voituriez and Dandonneau, 1974; Kawase and Sarmiento, 1985). This was reflected in the presence of a relatively shallow (< 100 m), steep thermocline (Fig. 2). In addition, the presence of the Guinea Dome was reflected in a very shallow (30 m) thermocline off the NW African coast (Fig. 2), which has been described in detail earlier (e.g. Voituriez and Dandonneau, 1974). The relatively high irradiance around 5° to 10° N resulted in the warmest surface waters found there (Fig. 2). In contrast, the waters occupying the gyre zone between 15°S and 25°S were characterized by a deep (down to 200 m), weak thermocline (Fig. 2). The southern limit of the transect was located close to the frontal zone between the Brazil and Malvinas currents, as indicated by the sharp horizontal gradient in the water properties there (Fig. 2). The surface waters at the gyre were also saltier, as a result of high evaporation rates there, and high salinity waters, resulting from the influence of Mediterranean outflow waters, were also located at the northern limit of the transect (Fig. 2). The combined changes in water column stability and shear stress along the transect resulted in major (four orders of magnitude) differences in the calculated vertical turbulent diffusivity coefficient at the thermocline, which was greatest at the Equator (Fig. 3).

The concentration of dissolved inorganic nutrients was closely associated to the distribution of water properties, with the isolines of nutrient concentrations closely following the isotherms (e.g. the 2.0 µmol NO_3^- and the 0.5 µmol PO_4^{3-} isolines roughly followed the 20°C isotherm, Fig. 4) and showing, therefore, a doming structure extending from 10°S to 20°N. The thickness of the nutrient-depleted upper layer changed greatly along the transect, from thin (30 m) layers at the Guinea Dome, to the very thick (200 m) nutrient-depleted layers in the South tropical gyre (Fig. 4).
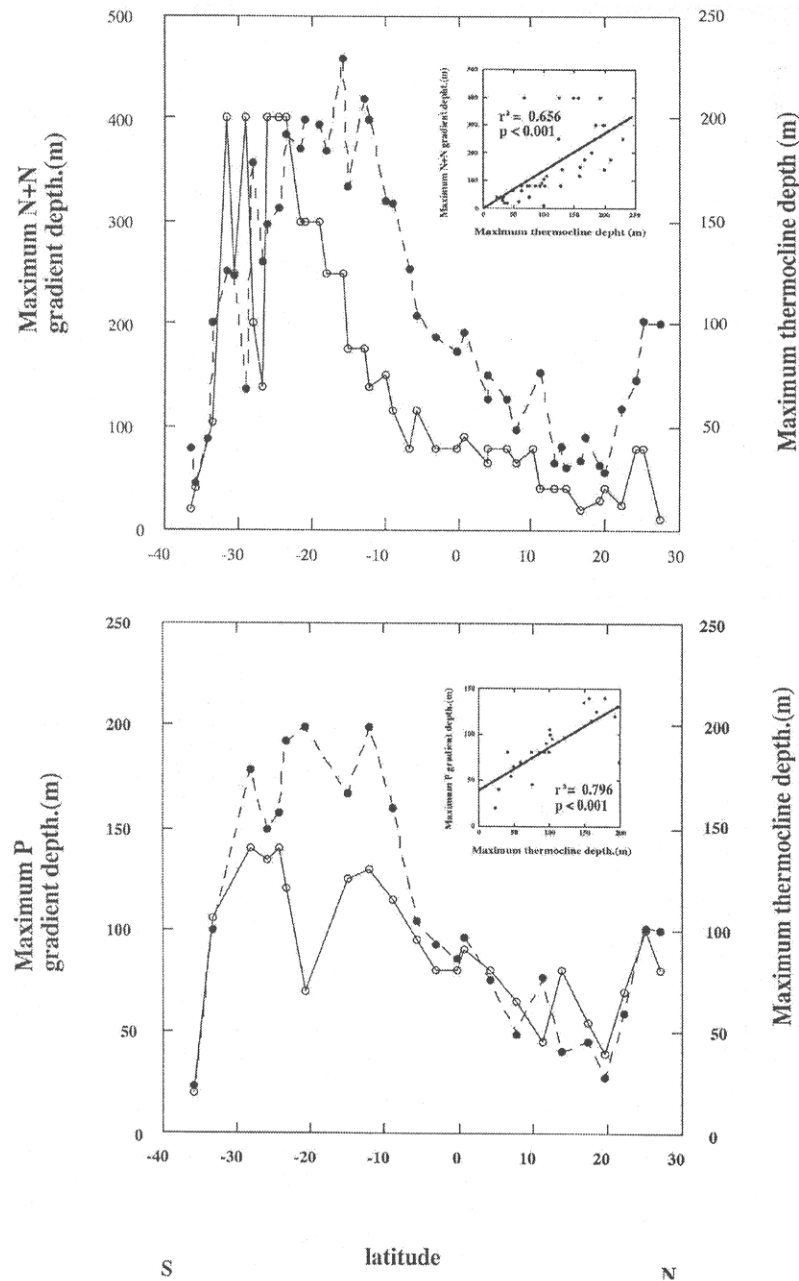
Fig. 5. The distributions of the depth of maximum vertical stability within the thermocline (broken line) and that where the vertical gradient in nutrient (nitrate plus nitrite, and phosphate, solid line) concentrations was greatest along the Central Atlantic. The insert shows the relationship between these two depths.
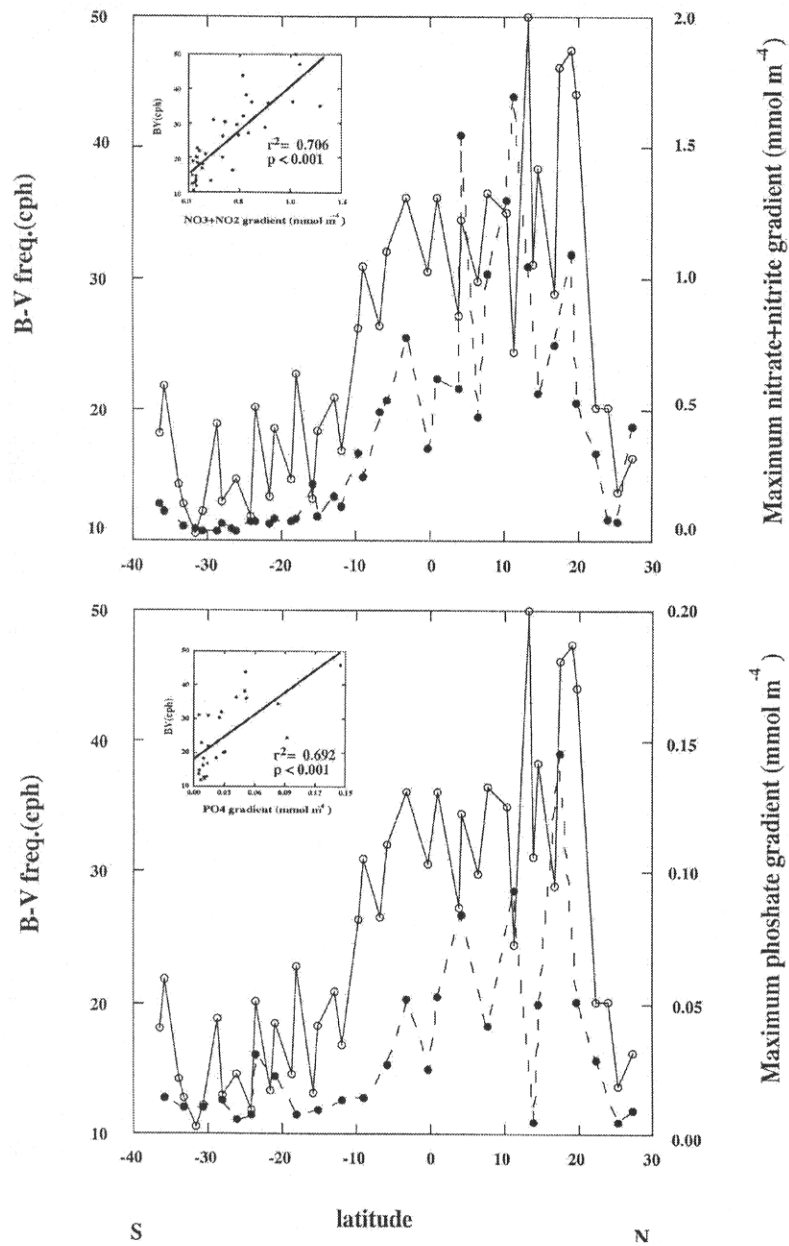
Fig. 6. The distributions of the maximum vertical stability within the thermocline (as the Brunt-Väisälä bouyancy frequency, in cycles per hour, solid line) and the gradient in nutrient (nitrate plus nitrite, and phosphate) concentration across the thermocline in the Central Atlantic (broken line). The insert shows the relationship between these two depths.

These changes reflected significant correlations between the depth where the nitrate and phosphate gradients were maximal and the depth of the thermocline (r^2 = 0.656 and 0.796, p < 0.01 respectively; Fig. 5). The nitrate (plus nitrite) and phosphate gradients across the thermocline ranged by three and two orders of magnitude, respectively, being greatest off the NW African coast and near the Equator (Table 1), where the vertical stability associated to the thermocline was also strongest. The strength of the nutrient gradients was, therefore, closely correlated to the strength of the thermocline, as indicated by a close correlation between the maximum Brunt-Väisäla frequency and the nitrate and phosphate gradients (r^2 = 0.706 and 0.692, p < 0.01, respectively; Fig. 6). The calculated upward diffusive flux of nitrate plus nitrite and phosphate into the biogenic layer was highest at the Equator, as a result of the shear stress introduced by the well defined Equatorial Undercurrent, and remained high throughout the extent of the dome (between 5°S and 20°N, Table 1).

The average ratio between dissolved inorganic nitrogen and phosphorus in the estimated vertical diffusive fluxes (14.4 ± 1.7) was similar to the Redfield ratio, but tended to be well below the Redfield ratio at the Southern end of the transect (Fig. 7), where the supply of phosphate to the biogenic layer was disproportionately high relative to that of nitrogen compared to the Redfield ratio. In contrast, the N:P ratio in the vertical diffusive flux of dissolved inorganic nutrients was well above the Redfield ratio at the center of the South subtropical gyre (10° S - 15°S, Fig. 7), indicating a severe deficiency in phosphorus in the upward nutrient supply to the biogenic zone there. The N:P supply ratio and the N:P ratio of dissolved inorganic nutrients in the biogenic layer were strongly correlated (r^2 = 0.87, P < 0.001; Fig. 8) indicating that 87 % of the variance in the dissolved inorganic N:P ratio within the biogenic layer was attributable to the differences in the ratios of the diffusive flux. As a consequence, the pool of dissolved inorganic nutrients within the biogenic layer was deficient in nitrogen in the southern end of the transect (26° to 36°S) and deficient in phosphorus in the center of the gyre (10°S to 15°S, Fig. 7). There was, however, a significant (Wilcoxon ranked sign test, P = 0.0032) tendency for the ratio of dissolved inorganicnitrogen
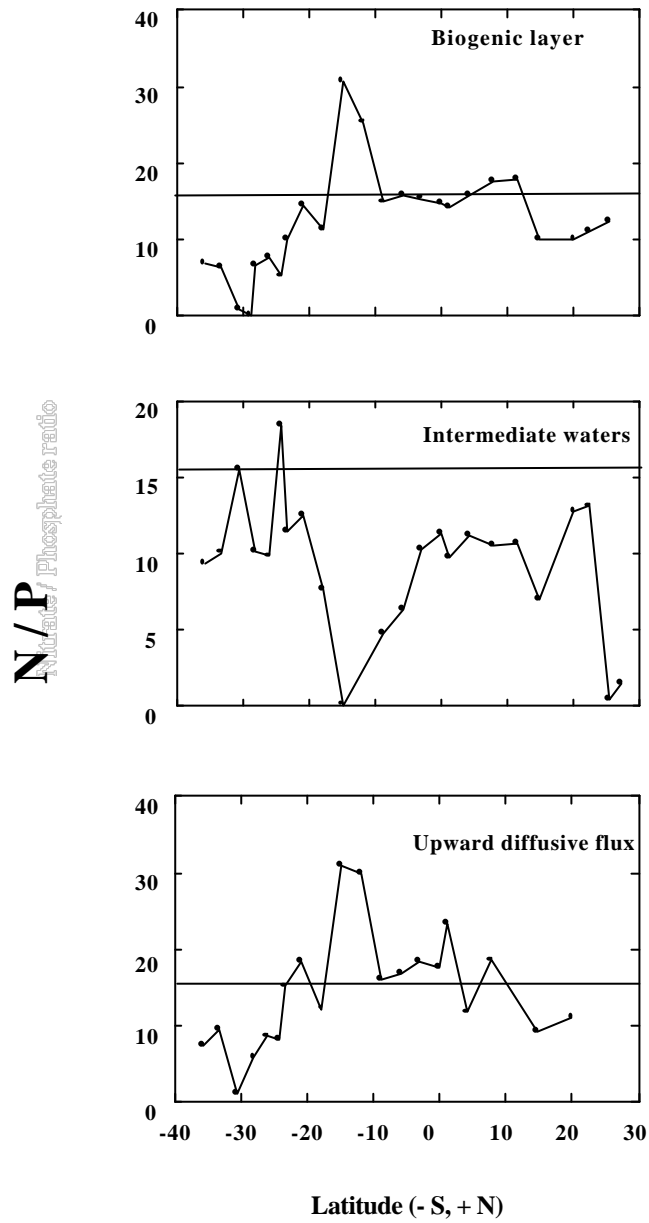
Fig. 7. The latitudinal distribution of the ratio of the pools of nitrate plus nitrite and phosphate within the biogenic layer, that in the intermediate waters between the thermocline and 1000 m depth, and that in the upward diffusive flux across the thermocline in the Central Atlantic.

and phosphorus within the biogenic layer to be lower than that in the diffusive flux, resulting in an average ratio (12.25 ± 1.4) somewhat below the Redfield stoichiometry, suggesting a tendency towards a relatively faster loss of nitrate compared to phosphate in the upper layer. The dissolved inorganic N:P ratio in the intermediate waters, operationally defined here as the layer extending from the bottom of the biogenic layer down to 1000 m, tended, however, to be below the Redfield ratio (mean ± SE = 9.37 ± 0.9; t-test, $P < 0.05$; Fig. 7), indicating these waters to be generally depleted in nitrogen, relative to P, when compared to the Redfield ratio. Moreover, the dissolved inorganic N:P ratio in the biogenic layer and that in the upward diffusive flux were weakly, but significantly negatively correlated to that in the intermediate waters ($r^2 = -0.58$, $P < 0.01$ for both).

The calculated residence time of phosphate and nitrate (plus nitrite) within the biogenic layer indicated that the diffusive flux was, in general, very slow relative to the size of the pools, and that the residence time of the phosphate and nitrate (plus nitrite) pools in the biogenic layer ranges from a few days to decades (Table 2). The residence time of phosphate tended to exceed (47 % longer, on average) that of nitrate (plus nitrite), suggesting that the phosphate pools within the biogenic layer were larger relative to the flux than those of nitrate (plus nitrite), which could be, therefore, more rapidly depleted as a consequence of biological uptake within the biogenic layer.
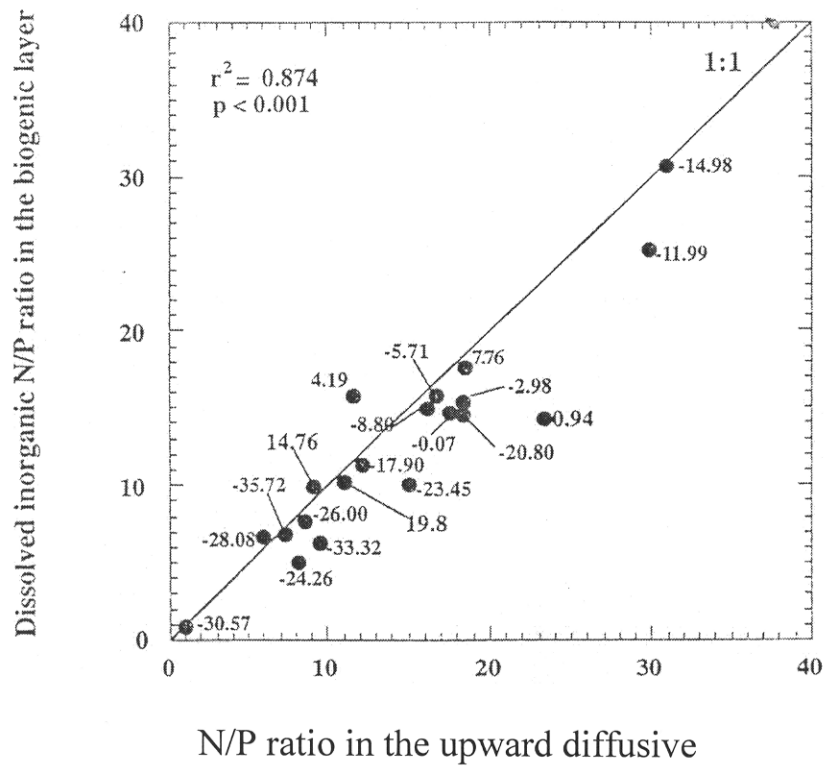
Fig. 8. The relationship between the the ratio of nitrate plus nitrite and phosphate in the upward diffusive flux across the thermocline and that in the size of the pools of these nutrients within the biogenic layer in the Central Atlantic. The solid line shows the 1 to 1 line, and the labels represent the latitude of the stations sampled (in decimal form, - S and + N).

2.5. Discussion.

The results presented confirm the oligotrophic nature of the Central Atlantic Ocean, with dissolved inorganic nutrient concentrations often close to the analytical detection limits within the mixed layer. In addition, the results demonstrate a very close correspondence between water flow distribution and dissolved inorganic nutrient concentrations along the Central Atlantic, resulting in parallel nutrient isolines and haloclines along the studied area. The large-scale patterns of nutrient concentration and biological productivity in the Central Atlantic are determined by the zonal current system. The zonal flow is driven by the easterly winds fueling at surface the South Equatorial Current to the west. The transport at depth is eastward in the form of the Equatorial Undercurrent at 0º and the North Equatorial Counter Current to the North. The former forms the Guinea Current in the eastern Atlantic (Voituriez and Dandonneau, 1974). Associated with the intense zonal is a weaker meridional pattern which is poleward on the mean above the Equatorial Undercurrent and equatorward at depth (Ekman divergence). The upwelling in low latitudes is formed as a result of the increased divergence of surface water in response to the strength of the South-East Trade Winds, resulting in the deeper water doming towards the surface and the thermocline sloping down towards the western coastal boundary. The upwelling of cold, nutrient-rich water which warms because of solar heating and becomes nutrient depleted due to biological uptake as it flows poleward explains the characteristic temperature minimum and nutrient maximum found at the equator (Figs. 2 and 3).

The strength and position of the thermocline was identified to be a major determinant of the position and sharpness of nutrient gradients across the thermocline, accounting for about 70 % of the variance in the magnitude and position of dissolved inorganic nutrient gradients across the thermocline. The thermoclines were sharpest and shallower at the equatorial and dome regions, which was associated to a similar pattern in the magnitude of dissolved inorganic nutrient gradients across the thermocline. The gradients across the thermocline of both nitrate and phosphate ranged

by about 30 fold across the transect, and were weakest in the gyre area. The depth where nitrate plus nitrate concentration gradients were sharpest tended to be located some 20 - 50 m below the depth of maximum vertical stability over most of the transect, whereas the maximum phosphate concentration gradient was located at the depth of maximum vertical stability throughout most of the transect, except for the south subtropical gyre, where it was located some 50 m below the thermocline. This suggests the presence of sinks for both nitrate and phosphate within the thermocline at the gyre area, whereas only sinks of nitrate were present below the thermocline north of the southern subtropical gyre.

The upward diffusive supply of nutrients to the mixed layer was positively related to the concentration gradient across the thermocline (Eq. 4) but inversely related to the stability of the water column (Eq. 3). Because both these properties are positively correlated across the transect, the latitudinal differences in calculated nutrient supply rates are buffered relative to those in nutrient gradients and the strength of the thermocline. Since these effects tended to cancel each other, the differences in nutrient supply rates are largely dependent on the shear strength at the thermocline. The calculated diffusive nitrate supply to the biogenic layer averaged 4.3 mmol N m^{-2} d^{-1}, exceeding by about three fold that predicted by recent models (Oschlies and Garçon, 1998), although showing a similar latitudinal trend. The new production possible with the estimated upward nutrient fluxes ranged from those characteristic of the unproductive oceanic deserts (e.g. Glibert et al., 1988) in the south subtropical gyre (1 mmol C m^{-2} d^{-1}), to moderately high rates (about 10 mmol C m^{-2} d^{-1}) in the northern end of the transect.

Since vertical and horizontal advection dominates nutrient fluxes by turbulent diffusion at the Equator (Carr et al., 1995; Chavez and Barber, 1987), the strong currents associated to the Equatorial undercurent (120 cm s^{-1} between 60 and 85 m) should lead to a fast advective export rate of plankton, thereby maintaining biomass well below the levels where resource supply could limit primary production. The low vertical eddy diffusivites ($< 10^{-4}$ m^{-2} s^{-1}) and low nutrient fluxes (< 0.1 and < 1 μmol

phosphate and nitrate $\bar{m}^2$ d^{-1}, respectively) in gyre areas concur with results previously published for comparable regions in the Pacific Ocean and the Sargasso Sea (cf.; Lewis et al., 1986; Altabet, 1989; Carr et al., 1995; Christian et al., 1997) and modeled values in the Central Atlantic (Oschlies and Garçon, 1998). These results highlight the importance of the Equatorial Undercurrent as a nutrient pump able to enhance primary production to values comparable to those at upwelling systems within the restricted area affected. Moreover, the effect of the nutrient pump at the Equator should also affect the adjacent areas, which should receive the excess nutrients exported.

Dissolved inorganic nitrogen and phosphorus fluxes across the thermocline were, on average, in good agreement with Redfield stoichiometry, but tended to be well above the Redfield ratio (i.e. P deficient) in the south subtropical gyre, and well below the Redfield ratio (i.e. N deficient) in the southernmost section of the transect. These differences accounted for most (87 %) of the variance in the ratio of dissolved inorganic nitrogen and phosphorus within the biogenic layer, but the N/P ratio of the upward diffusive flux and that in the biogenic layer were not positively correlated to the N/P ratio of the intermediate waters. This finding clearly indicates that the nutrient supply to the biogenic layer was closely coupled to the recycling of the downward flux, rather than to a supply from the intermediate waters. The tendency for the ratio of dissolved inorganic nitrogen and phosphorus within the biogenic layer to be lower than that in the diffusive flux together with the tendency for the nitrate residence times to be shorter than those of phosphate, suggests a tendency towards a relatively faster loss of nitrate compared to phosphate in the upper layer. The nutrient loss from the biogenic layer was dominated by a sinking flux of dissolved organic matter, likely deriving from nitrogen fixation (Vidal et al., 1999), which had a relatively high nitrogen: phosphorus ratio (median DON:DOP = 28, Vidal et al., 1999) in the studied waters. These observations point to the sinking loss of dissolved organic mater as the process responsible for a faster loss of N from the biogenic layer, accounting for the tendency towards a reduction in nitrate : phosphate ratios within the biogenic layer relative to those in the diffusive flux. An excess N within the thermocline of the tropical North

Atlantic has been already noted by Gruber and Sarmiento (1997), who assigned it to remineralization of nitrogen-rich organic matter from diazotrophic organisms.

The results obtained support recent studies of substantial N: P differences in the oligothrophic layers of the ocean (Fanning, 1992) relative to that reported by Redfield (1963). This variability must be mirrored by similarly substantial variability in the relative role of N and P as limiting nutrients in the ocean. Yet, the close correspondence between N:P ratios in the diffusive flux and those in the nutrient pools present in the biogenic layer suggests that plankton communities in the Central Atlantic are adapted to the incoming nutrient ratios, implying that the net removal ratio of N and P in those waters takes place at rates below the Redfield ratio. These results indicates that phosphorus deficiency is restricted to a limited area (10 - 15 ^{o}S), whereas nitrogen supply exert a greater control on primary production over most of the Central Atlantic Ocean.

The strong, well-defined Equatorial Undercurrent generated very substantial shear, which translated in diffusive nutrient fluxes about two orders of magnitude higher at the Equator than anywhere else along the transect. The diffusive nutrient flux was weakest at the southern subtropical gyre, and tended to be relatively higher in the dome area, hence leading to important differences in the areal new production that could be supported from these nutrient inputs. The potential new production ranged about an order of magnitude from the gyre area (about 1 mmol C $\bar{m}^{2}$ d^{-1}) to the northern part of the transect (about 10 mmol C m^{2} d^{-1}), and was very large (> 500 mmol C $\bar{m}^{2}$ d^{-1}) at the well-defined area of the Equatorial Undercurent, where this high potential new production was not realized. The biogenic layer was much deeper in the south subtropical gyre than in the dome area, so that the volumetric primary production that could be supported by the diffusive nutrient flux in the dome area was substantial. In contrast, the thick biogenic layer (up to 200 m) and low areal nutrient flux in the gyre area are conducive to the very low potential volumetric new production rates there. These results are consistent with the distribution of nitrate uptake rates in the area (Planas et al., 1999), which are, however, well above the upward diffusive nutrient

supply at the gyre areas, where episodic nutrient inputs must be important (Planas et al., 1999; Vidal et al., 1999).

In conclusion, the results presented indicate that the vertical conveyor belt of nutrients in the upper waters operates relatively independently of the underlying waters in the Central Atlantic, except at the Equator where the divergence drives the upwelling of the intermediate waters to maintain a direct upward supply of nutrients. The nutrient conveyor belt is able to operate at nitrate to phosphate ratios well away from the Redfield ratio, so that both the biogenic and the biolithic (deeper) components should be able to adapt to local variation about the Redfield ratio. Moreover, there was a tendency for a relatively faster loss of nitrate compared to phosphate from the biogenic layer, which likely accounts for the observation of a general N deficiency in the Central Atlantic, with P deficiency being apparent only at the south subtropical gyre.

Station	Latitude	Longitude	PO4 gradient (mmol m^4)	N+N gradient (mmol m^4)	F_{PO4} (mmol m^2 d^{-1})	F_{N+N} (mmol m^2 d^{-1})	N/P upper ratio	N/P lower ratio	N/P fluxe	K_Z (m^2 s^{-1})	Depth of max.N+N gradient (m)	Depth of max.P gradient (m)	Thermocline depth (m)
1	27.21	-17.62	1.97E-03	0.04				1.48			10	80	100
3	25.17	-19.05	3.93E-03	0.05		0.02	12.44	0.57			80	100	101.5
4	24.15	-19.11		0.06		0.02				4.40E-06	80		73.3
5	22.25	-18.25	-2.75E-03	2.56E-04	-1.16E-03	1.09E-04	10.91	13.15		4.90E-06	25	70	59
7	19.83	-18.26	1.95E-02	0.22	2.04E-01	2.26	10.11	12.82	11.09	1.21E-04	40	40	28.3
8	19.16	-18.26		0.33		1.74				6.08E-05	30		32.2
9	17.42	-18.25	1.64E-02		6.92E-01					4.88E-04		55	45.2
10	16.75	-18.25		0.21		0.24				1.35E-05	20		34.2
11	14.76	-18.00	1.96E-02	0.18	2.41E-01	2.22	9.92	7.03	9.19	1.43E-04	40	150	30.2
12	14.00	-19.00	1.61E-02		1.07E-02					7.70E-06		80	40.6
13	13.24	-19.64		0.26		1.25				5.45E-05	40		33.3
14	11.31	-20.44	9.07E-04	-0.15	1.02E-01	-16.96	17.83	10.76		1.30E-03	40	45	76.3
15	10.29	-20.81		0.25		1.53				7.18E-05	80		
16	7.77	-21.74	1.42E-02	0.26	8.02E-02	1.48	17.50	10.59	18.50	6.53E-05	65	65	49.3
17	6.63	-22.19		0.14		19.07				1.60E-03	80		63.3
18	4.20		1.12E-02	0.13	6.59E-01	7.65	15.81	11.20	11.61	6.78E-04	80	80	75.3
19	4.08	-23.53		0.18		7.03				4.58E-04	65		63.5
20	0.95	-24.41	7.08E-03	0.16	1.45E-01	3.38	14.26	9.74	23.27	2.38E-04	90	90	96.4
21	-0.07	-24.72	6.41E-03	0.11	6.00E+00	105.01	14.60	11.35	17.50	1.08E-02	80	80	86.3
22	-2.99	-26.22	7.12E-03	0.13	3.39E-01	6.21	15.27	10.31	18.31	5.51E-05	80	80	93.4
23	-5.72	-27.06	1.40E-02	0.23	9.39E-02	1.57	15.80	6.37	16.71	7.77E-05	115	95	104.5
24	-6.58	-27.53		0.19		1.06				6.49E-05	80		127.4
25	-8.80	-28.30	7.66E-03	0.12	5.08E-02	0.82	14.86	4.81	16.10	7.67E-05	115	115	159.5
26	-9.75	-28.63		0.16		9.04				6.45E-04	150		160
27	-11.99	-29.74	1.62E-03	0.05	6.99E-05	0.00	25.24		29.82	5.00E-07	140	130	199
28	-12.77	-29.74		0.10		3.51				4.08E-04	175		210
29	-14.98	-31.04	2.90E-03	0.09	1.85E-02	0.57	30.70	0.19	30.96	7.36E-05	175	125	166.5
30	-15.69	-31.34		0.12		4.84				4.75E-04	250		229
31	-17.90	-32.22	5.92E-03	0.07	9.99E-02	1.22	11.24	7.72	12.21	1.95E-04	250	160	184.6
32	-18.78	-32.57		0.06		1.30				2.39E-04	300		197
33	-20.80	-33.37	3.66E-03	0.07	1.81E-02	0.33	14.46	12.51	18.32	5.72E-05	300	70	199
34	-21.65	-34.07		0.05		0.63				1.39E-04	300		185
35	-23.45	-35.75	1.95E-03	0.03	3.52E-02	0.53	9.97	11.43	15.10	2.09E-04	400	120	192
36	-24.27	-36.35	2.51E-03	0.02	2.58E-03	0.02	5.08	18.51	8.14	1.19E-05	400	140	157
37	-26.00	-38.17	2.72E-03	0.02	1.31E-02	0.11	7.65	9.88	8.52	5.58E-05	400	135	149
38	-26.58	-38.71		-6.19E-04		-0.01				2.15E-04	140		130
39	-28.09	-40.14	2.68E-03	0.02	4.69E-02	0.28	6.66	10.21	5.92	2.03E-04	200	140	178.6
40	-28.84	-40.84		0.02		0.27				1.58E-04	400		68
41	-30.58	-42.54	4.28E-03	4.24E-03	1.37E-01	0.14	0.81	15.49	0.99	3.69E-04	250	250	124
42	-31.59	-43.56		0.03		1.98				8.17E-04	400		126
43	-33.32	-45.27	1.25E-03	0.01	1.07E-01	1.02	6.31	10.05	9.52	9.98E-04	105	105	100.3
44	-34.01	-46.01		0.02		0.34				2.56E-04			44.4
45	-35.73	-47.70	5.03E-03	0.04	8.15E-02	0.59	6.77	9.40	7.28	1.87E-04	40	20	23
46	-36.35	-48.39		0.04		0.38				1.07E-04	20		39.4

Table 1. Physical estimates and nutrient ratios for each station along the transect. Nutrient gradient was estimated within the upper layer. Nutrient fluxe (F_{PO4} and F_{N+N}) were calculated following equation (4). The N/P upper ratio determines the nutrient stoichiometry of the upper layer whereas the N/P lower ratio indicates the N+N and PO4 relationship between the base of the upper layer and the first 1000 m. depth.The coefficient of diffusivity (K_Z in m^2 s^{-1}) was calculated at the base of the euphotic layer. The depth where the nitrate(+nitrite) and phosphate gradients were maximal (m) are also showed as well as the depth of the thermocline (m).

Station	Latitude	Longitude	M_{PO4} (mmol m^2)	M_{N+N} (mmol m^2)	T_{PO4} (d^{-1})	T_{N+N} (d^{-1})	PO4 Residence time (d)	N+N Residence time (d)	New Production (mmolC m^{-2} d^{-1})
1	7.21	-17.62	10.28	102.65					
3	25.17	-19.05	13.56	92.28					
4	24.15	-19.11	32.14	174.04		1.32E-04		7598.98	0.15
5	22.25	-18.25	59.25	483.27	-1.96E-05	2.25E-07	-50926.38	4450538.46	7.00E-04
7	19.83	-18.26	11.40	271.48	1.79E-02	8.31E-03	55.99	120.29	14.95
8	19.16	-18.26	29.66	27.49		6.34E-02		15.78	11.54
9	17.42	-18.25	17.94	11.47	3.86E-02		25.94		
10	16.75	-18.25		204.83		1.18E-03		849.77	1.60
11	14.76	-18.00	13.69	233.25		9.51E-03		105.13	14.70
12	14.00	-19.00	15.37	14.89	6.98E-04		1432.27		
13	13.24	-19.64	18.42	189.77		6.57E-03		152.29	8.26
14	11.31	-20.44	71.62	71.57	1.43E-03	-2.37E-01	701.04	-4.22	-112.37
15	10.29	-20.81		1389.56		1.10E-03		905.78	10.16
16	7.77	-21.74	4.99	287.53	1.61E-02	5.16E-03	62.15	193.73	9.83
17	6.63	-22.19	18.87	197.23		9.67E-02		10.34	126.32
18	4.20	-23.21	11.46	243.26	5.75E-02	3.14E-02	17.40	31.82	50.65
19	4.08	-23.53	18.90	57.22		1.23E-01		8.14	46.58
20	0.95	-24.41	39.19	551.85	3.71E-03	6.13E-03	269.49	163.11	22.41
21	-0.07	-24.72	28.00	171.73	2.14E-01	6.11E-01	4.67	1.64	695.69
22	-2.99	-26.22	29.57	178.53	1.15E-02	3.48E-02	87.20	28.75	41.14
23	-5.72	-27.06	32.79	275.49	2.86E-03	5.70E-03	349.14	175.49	10.40
24	-6.58	-27.53	18.35	670.40		1.58E-03		633.90	7.01
25	-8.80	-28.30	69.79	677.38	7.27E-04	1.21E-03	1374.80	828.91	5.41
26	-9.75	-28.63	18.46	28.06		3.22E-01		3.10	59.88
27	-11.99	-29.74	95.28	1357.68	7.34E-07	1.54E-06	1362390.82	651036.17	0.01
28	-12.77	-29.74	10.57	689.97		5.08E-03		196.82	23.22
29	-14.98	-31.04	39.30	44.65	4.70E-04	1.28E-02	2127.30	78.07	3.79
30	-15.69	-31.34	7.25	439.47		1.10E-02		90.79	32.07
31	-17.90	-32.22	41.71	47.22	2.40E-03	2.58E-02	417.53	38.71	8.08
32	-18.78	-32.57	17.58	258.82		5.02E-03		199.16	8.61
33	-20.80	-33.37	48.13	125.26	3.76E-04	2.65E-03	2658.93	377.80	2.20
34	-21.65	-34.07	17.58	60.50		1.04E-02		95.76	4.19
35	-23.45	-35.75	42.39	346.80	8.31E-04	1.53E-03	1203.76	652.34	3.52
36	-24.27	-36.35	15.22	242.95	1.69E-04	8.64E-05	5902.09	11577.03	0.14
37	-26.00	-38.17	16.16	339.48	8.10E-04	3.29E-04	1234.69	3043.75	0.74
38	-26.58	-38.71	27.82	3.81		-3.02E-03		-331.64	-0.08
39	-28.09	-40.14	22.79	32.06	2.06E-03	8.67E-03	485.42	115.29	1.84
40	-28.84	-40.84	17.10	163.76		1.67E-03		598.61	1.81
41	-30.58	-42.54	15.48	152.52	8.82E-03	8.87E-04	113.33	1126.86	0.90
42	-31.59	-43.56	14.55	159.95		1.24E-02		80.91	13.10
43	-33.32	-45.27	12.03	181.73	8.93E-03	5.63E-03	111.93	177.60	6.78
44	-34.01	-46.01	16.21	151.22		2.27E-03		441.19	2.27
45	-35.73	-47.70	3.99	138.73	2.04E-02	4.27E-03	48.98	233.92	3.93
46	-36.35	-48.39	25.54	353.97		1.07E-03		933.63	2.51

Table 2. The integrated nutrient (M_{PO4}, M_{N+N}) and the turnover rates (T_{PO4}, T_{N+N}) were estimated following (5) and (6) respectively. Residence time was calculated as T_N^{-1}. All the parameters were representative of the upper layer. Corresponding new production (in mmol C m^{-2} d^{-1}) was estimated with the Redfield ratio of 6.6C: 1N.

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Nutrient (N,P and Si) and carbon partitioning in the stratified NW Mediterranean

Nutrient (N, P and Si) and carbon partitioning in the stratified NW Mediterranean

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Abstract

The distribution of nutrients and carbon in the different pools present in the three functional layers (the upper, biogenic layer, the thermocline layer, and the deeper, biolythic layer) of the stratified NW Mediterranean Sea was examined. The stoichiometry between dissolved inorganic nutrients, which had low concentrations in the surface waters, indicated a deficiency in nitrogen, relative to phosphorus, and an excess nitrogen relative to phosphorus within the thermocline, as well as a general silicate deficiency relative to both N and P, even extending to the biolythic layer. The dissolved organic matter was highly depleted in N and, particularly, in P relative to C, with average DOC/DON ratios >60 and DOC/DOP ratios >1500 in all three layers. The particulate pool was also depleted in N and P relative to C, particularly in the biolythic layer. The concentration of biogenic silica was low relative to C, N and P, indicating that diatoms were unlikely to contribute a significant fraction of the seston biomass. Most (>80%) of the organic carbon was present as dissolved organic carbon. Total organic N and P comprised 50–80% of the N and P pool in the biogenic layer, and decreased with depth to represent 10–25% of these nutrient pools in the biolythic layer. The high total N:P ratios in all three depth layers (N/P ratio >20) indicated an overall phosphorus deficiency in the system. The high P depletion of the dissolved organic matter must derive from a very rapid recycling of the P-rich molecules within DOM, and the increasing C/N ratio of DOM with depth indicates that N is also recycled faster than C in the DOM. Because of the uniform depth distribution of the total dissolved nitrogen concentration, the increase in the percent inorganic N and the decline in the percent dissolved organic N with depth indicates that there must be biological transformations between these pools, with a dominance of DON production in surface waters and remineralisation in the underlying layers, from which dissolved inorganic nitrogen is supplied back to the biogenic layer. Downward fluxes of DON and DOC were estimated at 200–250 μmol N m^{-2} d^{-1} and 1.4–2.1 mmol C m^{-2} d^{-1}, respectively, while there should be little or no export of P as dissolved organic matter. The downward DON flux exceeded the diffusive DIN supply of about 145 μmol N m^{-2} d^{-1} to the biogenic layer, suggesting that allochthonous N inputs must be important in the region.
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Keywords: Nutrients; Silica; DOC; Stoichiometry; NW Mediterranean

1. Introduction

Nutrient, particularly phosphorus, limitation plays an important role in controlling biological processes in the oligotrophic Mediterranean Sea (e.g. Fiala et al.,

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1976; Berland et al., 1980; Bethoux and Copin-Montégut, 1988; Thingstad and Rassoulzadegan 1995). Provided the low nutrient supply, the maintenance of plankton in the Mediterranean depends on nutrient recycling (Thingstad and Rassoulzadegan, 1995; Thingstad et al., 1998; Zweifel et al., 1993; Bethoux et al., 1992), particularly in summer when the development of the seasonal thermocline separates the pelagic ecosystem in three distinct layers, (1) the upper, biogenic layer, where photosynthesis supports the formation of organic matter; (2) the thermocline area, where biogenic processes associated with a deep-chlorophyll maximum established at or near the thermocline coexist with an intense remineralisation of organic matter that supplies nutrient to maintain biogenic processes; and (3) the lower, biolythic layer, where heterotrophic biological processes are maintained by organic matter exported from the overlying layers. These different layers are coupled by the downward transport of particulate and dissolved organic matter (POM and DOM, respectively), and the upward transport of inorganic nutrients from the biolythic layer.

While nutrient supply sets an upper limit to the biological production in Mediterranean waters, the planktonic organisms exert a tight control on the elemental distribution, affecting the chemical composition of both dissolved and particulate matter (Redfield, 1934; Redfield et al., 1963). While the stoichiometry between dissolved inorganic components in the biolythic zone and particulate organic compounds in the biogenic layer appears to be

described adequately by the Redfield ratio of 106 C:16 N:1 P, that of dissolved organic matter seems to deviate greatly from the Redfield ratio (Jackson and Williams, 1985; Jackson, 1988; Butler et al., 1979). A reliable account of the total stoichiometry of nutrient elements in the ocean must incorporate the dissolved organic pool (Jackson and Williams, 1985), because this is the major reservoir of both organic carbon and nutrients in the biogenic layer, particularly in oligotrophic waters (Eppley et al., 1977; Butler et al., 1979; Orret and Karl, 1987; Vidal et al. 1999). The labile and semi-labile fractions of DOM can be major sources of N and P for planktonic food webs via bacterial assimilation (Thingstad and Rassoulzadegan, 1995). In addition, the downward fluxes of dissolved organic carbon and nitrogen may comprise an important fraction of the export of new production from the biogenic layer of oligotrophic waters (e.g. Toggweiler, 1989; Vidal et al., 1999), rendering dissolved organic matter an important link between the biogenic and the biolythic layers.

The examinations of nutrient pools in oceanic waters rarely consider all three components (particulate and dissolved organic and inorganic, together with their distribution across the three distinct layers of the stratified ocean) simultaneously. As a consequence, our knowledge on the partitioning of nutrients between these pools is still poor. Our goal here is to contribute to bridging this gap by examining the distribution of C, N, P and Si in their particulate, dissolved organic, and inorganic forms in the three distinct layers of the stratified NW Mediterranean,
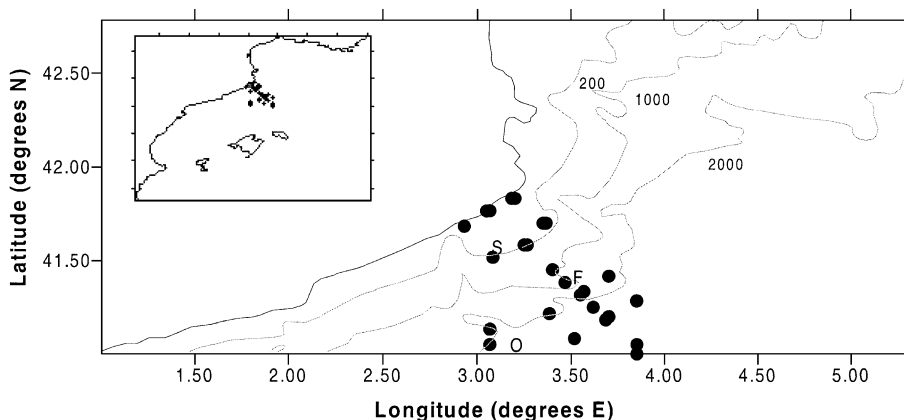

Fig. 1. Map showing the study area, with the position of the stations sampled and that of the stations S (shelf), F (front), and O (Oceanic) studied in greater detail.
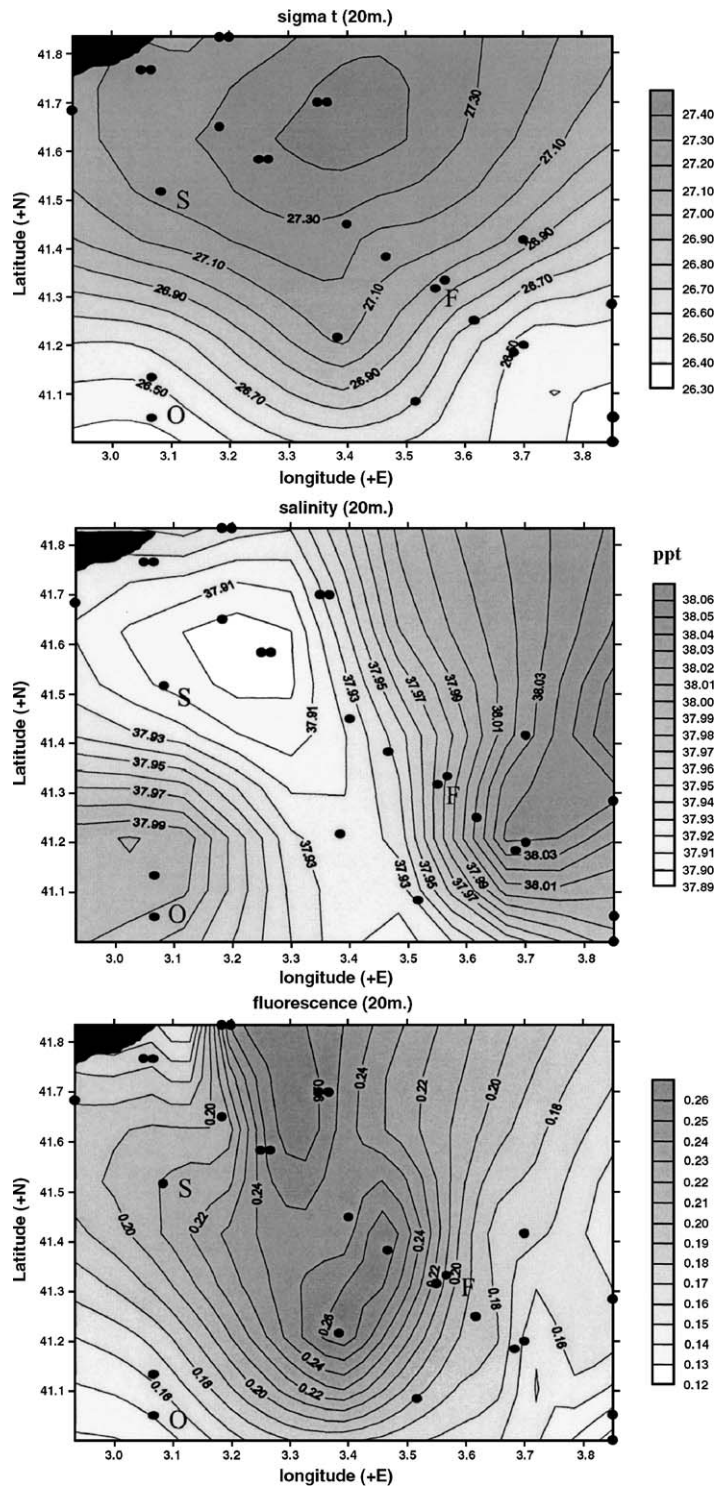
Fig. 2. Contour plot describing the distribution of salinity, density anomaly (sigma t) and fluorescence in subsurface (20 m depth) waters.

where studies concerning all nutrient pools are particularly scarce. We do so based on data collected on a cruise in the NW Mediterranean at the end of the summer of 1996. The examination of the ratios between C, N, P and Si was used to draw inferences on the recycling of nutrients and their possible role as limiting factors for primary production.

2. Methods

The study was conducted in the north-western Mediterranean Sea during the 'FRONTS 96' cruise (September 1996) on board the Spanish research vessel B/O 'Garcia del Cid'. A network of stations along the Catalan-Balearic Sea was occupied (Fig. 1).
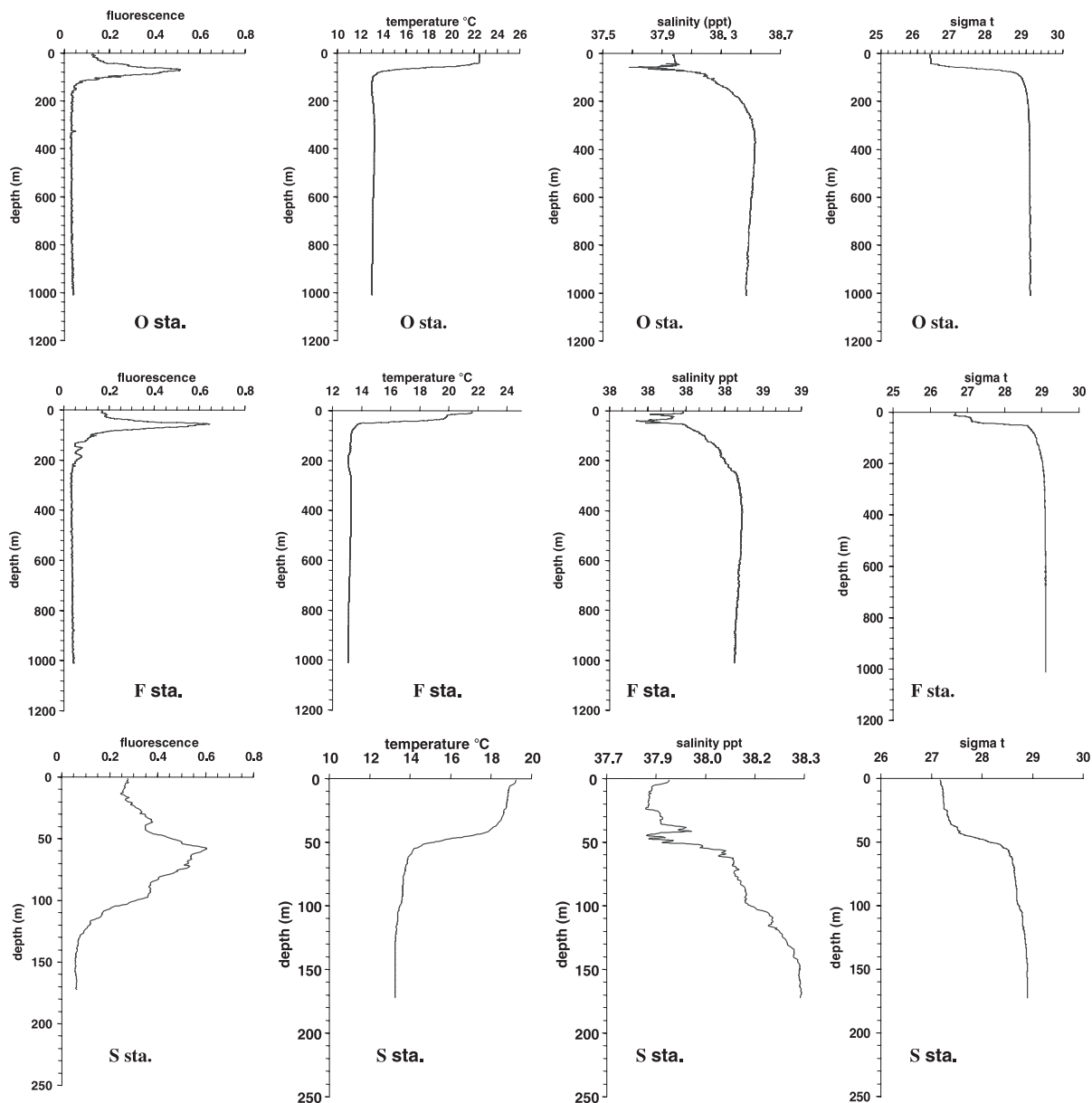
Fig. 3. Vertical profiles of fluorescence, temperature salinity, and density anomaly (sigma t) for O (oceanic), F (frontal) and S (shelf) stations.

Detailed studies were conducted at three stations, each representing the shelf (S station) and oceanic (O station) waters, as well as the frontal (F station) area separating shelf from oceanic waters (Fig. 1).

Conductivity-temperature-pressure (CTD) data were obtained with a vertical resolution of 10 cm. Salinity and temperature estimates derived from CTD casts were calibrated along the cruise using direct salinity measurements from a Guildline Autosal salinometer calibrated with IAPSO standard seawater (34.993 ppt) and a highly accurate reversible thermometer, respectively. Vertical profiles of salinity and temperature derived from the (corrected) CTD data were screened for errors. CTD casts reached near the

sediment surface, in shallow stations, or down to a maximum of 1000 m. Water samples were collected using Niskin bottles attached to a rosette system. The depths for discrete water sampling were selected from the features emerging from examination of the CTD and fluorometer profiles and included, at least, the surface mixed layer, the deep Chlorophyll-a maximum (if present), the minimum potential temperature (Mediterranean Winter Water = MWW), and the deep maximum temperature and salinity (Levantine Intermediate Water = LIW). Samples collected below 150 m in the shelf zone and below 375 m in the oceanic zone were located between the LIW and the Mediterranean Deep Water (MDW) (Lacombe and Tcher-
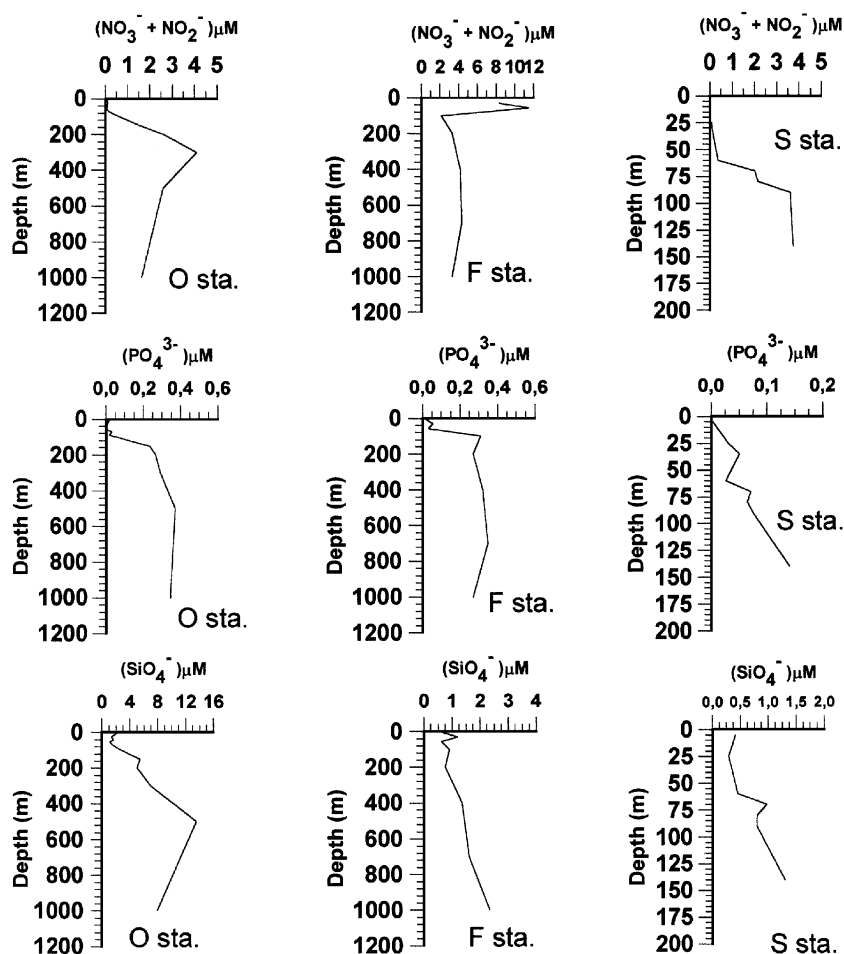
Fig. 4. Vertical profiles of dissolved inorganic nitrogen, phosphorus and silica for O (oceanic), F (frontal) and S (shelf) stations. The maximum depth sampled at the S station was 200 m.

nia, 1972). Subsamples were drawn from the Niskin bottles into pre-cleaned polyethylene bottles. Samples for dissolved inorganic and organic nutrients were preserved with chloroform and immediately frozen for later analysis. Dissolved inorganic nutrients were measured spectrophotometrically using a 10-cm cuvette cell according to standard methods (Hansen and Koroleff, 1999). The detection limit of dissolved nutrient concentrations was 0.01 μM for PO_4^{3-} and $Si(OH)_4$, and 0.02 μM for $NO_3^- + NO_2^-$. Precision was better than 0.05 μM for all nutrients, as indicated by analyses of replicate samples. Samples for particulate organic carbon (POC), nitrogen (PON) and phosphorus (POP) were collected onto pre-combusted (450

°C) Whatman GF/C glass fibre filters and kept frozen. Samples for POC and PON were exposed to concentrated HCl fumes for 30 min to remove the dissolved inorganic carbon, which may interfere with the analysis. Measurements were carried out using a Perkin-Elmer 240 CHN analyser. Samples for POP determinations were oxidised in acidic persulphate solution. The particulate phosphate was then analysed as soluble reactive phosphorus following the methods outlined in Murphy and Riley (1962). Total dissolved phosphorus and nitrogen were measured by the persulphate oxidation method of Murphy and Riley (1962) and the methods of Solórzano and Sharp (1980), respectively. Dissolved organic nitrogen
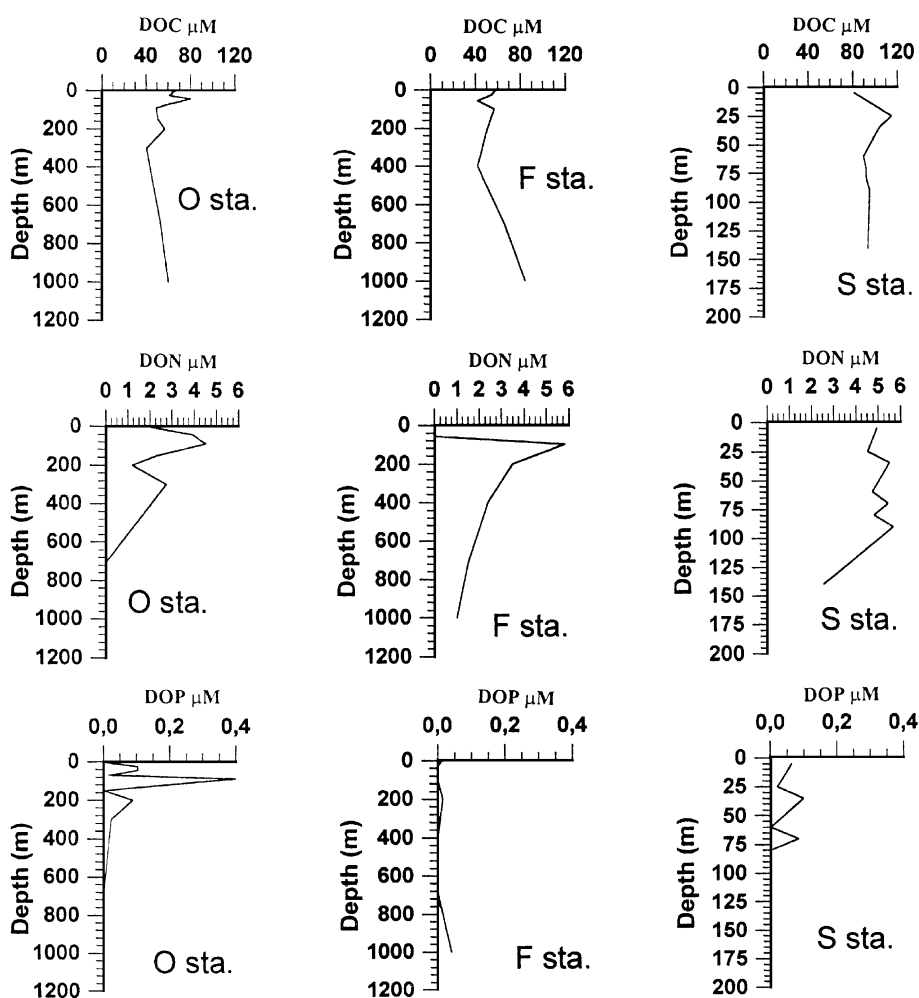
Fig. 5. Vertical profiles of DOC, DON, and DOP for the O (oceanic), F (frontal) and S (shelf) stations.

(DON) and phosphorus (DOP) concentrations were estimated as the difference between the total nutrient and dissolved nutrient concentrations.

Samples to estimate the concentration of silica of biological origin (Si_{bio}) were filtered onto 47 mm polycarbonate Nucleopore filter (0.6 μm pore size) and dried for 12 h at 60 °C. The determination of biogenic silica followed the NaOH/HF digestion me-

thod (Ragueneau and Tréguer, 1994). Samples for DOC analyses were immediately filtered (burned Whatman GF/F filters, not washed) and stored in acid-washed, precombusted glass vials. The 10 ml-subsamples were acidified by 40 μl 2 N HCl and sealed with acid-rinsed Teflon lining. The samples were stored in the dark and kept cold. DOC was measured by Pt-catalysed high temperature combus-
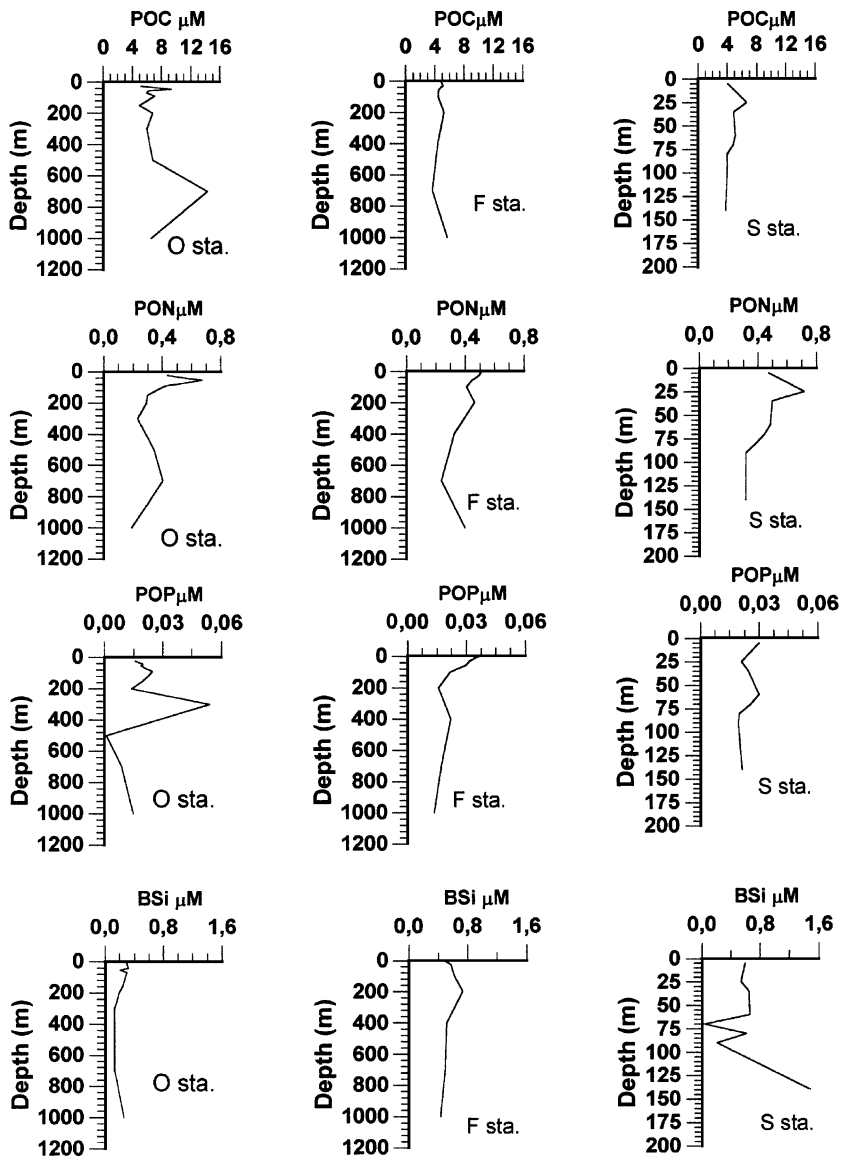
Fig. 6. Depth distribution of the particulate organic matter pool and biogenic silica for the O (oceanic), F (frontal) and S (shelf) stations.

tion in a Shimadzu TOC-5000 with autosampler injection, after being sparged with the carrier gas for 6 min at 75 ml/min (Sharp et al., 1993). A 4-point calibration curve between 50 and 200 μM was prepared for each series of measurements and its slope was used to calculate sample concentrations. Ten blank samples with MilliQ-water and added acid were evenly distributed within each series and the mean

blank area of about 750 (CV lower than 15% for 50 μl injections) was subtracted from each sample before calculation. Duplicate samples were analysed with a coefficient of variation lower than 7%. Measurements of DOC in blank and deep ocean water reference materials supplied by J. Sharp (pers. comm., 1997) were included in each series as a quality control. A deviation in concentration of more than 6 μM (n = 3)
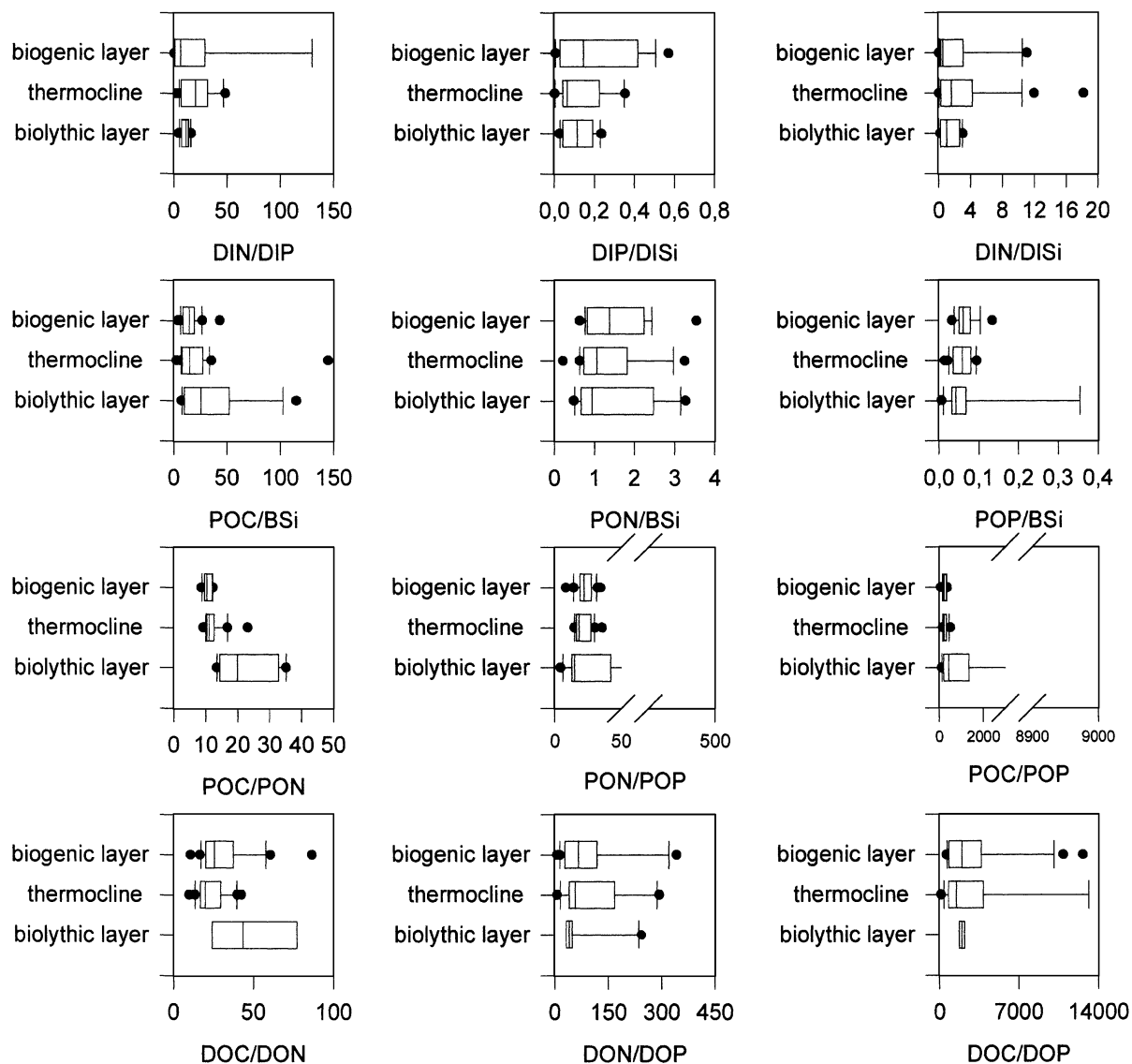
Fig. 7. Box plots showing the distribution of the carbon to nutrient ratios of dissolved inorganic, dissolved organic and particulate organic pools for the biogenic, thermocline and biolythic layers. The central line indicates the median value, the boxes encompass the lower and upper quartiles of the distribution, the lines encompass 95% of the data and the solid circles indicate observations beyond the 95% limits.

from the stated 44 μM in the deep ocean water released a re-run of our samples. Our blank samples had an average area of 117 ± 39 (SD, n = 25) higher than the reference blank. Thus, the presented DOC concentrations are probably underestimated by about 3 μM.

3. Results

The horizontal salinity distribution revealed the presence of a relatively low salinity core (about 37.91 at 20 m) associated to the Palamós Canyon. The locations of the canyon and the Catalan front (Estrada and Margalef, 1988) drive the distribution of the water stratification (Fig. 2). The circulation and upwelling associated with the canyon also contribute to supply nutrients to the euphotic zone (cf. Granata et al., 1999) as indicated by maximum fluorescence values along the northern boundary of the canyon (Fig. 2). The water column was strongly stratified (Fig. 3).

Dissolved inorganic nutrients were generally low in the biogenic layer (Fig. 4), although unusually high nitrate + nitrite concentrations of 8–12 μM were observed in surface waters of the Frontal station, suggesting high nitrogen inputs in the frontal zone prior to the cruise. These nitrate + nitrite concentrations are abnormally high, and a possible contamination cannot be ruled out. Yet, contamination is an unlikely explanation, since analyses of independent samples from adjacent stations (Fig. 1) yielded similarly high nitrate + nitrite concentrations in surface waters. Phosphate was below or near detection limit in the surface waters, and rose to about 0.4 μM in the biolythic layer (Fig. 4). Silicate concentrations varied substantially across stations, with the shelf and frontal waters having lower concentrations (0.4–2 μM) than the oceanic stations, where the concentration reached a maximum of 14 μM below 400 m.

Dissolved organic carbon and nitrogen concentrations were highest in the mixed layer (80–120 μM DOC, 5–6 μM DON) at the shelf and oceanic stations (Fig. 5). DOP concentrations were low at all stations, except for some relatively high (0.4 μM) values just below the thermocline at the oceanic station (Fig. 5). The range of particulate organic nutrients was similar across stations (Fig. 6), although it showed greater variability with depth in the oceanic and shelf stations

than at the front. The concentration of Si_{bio} was highest in shelf waters and declined towards the open sea (Fig. 6).

The elemental ratios differed greatly among pools and across layers. The ratio of dissolved inorganic nutrients indicated a deficiency in nitrogen, relative to phosphorus in the biogenic layer — where these estimates are subject to considerable uncertainty due to low concentrations — and, conversely, an excess of nitrogen relative to phosphorus within the thermocline relative to the Redfield N:P ratio of 16, which was found in the biolythic layer (Fig. 7, Table 1). There was, on average, a general silicate deficiency relative to both N and P extending even into the biolythic layer (Table 1), compared to the global ratios of these elements (16 N: 16 Si: 1 P, Redfield et al., 1963; Brzezinski, 1985). The dissolved organic matter was highly depleted in P relative to N and C in all layers (Fig. 7), with average DOC/DON ratios >25 and DOC/DOP ratios >1500 in all three layers of the water column (Table 1). The stoichiometric ratios of the particulate pool also showed evidence of N and P depletion relative to C in all layers, which was particularly high in the biolythic layer, whereas N was present in near-Redfield balance relative to P in the particulate pools of the biogenic and thermocline layers (Fig. 7, Table 1). The concentration of Si_{bio} relative to C, N and P was low (Fig. 7, Table 1), indicating that diatoms were unlikely to contribute significantly to the seston biomass.

The partitioning of the nutrient stocks between different pools is best represented by the percentage of the total stock they comprise (Fig. 8). Most (>80%) of the organic carbon was present as dissolved organic carbon, with POC representing a minor percentage throughout the water column (Fig. 8). In contrast, the importance of organic matter as a reservoir of N and P

Table 1

Average nutrient ratios in the different pools (dissolved and particulate) and layers of the stratified NW Mediterranean considered

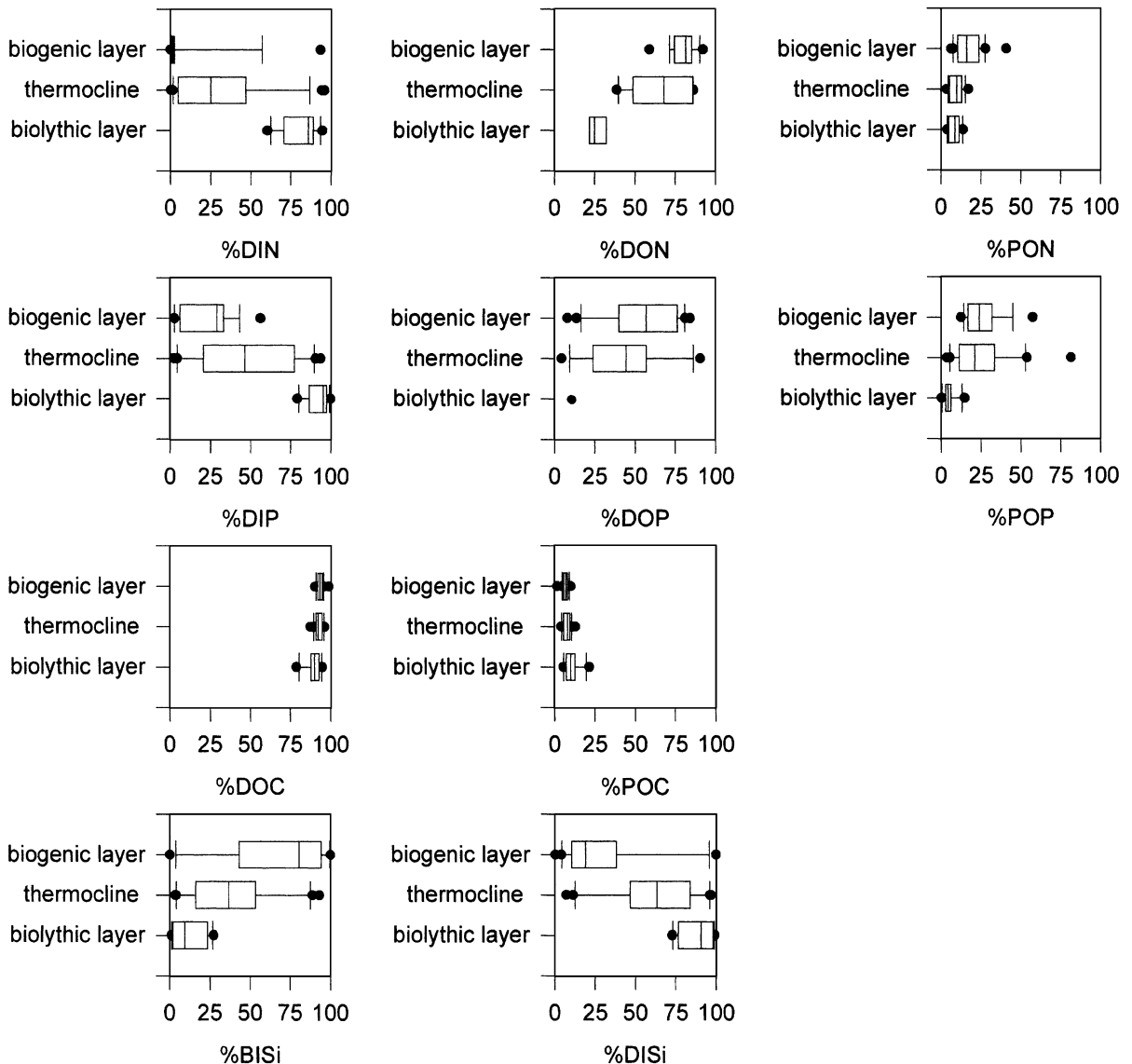| zone	DIM N : Si : P	DOM C : N : P	POM C : N : Si : P
biogenic	6 : 7 : 1	1984 : 66 : 1	232 : 22 : 16 : 1
thermocline	20 : 17 : 1	1510 : 58 : 1	220 : 18 : 16 : 1
biolythic	11 : 10 : 1	1974 : 25 : 1	426 : 15 : 25 : 1

Fig. 8. Box plots showing the distribution of the relative distribution of the total nutrient stock among different pools for the biogenic, thermocline and biolythic layers. The central line indicates the median value, the boxes encompass the lower and upper quartiles of the distribution, the lines encompass 95% of the data and the solid circles indicate observations beyond the 95% limits.

declined greatly with depth, from some 50–80% in the biogenic layer decreasing to 10–25% in the biolythic layer, parallel to the rise with depth of the contribution of importance of dissolved inorganic compounds to the reservoir of N and P (Fig. 8). A similar shift with depth was evident in the partitioning between the particulate and dissolved silica pools (Fig. 8).

4. Discussion

The hydrographic structure during the cruise showed a ridged-shaped elevation of the isopycnals as a result of the divergence between the southwest-flowing coastal current on the Catalan side and a flow to the north-east on the Balearic Islands side, which

determines the presence of a dome resulting in a front parallel to the Catalan coast (Font et al., 1988). The origin of the low salinity core on the slope of the Palamós canyon is the advection of shelf water from the Gulf of Lions by the Liguro-Provençal-Catalan (or Northern) Current (Le Vouch et al., 1992). The higher salinity values are formed by upwelling of Levantine Intermediate Water, which is traced by the pattern of fluoresence. A fluorescence maximum was associated with the front area, where doming pycnoclines lead to a high nutrient supply (Estrada and Margalef, 1988). Dissolved inorganic nutrient concentrations were otherwise low, and the results suggested a general silicon deficiency, consistent with the dominance of autotrophs other than diatoms (i.e. picoplankton, Agustí et al., 1998; Agawin et al., 1998) in the stratified NW Mediterranean.

DOC and DON concentrations were comparable to those observed earlier for the NW Mediterranean (Copin-Montégut and Avril, 1993; Doval, 1999), and comprised the bulk of the organic carbon and nitrogen, as observed for other oligotrophic waters (Bethoux and Copin-Montégut, 1988; Minas et al., 1988; Copin-Montégut and Avril, 1993; Doval, 1999; Vidal et al. 1999) and some more productive waters (Søndergaard et al., 2000) in the past. The relatively high DOC concentration of up to 90 μM in the deep (1000 m) waters at the frontal stations is consistent with earlier observations in this area (Cauwet, 1983; Cauwet et al., 1997), suggesting that the acumulation of DOC is a characteristic feature of the deep water mass located in the frontal region.

The total N:P ratios far exceeded the Redfield ratio in all three depth layers (N/P ratio >20), suggesting an overall system phosphorus deficiency, which is consistent with reports of P limitation in the Mediterranean Sea (Minas et al., 1988; Jacques et al., 1973; Thingstad and Rassoulzadegan, 1995; Thingstad et al., 1998). Such P deficiency is, however, not evident from either the dissolved inorganic nutrients, where P is in excess relative to N in the biogenic zone, or the particulate organic matter, where the observed N/P ratios are comparable to the Redfield ratio (Table 1). The P deficiency, however, becomes evident when the highly P-depleted dissolved organic pool (N/P ratio >50) is considered. P depletion of dissolved organic matter has already been reported elsewhere (e.g. English Channel, Butler et al., 1979; Pacific Ocean, Jackson and Wil-

liams, 1985; Central Atlantic, Vidal et al., 1999) and seems to be a general feature of dissolved organic matter. Because DOP production rates are high in oligotrophic environments (e.g. Cañellas et al., 2000), the P depletion of the dissolved organic matter must derive from a very rapid recycling of the P-rich molecules within DOM (Orret and Karl, 1987; Thingstad, 1993; Thingstad and Rassoulzadegan, 1995). The increasing C/N ratio of DOM with depth indicates that N is also recycled faster than C in the DOM (Fig. 6).

The distribution of total nitrogen was rather uniform with depth, with PON representing a modest (median < 20%) fraction of the total N pool. Examination of the relative distribution of the nutrient pools revealed an opposite trend in the contribution of the dissolved inorganic and dissolved organic pools with depth (Fig. 7). Because of the uniform depth distribution of the total dissolved nitrogen concentration, this pattern indicates that the net result of biological activity in the waters studied is the production of dissolved organic nitrogen in the biogenic layer and its subsequent remineralisation in the aphotic, biolythic layer. These changes are mediated by the biota contained within the relatively modest particulate organic N pool. High phytoplankton DON and DOP production rates, often exceeding 50% of the N and P uptake (Bronk et al., 1994; Cañellas et al., 2000) have indeed been reported for oligotrophic marine waters, where DOC production rates can also be high, e.g. >30–40% of photosynthetically-produced C (Williams, 1995; Søndergaard et al., 2000). Exudation by healthy cells, representing < 10% of the primary production (Carlson and Ducklow, 1995) does not appear to be sufficient to account for such high DOM production rates, so other processes must be invoked to explain the apparently high DOM production. Recent results point to high phytoplankton lysis rates in oligotrophic marine waters (e.g., 0.8 d^{-1} and 0.45 d^{-1} in the open and coastal NW Mediterranean, Agustí et al., 1998; Agustí and Duarte, 2000, respectively; and 0.95 d^{-1} in the NE subtropical Atlantic, Agustí et al., 2000) as an important source of DOM, which must be accompanied by a relatively inefficient recycling by bacteria to allow its accumulation.

The inorganic nutrients produced upon mineralisation of DON in the deeper water column is re-supplied to the biogenic layer. We hypothesise that DON is an important link between the biogenic and biolythic

layer. This was tested by calculating the downward flux of DON by gradient-driven diffusive transport (cf. Lewis et al., 1986; Vidal et al., 1999) as the product between the mean DON gradient across the thermocline in the frontal and oceanic stations (68 and 83 μM N m^{-1}, respectively), and an assumed vertical turbulent diffusion coefficient of 3 m^2 d^{-1} for the stratified NW Mediterranean (Copin-Montégut and Avril, 1993). The calculated downward fluxes (200 and 250 μmol N m^{-2} d^{-1} for the frontal and oceanic stations, respectively) are high compared to the average PON export from the biogenic layer in the NW Mediterranean (140 μmol N m^{-2} d^{-1}, Miquel et al., 1994). In contrast, there was no measurable DOP gradient across the thermocline, indicating the export of P as dissolved organic matter to be small, consistent with results from oligotrophic waters elsewhere (Vidal et al., 1999). These calculations indicate that the downward flux of DON is a dominant source of nitrogen export from the biogenic layer of the NW Mediterranean (about 60% of the downward N transport). Our calculations — using a similar approach of that used to calculate the DON flux — of the downward DOC transport, yielded estimates of 1.4 and 2.1 mmol C m^{-2} d^{-1} for the Frontal and Oceanic stations, respectively, somewhat greater than previous estimates (0.82 mmol C m^{-2} d^{-1}, Copin-Montégut and Avril, 1993), but comparable to the estimates of the average POC export from the biogenic layer (1.08 mmol C m^{-2} d^{-1}, Miquel et al., 1994). These estimates support previous findings that DOC transport comprises half of the organic C export from the biogenic layer of the NW Mediterranean Sea. The sum of the DOC flux estimated here (average 1.7 mmol C m^{-2} d^{-1}) and the average POC flux (1.1 mmol C m^{-2} d^{-1}) in the NW Mediterranean yield an estimate of TOC flux which is in the low range of the estimated new production in the region (4 mmol C m^{-2} d^{-1}, Minas et al., 1988, and 2.7–7.9 mmol C m^{-2} d^{-1}, Bethoux, 1989). This contrast suggests that new production must be much higher during the winter mixing period than that we found during the stratified period, consistent with the nutrient limitation experienced in the stratified period.

Based on the vertical gradient of dissolved inorganic nitrogen of the oceanic station (48 μM N m^{-1}), a supply of 145 μmol N m^{-2} d^{-1} to the biogenic layer was estimated. Hence, the export of N from the biogenic layer appears to exceed the re-supply of inorganic nitrogen across the thermocline, suggesting that allochthonous N inputs must be important in the region. Indeed, the atmospheric N deposition in the NW Mediterranean averages 120 μmol N m^{-2} d^{-1} (Erdman et al., 1994), which would represent about half of the total N supply, and is sufficient to compensate for the excess downward N export as DON. The imbalance of N exchange across the thermocline suggests that there must be lateral export or burial of N. The diffusional phosphate supply to the biogenic layer, based on the vertical gradient of phosphate for the oceanic station was estimated at 7.2 μmol P m^{-2} d^{-1}, which must be balanced by a similar particulate export, since there was no evidence for a downward flux of DOP at the station.

5. Conclusions

Our results show that dissolved organic components dominate the pools of N, P and organic C in the biogenic layer of the NW Mediterranean, shifting to a dominance of dissolved inorganic nutrient pools below the thermocline. These patterns result from the active elemental cycling between dissolved organic and inorganic pools as catalysed by a small pool of living organisms together with gradient-driven vertical fluxes of DOM from the biogenic layer to the biolythic layer and a reverse upward gradient-driven flux of dissolved inorganic nutrients. The downward export of N appears to exceed the upward diffusional flux of dissolved inorganic nitrogen, and is probably balanced by a significant input of atmospheric N to the biogenic layer. In contrast, the downward transport of P is much reduced and limited to the modest particulate flux. P appears to be rapidly and preferentially recycled from DOM. Hence, the system shows clear symptoms of P limitation, so that the high excess export of N from the biogenic layer does not represent a reliable estimate of the new production of the system, which must be estimated in terms of the export of P, the limiting nutrient, in the NW Mediterranean.

Acknowledgements

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Nutrient dynamics and ecosystem metabolism in the Bay of Blanes (NW Mediterranean)

By

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4.1. Abstract.

The dynamics of the nutrient pools and their stoichiometry as well as their control by ecosystem metabolism (benthic and planktonic) and benthic-pelagic exchanges (sedimentation rates and sediment-water fluxes) were examined in the Mediterranean littoral (Blanes Bay, NE Spain). Dissolved organic nitrogen comprised about half of the nitrogen present in the water column and the carbon pool dominated by the inorganic pool (95% of the carbon present in the water column). The dissolved and particulate organic pools were deficient in P, indicating a rapid recycling of P from organic matter. The pelagic compartment was heterotrophic, supported by significant allochthonous inputs of land material, which also contributed greatly to the sedimentary inputs (37% of total sedimenting carbon). In contrast, the benthic compartment was autotrophic, with the excess net benthic community production balancing the deficit in pelagic community production, leading to a system in metabolic equilibrium. Sedimentary inputs of phosphorus and silicon were compensated by sediment release of phosphate and silicate, whereas nitrogen was lost or accumulated in the sediment compartment. Carbon inputs to the benthic compartment also exceeded requirements, due to the allocthonous subsidies to the system, so that the benthic compartment stored or exported organic carbon.
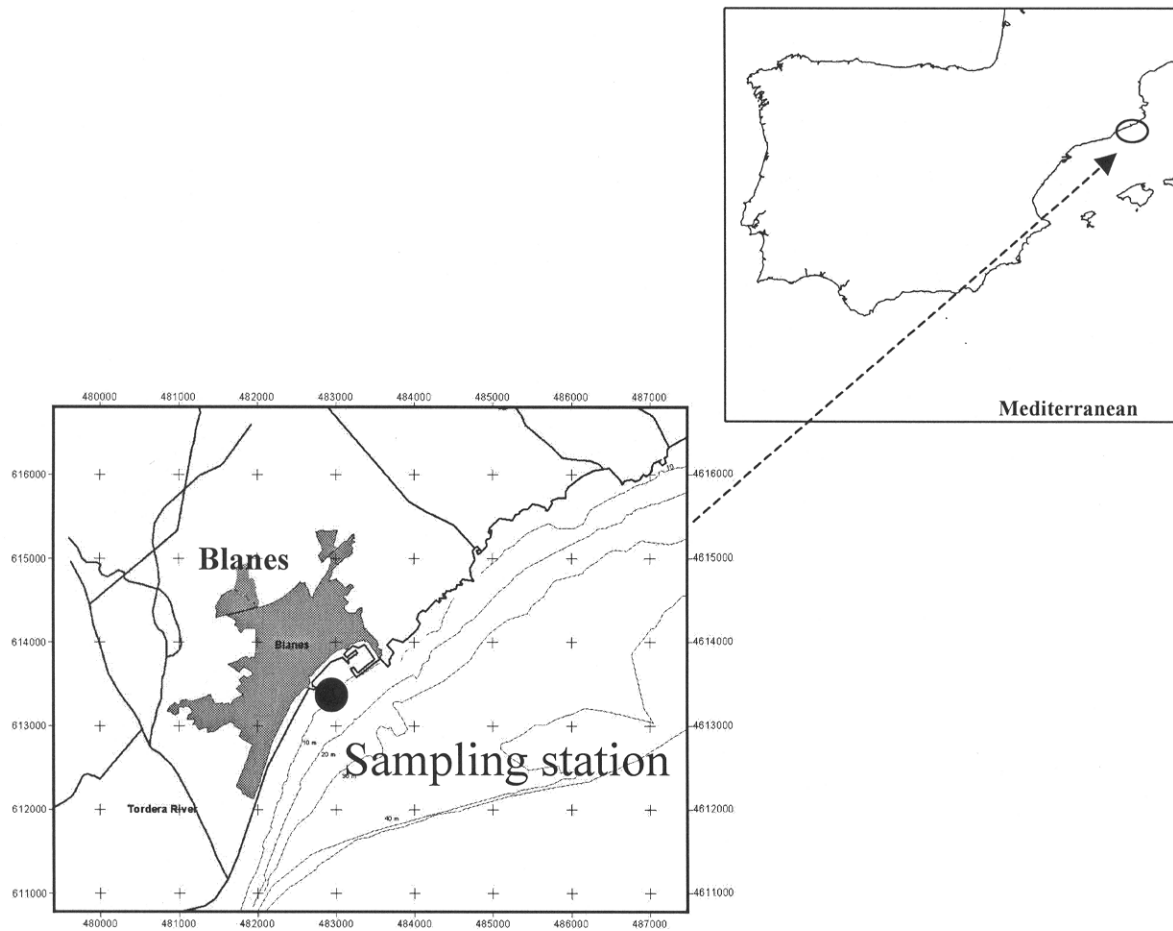
Fig.1. Location of the study area.

4.2. Introduction.

Widespread eutrophication has stimulated research on nutrient dynamics in coastal ecosystems (e.g. refs. Estuaries; Justic et al., 1995, Nixon et al., 1995, Vidal et al., 1999a). However, the resulting knowledge is biased towards nutrient-rich or eutrophied systems (Rosenberg, 1985; Westernhagen et al., 1986; Andersson and Rydberg, 1988: Vidal et al., 1999a), and accounts of nutrient dynamics in oligotrophic coastal waters are still few. Whereas nutrient dynamics in eutrophied coastal systems are dominated by allochthonous inputs (Marchetti, et al., 1989; Turnner and Rabalais, 1991) nutrient dynamics in oligotrophic coastal waters are strongly dependent on internal recycling processes (Thingstad and Sakshaug., 1990; Ittekkot et al., 1991). These are linked to respiratory processes, much of which occur in the sediments (Heip et al., 1995), thereby requiring a close coupling between benthic and pelagic nutrient cycling and metabolism. The extent of the coupling between benthic and pelagic nutrient cycling and metabolism may change seasonally because of the possible stimulation of respiration rates with increasing temperature, which may lead to faster nutrient recycling. However, winter mixing may also supply nutrients entrained from deeper offshore waters to the coastal zone of temperate regions, relaxing the dependence on remineralization as a source of nutrients in winter.

The Mediterranean littoral zone is still largely oligotrophic, although eutrophication problems are also increasing in the Mediterranean (Vidal et al., 1999a, UNEP 2000). Moreover, because of the characteristic episodic nature of rainfall in the Mediterranean region (e.g. Duarte et al., 1999), nutrient inputs from land occur in pulses (Bavestrello, 1995; Buscail et al., 1995), rather than as a continuous supply. As a consequence, the nutrient dynamics of the Mediterranean coastal waters are dependent on the complex interplay between land inputs, marine supply in winter and recycling processes from the sediments. Each of these sources involve a differential partitioning between the various nutrient pools involved, with land-derived inputs delivering both organic and inorganic nutrients (Meybeck, 1982; Cauwet and Martin, 1982; Cauwet et

al., 1990), and marine supply and sediment release delivering inorganic nutrients. As a consequence, the concentrations of nutrients, their stoichiometric balance and the partitioning between inorganic and organic (dissolved and particulate) nutrient pools all may vary significantly over time, likely leading to complex biogeochemical dynamics in Mediterranean littoral waters.

Here we contribute to our knowledge on nutrient dynamics in temperate oligotrophic coastal waters by assessing the nutrient partitioning in the Bay of Blanes (NW Mediterranean). To this end we describe the dynamics of the nutrient pools and their stoichiometry as well as their control by ecosystem metabolism (benthic and planktonic) and benthic-pelagic exchanges (sedimentation rates and sediment-water fluxes).
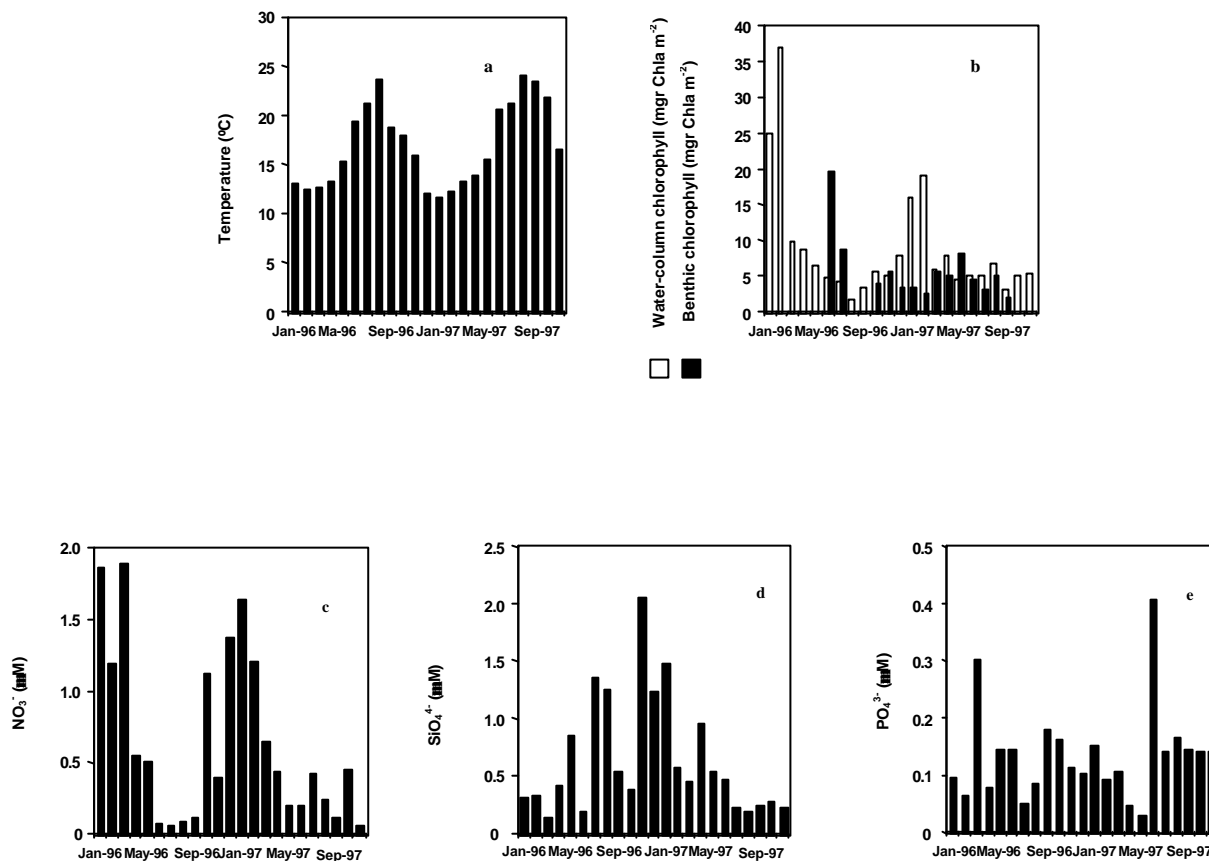
Fig.2. Temporal variation of mean monthly dissolved inorganic nutrient concentrations, surface seawater temperature and water-column and benthic chlorophyll a concentration in the Blanes Bay for 1996 to 1997.
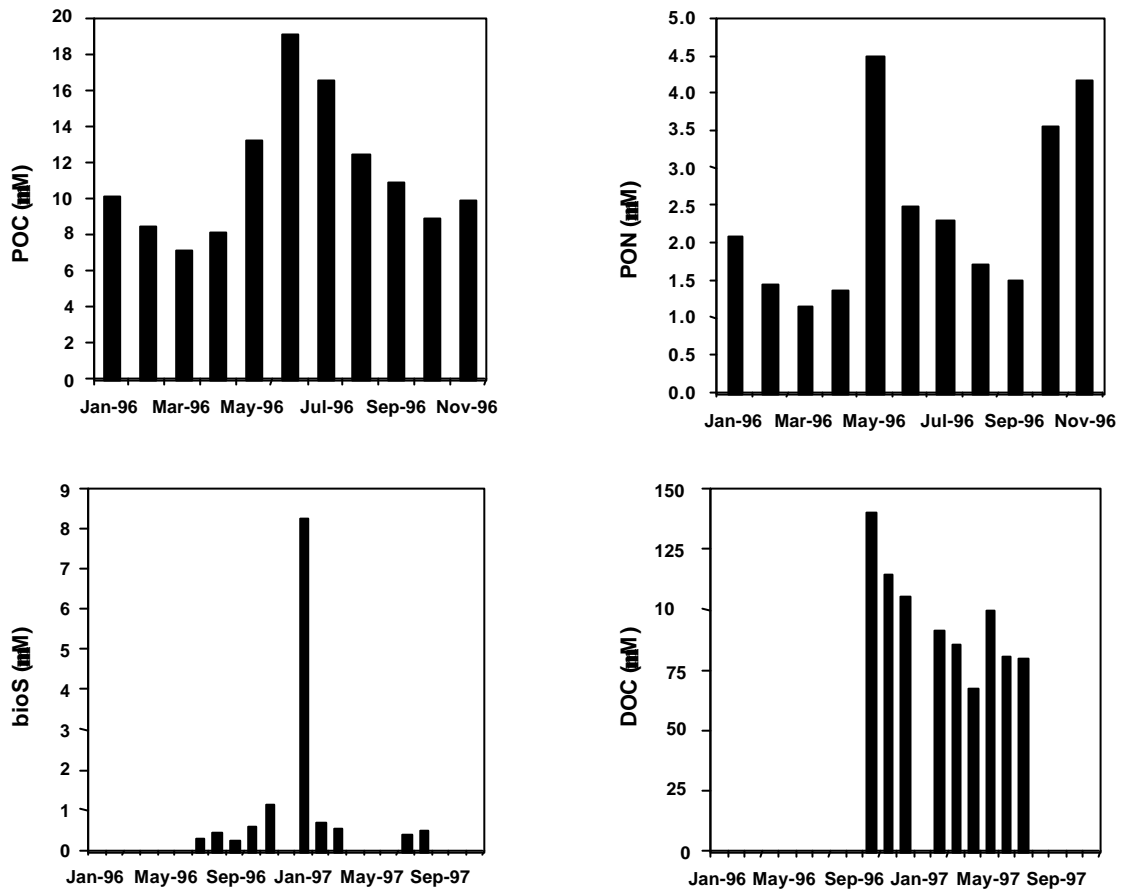
Fig.3. Temporal variation of mean monthly particulate organic matter and DOC concentrations in the Blanes Bay for 1996 to 1997.

4.3. Methods.

The study was conducted in the Bay of Blanes located in the North Western Mediterranean Spanish coast (Fig. 1; 41°40,19' N 2°47,11' E), an open, oligotrophic coastal area (cf. Duarte et al., 1999, Lucea et al., 2003). The Bay of Blanes receives terrestrial inputs from the Tordera River as well as urban runoff from the town of Blanes, which receives a high number of tourists during summer. The sediments in the Bay are sandy, with an average organic content of 0.4 % of the dry weight and remain oxic to a depth of about 10 cm (Marbá and Duarte, 2001).

Subsurface water samples were collected weekly between 1996 and 1997 from a permanent station 1 km offshore, at a depth of 15 m, where the water column remains well mixed throughout the year (Lucea et al., 2003). Samples, collected on acid-washed polyethylene bottles were taken to the laboratory and processed within 30 min. from collection. Samples for dissolved nutrient analyses were immediately frozen. Samples for particulate organic carbon (POC), nitrogen (PON) and phosphorus (POP) analyses were filtered (about 2 L) through pre-combusted Whatman GF/C glass fiber filters and stored frozen for subsequent analyses. A variable water volume (50 to 250 ml, depending on phytoplankton biomass) was filtered through 0.45 µm Whatman GF/F filters for spectrofluorometric analysis of chlorophyll <u>a</u> concentration (Parsons et al, 1984). The filters were homogenised and kept refrigerated for ca. 6 h in the dark while pigments were extracted in 90% acetone. Fluorescence was measured, following extraction, in a Turner Designs fluorometer calibrated with pure chlorophyll a (Sigma Co.)

The downward particle flux was measured at monthly intervals between October 1996 and November 1997 using sediment traps. The sediment traps consisted of 6 cylindrical PVC tubes (cross-sectional exposed area = 31.81 cm^2) with an aspect ratio of 7.8 to prevent resuspension losses (Hargrave and Burns, 1979, Blomquist and Hakanson, 1981). The array of traps were mounted on a 1 m diameter stainless steel circular frame inserted into the sediments so as to raise the mouth of the traps 1.5 over

the sediment surface. This placed the traps, above the depth where sediment is resuspended during storms (Gacia et al. 1999, 2002). The traps were deployed by SCUBA divers for a total period of seven days, and capped before returning them to the surface. At the laboratory the content of the tubes was filtered through 25 mm pre-combusted GF/F filters, which were subsequently dried for 24 h at 60° C for POC, PON, POP and biogenic silica (bioSii) determinations. Each month SCUBA divers collected 10 plexiglass corers ($\varnothing$ = 4.3 cm; H = 33 cm) containing 8 cm of sediments, which were brought to the laboratory to estimate sediment-water fluxes. Flux measurements were conducted by incubating the cores at in situ temperature for 24 h, with 5 cores exposed to light (200 μmol photon m^{-2} s^{-1}, the average incident irradiance on the sediments at the sampling station), while the rest were kept in the dark. Dissolved inorganic PO_4^{3-}, NO_3^- $Si(OH)_4$ and O_2 fluxes were determined from the changes in concentrations from the beginning to the end of the incubation period.

Samples for nutrient analysis were thawed and PO_4^{3-} and $Si(OH)_4$ concentrations were measured spectrophotometrically using a 10-cm cuvette cell (Koroleff, 1976; Grasshoff, 1983). $NO_3^- + NO_2^-$ concentrations were determined by the colorimetric methods of Grasshoff (1983). The detection limit of dissolved nutrients concentrations were 0.01 μmol L^{-1} for spectrophotometric determinations of PO_4^{3-} and $Si(OH)_4$, and 0.02 μmol L^{-1} for $NO_3^- + NO_2^-$ analyses. Filters for POC and PON analyses were exposed to concentrated hydrochloric acid fumes for 30 min to remove any inorganic carbon, which may interfere with the analysis. Measurements were carried out using a Perkin-Elmer 240 CHN analyzer. Samples for POP determination were oxidized in acidic persulphate solution and then analyzed as soluble reactive phosphorus following the methods outlined in Murphy and Riley (1962) and Solórzano and Sharp (1980).

The samples for δ^{13} C-POC analysis were filtered through pre-combusted (3 hours at 500 °C) GF/F filters and frozen. Filters for δ^{13} C-POC analyses were fumed with concentrated hydrochloric acid overnight to remove calcium carbonate, dried and loaded into small pre-combusted quartz tube with Cu, CuO and Ag foil and subsequently placed inside a larger quartz tube, which was evacuated and sealed. The

samples were combusted at 900ºC for 3 hours, the resulting gases were distilled cryogenically in a vacuum line and the separated CO2 was analysed on a VG SIRA II mass spectrometer. Precision δ^{13}C-POC of an internal laboratory standard was 1 SD = 0.06 ‰. The carbon isotope ratios are reported in per mil (‰) relative to PDB standard:

$$\delta^{13}C = [(R_{sample} / R_{standard}) - 1] \times 1000$$

where R = ^{13}C:^{12}C ratio.

Dissolved organic nitrogen (DON) and phosphorus (DOP) concentrations were estimated as the difference between total dissolved nutrient pool and the dissolved inorganic plus particulate organic nutrient pools. Biogenic silica samples were filtered through a 47 mm polycarbonate Nuclepore filter (0.6µm pore size) and dried 12 hours at 60°C. The determination of biogenic silica followed the NaOH / HF digestion method (Ragueneau and Tréguer, 1994).

Samples for DOC analyses were filtered through precombusted (400 °C for 2 h) GF/F filters, and kept frozen until analysis. DOC was determined on 2 ml samples, after acidification with 10µl 85% H_3PO_4 and sparging with N_2 for 5 minutes, by high temperature oxidation using an MQ1001 TOC Analyzer (Qian and Mopper 1996).

Dissolved inorganic carbon (DIC) was determined by a potentiometric method using a Mettler DL21 titration device. A know amount of seawater was placed into a conic flask where it was titrated using an automatic titrator with a solution of Hydrochloridric acid (0.1N) at in situ temperature. The acid was prepared with an accuracy of 0.0002 mol L^{-1} to which sodium dichloride and sodium sulfate were added to approximate the solution to the ionic strength of sea water, so as to maintain the activity coefficients constant during the titration (Grasshof, 1983). The volume dispensed by the burette (ml) and the signal (mv) were recorded automatically to calculate the first derivative (mv/ml) which represents the biggest gradient in the flat

part of the titration curve. The reagent consumption for the CO_3^- (V_1) and 2 CO_3^- + HCO_3^- (V_2) titration points was recorded. Then DIC was calculated as :

$$\Sigma\, CO_2 = \ (V_2 - V_1) \cdot [HCl] / (ml\ of\ sample)$$

Planktonic community metabolism was estimated weekly along the two-year study. Water samples were carefully siphoned into fifteen 125 ml narrow-mouthed Winkler bottles. Five of the bottles were immediately processed to measure the initial oxygen content present in the samples, five transparent ones were incubated for 24 hr in the light (200 µmol photon $m^{-2}\ s^{-1}$) and the remaining five were incubated for 24 hr in the dark at *in situ* temperature. Dissolved oxygen concentration was measured using high-precision Winkler titration after Carrit and Carpenter (1966), using a Metrohm-682 Autotritator for the potentiometric (redox electrode) end-point detection (Oudot et al., 1988). The average coefficient of variation of the dissolved oxygen concentration was about 0.35 % and the resulting detection limit for net production and respiration was about 0.0009 mg $O_2\ L^{-1}\ h^{-1}$. Oxygen evolution rates were converted into carbon incorporation assuming a photosynthetic quotient of 1, and were converted to daily values using the observed corresponding photoperiod. Respiration rates (R) were determined from the oxygen change in the dark bottles, net community production (NCP) was determined from the oxygen change in the clear bottles, corrected for photoperiod, and gross primary production (GPP) was calculated as the sum of R and P. The same procedure was followed for determining benthic community metabolism from core incubations.
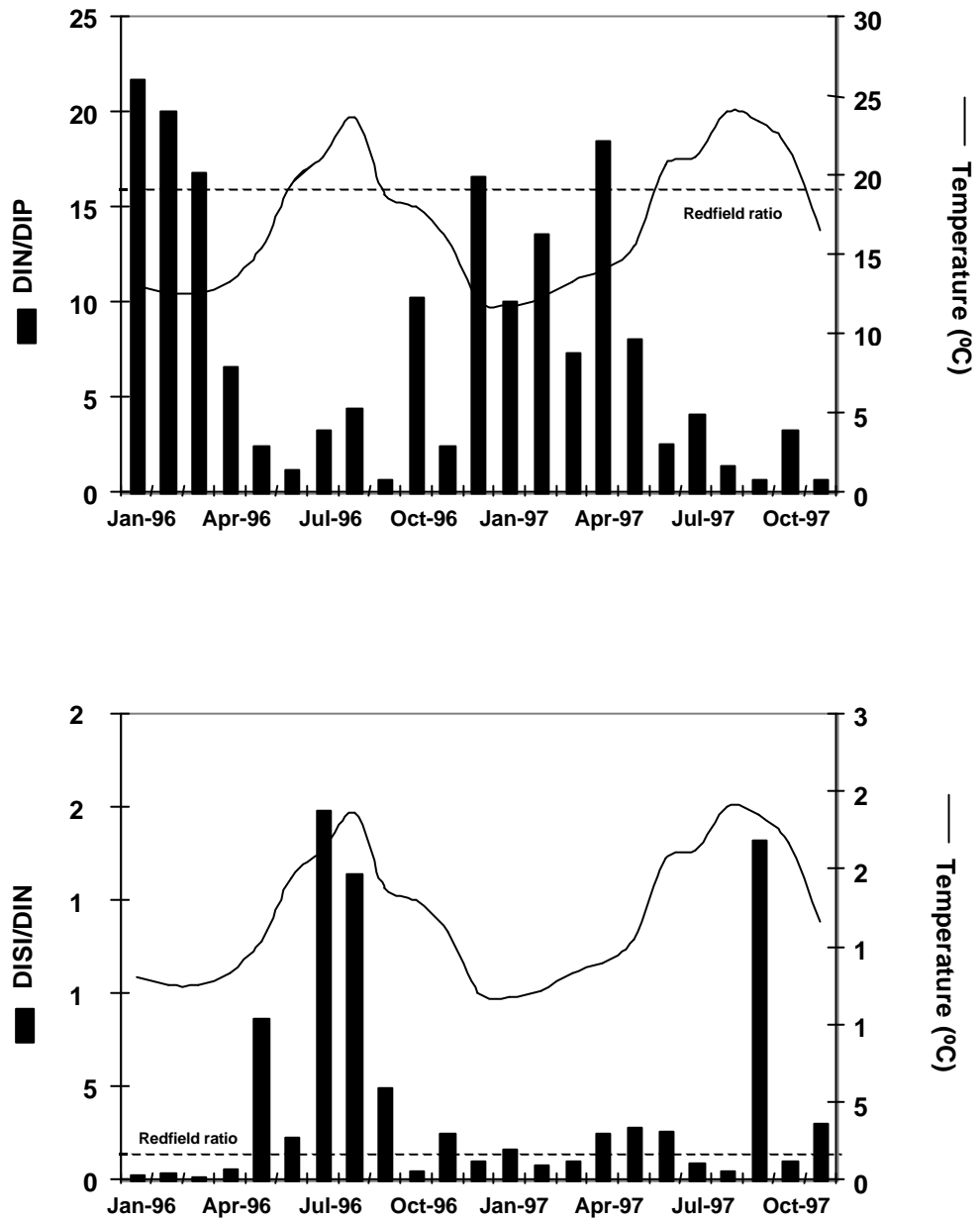
Fig.4. Temporal variation of of mean monthly of dissolved inorganic nutrient ratios in the Blanes Bay. The solid lines indicate the Redfield N/P and Si/N ratios.
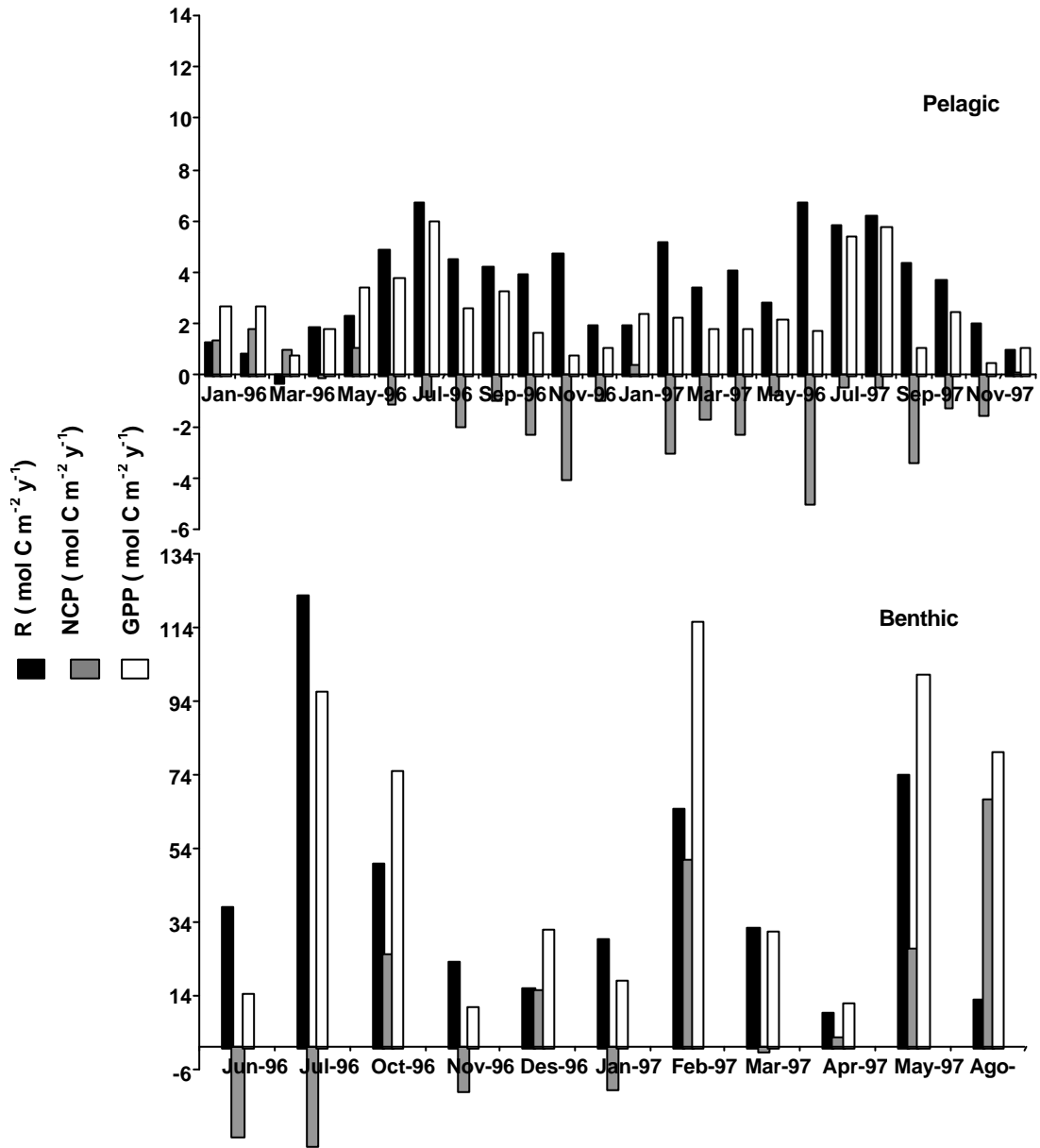
Fig.5. Mean monthly respiration (R), gross primary production (GPP) and net community production (NCP) for the pelagic and benthic compartments at the Bay of Blanes.

4.4. Results.

Seawater temperature varied from a mean winter minimum of 11.1 °C to a mean summer maximum of 24.4 °C (Fig. 2a). CTD profiles showed that the water column at the sampling station remained well mixed in this exposed location throughout the study, as the summer thermocline was established at about 40 m depth. The chlorophyll a concentration showed winter maxima in the water column (Fig. 2b), corresponding to the main algal bloom in these waters (Duarte et al. 1999). The sediments, which remained well-illuminated due to the high transparency of these waters (Duarte et al. 1999), throughout the study, also supported an important microphytobenthic community, with chlorophyll values half, on average, those of the water column, but that were comparable or even exceeded those in the water column during spring and summer (Fig. 2b).

Dissolved inorganic nutrients varied greatly during the study, with high (up to 2 µM) nitrate concentrations during winter and low values (< 0.5 µM) during summer (Fig. 2c). Silicate concentrations remained low (< 1.0 µM) throughout 1997, but reached high concentrations (up to 2 µM) values during winter and summer 1996 (Fig. 2d). Phosphate concentrations remained low (< 1 µM) throughout the study (Fig. 2e). The average dissolved inorganic nitrogen concentration was similar to that of silicate at 0.6 µM, while phosphate concentration averaged 0.13 µM the study period (Table 1). The ratio between silicate and dissolved inorganic nitrogen also showed a clear seasonality, with generally low values (< 4), except for sporadic high values (up to 40) during summer, when the nitrate pool is depleted. These patterns resulted in consistent changes in the inorganic nutrient ratios, with the N: P ratio being close to the Redfield ratio of 16 during the winter period and much lower (< 5) between May and August (Fig. 4).

The elemental concentrations in the particulate pool lower during winter, and reached the maximum values during spring and late summer, with biogenic silicon concentrations showing a clear peak in early winter (Fig. 3). The average concentration

of bioSi was similar to that of PON (Table 1), but the particulate material was relatively enriched in N relative to C and P, having a lower C/N and higher P/N than the dissolved inorganic pool (Table 1). DOC concentrations ranged from a minimum of 67 μM in winter to a maximum value of 160 μM observed in spring and late summer. DOC values were comparable to open ocean concentrations. DON and DOP concentrations showed a similar pattern, with the average DON and DOP concentrations being comparable to those of PON and POP (Table 1). In contrast, carbon was dominated by the inorganic pool, which comprised, on average 95 % of the carbon present in the water column (Table 1), phosphorus showed a balanced partitioning between the three different pools (Table 1), and nitrogen was dominated by the dissolved organic pool, which comprised about half of the total nitrogen present in the water column (Table 1). The stable carbon isotope value of the POC material was –26.02 ‰ (Table 1), similar to values of Mediterranean land vegetation (Dauby, 1989), suggesting an important contribution of land-derived material.

The benthic community was autotrophic (P > R) in all but two of the experiments conducted. Gross benthic primary production rates were highest (up to 98 mmol C m^{-2} d^{-1}) in summer and low in winter, except for a highly productive event recorded in February 1997 (Fig. 5). Respiration rates were also highest in summer with the P/R ratio averaging 1.5 ± 0.5, yielding an annual gross primary production of 30.06 ± 11.86 mol C m^{-2} y^{-1}, a respiration rate of 23.83 ± 9.21 mol C m^{-2} y^{-1} and a net benthic community production of 6.23 ± 4.08 mol C m^{-2} y^{-1}. In contrast, the pelagic compartment was heterotrophic, with a seasonal pattern characterized by high respiration and gross production rates in summer and lower values in winter (Fig. 5). Planktonic respiration exceeded gross primary production in 40 % of the 24 experiments conducted during the measurement period (one per week), with an average P/R ratio of 0.64 ± 0.07. The annual pelagic gross primary production (12.90 ± 1.85 mol C m^{-2} y^{-1}) was three-fold lower than the benthic gross primary production, the annual pelagic respiration rate was 21.57 ± 2.06 mol C m^{-2} y^{-1} and the net planktonic community production was calculated at –8.67 ± 1.79 mol C m^{-2} y^{-1}, indicative of an heterotrophic

planktonic compartment. Overall, the system was in metabolic balance, as the integrated system gross primary production (pelagic + benthic) of 42.9 $\pm$ 5.20 mol C m^{-2} y^{-1} was in close metabolic balance with the integrated respiration rates of 45.4 $\pm$ 4.75 mol C m^{-2} y^{-1}.

In addition to the organic inputs derived from its significant gross production, the benthic compartment received an important sedimentary input of organic matter. The bulk sedimentation was relatively low from the beginning of February to mid-September (Fig 6), averaging 14 g DW m^{-2} d^{-1}, but increased significantly in October, reaching a maximum in November of 96 g DW m^{-2} d^{-1} with a local maxima of 33 g DW m^{-2} d^{-1} in April. These peaks coincided with periods of intense rain and discharge from the Tordera River to the Bay of Blanes. The corresponding POC flux ranged one order of magnitude from 0.16 mmol C m^{-2} d^{-1} to a maximum of 60 mmol m^{-2} d^{-1}, and an average flux of 27.3 mmol C m^{-2} d^{-1} (Fig. 6). The depositional fluxes of PON, POP and bioSii were temporally variable (Fig. 6), and showed a stoichiometry indicative of a sedimentary flux relatively enriched in organic carbon and, particularly bioSi, and depleted in nitrogen and phosphorus relative to the sestonic pool (Tables 1 and 2). The stable carbon isotope values in the material collected in the sediment trap (Table 2) were similar to that in the seston, and tended to be lighter, resembling that of land-derived carbon (-26.02 $\pm$ 0.02 ‰), during periods of high sedimentary flux, which corresponded with high river discharge.

Benthic remineralization processes released nutrients to the water column, despite the net autotrophic metabolism of the sediment community. The sediment nutrient efflux varied by two-orders of magnitude along the study for silicate, phosphate and ammonium (Fig. 7). The sediment efflux of silicate and phosphate followed similar patterns, with high release rates following the collapse of the winter phytoplankton bloom (Figs. 2 and 7). The ammonia efflux showed a contrasting pattern (Fig. 7), and an annual release rate comparable, on average, to that of silicate (Table 2). The annual release rates of phosphate and silicate closely matched (t-test, $P < 0.05$) those of the depositional fluxes (Tables 2), but the release rate of nitrogen was only 0.6 % of the

nitrogen deposition, showing a high retention of nitrogen in the benthic compartment or nitrogen loss through denitrification. Similarly, the benthic respiration rate was only 34 % of the organic carbon deposition rate (Tables 2), suggesting an important carbon burial in the sediments.
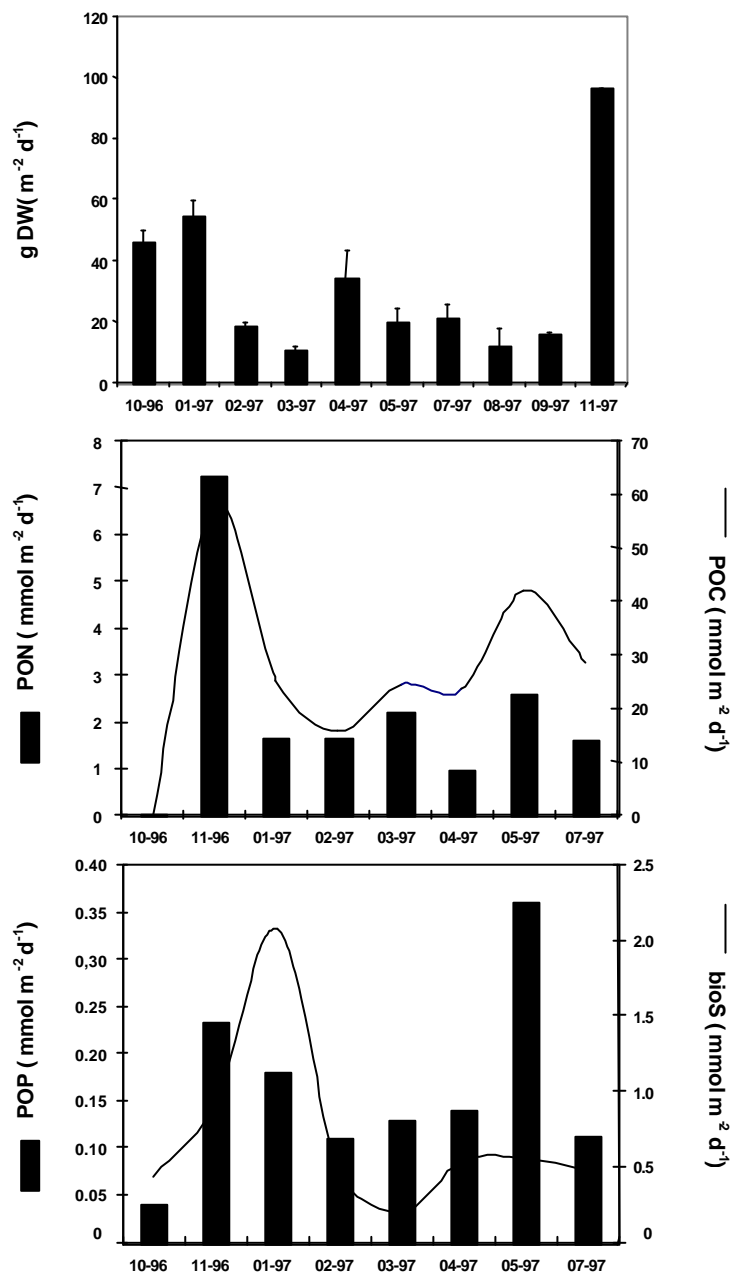
Fig.6. Total depositional fluxes of material (as dry weight DW), POC, PON, POP and bioS at the Bay of Blanes.
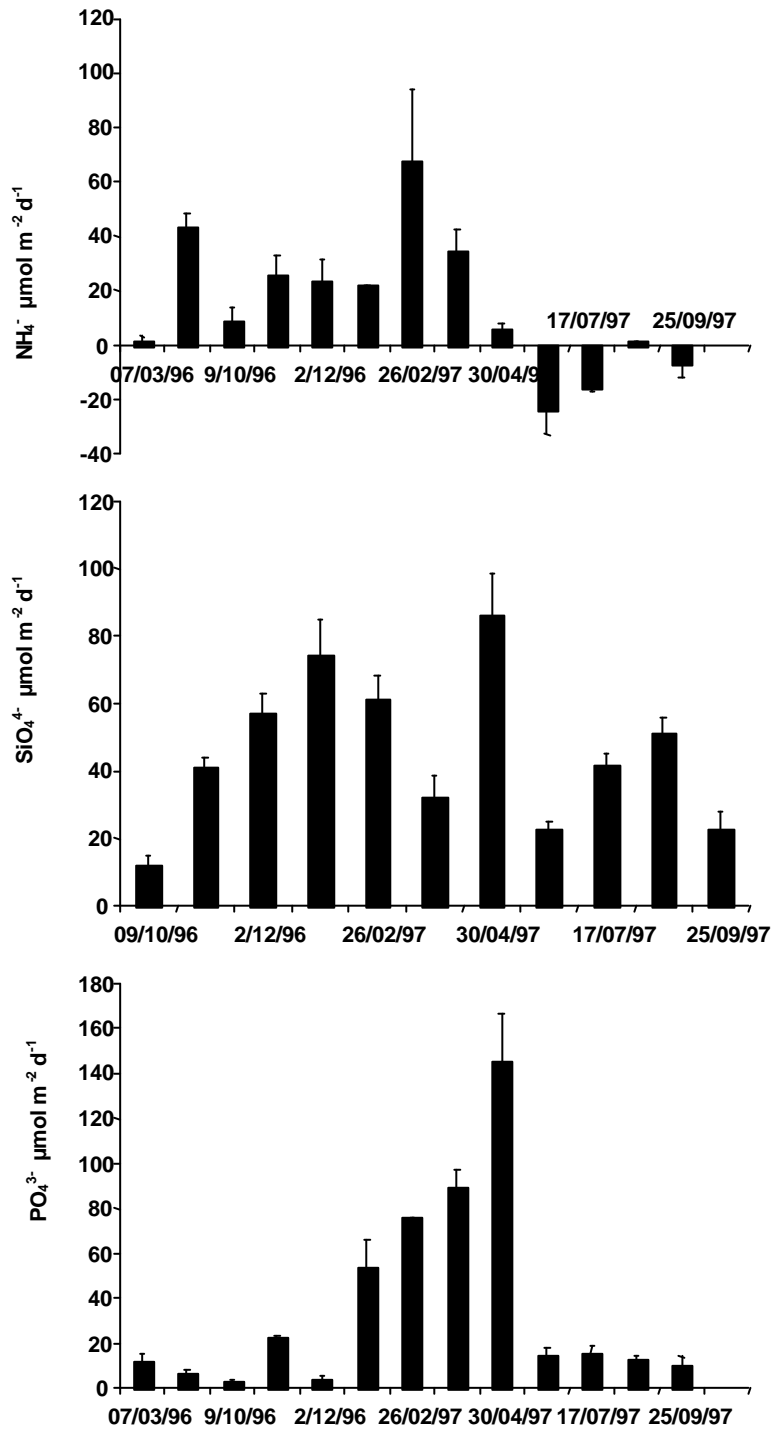
Fig. 7. Mean monthly sediment-water dissolved inorganic nutrient fluxes at the Bay of Blanes. Negative values denote uptake by the sediment compartment.

4.5. Discussion.

The average elemental ratios in the total particulate and dissolved organic pools are indicative of acute phosphorus depletion in the Bay of Blanes (see Table 1), a general feature of the biogenic layer in the Mediterranean sea (Berland et al., 1980; Minas et al., 1988; Krom et al., 1999; Lucea et al., 2003), although this is not reflected in the DIN:DIP ratios, which are relatively low. This situation varied seasonally, with no evidence of such deficiency in winter (see Fig. 2) when water mass circulation, influenced by the inflow of cold and low-salinity waters advected from continental slope waters off the Blanes canyon (Rojas et al., 1995; Masó and Tintoré, 1991; Granata et al. 1999), entrains deeper waters into the littoral zone yielding DIN:DIP ratios higher than the average value of 7. Relatively high amounts of silica delivered by episodic land inputs enhance the relative phosphorus depletion, showing an important role of events of high rainfall and river discharge in determining the nutrient partitioning within the water column.

In addition to the important role of the exchanges and inputs with land and the ocean, the local stoichiometric C, N, P and Si ratios in the Blanes Bay are also dependent on local ecosystem processes. Nutrient assimilation by phytoplankton is responsible for the decline of nutrients from winter to summer. On the other hand, organic matter release by phytoplankton cell lysis (Agustí and Duarte, 2000), would yields an increase in dissolved organic carbon from spring to summer and a greater importance of dissolved organic over inorganic nutrients in the water column during summer. The elemental ratios in the dissolved organic pool (DOC: DON = 35 and DOC: DOP = 875) are indicative of a faster recycling rates for nitrogen and, particularly phosphorus, than for carbon. The DOC: DON: DOP ratio in the Bay of Blanes was somehow lower than the average ratio reported for the biogenic layers of the NW Mediterranean (DOC: DON: DOP = 1984:66:1; Lucea et al. 2003) which suggests a more labile pool of DOM and consequently more oxidative activity in the waters of the Bay of Blanes than in the open ocean.

Indeed, the pelagic component in the Bay of Blanes was found to be heterotophic on an annual balance (Fig. 8), as reported for this system in the past (Satta et al. 1996). Planktonic respiration rates were particularly high in the summer (see Fig. 4), also consistent with previous results (Satta et al. 1996), suggesting an important recycling of nutrients within the water column. The faster recycling of nutrients relative to carbon in the organic matter pool is also reflected in the differences in the average elemental ratios between the sestonic material, with POC: PON ratios similar or below the Redfield value of 6.6 (cf. Table 1) in summer, when nitrate is depleted, compared to the average POC: PON ratio of the sedimenting material of 12 (see Table 2). This situation coincides with the seasonal minimum in sedimentation values (Fig. 6).

Indeed, the finding of a significant sedimentary flux despite the observation of an heterotrophic planktonic component implies an important allochthonous input of materials to support this sedimentary flux. The estimated annual organic carbon deposition at Blanes Bay (Table 2) was comparable to carbon sedimentation rates reported for the continental shelf margin of the Northwestern Mediterranean (15.2 mol C m^{-2} yr^{-1}, Buscail et. al., 1990; 30.4 mol C m^{-2} yr^{-1}, Monaco et al,. 1990). The likely source of organic matter is the adjacent terrestrial compartment through the inputs by the Tordera River. This suggestion was tested through the evaluation of the sources of organic carbon as derived from the stable isotope signatures. The average isotopic composition of organic carbon in the sedimentary flux deposition was more negative (-23.74 ± 0.92 %o $^{13}\delta$ C) than those found in sediment trap studies in the Western Mediterranean basin (Dauby et al., 1995) and than those characteristic of Mediterranean phytoplankton (Dauby 1989; Fontugne et al., 1981 and Faganeli et al., 1994). These comparisons suggested an important terrestrial component in the sedimentary flux. We calculated the relative contribution of planktonic material and terrestrial-derived material (see Table 1) to the settling carbon flux (see Tables 2) using the equation:

$$\delta^{13}C_{\text{sediment trap}} = \delta^{13}C_{\text{terrestrial}} \cdot f + \delta^{13}C_{\text{plankton}} (1-f)$$

where f is the fraction derived from terrestrial land-derived material., and $\delta^{13}C_{plankton}$ is the stable carbon isotope signature of Mediterranean phytoplankton, taken to be –22.4 ‰ (Dauby, 1989).   These calculations indicated that terrestrial materials contributed 37 % of the total sedimentary flux, also accounting for the highest carbon: nutrient ratios in the trapped material (Table 2) relative to those in the water column (Table1). These calculations support our conclusion that the net heterotrophic nature of the planktonic compartment of the Bay of Blanes is, therefore, driven by land inputs.

The total organic carbon inputs to the sediment (sedimentation rate + gross community production) was estimated at 39.9 mol C m^{-2} y^{-1} while respiratory losses in sediments removed 23.8 mol C m^{-2} y^{-1} resulting in a net accumulation (and export) of 16.1 mol C m^{-2} y^{-1} in the sediments at the Bay of Blanes (Fig. 7). Whereas much of the organic carbon deficit of the pelagic compartment in the Bay of Blanes may be met by transference of the excess production of the benthic community, the net accumulation of organic carbon in the sediments requires inputs from land. Because some of the land inputs of organic carbon may be exported out of the Bay of Blanes, these are estimated at between a minimum of 7.5 mol C m^{-2} yr^{-1} and 18.6 mol C m^{-2} yr^{-1} if the respiratory surplus of the pelagic compartment is fueled by allochthonous inputs.

Although our results reveal important storage rates of organic carbon in the sediments at the Bay of Blanes, these still act, at the annual time scale, as nutrient sources, releasing as much Si and P as that delivered to the sediments by sedimentation processes. In contrast, however, the release rates of N, as ammonia, represented a minor fraction (0.6 %) of the depositional flux, indicative of high retention of nitrogen or, alternatively, denitrification losses. The N retention in the sediments could be expected to be conducive to N deficiency in the dissolved inorganic water pool, consistent with observations, and, therefore, a dependence on external inputs, through riverine inputs, to which N supplied by mixing and entrainment of deeper off-shore waters adds in winter. The dissolved and particulate organic pools were, in contrast, deficient in P, indicating a rapid recycling of P from organic matter, consistent with

observations in other oligotrophic ecosystems (Vidal et al., 1999 b, Cañellas et al., 2000).

In conclusion, our study reveals that the Bay of Blanes remains oligotrophic despite substantial inputs of organic carbon and, therefore, nutrients from land. The reason for the lack of eutrophication symptoms is twofold, (1) the high dilution rate of Blanes Bay waters with off-shore waters resulting from the dynamics imposed by the adjacent submarine canyon (Rojas et al., 1995; Masó and Tintoré, 1991; Granata et al., 1999), and (2) the heterotrophic nature of the planktonic community which prevents the accumulation of organic matter in the system. In contrast, the benthic compartment is autotrophic and receives important sedimentary inputs of organic carbon, with an important contribution of land-derived carbon. Although Si and P are remineralised and released back to the water column, C and N are retained, leading to a significant rate of organic C storage in the sediments and a deficiency of N in the dissolved nutrient pool in the overlying water column. These results highlight the important coupling between the benthic and water column compartments in determining the metabolism and biogeochemical behavior of oligotrophic littoral ecosystems.
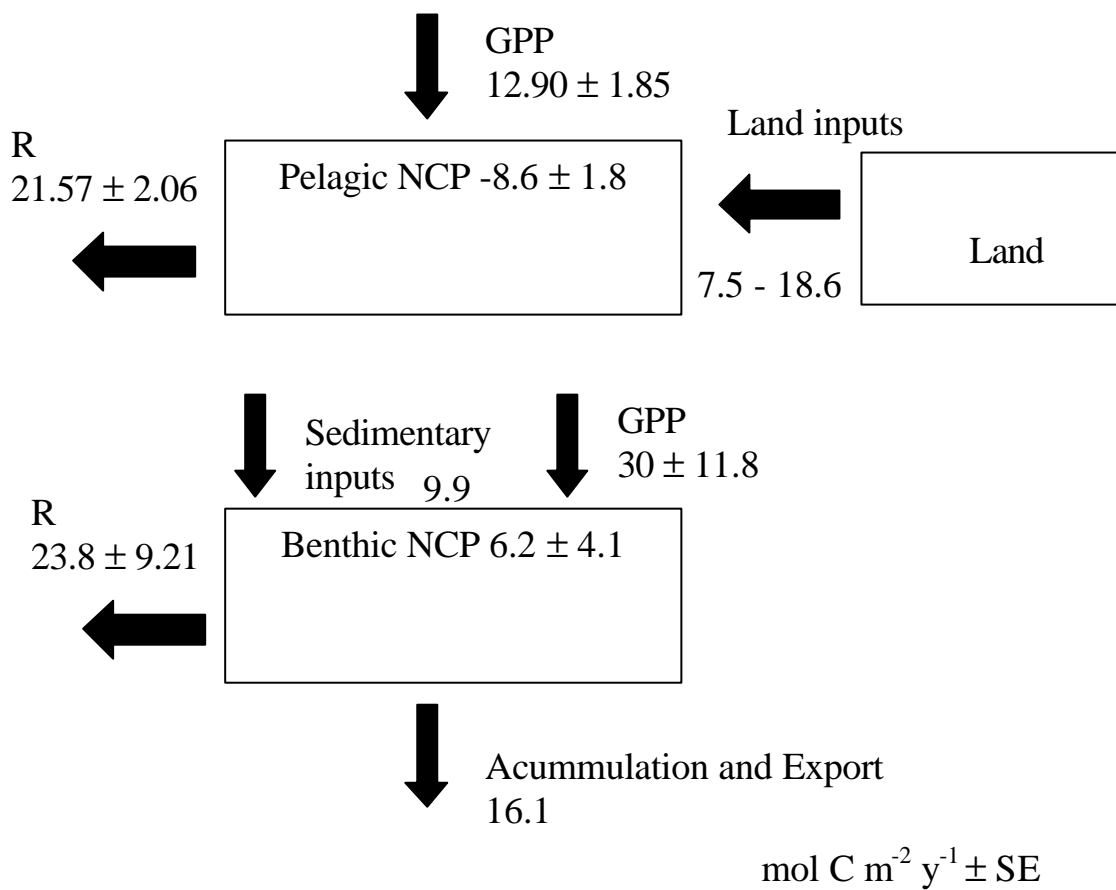
Fig. 8. A summary depiction of the carbon fluxes in the Blanes Bay (NW Mediterranean).

Table 1. Average (μmol l^{-1} ± SE) nutrient concentrations and average ratios for particulate organic matter, dissolved inorganic and dissolved organic matter pools in the Bay of Blanes (1996-1997). Isotopic composition values are expressed in ‰.

Element	DIM	DOM	POM	Elemental ratios	Average ratios	Pool ratios	Average ratios
C	2409.8 ± 41.5	91.94 ± 5.7	11.17± 0.75	DIN/DIP	7	DIC:DOC	26
N	0.64± 0.09	2.59 ± 0.4	2.12 ± 0.18	DISi/DIN	4	DIN:DON	0.25
P	0.13± 0.02	0.1± 0.02	0.12 ± 0.01	DOC/DON	35	DIP:DOP	1.3
Si	0.61± 0.08			DON/DOP	24		
bioS			1.89 ± 0.78	POC/PON	6		
				PON/POP	22		
				DISi:DIN:DIP	14:7:1	DIC:DOC:POC	219:8:1
				DOC:DON:DOP	857:24:1	DIN:DON:PON	0.32:1.3:1
				POC: bioS:PON:POP	124:31:22:1	DIP:DOP:POP	1:0.8:1
$\delta^{13}C$ (land derived material)			-26.02 ± 0.02				

Table 2. Fluxes of sedimentary material collected with sediment trap in the Bay of Blanes (1996). Isotopic composition values are expressed in ‰. Average nutrient fluxes of sedimentary record in Blanes Bay (1996).

	Sediment trap fluxes Average ± SE	Sedimentary flux ratio	Sediment trap $\delta^{13}C$ Average ± SE
POC (mol m^{-2} yr^{-1})	9.96 ± 2.4		-23.74 ± 0.92
PON (mol m^{-2} yr^{-1})	0.81 ± 0.30		
POP (mol m^{-2} yr^{-1})	0.06 ± 0.01		
bioS (mol m^{-2} yr^{-1})	0.19 ± 0.07		
DW (gr m^{-2} yr^{-1})	9349.22 ± 2823		
POC: bioS :PON:POP		159:50:12:1	
POC:PON		12	

Sediment fluxes (mmol m^{-2} yr^{-1})	Average ± SE
PO_4^{3-}	13.05 ± 4.81
SiO_4^{4-}	16.61 ± 2.42
NH_4^+	5.19 ± 2.68

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The effect of nutrient additions on the partitioning of nutrients in an experimental coastal Mediterranean system

By

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5.1. Abstract.

The hypothesis that the importance of dissolved organic matter (DOM) as a reservoir of C, N, and P declines, relative to that of the particulate pool, with increasing nutrient inputs was tested using mesocosms exposed to a gradient of nutrient inputs in the Spanish Mediterranean. The nutrient additions included a treatment equivalent to the loading in the coastal ecosystem studied (5 mmol N m^2 d^{-1}), and mesocosms receiving half , 2-, 4-, 8-, and 16-fold this value, as well as a mesocosm to which no nutrients were added. Nutrients were added at ratios of 20 N (as ammonium) : 7 Si : 1 P. The initial concentration of dissolved inorganic nutrients was very low (dissolved inorganic nitrogen < 0.05 µM, phosphate < 0.01) and comprised, together with the particulate pool < 25 % of the total N and P in the system, with the bulk N and P in the system present as DOM (> 75 %). Particulate and dissolved organic matter was depleted in N (C/N ratio > 15) and, particularly, P (C/P ratio > 1000), indicative of a strongly nutrient, particularly phosphorus, deficient ecosystem. Experimental nutrient additions lead to a parabolic change in C/N and C/P ratios in the dissolved organic matter with increasing nutrient inputs, which approached the Redfield stoichiometry at nutrient inputs > 8 fold above the ambient loading. The relative size of the dissolved inorganic nutrient pools (about 20 % of the N and P) did not vary, but there was a tendency towards an increase in the relative size of the particulate pool at the expense of a decrease in the relative importance of DOM as a reservoir of N, P and C, with increasing nutrient inputs. The production of nutrient-depleted organic matter at low nutrient inputs likely prevents efficient recycling, leading to the dominance of nutrients in DOM in the system.

5.2. Introduction.

Dissolved organic matter (DOM) is a major reservoir of nutrients and carbon in the sea (Druffel et al. 1992, Hedges 1992), particularly in unproductive waters, where the pools of dissolved organic carbon (DOC) and nutrients are dominant (e.g. Hedges 1992, Wheeler et al. 1997, Doval et al. 1999, Vidal et al. 1999). The greater importance of DOM in unproductive compared to productive systems is paradoxical, for it suggests that the plankton communities in nutrient-poor environments should be less efficient in conserving the nutrients incorporated by phytoplankton than those in nutrient-rich waters. Yet, the apparent tendency towards a dominance of DOM as a nutrient reservoir in oligotrohic systems relies on scattered information from the relatively few reports that include simultaneous data on dissolved and particulate nutrient pools. Moreover, this pattern may reflect a covariation between nutrient availability and other traits influencing the biota (e.g. temperature, cf. Li, 1998, Agawin et al. 2000) rather than responses to a gradient of nutrient supply across an oligotrophic to eutrophic gradient. There is, therefore, a need to test the changes in the partitioning of nutrients among different pools (inorganic and dissolved and particulate organic) in response to a gradient of nutrient supply.

We here test the hypothesis that the importance of dissolved organic matter as a reservoir of C, N, and P declines, relative to that of the particulate pool, with increasing nutrient inputs. We do so on the basis of the examination of the changes in the concentration of nutrients (nitrogen and phosphorus) and carbon in different pools in response to experimental nutrient additions to experimental mesocosms moored in the Spanish Mediterranean (Blanes Bay, NE Spain).

5.3. Methods.

The experiment was conducted between June 18 and July 8, 1997 near the town of Blanes (NE Spain). The experimental design involved a gradient of nutrient additions to mesocosms following a geometric series of nutrient inputs centered at the nutrient loading calculated for the Bay of Blanes. The background loading is estimated to be about 5 mmol N m^{-2} d^{-1} in summer (Duarte et al. 2000a), and is hereafter referred to as a "business as usual" control. Phosphorus and silicon were added to maintain their average summer stoichiometry with nitrogen in the sedimentary flux (20 N : 7 Si : 1 P, Duarte et al. 2000a). Nitrogen was added as ammonium, the dominant form of summer nitrogen inputs to the Blanes Bay (Duarte et al. 2000a).

We used a series of 7 large (nominal and effective volume 50 and 33 m^3, respectively) mesocosms, consisting of 14 m high bags and a 4.2 m^2 cross-sectional area. One of the units (that receiving twice the "business as usual" nutrient loading), was found to be damaged four days prior to the end of the experiment. Nutrients, as solutions of $ClNH_4$, KH_2PO_4, and Na_2SiF_6, were added to the mesocosms in alternate days, following the collection of samples. The nutrient additions included a treatment equivalent to the "business as usual" control, a treatment equivalent to half of that nutrient input, and enriched nutrient additions equivalent to 2-, 4-, 8-, and 16- fold the "business as usual" control. In addition, we also examined the changes in nutrient pools in a mesocosm to which no nutrients were added. The nutrient input to this mesocosm unit, derived from wet and dry atmospheric deposition, was determined from short-term nutrient mass balances to be about 0.005 μM N d^{-1} (Duarte et al. 2000a). Additional details on the experimental design are reported by Duarte et al. (2000a).

Integrated water samples (0 - 13 m) to determine nutrient concentrations and phytoplankton biomass, as chlorophyll a concentration, were collected in alternate days at 7:00 am. Briefly, a washed and pre-rinsed hose (internal diameter 8 cm) with a ballasted end connected to the surface by a piece of rope was slowly deployed down to 13 m depth, and the ballasted end subsequently retrieved to the surface while maintaining the opposite end higher up as to prevent backflow, delivering the contents to an acid-washed 30 L carboy. The first sampling event occurred prior to the first nutrient addition, as to allow identification of any possible initial difference not attributable to the treatment. The water samples were transported to the laboratory for analysis within 30 min. At four days interval, the sedimentary material accumulated in the cone-shaped bottom was pumped out of the mesocosm, using a hand pump connected to the bottom of the mesocosm by a hose.

The concentrations of dissolved inorganic phosphorus (DIP), ammonium and silicate were measured spectrophotometrically following standard methods (Hansen & Koroleff 1999), using a 10-cm cuvette cell when necessary to increase the detection limit. Samples for dissolved organic N and P determinations were immediately filtered through previously rinsed, 0.2 µm cellulose ester filters mounted on polyvinyl chloride Millex units (Millipore). Controls with distilled water showed that the filtration step yielded no measurable contamination of the samples. Filtered samples were recovered in polycarbonate bottles, which had been cleaned by filling them with distilled water and running the oxidation procedure several times, soaking in diluted HCl and rinsing with distilled water. Independent samples were analyzed for total dissolved nitrogen (TDN) and phosphorus (TDP). TDN and TDP concentrations were determined after oxidation at 120 $^{\circ}$C for 30 min of the filtrate of 20 ml subsamples in alkaline and acidic persulfate, respectively, and subsequent analyses of dissolved nitrate and phosphate (Hansen & Koroleff

1999). EDTA standards revealed a recovery of DON of 97 %. The analytical precision, estimated as the standard deviation of replicated samples, was 0.26 and 0.014 µM for TDN and TDP, respectively. DON and DOP were calculated as the difference between total and inorganic N and P. The detection limits of dissolved nutrient concentrations were 0.005, 0.05, 0.01 µM for $NO_3^- + NO_2^-$, NH_4^+, and PO_4^{3-}, respectively.

Samples (1 to 4 L, depending on seston concentration) for particulate organic carbon (POC) nitrogen (PON) and phosphorus (POP) determinations were filtered through precombusted (450 ^{o} C, 2 h) GF/F filters, kept frozen until analysis. POC and PON were analysed in a Carlo-Erba analyser after thawing the filters in an atmosphere of HCl fumes to remove carbonate and then drying at 60 ^{o} C. POP was determined following oxidation, at 120 ^{o} C for 1.5 h of the sample in acidic persulfate and subsequent analysis of the dissolved phosphate (Hansen and Korolef 1999).

Samples for analysis of dissolved organic carbon (DOC) were immediately filtered (burned Whatman GF/F filters) and 10 ml was collected directly in burned glass vials, added 100 µl 2M HCl, sealed and stored in the dark for later measurement. DOC was measured by Pt-catalyzed high temperature combustion in a Shimadzu TOC-5000 with autosampler injection and after sparging with the carrier gas for 6 minutes at 75 ml/min (Sharp et al. 1993). A 4-point calibration curve between 50 and 200 µM was prepared for each series of measurement. A mean blank area (n = 10) was subtracted before the slope of the calibration curve was used to calculate sample concentrations. Blanks were prepared with the water used for the standard solutions and evenly distributed within each series of measurements. Measurements of the DOC reference materials (blank and deep ocean water) prepared and supplied by Jonathan Sharp (personal communication, 1997) were included in each series. A statistically significant deviation (10% level,

t-test) in concentration of deep ocean water of more than 6 µM (n = 6) from the stated 44 µM led to a second analysis of the samples. Our blank samples (MQ-water) were on the average 3 ± 1 µM (SD, n = 25) higher than the reference blank, which is the reason to accept a deviation of 6 µM. In consequence the presented DOC concentrations might be slightly biased to the low side.

A variable water volume (50 to 500 ml, depending on phytoplankton biomass) was filtered through Whatman GF/F filters for fluorometric analysis of chlorophyll a concentration (Parsons et al. 1984). The filters were homogenised and kept refrigerated in the dark while pigments were extracted in 90% acetone for ca. 6 h. Fluorescence was measured in a Turner Designs fluorometer calibrated with pure chlorophyll a (Sigma Co.).
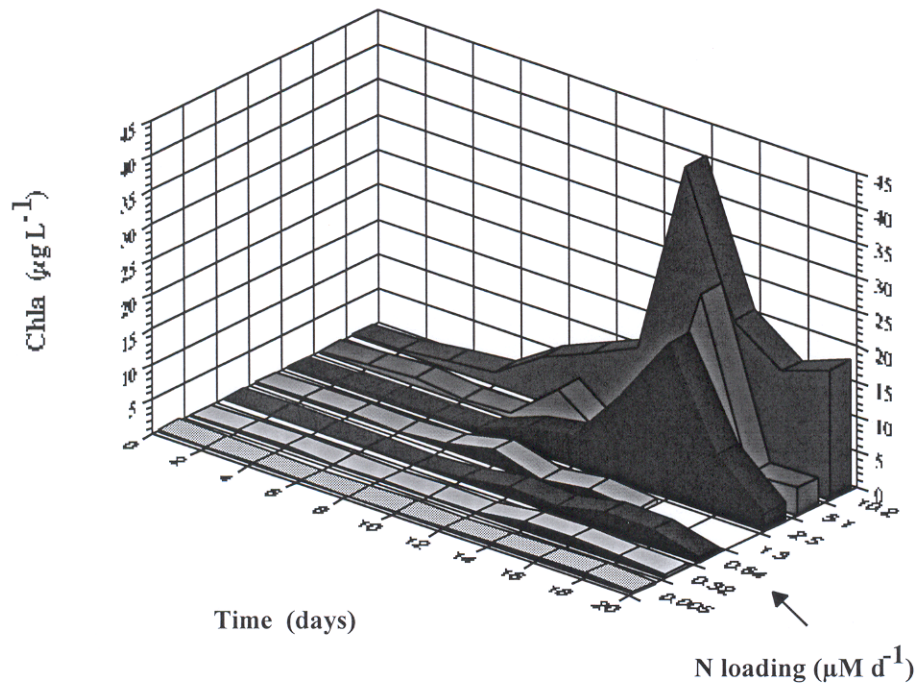
Fig. 1. The response of phytoplankton biomass, as the chlorophyll a concentration, in mesocosms enclosing receiving increasing nutrient inputs.

5.4. Results.

The biomass of the phytoplankton community increased with increasing nutrient inputs, following a time lag of about 4 to 12 days, reaching a maximum chlorophyll a concentration of 40.8 µg/L at the greatest nutrient inputs (Fig. 1). The initial concentration of dissolved inorganic nutrients was very low (dissolved inorganic nitrogen < 0.05 µM, phosphate = 0.01, Fig. 2), and among the lowest values recorded in a 7-year monitoring program in Blanes Bay (Duarte, unpubl. data). Initial particulate and dissolved organic nitrogen concentrations at 1.52 and 2.5 µM N, respectively, were substantially higher than inorganic N (; Figs. 3 and 4), and DON comprised 88 % of the total nitrogen. This was also the case for phosphorus, for which the initial POP and DOP concentrations (0.022 and 0.15 µM P) exceeded that of dissolved inorganic P by two and ten fold, respectively (Figs. 3 and 4). Hence, the dissolved inorganic and the particulate pools comprised < 10 % of the total N and P (4.5 % and 8.1 % respectively) and about 15 % of the total N and P (17.5 % and 14.2 % respectively) in the system at the onset of the experiment. About 78 % of the N and P in the system was, therefore, in DOM, and DOC also comprised most (84 %) of the organic carbon present in the system at the onset of the experiment.

The added nutrients were rapidly assimilated by the community, so that accumulation of the added dissolved inorganic nutrients was only observed after increasing nutrient inputs 4 (for ammonium and silicate) to 8 (for phosphate) times the "business as usual" value (Fig. 3). After an initial period (4 to 8 days), the communities present in the mesocosms built a biomass sufficient to lower the accumulated nutrient pools in the mesocosms receiving nutrient inputs elevated > 4 fold above the ambient levels. As a result, the dissolved inorganic nutrient concentrations were low (< 0.5 µM for ammonium, < 1 µM for dissolved inorganic

nitrogen, < 0.2 µM for phosphate) in all mesocosms by the end of the experiment (Fig. 2). DON concentrations increased from the initial concentrations of 2.5 µM shortly (4 days) after the initiation of nutrient inputs but decreased subsequently to reach levels below the initial ones (Fig. 4). DOP concentrations also showed a declining trend, following an initial increase at high nutrient dosage, along the experiment, except in the mesocosm receiving the highest nutrient dose, where DOP concentrations increased over the last week of the experiment to reach values four-fold greater than those encountered at the onset of the experiment (Fig. 4).

DIC comprised most of the carbon present in the mesocosm, but was reduced as nutrient inputs increased, indicating an important draw-down by photosynthetic uptake (Fig. 2). As a result, the POC pool increased greatly with increasing nutrient inputs (Fig. 3). Yet, DOC, which comprised about 6 times more carbon than that present in particulate organic form at the onset of the experiment, remained relatively constant, except for an increase at a rate of 17.8 ± 1.7 µmol C L^{-1} d^{-1} over the last 2 weeks of the experiment in the mesocosm receiving the highest nutrient load (Fig. 4).

The very low concentration of dissolved inorganic nutrients at the onset of the experiment were mirrored by the nutrient depleted organic pools, with average C/N ratios of 18 and 15.8 and average C/P ratios of 1101 and 1031 in the dissolved and particulate organic pools, respectively. Phosphorus was in deficit relative to nitrogen in all pools at the onset of the experiment, with atomic N/P ratios of 31, 104 and 69 in the dissolved inorganic and organic, and the particulate organic pools, respectively. Hence, the nutrient pools at the onset of the experiment deviated greatly from the general Redfield stoichiometry, indicative of a strongly nutrient deficient ecosystem, particularly with respect to phosphorus. Experimental nutrient additions altered this situation, leading to a parabolic change in C/N and C/P ratios in the dissolved organic matter with increasing nutrient inputs (Fig.5). The nutrient

ratios in the particulate organic fraction substantially exceeded the Redfield stoichiometry until nutrient inputs were increased > 8 fold above the "business as usual scenario", and reached values closer to Redfield stoichiometry at higher nutrient inputs (Fig. 5). The deviation of nutrient ratios from the Redfield stoichiometry was often highest at nutrient inputs ranging between the "business as usual" and 4 fold higher loadings (Fig. 5). The C/N and N/P ratios in the sedimentary material removed from the mesocosms averaged 17.7 and 8.3, respectively, and tended to decrease with increasing nutrient inputs to reach average C/N ratios of 13.5. However, the average N/P ratio in the material did not change with increasing nutrient inputs.

Increased nutrient inputs led to a shift in the partitioning of nutrients between the dissolved inorganic and organic, and the particulate organic pools by the end of the experiment (Fig. 6). While the relative size of the dissolved inorganic nutrient pools (about 20 % of the N and P) did not differ substantially with increasing nutrient inputs, there was a tendency towards an increase in the relative size of the particulate pool at the expense of DOM as a reservoir of N , P and C (Fig. 6). These tendencies were significant ($P < 0.05$) in all cases except for the decrease in the relative size of the DOP pool with increasing nutrients, which was not statistically significant ($P = 0.24$). Despite these tendencies, the DOC pool remained the dominant pool of organic carbon, whereas most of the N and P were contained in the particulate organic matter at the highest nutrient inputs tested (Fig. 6).
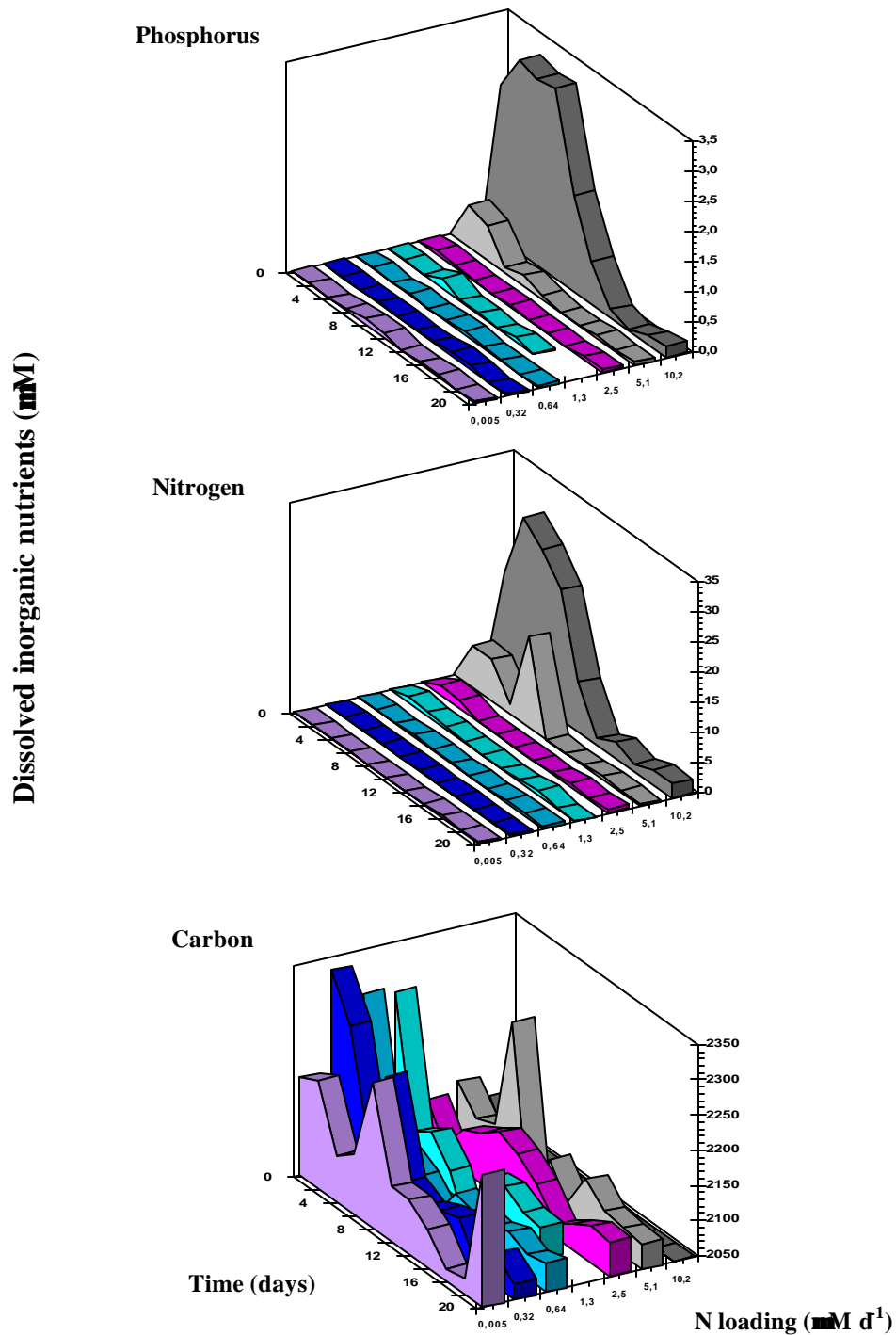
Fig. 2.   The response of dissolved inorganic nutrient and carbon concentrations in mesocosms enclosing receiving increasing nutrient inputs. Dissolved inorganic phosphorus concentration from Duarte et al. (2000a).
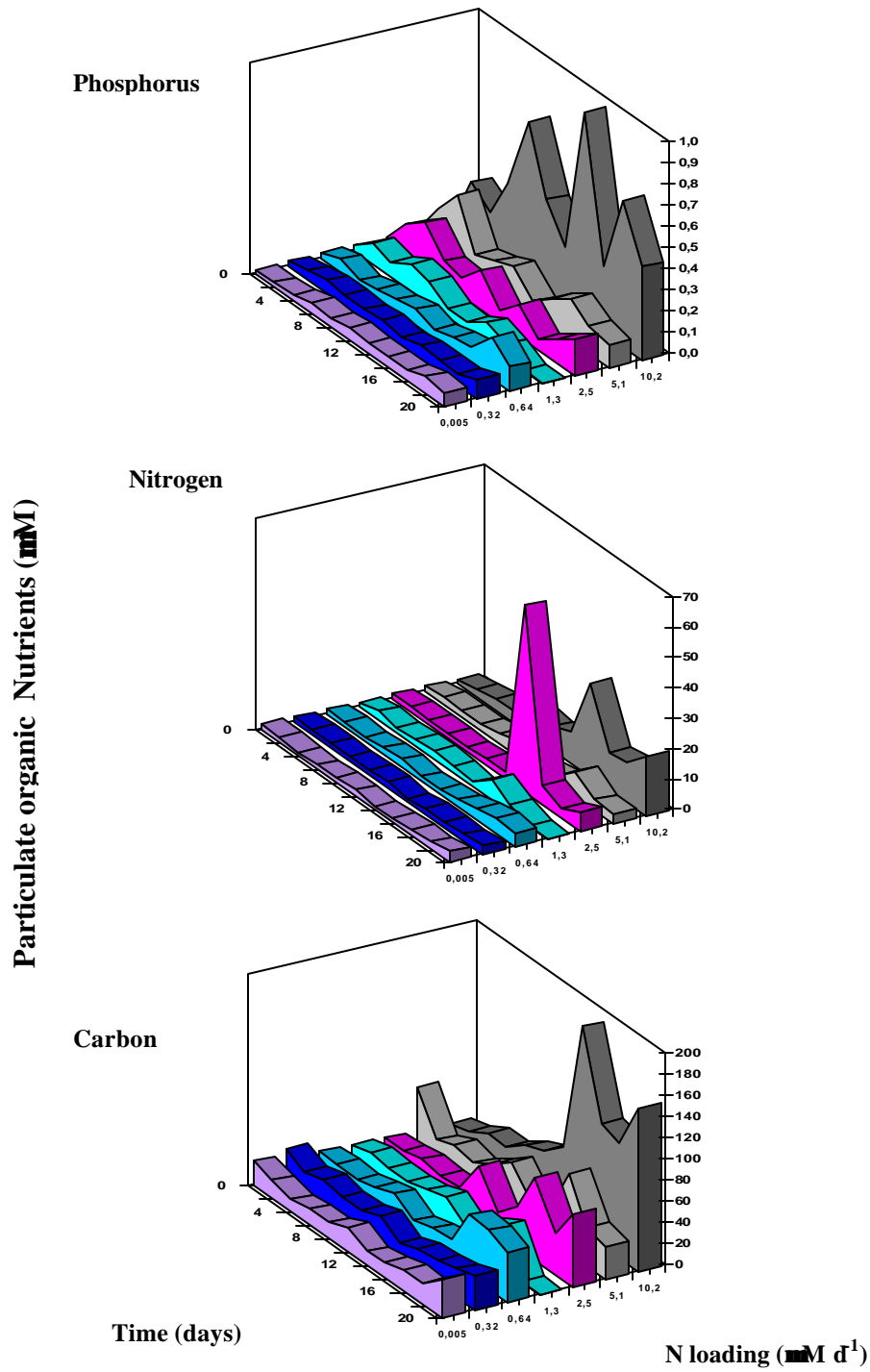
Fig. 3.   The response of particulate organic nutrient and carbon concentrations in mesocosms enclosing receiving increasing nutrient inputs.
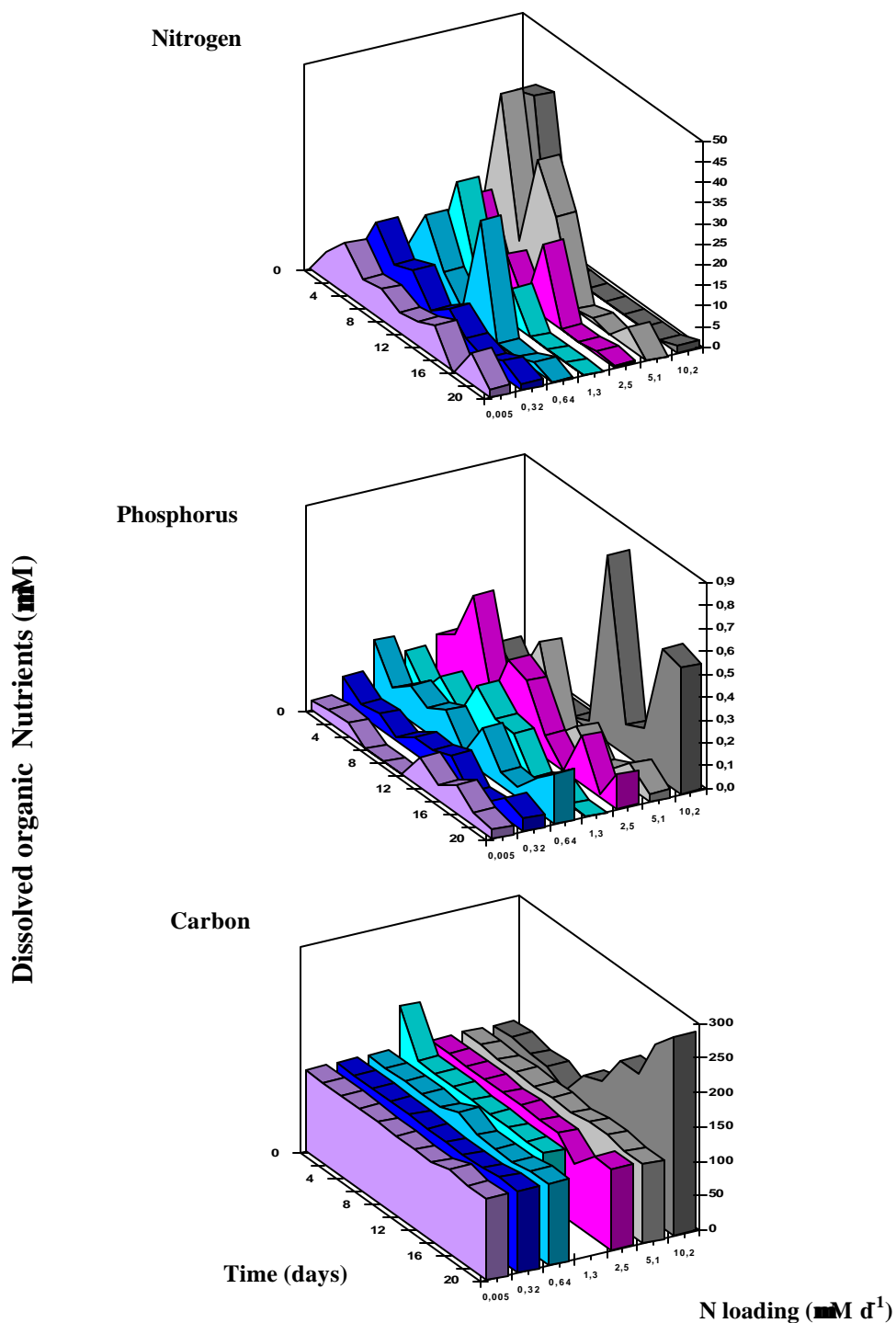
Fig. 4.   The response of dissolved organic nutrient and carbon concentrations in mesocosms enclosing receiving increasing nutrient inputs.

5.5. Discussion.

The Mediterranean waters investigated were acutely oligotrophic at the onset of the experiment, representative of the nutrient-poor conditions that characterise Mediterranean waters in the summer season. In agreement with previous reports from oligotrophic waters (e.g. Doval et al. 1999, Vidal et al. 1999), dissolved organic matter was the dominant pool of nutrients and organic carbon in the nutrient-poor area investigated. The concentrations of nutrients, particularly that of N, in the dissolved organic matter was sufficiently high as to be able to fuel the planktonic community well above their normal biomass if this nutrient pool was available. That this was not the case, and that the planktonic community was strongly nutrient-limited (Duarte et al. 2000a) suggest that the dissolved organic nutrient pool was unavailable to the community, thereby accumulating in the system to comprise > 75 % of the N, P and organic C in this oligotrophic ecosystem. The nutrient deficiency was alleviated by the increased nutrient inputs, which accumulated as dissolved inorganic nutrients until the community developed sufficient biomass as to be able to remove them. Dissolved inorganic nutrients were efficiently removed after this initial response occurred, which reduced the nutrient input per unit biomass, eventually driving the community back to a situation of nutrient limitation. The bulk of the nutrients added (> 70 %) were lost through sedimentation of particulate organic matter, which was pumped out from the mesocosms at four-day intervals.

The distribution of nutrients at the onset of the experiment was indicative of a strongly nutrient-limited system, where both dissolved inorganic N and P, but particularly the latter, were deficient compared with the standard Redfield stoichiometry. This situations is consistent with reports of a general tendency for phosphorus limitation in the Mediterranean planktonic ecosystem (Krom et al.

1991; Thingstad and Rassoulzadegan 1995; Thingstad et al. 1998; Zohary and Robarts 1998). The particulate organic pool, which comprised a modest fraction of the nutrients (< 20 %) was also characterised by exceedingly high C/P and C/N ratios, as a result of the dominance of detrital material (Duarte et al. 2000b). Nutrient-depleted organic matter pools are highly resistant and can only be degraded significantly if an external nutrient supply is available (e.g. Zweifel et al. 1993). The phytoplankton communities present in the oligotrophic waters examined have been reported to be major sources of DOM, derived from high cell lysis rates in summer (Agustí and Duarte 2000). Provided the very low nutrient concentration at the onset of the experiment, it is clear that organic matter would accumulate, albeit slowly given the low primary production, as DOM, thereby accounting for the dominance of this pool relative to the POM pool at low nutrient supply.

The nutrient ratios in the different pools, particularly the particulate pool, only approached the Redfield ratio at the highest nutrient additions (> 20 mmol N $m^{-2} d^{-1}$), despite the "business as usual" nutrient loading being already considerable (5 mmol N $m^{-2} d^{-1}$). The apparent refractory nature of the dissolved organic matter pool is associated with very high C/nutrient ratios (C/N = 18, C/P = 1101) in the DOM, which are likely to result in a very ineffective use and growth yield of bacteria (Zweifel et al. 1993, Cherrier et al. 1996). The DOM was particularly poor in phosphorus, consistent with the general phosphorus deficiency in Mediterranean waters (Krom et al. 1991; Thingstad and Rassoulzadegan 1995; Thingstad et al. 1998; Zohary and Robarts 1998). The accumulation of dissolved organic matter in oligotrophic waters suggests that recycling processes are limited by nutrient availability, particularly that of P (e.g. Zweifel et al. 1993; Rivkin and Anderson 1997), which has been found to limit bacteria in oligotrophic marine waters (Rivkin

and Anderson 1997, Cotner et al. 1997), including the Mediterranean Sea (cf. Thingstad and Rassoulzadegan 1995, Thingstad et al. 1998).

The high initial increase in DON concentrations at the highest nutrient additions is puzzling, since it would require that most of the added inorganic nitrogen be cycled to organic form by a comparatively small autotrophic community, and be rapidly released to the water. Phytoplankton mortality rates were high (> 1 day^{-1}, Agustí and Duarte 2000) at the onset of the experiment, providing a mechanism for the loss of the primary production to DON, and the maintenance of a modest biomass. Dissolved organic nitrogen was rapidly consumed following an initial increase. In contrast, DOC concentrations remained relatively uniform and only increased in the mesocosm receiving the highest nutrient loading. This increased occurred at a rate of 17.8 ± 1.7 µmol L^{1} d^{-1}, corresponding to about 10 % of the primary production. These results are in contrast with those of mesocosm experiments in Atlantic waters (Søndergard et al. 2000), where very high DOC accumulation rates were observed even at low nutrient inputs compared to those applied here.

The experimental results do not necessarily represent steady-state conditions, since some nutrient pools were clearly increasing (e.g. DOC and PON) at the time the experiment was terminated. Continuation of the experiment over longer time scales, as to assess responses at the seasonal scales that largely drive the shifts between nutrient pools in temperate coastal waters, such as the seasonal increase in DOC in coastal waters (Williams 1995), would have introduced confounding factors from wall effects. This limitation in the time scale over which the experiment was run, a common limitation of mesocosm experiments (Duarte et al. 1997), implies that the results observed are not readily extrapollatable over longer time scales.

The results presented clearly support the hypothesis of a shift from a dominance of nutrients in the DOM pool to an increasing importance of the POM pool with increasing nutrient inputs. The percentage of nutrients in dissolved organic form, as well as the absolute concentration of dissolved organic nitrogen, tends to decline with increasing nutrient inputs, consistent with the observed contrast in the importance of particulate and dissolved pools in oligotrophic vs. eutrophic waters (e.g. Slawyk and Raimbault, 1995; Karl et al. 1997, Wheeler et al. 1997, Doval et al. 1999, Vidal et al. 1999, Packard et al. 2000). Oligotrophic waters are characterised by a dominance of nutrients, particularly N, in dissolved organic form, likely because the dissolved organic matter there is still so poor in nutrients that bacteria require external nutrient sources to recycle it. In contrast, high nutrient inputs result in a dominance of particulate nutrients. The shift in the relative partitioning of organic nutrients between the dissolved and the particulate pools with increasing nutrient inputs implies a shift in the form of carbon export from DOM accumulation and subsequent gradient-driven export to sinking fluxes with increasing nutrient inputs. Moreover, a dominance of nutrients in DOM form allows a long-range horizontal export of carbon and nutrients. In addition, the dominance of particulate organic nutrients in waters receiving high nutrient inputs implies that these should be available to be transferred to protists and metazoans. In contrast, the high fraction of the nutrients and carbon contained in DOM in oligotrophic waters can only be transferred up the food web through bacterial use, thereby accounting for differences between the importance of the microbial loop in oligotrophic waters and the dominance of the linear food web in more productive waters (Legendre and Rassoulzadegan 1995). The important consequences of the shift in the partitioning of carbon and nutrients between dissolved and particulate organic pools indicates that further progress in our understanding of the processes

responsible for this pattern will substantially enhance our capacity to predict the fate of carbon and nutrients in pelagic waters.
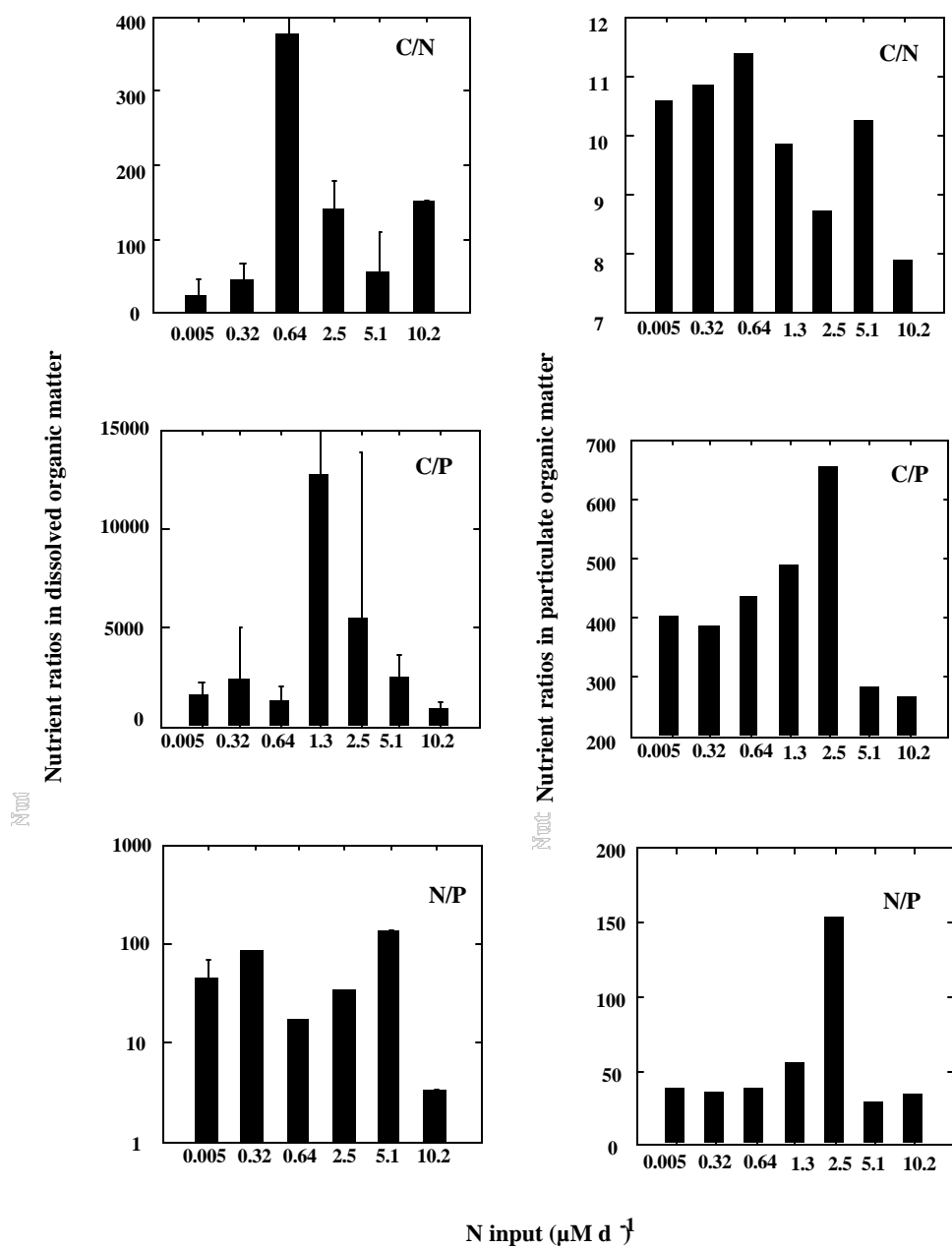
Acknowledgments

Fig. 5. The average C/N, C/P and N/P ratios of the dissolved and particulate organic pools in the mesocosms between days 14 to 20 after the initiation of the experiment. Error bars represent the standard error of the average values.
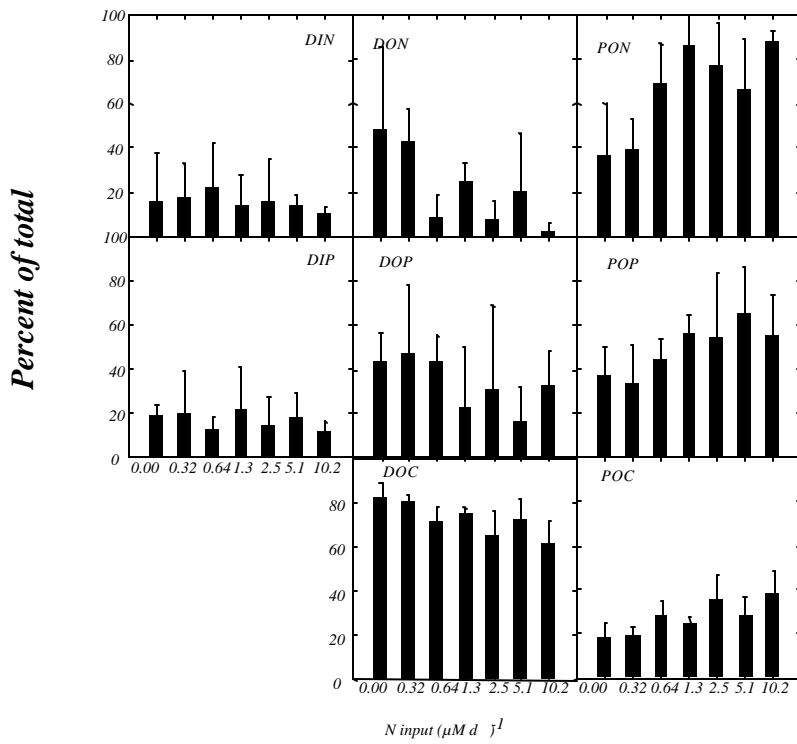
Fig. 6. The average percentage of N, P and C in the dissolved (inorganic and organic) pools in the mesocosms between days 14 to 20 after the initiation of the experiment.   Dissolved inorganic carbon is not represented, for its very large size compared to the other pools would obscure any patterns present there. Error bars represent the standard error of the average values.

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6.1. General discussion.

The close correspondence found in the Central Atlantic between water mass distribution (See Fig. 2 and 4, Chapter 2) and dissolved inorganic nutrients, as well as the positive correlation found between the upward diffusive supply of nutrients to the mixed layer and the strength of the thermocline (See Fig. 6, Chapter 2) clearly indicate that nutrient fluxes depend on density variations within the thermocline. Parallel to physical processes, the nutrient uptake rates drive the nutrient gradients. The resulting variability in new production values estimated from nutrient fluxes (see Table 2, Chapter 2) in the Central Atlantic provides evidence of the close link between physical and biochemical processes as drivers of biogeochemical fluxes in marine ecosystems. Recycling within the biogenic layer must be faster for nitrate as the estimated residence times were shorter than those for phosphate.

Dissolved inorganic nitrogen and phosphorus concentrations within the biogenic layer were close to Redfield stoichiometry through the transect, except for the subtropical gyre zone (10ºS to 15ºS. Fig. 7, Chapter 2) where nitrogen supply exceeded the expected phosphorus supply and consequently the N:P ratios within the biogenic layer were well above the Redfield ratio. The rest of the southern transect was deficient in nitrogen. No significant correlation was found between the N/P ratio in the intermediate layer and those in the upward flux, which indicates that changes in N/P input ratios result from variable recycling activity rather than a nutrient influx from intermediate waters. The close correspondence between nitrate:phosphate ratios in the diffusive flux and those in the biogenic layer suggest that the planktonic metabolism adapts to local variations about the Redfield ratio within the biogenic area (nutrient assimilation) and the thermocline (remineralization). Except for those areas subject to advective supply of nitrate to the euphotic zone due to upwelling, nitrogen supply exerts a dominant control over primary production along most of the Central Atlantic Ocean, while the south subtropical gyre appears to be phosphorous deficienct, in agreement with results derived form P uptake experiments (Cañellas et al. 2000).

The Guinea Dome and the Atlantic Equatorial area are characterized by relatively high diffusive nutrient fluxes driven by physical forces, with N/P values close to the Redfield ratio and a high potential new production (Table 1, Chapter 2). Advective flows of nutrients supplied by the Equatorial upwelling are the major mechanism of nitrate supply in the vicinity of the Equator, whereas away from the equator where vertical velocities are very small and surface waters are nutrient-poor, nutrient fluxes driven by turbulent diffusion are the main source of nitrate (Carr et al., 1992; Chavez and Barber, 1987). The advective components of nutrient inputs have not been incorporated into the estimates of nitrate supply to the euphotic layer in the equatorial and doming region, so that the rates derived from diffusive vertical flux of nitrate estimated there are conservatives ones. On the other hand, the diffusivity estimates used for the Equatorial area reveal potential new production values higher than those based on microstructure measurements in the equatorial Pacific (Carr et al., 1995; Chavez and Barber, 1987). Indeed, the estimates of the vertical diffusivity coefficient derived here integrate fields that have characteristic time scales of weeks, whereas microestructure measurements yield estimates of diffusivity coefficients with characteristic time scales of hours to days only. Hence, while the latter may be more precise they are certainly less representative of mid-term conditions.

The potential new production estimated from vertical nutrient fluxes ranged about an order of magnitude from the gyre area (about 1 mmol C m^{2} d^{-1}) to the northern part of the transect (about 10 mmol C m^{2} d^{-1}). The observed nitrate uptake rates in the gyre area (Planas et al., 1999) were higher than our physical estimate of new production. The discrepancy between new production rates estimated from physical supply and that estimated from nitrate incorporation into particles suggests atmospheric input, through remineralization of dissolved nitrogen-rich organic matter from diazotrophic organisms (Gruber and Sarmiento, 1997) together with direct inputs of reactive nitrogen, and provides evidence of the important role that dissolved organic nitrogen play in the upper layers of the Central Atlantic.

Indeed, the important role of the dissolved organic pool as a major reservoir of N in oligothrophic environtments is a consistent result of the research presented here. The stratified oligothrophic NW Mediterranean was chosen as the scenario to test the existence of dissolved organic matter stoichiometric relations in balance with the particulate and dissolved inorganic pools. Indeed, dissolved organic matter takes an intermediate position between the inorganic nutrient removal and recycling processes described by the general equation:

$$x \, CO_2 + y \, HNO_3 + z \, H_3PO_4 + w \, H_2O \Leftrightarrow (CH_2O)_x \, (NH_3)_y \, (H_3PO_4)_z + k \, O_2$$
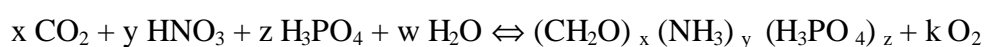
Producers and consumers of organic matter interact in the marine food web when the former take dissolved inorganic nutrients at certain ratio (x:y:z) while the latter recycle nitrogen and phosphorus back into dissolved form in deeper water. The inclusion of DON and DOP with DIN and DIP to form total dissolved nitrogen (TDN) and total dissolved phosphate (TDP) link the organism PON:POP values with DIN:DIP values (Jackson and Williams, 1985) and supports the "Redfield" nutrient model (Redfiled, 1963) in euphotic zones of the Pacific ocean.

But, does production and consumption of dissolved nutrient occurs in ratios that differ from the ratios of the concentrations of those nutrients in the surrounding waters of the oligothrophic Mediterranean ocean and which are the potential causes of such deviation?. The overall phosphorus deficiency found exerts a tigth control in the total nutrient partitioning within the system. Except for the biogenic layer where P is in excess relative to N, total N:P dissolved inorganic ratios far exceeded the Redfield ratio in thermocline and biolithic layers of the Mediterranean Sea (Table 1, Chapter 3). P deficiency is particularly accute in the Mediterranean summer (Bethoux and Copin-Montegut, 1988; Thingstad et al., 1998), when surface layers are well statrified. The opposite patterns between the N contained in the dissolved inorganic and organic pools with depth in the Mediterranean Sea (Fig. 8, Chapter 3) suggest that the net result of biological activity is the prevalence of DON production in surface

waters and remineralization in the biolythic layer. The dominant chemical form of reduced nitrogen and phosphorus shifts towards dissolved organic matter forms within the euphotic zone as DON, DOP concentrations increase and NO_3^- and PO_4^{3-} concentrations decrease. The particulate pool (< 30% of the total nutrient pool) mediates the chemical changes among different nutrient chemical forms. High DON and DOP production often exceeding 50% of the N and P uptake rates have indeed been reported for oligotrophic marine waters (Bronk et al., 1994; Cañellas et al., 2000). Exhudation by healthy cells, repesenting < 10 % of the production (Carlson et al., 1992) does not appear to be sufficient to account for such high DOM production rates, so that other processes must be involved. Indeed, recent results point to high phytoplankton lysis rates in oligotrophic marine waters in the open and coastal NW Mediterranean (Agustí et al., 1998; Agustí and Duarte, 2000) and in the NE subtropical Atlantic (Agustí et al., 2000).

Comparative trends of C/N in particulate and dissolved organic pools reveal an accumulation of dissolved and particulate organic carbon at biolythic layer (Fig. 7, Chapter 3). The POC/PON (C/N:28) at depth were higher than those found in upper layers (C/N:11) suggesting a differential particulate organic carbon transport relative to nitrogen within the water column. Reported POC concentrations vary from 4μM to 16μM which are common winter values in the Mediterranean Sea (Miquel et al., 1993; Cauwet el al.,1997). We report DOC concentration values around 90 μM at about 1000-m depth which implies the presence of a water mass richer in DOC, as also reported by Cauwet et al.(1997,1983). Recent studies point to DOC export from upper layers of the Pacific ocean during the formation of intermediate waters as responsible for higher DOC concentrations at 1000 meters depth (Hansell et al., 2002). High DOC values found at 1000-2000 meters depth in a central region of the stratified Mediterranean Sea (Santinelli et al., 2002) were attributed either to physical processes and oxidative activity on fast sinking particles and sedimentary material (La Ferla and Azzaro, 2001).

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The averaged DON downward fluxes are high compared to the average PON export from the biogenic layer in the NW Mediterranean (140 µmol N m^2 day^{-1}, Miquel et al., 1994). A budgetary approach of diffusional nutrient supply and downward transport of N and C species within thermocline for the Mediterranean system are schematized in Figure 1. The downward flux of DON is the dominant source of nitrogen export from the biogenic layer of the NW Mediterranean (about 60 % of the downward N transport). Since diffusional DIN supply to the biogenic layer yields an estimate supply of 145 µmol N m^2 d^{-1} to the biogenic layer, allocthonous inputs must balance the excess of N downward flux. The sum of the sinking DOC flux and the average POC flux in the NW Mediterranean yields an estimate of 2.8 mmol C m^{-2} d^{-1} which must represent the new production in the area and exceeds the new production estimated from DIN supply to the biogenic layers (1.7 mmol C m^2 d^{-1}), further pointing to atmospheric N supply. Eppley and Peterson (1979) defined new production as primary production supported basically by the fresh upwelled nutrients supply to the biogenic layer, while atmospheric and terrestrial inputs were considered as secondary terms. In our case the new production values calculated from internal nutrient supply account for only half of the new production, suggesting atmospheric inputs to be important to support new production in the Mediterranean.
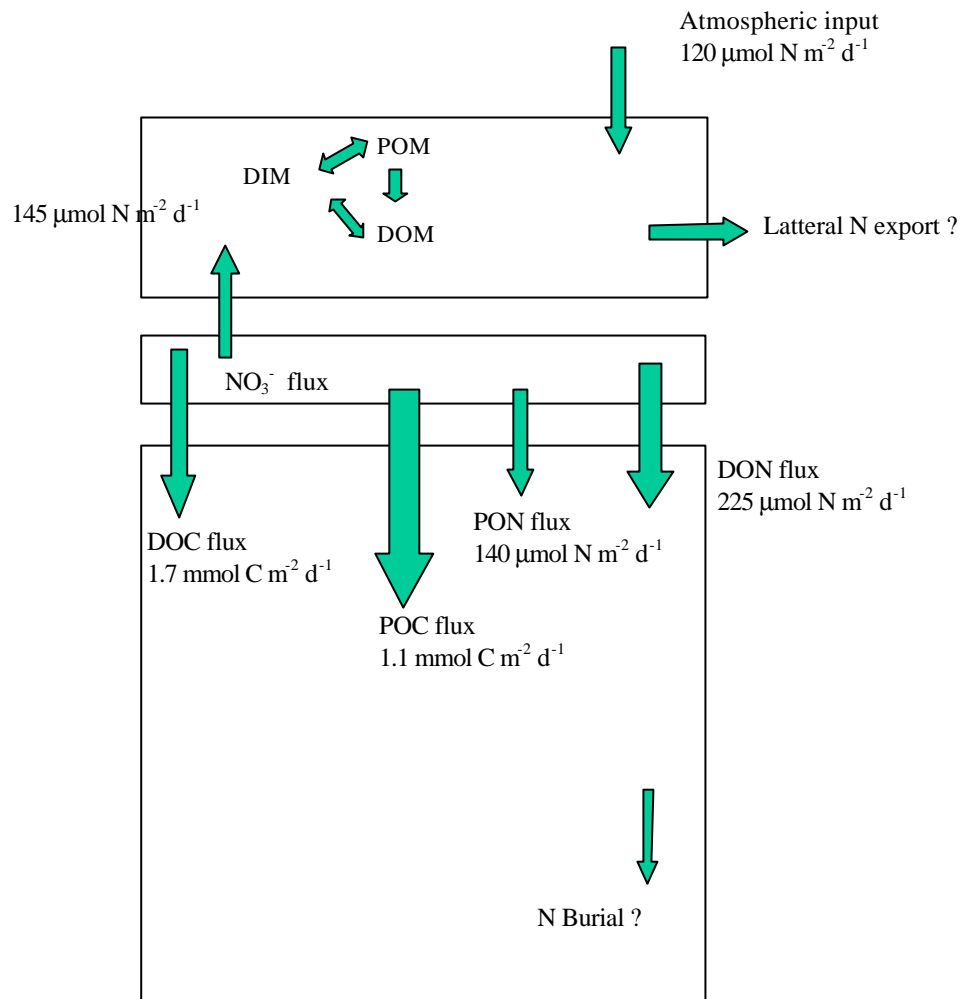
Fig. 1. Nitrogen and Carbon budgets of the NW Mediterranean derived from chapter 3. Large arrows size indicates relative size Carbon and Nitrogen diffusional fluxes within the thermocline layer estimated by assuming gradient transport and using a turbulent coefficient of Kz =3 m^2 day^{-1} (Copin-Montégut et al., 1993) calculated during a stratified period.

This study supports previous findings of high rates of marine nitrogen fixation in the Mediterranean Sea (Gruber and Sarmiento, 1997; Bethoux et al., 1992). The physical estimate of new production values in the oligothrophic NW Mediterranean derived from upward nitrate fluxes are towards the low range of the estimated new production in the Central Atlantic (1-10 mmol C m^2 d^{-1}) but of the same range of magnitude of those found in the nutrient depleted South Atlanthic gyre area.

Chapters two and three address the biochemical processes driving nutrient distribution in the oligothrophic Atlanthic and Mediterranean open ocean. However, coastal oligotrophic locations receive significant terrigenous inputs and may, therefore, behave quite different. The bay of Blanes, located in the North Western Mediterranean basis, is characterized by significant land inputs from the Tordera river and shallow-deeper waters exchanged in winter as a result of the water mass circulation of the Blanes canyon (Masó and Tintoré, 1991). The nature of the Blanes canyon forces the surface water mass circulation to be northeastwards on the shelf, enhancing its water exchange with the continental slope (Rojas et al., 1995). Indeed, dissolved inorganic ratios are relatively low in summer (DIN/DOP < 7) but higher in the winter season (DIN/DOP ≥ 10), similar to the stoichimetry found in biolythic layers of the stratified NW Mediterranean (Table 1, Chapter 3). The nutrient dynamics of this oligotrophic coastal site not only depend on marine supply and terrestrial inputs but local processes such as phytoplancton cell lysis (Agustí and Duarte, 2000) contribute to increase the dissolved organic content in the water column.

Lower elemental rations in dissolved organic matter (DOC:DON:DOP:857:24:1) relative to the biogenic layers ratio of the stratified NW Mediterranean (Table 1, Chapter 4) indicate an important oxidative activity in the shallow waters of Blanes Bay. Faster nutrient recycling in the organic matter pool is reflected in the average POC:PON ratio of seston, similar or below the Redfiled ratio (6.6), relative to the average POC:PON ratio of 12 of the material collected in sediment traps. Some fraction of POC within water column is converted to DOC during the microbial degradation and another fraction of refratory material as

accumulates in the DOC pool (Cauwet et. 1997). In fact, the highest DOC concentrations recorded (60 - 140 μM) correspons to the lowest POC concentrations found in the area. Although the planktonic community in the coastal area is heterotrophic (net community production: -8.6 mol C m^2 y^{-1}), the total organic carbon inputs to the sediment (Fig. 8, Chapter 4) lead to a net deposition of carbon. This requires terrestrial inputs, derived from suspended particles from riverine discharge (the nearby Tordera river), resulting in high deposition fluxes in spring and winter (Fig. 6, Chapter 4). In contrast, the benthic compartment is autotrophic net community production: 6.2 mol C m^{-2} y^{-1}) and receives important sedimentary inputs of organic carbon, with an important contribution of land-derived carbon. Indeed terrestrial materials contribute 37% of the total sedimentary flux, as derived from stable isotope signatures of sediment trap material collected at the study site. Despite substantial inputs of organic carbon and nutrients from land, the Bay of Blanes remains oligotrophic with no evidence of eutrophication effects.

Chapter 5 addresses the hypothesis that the importance of dissolved organic matter pool (DOM) as a reservoir of C, N and P declines, relative to that of the particulate pool, with increasing nutrient inputs. The planktonic community in the system was strongly nutrient limited and increased nutrient inputs led to a shift in the partitioning of nutrients between the dissolved inorganic and organic, and the particulate pools. The accumulation of dissolved organic matter in the system evidences the refractory nature of DOM pool associated with very high C/N ratios (Fig. 5, Chapter 5) and suggests that recycling processes are limited by the nutrient availability of the system. Nutrient partitioning in oligotrophic waters is characterized by a dominance of DOM since bacteria require external nutrient sources to recycle it.

Nutrient-enhanced primary productivity in coastal systems is often a signal of eutrophication (Anderson and Rydbert, 1988; Justic et. al, 1987). Eutrophication processes have increased over the last several decades in many coastal areas, particularly those affected by rivers inflows. Nitrogen and phosphorus have increased substantially as a result of fertilizers and detergent use in the watershed. The experiment demonstrates that nitrogen inputs

> 40 mmol N m $^{-2}$ d^{-1} in the system result in a considerable increase of organic nutrient pool. However, the Blanes Bay remains oligothrophic because of the high water exchange with off-shore waters as a consequence of enhanced exchange induced by the adjacent submarine canyon (Rojas et al., 1995; Granata et al., 1999) and the heterotrophic nature of the planktonic community (Satta et al ., 1996) that prevents organic matter accumulation in the system.

The export of primary production from the surface water is a key component of the ocean carbon cycle since it leads to a drawdown of atmospheric carbon (Sarmiento and Siegnthaler, 1992). The C:N:P stoichimetry adressed in Chapters 2 and 3 represents the ratio at which photosynthesis and consequently the carbon export takes place. Southern Subtropical Atlantic Gyre and stratified oligotrophic Mediterranean systems fix dissolved inorganic N and P in a proportion lower than expected from the Redfield ratio (see Table 1, Chapter 2 and Table 1, Chapter 3). This non-Redfield behaviour ocurring in biogenic layers suggests a chemical impact of marine organisms (Vidal et al., 1999; Agustí et al., 2000; Agustí et al., 1998; Agustí and Duarte, 2000). Biogenic and thermocline layers of nutrient remineralization in the Atlantic waters have different dissolved inorganic N:P ratios relative to those found in biolythic layer suggesting that nutrients produced or consumed are not in balance with their environment (Fig. 7, Chapter 2). DIN:DIP ratios in the thermocline layer of the stratified NW Mediterranean exceeded the Redfield ratio. Potential causes are the influx of Levantine Intermediate Water where doming pycnoclines lead to a high nutrient supply (Fig. 2, Chapter 3) and atmospheric N deposition (Migon et al., 1989; Erdman et al., 1994). Nitrogen fixation could also produce the subsurface maximum N:P. Aerobic decomposition of the sinking organic matter whithin the thermocline might then produce a buildup of N relative to P.

Substantial variability in nitrate:phosphate ratios has been reported between the pthotic zone and 500 m in the west central North Atlantic [N:P ~ 17-50 , Fanning, 1992; ~ 12-22 mol N (mol^{-1} P), Takahashi et al., 1985]. Krom et al. (1991) reported Mediterranean N:P

ratios ranging from 19 to 23 mol mol $^{-1}$. Despite the large N:P differences found in upper ocean waters, all data distributions approach a "constant" N:P ratio of about 15 mol mol $^{-1}$ toward the deep waters where nutrient remineralization predominates. An average deep-sea N:P ratio of 15 has been also reported for the world ocean (Tyrell and Law, 1997). Deviations from this value (N.P ~ 1.5-3.0) has been attributed to anaerobic decomposition by denitrifying bacteria.

Data on simultaneous determination of carbon, nitrogen and phosphorus in particulate matter within surface layers are still few (Cauwet et al., 1997; Bishop et al., 1980; Copin-Montegut and Copin-Montegut., 1993). The available information point to an increase in the C:N ratio from ~ 5 to 8, to values between 8 and 15 or greater in POM at depth. The average POC:PON ratio of seston in the open Mediterranean (Table 1, Chapter 3) is higher than the Redfield ratio (6.6) and increases with depth which indicates an increase in export carbon flux from surface ocean to depth. Biogenic layers in the coastal waters studied show values of particulate carbon relative to nitrogen export similar or below the Redfield ratio despite the influence of organic terrigenous inputs (Table 1, Chapter 4).

Overall dissolved organic matter pool in the NW Mediterranean (Table 1, Chapter 3; Table 1, Chapter 4) deviates substantially from Redfield stoichiometry. Relationships between DOC, DON and DOP indicated that N and P were preferentially remineralized relative to carbon. Biolythic layers, with DOC:DON ratios of about 78, deviate even further from Redfield. DOM export may be an important component of the biological C pump (Kumar et al, 1990; Ducklow et al., 1995). The results presented here demonstrates that, due to the importance of the dissolved organic pool, the amount of carbon removed from biogenic layers exceeded the amount predicted based on the nitrate removal and the Redfield ratio (Redfield, 1963). DOC and DON concentration values, carried out using high temperature oxidation methods and persulfate methods respectivelly in this study, are similar to those measured with recent high temperature oxidation methods in coastal sites (C:N ~ 15-38, Hansell et al., 1993; C:N ~ 24-55, Hopkinson et al., 1997) , upper water column of Arctic ocean (C:N ~ 9-25

Wheeler et al., 1997) and NW Mediterranean sea (C:N ~ 20, Wheeler et al., 1997). Persulfate and high temperature oxidation methods have been found to be comprable in oxidation efficiency, precision and limits of detection (Bronk, 2000).

The partitioning of chemical species (C, N; P and Si) among different pools was addressed in chapters 3, 4 and 5. Examination of the percentage of nutrients among the different pools allowed us to test the role that DOM acts as a major nutrient reservoir in oligotrophic systems. DOC represents the second largest pools of carbon in the ocean (Hansell and Carlson, 1998 a), second to the very large pool of dissolved inorganic carbon. DOC concentrations in the surface ocean contain the highly biologically labile fraction of DOC that supports microbial heterotrophic processes in the ocean (Carlson and Ducklow, 1992). Vertical profiles of DON generally show a surface enrichment relative to concentrations deeper in the water column. This has been observed in a wide number of environments including the equatorial Pacific (Libby and Wheeler, 1997); Bering Sea (Hansell, 1993), Santa Monica Basin (Hansell et al., 1993), the oligotrophic North Pacific (Maita and Yanada, 1990), the North Pacific (Abell et al., 2000). Vidal et al. (1999) calculated DON fluxes from vertical profiles of DON concentrations and estimates of the vertical eddy diffusion coefficient. They found that surface DON did appear to be transported to depth at times, however, the direction of the dominant flux varied along the north-south transect. Vertical distributions of TDN (total dissolved nitrogen) and DON in the Pacific Ocean indicated a high contribution of DON in the surface mixed layers (> 90% of the TDN concentration; Maita and Yanada, 1993). In biogenic layers of the open NW Mediterranean sea > 75% of the organic nitrogen material was present in the dissolved organic phase, while < 30% of the organic material was present in the particulate phase (Fig. 8, Chapter 3). Most (> 80%) of the organic carbon was present as dissolved organic carbon, with POC represented less than 10% of the organic carbon present in biogenic layer.

Similar patterns were found in coastal waters (Table 1, Chapter 4) where dissolved organic material dominated the carbon and nitrogen pools (DOC: > 88 %; DON: > 66%).

The coastal zone is one of the most productive regions of the World Ocean and its contribution to the release of DOM should be also significant in the global budget. The importance of DOM distributions and dynamics is receiving growing attention, as evidence that dissolved organic substrates are important intermediate steps in rapid cycling of bioactive elements within ocean (Thingstad and Rassoulzadegan, 1995; Zweifel et al., 1993).

The mesocosms experiment carried out at the oligothrophic waters of NW Mediterranean allowed an improved understanding of the role of nutrient supply in regulating the importance of DOM. At the onset of the experiment the dissolved organic nutrient pool was unavailable to the community, thereby accumulating in the system to comprise > 75 % of the N, P and organic C. Nutrient-depleted organic matter pools were highly recalcitrant and were only degraded when the external nutrient supply was available for recycling processes. At low nutrient inputs oligotrophic waters were characterised by a dominance of dissolved organic form (~ 40-50% for DON and DOP), likely because the dissolved organic matter there is still so poor in nutrients that bacteria require external nutrient sources to recycle it. At the end of the experiment 80% of organic material was present as particulate organic form. The shift in the relative partitioning of organic nutrients between the dissolved and the particulate pools with increasing nutrient inputs implies a shift in the form of carbon export from DOM accumulation and subsequent gradient-driven export to sinking fluxes with increasing nutrient inputs.
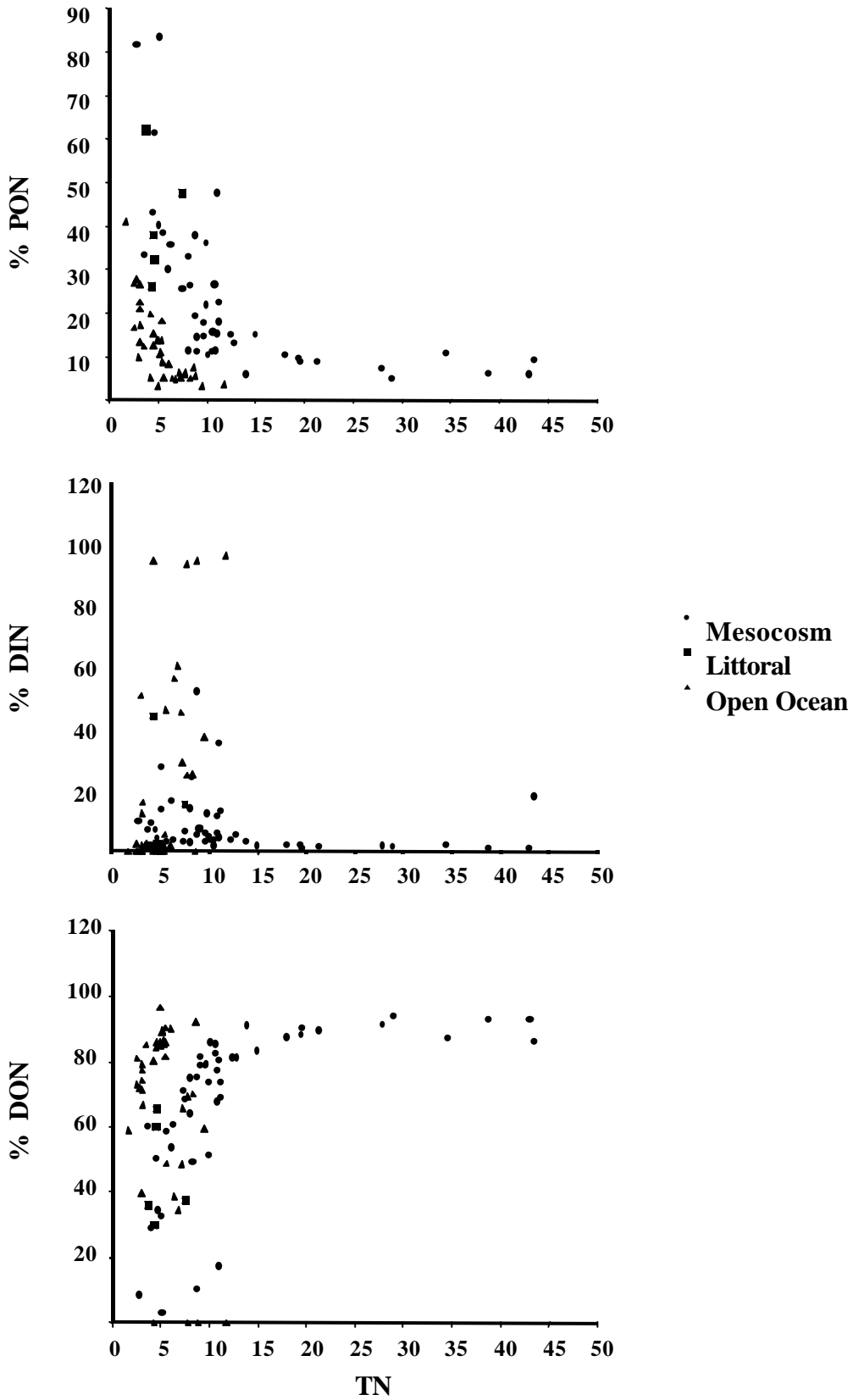
Fig. 2. Global Nitrogen partitioning in the oligrothrophic waters of the study.
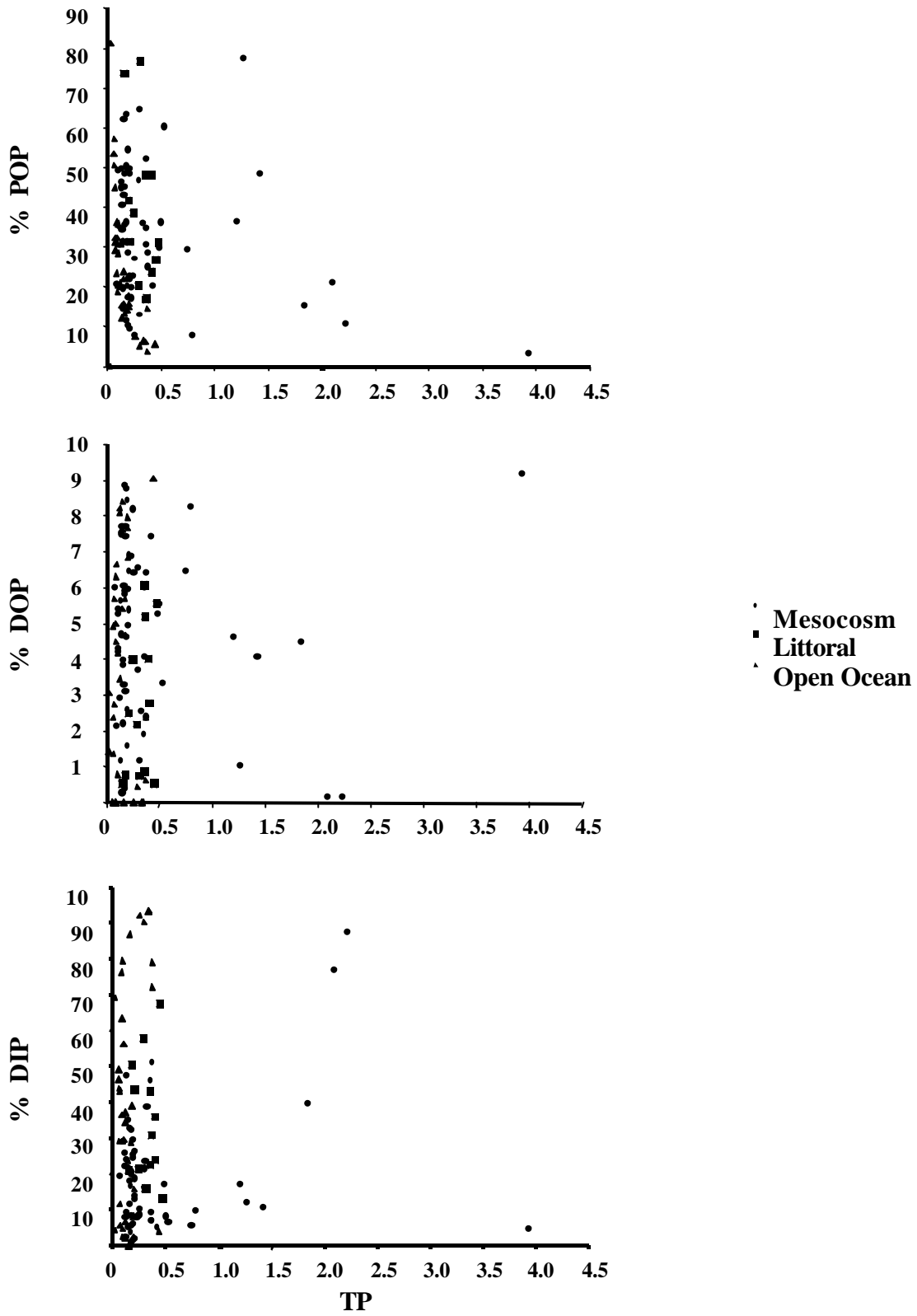
Fig. 3. Global Phosphorus partitioning in the oligrothrophic waters of the study

The patterns in nutrient partitioning among pools emerging from the results presented are best shown through a concerted analysis of the various studies across contrasting oceanic and littoral ecosystems herein contained. The contribution of each form of N and P (dissolved and particulate organic and dissolved inorganic) to the total nutrient pool (TN, TP) can be examined (Fig. 2 and Fig. 3, in this chapter) for total nitrogen (TN) and total phosphorus (TP) respectively. TP concentrations in this thesis range from 0 to 0.5 µM and maximum values of up to 2 µM were observed during the mesocosm experiment. Phosphorus partitioning ranges from 0 to 100 % for DIP, DOP and POP values. No distribution pattern is observed in all the three systems considered (Mesocosm, Littoral and Open Ocean). In contrast, the TN concentration ranges from 2.5 and 15 µM that confirms the phosphorus deficiency found in oligothrohic waters in the NW Mediterranean Sea. The lowest percentatge of the TN pool correspond to dissolved inorganic nitrogen forms (less than 40%) that remained low even at the high concentrations of TN reached in the mesocosm experiment. The partition of N between DON and PON pools followed opposite patterns. Organic matter was mainly present in dissolved form (> 40% to 90%) rather than particulate form which comprises less than 40% of TN. The importance of DON as the major component of the total nitrogen pool relative to DOP is reflected in the exponential increase in the percent contribution of DON and the corresponding decline in that of PON with increasing nutrient pools within the system. Yet, the contribution of the DON pool in the oligothrophic Mediterrean is inversaly proportional to the DOC/DON ratio (Fig. 4, in this chapter) as higher contributions of DON corresponded to low DOC/DON ratios.

The relative importance of different nitrogen pools was examined in relation to the total nutrient stoichiometry of the oligotrophic system (TOC/TN, figure 5). Lower percentages of PON, DON and DIN (< 20%) corresponded to TOC/TN values lower than 20. Maximum PON percentages (80-90%) are found at TOC/TN values around 20. The contribution of PON to the TN tends to decrease at TOC/TN ratios higher than 20. In contrast, maximum contribution of DON to the TN is shown at TOC/TN values lower than 20. The shift of the

dominance of DON towards PON at TOC/TN values higher than 20 on the oligothrophic areas of the study, gives evidence of increasing carbon export fluxes in a system dominated by particulate pool and points to the effect of the biota on the gradient-driven export to sinking carbon fluxes in the ocean.

The broad range of conditions encompassed in the research presented allows the formulation of general nutrient ratios (Table 1). The studies reporting the total nutrient stoichimetry (C, N, P and Si) for the various pools (DIM, DOM and POM) are few. Hence, the nutrient ratios obtained in this study for the oligothrophic waters of NW Mediterranean are compared to those found in surface waters of other geographical locations. The ratio of dissolved inorganic nutrients reported in the research presented is indicative of a general deficiency in nitrogen relative to phosphorus in the systems studied compared to the global ratios reported in literature. There is also, on average, a general N and P deficiency relative to silicate compared to the global ratios of these elements (16 N: 16 Si : 1 P , Redfield et al., 1963). The dissolved organic matter was highly depleted in P relative to N and C at all locations investigated and the resulting C:N ratio (11) of this study in the particulate pool deviates greatly from the literature values which aproximates Redfield value (5.5-6). Deviation of the elemental ratios from that postulated by Redfiled are a consequence of biological processes, which also exert a tight control on the paritioning of nutrient elements between different pools in oligothrophic waters.
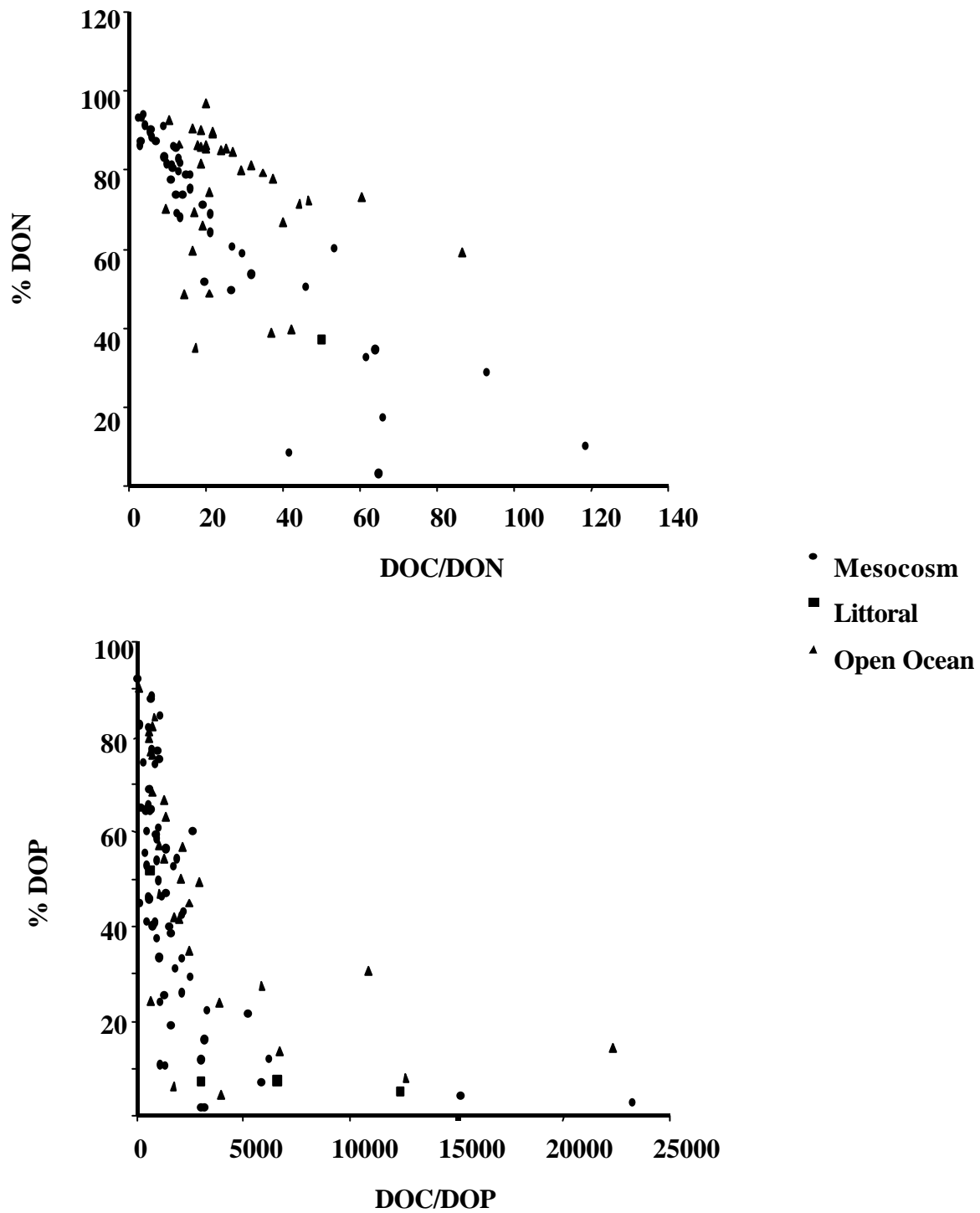
Fig. 4. DON and DOP partitioning versus dissolved organic C and P ratios in the oligothrophic waters of the study.
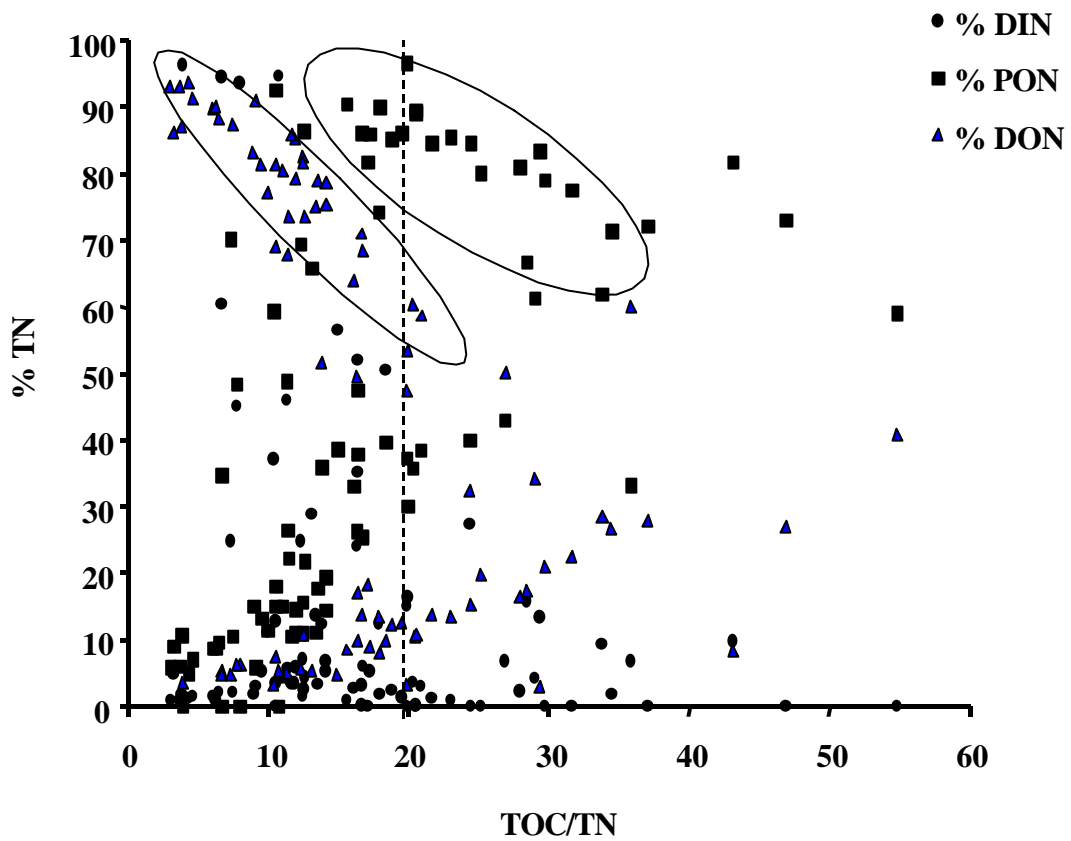
Fig.5. Distribution patterns of DIN, PON and DON relative to TOC/TN ratios in the oligothrophic waters of the study.

Table 1. Global C, N, P and Si stoichimetry in DIM, DOM and POM pools from this study and literature results.

DIM	DOM	POM
DIN: DISI: DIP	**DOC: DON: DOP**	**POC: PON: POP: BSi**
This study	This study	This study (C:N:P)
6: 12: 1	1058: 51: 1	47: 4: 0.15: 1
Literature [1]	Literature [3]	Literature [5] (C: N: P)
13:33:1 (Antartic Ocean; Weddel Sea)	553: 39: 1 (Pacific Ocean)	99: 18: 1 (Indian Ocean ;surface)
Literature [2]	Literature [4]	83: 15: 1 (Antarctic Ocean ;surface)
12: 14: 1 (NE Pacific Ocean)	400-800: 11-15: 1 (Georges Bank Region, continental shelf NE U.S.A)	72:13: 1 (NW African.upwelling region ;0-75m)
		103: 16: 1 (Mediterranean Sea ;surface)

[1] Leynaert, A., Nelson, S.M., Queginer, B., Treguer, P. 1993. The silica cycle in the Antarctic Ocean: Is the Weddell Sea atypical?. Marine Ecology Progress Series.96:1-1.

[2] Wash, T. 1989. Total Dissolved Nitrogen in Seawater: a New High-temperature Combustion Method and a Comparison with Photo-Oxidation. Marine Chemistry. 26: 295-311.

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[4] Jackson,G.A.and Williams,P,M. 1985. Importance of dissolved organic nitrogen and phosphorus to biological nutrient cycling. Deep-sea Research. 32:223-235.

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6.2. Conclusions.

1. The average ratio between dissolved inorganic nitrogen and phosphorus in the estimated vertical diffusive fluxes within the biogenic layer along the Central Atlantic Ocean (14.4 ± 1.7) was similar to the Redfield ratio, but tended to be well below the Redfield ratio at the southern end of the transect and above the Redfield ratio at the center of the South subtropical gyre (10° S - 15°S). The N:P supply ratio and the N:P ratio of dissolved inorganic nutrients in the biogenic layer were strongly correlated ($R^2 = 0.87$, $P < 0.001$), but were not positively correlated to that in the intermediate waters. The results indicate that vertical nutrient cycling is able to operate at nitrate to phosphate ratios well away from the Redfield ratio, so that both the biogenic and the biolythic components adapt to local variation about the Redfield ratio.

2. Most (> 80 %) of the organic carbon in oligotrophic waters was present as dissolved organic carbon, with POC representing a minor percent of total organic C throughout the water column of the stratified NW Mediterranean Sea. Organic N and P comprised 50 – 80 % of the N and P pool in the biogenic layer, and decreased with depth to represent 10 – 25 % of these nutrient pools in the biolythic layer. Because of the uniform depth distribution of the total dissolved nitrogen concentration, the increase in the percent inorganic N and the decline in the percent dissolved organic N with depth indicates that there must be a dynamic equilibrium between the biological transformations between these pools with depth, with a dominance of DON production in surface waters and remineralization in the underlying layers. The downward DON flux exceeded the diffusive DIN supply of about 145 µmol N m^{-2} day^{-1} to the biogenic layer, suggesting that alocthonous N inputs must be important in the region.

3. Dissolved organic nitrogen comprised about half of the nitrogen present in the water column and the carbon pool dominated by the inorganic pool (95% of the carbon present in the water column) in the Mediterranean littoral. The pelagic compartment was heterotrophic, supported by significant allochthonous inputs of land material, which also contributed greatly to the sedimentary inputs (37% of total sedimenting carbon). In contrast, the benthic compartment was autotrophic, with the excess net benthic community production balancing the deficit in pelagic community production, leading to a system in metabolic equilibrium. Carbon inputs to the benthic compartment also exceeded requirements, due to the allocthonous subsidies to the system, so that the benthic compartment stored or exported organic carbon.

4. Experimental nutrient additions carried out in the oligothrophic areas of the NW Mediterranean littoral lead to a parabolic change in C/N and C/P ratios in the dissolved organic matter with increasing nutrient inputs, which approached the Redfield stoichiometry at nutrient inputs > 8 fold above the ambient loading. The relative size of the dissolved inorganic nutrient pools (about 20 % of the N and P) did not vary, but there was a tendency towards an increase in the relative size of the particulate pool at the expense of a decrease in the relative importance of DOM as a reservoir of N, P and C, with increasing nutrient inputs. The production of nutrient-depleted organic matter at low nutrient inputs likely prevents efficient recycling, leading to the dominance of nutrients in DOM in the system.

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