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Title: Submicron Particulate Organic Nitrogen and Dissolved Organic Nitrogen in the Central Equatorial Pacific

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A persulfate wet-oxidation method was developed for measuring particulate organic nitrogen (PON) on 0.2 μm membrane filters and used to determine PON distributions in the central equatorial Pacific. The suitability of 6 types of membrane filters for persulfate oxidation was evaluated on the basis of filter blanks, chemical interference and adsorption characteristics. Concurrent comparisons were made using the bicinchoninic acid (BCA) protein assay. The 0.2 μm TF-200, teflon filters had low background N, no chemical interference with either assay, low adsorption of dissolved nitrogen and a higher trapping efficiency than glass fiber filters (Whatman GF/F).

The persulfate oxidation method was used to measure both >0.2 μm PON and dissolved organic nitrogen (DON) in the equatorial Pacific. For samples taken in the

mixed layer, the use of 0.2 μm filters resulted in PON estimates that were 60% higher than those obtained with traditional protocols using glass fiber filters. There was a strong PON gradient in the lower euphotic zone and no significant difference between >0.2 μm and GF/F PON estimates below the euphotic zone. Submicron PON (determined as the difference between >0.2 μm and GF/F PON) made up a significant portion of total PON (>0.2 μm) in the mixed layer. The sharp gradient and negligible sinking rate of submicron PON implies a strong diffusive flux of this material out of the mixed layer. Measurements of DON, chlorophyll and NO_3^- showed poleward increases in DON and detrital/heterotrophic N coincident with decreases in NO_3^-. These observations suggest that meridional advection of DON and detrital PON is an important mechanism for movement of nitrogen away from the point of equatorial upwelling. During an equatorial time series station, correlations were found between changes in the relative amounts of submicron and GF/F PON and changes in the concentrations of NO_3^-, NH_4^+ and Chl *a*. The dynamic nature of these particulate and dissolved constituents presumably reflect ongoing biological processes as the upwelled water "ages". Our results suggest that both submicron PON and DON are important nitrogenous components of equatorial Pacific water. More detailed information on the mechanisms of production and degradation of these pools of organic nitrogen is needed for a complete description of the equatorial nitrogen cycle.

Submicron Particulate Organic Nitrogen and Dissolved Organic Nitrogen in the Central Equatorial Pacific

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Chapter 1

INTRODUCTION

Particulate organic nitrogen (PON) is a bulk property of seawater commonly measured in oceanographic research. PON consists of a mixture of living and detrital particles. Coincident measurements of dissolved organic nitrogen (DON) are often made and it has been noted that the delineation between these two components is arbitrary (Sharp, 1973). The distinction between the dissolved and particulate phases is empirically determined by choice of filter. Criteria used for filter selection vary widely and depend not only on the particle size of interest but also on the chemical characteristics of the particles and the subsequent analysis to be conducted (Buffle et al., 1992). For biological studies, an important criterion is efficient trapping of biogenic particles. The two constituents most often measured in particulate material for biomass determinations and for normalization of rate measurements are carbon and nitrogen.

PON has historically been considered a minor component of the total N pool. In the open ocean, average estimates of the contribution of PON to total N range from < 1% in nutrient rich waters at depth to ~ 10% in nutrient depleted surface waters (Sharp, 1983). Despite its low concentrations, PON is a dynamic component of the N pool and the determination of PON standing stocks and the vertical and horizontal flux of PON is necessary for a detailed description of the biogeochemical cycle of nitrogen. DON concentrations usually exceed PON concentrations, and often exceed dissolved inorganic nitrogen concentrations in the upper 50-100 m of the water column. The role of DON in nitrogen fluxes is poorly understood due to a paucity of data and to a recent controversy over the accuracy of analytical techniques used in past studies (Suzuki et al., 1985; Hedges and Farrington, 1993). Nonetheless, recent biological studies (e.g. Eppley and Renger 1992) and regional modeling efforts (Najjar et al. 1992) support the notion of an important role of DON in the nitrogen cycle especially in the upper water column.

A variety of sampling techniques have been used to collect or measure PON. These methods include both discrete sampling (bottle and *in situ* pumps) and continuous (optical) profiling. Discrete water samples (1 to 30 L) collected by deployment of multiple bottle rosettes are used most often for coincident sampling of a suite of chemical and biological parameters including inorganic nutrients and a variety of biomass indicators. Less abundant large particles are missed by routine Niskin and Go-Flo bottles collections, but can be collected by large volume (~1000 L) *in situ* pump systems. Optical instruments are able to estimate particulate organic material (POM, analogous to PON) by measuring beam attenuation, changes in

adsorption and scattering characteristics, and provide a continuous profile of POM. If properly calibrated, beam attenuation measurements yield a high resolution representation of the vertical POM distribution. For small particles (< 50 μm), water collection with multiple bottle rosettes remains the most widely used technique to obtain samples for direct analysis of PON and dissolved constituents.

For the last decade particulate material has been collected on glass fiber filters for subsequent POC/PON analysis. The two most commonly used filters are Whatman GF/C and GF/F with nominal pore sizes of 1.2 and 0.7 μm respectively. It is now clear, however, that an abundance of living and non-living submicron particles are present in seawater. Autotrophic picoplankton (Waterbury et al., 1979; Johnson and Sieburth, 1979; Li et al., 1983; Chisholm et al., 1988), heterotrophic bacteria (Lee and Fuhrman, 1987; Fuhrman et al., 1989; Cho and Azam, 1990) and viral particles (Bergh et al., 1989; Proctor and Fuhrman, 1990) are plentiful and a major component of living biomass in open ocean waters. Biogenic submicron detrital particles are also abundant in seawater. Particle abundance in the 0.38-1.0 μm size class is approximately 10^8 particles/ml, and less than 5% of these submicron particles are accounted for as picoplankton cells (Koike et al., 1990). Despite the recognized importance of picoplankton and the abundance of associated submicron particles, techniques for direct analysis of their contribution to POC and PON have not been developed.

Analysis of PON collected on filters is accomplished by oxidation of the organic material and the subsequent measurement of the oxidized N species. A variety of dry-combustion (e.g., CHN analysis) and wet-oxidation (e.g., persulfate

digestion) techniques have been used. Recent work has indicated that the persulfate wet-oxidation technique is comparable to CHN analysis (Raimbault and Slawyk, 1991). The research that follows in Chapters II & III was undertaken using persulfate wet-oxidation for the analysis of both particulate and dissolved organic nitrogen in bottle-collected water samples from the equatorial Pacific.

The major goal of this study was to develop a method for direct measurement of submicron PON using 0.2 μm membrane filters and to compare the 0.2 μm PON estimates with conventional GF/F estimates. The spatial distributions of the >0.2 μm, >0.7 μm and submicron (0.2 to 0.7 μm) PON were determined and compared in the central equatorial Pacific. Additional samples were taken for the analysis of NO_3^-, DON and Chl *a*. Correlations between spatial and temporal variations in the concentrations of these inorganic and organic constituents are used to evaluate the relative importance of PON and DON in recently upwelled water at the equator and for qualitative evaluation of vertical and horizontal nitrogen fluxes. Method development is detailed in Chapter II. The field work is presented and discussed in Chapter III. A summary of the conclusions is given in Chapter IV. Compilations of the data from each sampling station are listed in Appendices 1 & 2.

Chapter II

A WET-OXIDATION METHOD FOR DETERMINATION OF PARTICULATE ORGANIC NITROGEN ON GLASS FIBER AND 0.2 µm MEMBRANE FILTERS

Particulate organic carbon and nitrogen are routinely measured with glass fiber filters (Whatman GF/F; Feely et al., 1991). These filters miss approximately 30% of PON and 40% of POC in open ocean waters (Altabet, 1990). Biological biomass measurements may also be underestimated as a consequence of using glass fiber filters. It is estimated that GF/F filters miss ~50% of the bacteria in natural assemblages (Lee and Fuhrman, 1987; Kirchman et al., 1989). Similarly, in oligotrophic regions where autotrophic picoplankton are important, it is estimated that GF/F filters fail to retain 40% (Phinney and Yentsch, 1985) to 74% of the Chl *a* (Dickson and Wheeler, 1993). The importance of picoplankton to the total biomass in oceanic waters has been well-documented (Li et al., 1983; Herbland et al., 1985; Lee and Fuhrman, 1987; Cho and Azam, 1988; Fuhrman et al., 1989; Li et al. 1992), and it has been suggested that 0.2 µm filters would yield more accurate estimates of picoplankton biomass (Li et al., 1983; Fuhrman et al., 1989). Submicron filters would also retain the numerous nonliving submicron particulates

associated with the picoplankton (Koike et al., 1990). Since glass fiber filters fail to retain a significant portion of picoplankton and other submicron particles, their use has led to an underestimate of PON, POC and Chl *a*.

The main goal of this study was to assess the suitability of a variety of 0.2 µm membrane filters for PON analysis. The filters were evaluated based on N background, interference with chemical analyses, and N adsorption. A secondary goal of this study was to determine if the bicinchoninic acid (BCA) protein assay (Smith et al., 1985) could be used as a suitable alternative for PON analysis. The BCA protein assay is a fast, relatively simple technique and is well-suited for shipboard oceanographic applications. Only 1 of the 6 types of membrane filters tested was suitable for both the persulfate and BCA assays; this was the TF-200, teflon filter. In direct comparison with the persulfate wet-oxidation method, the BCA protein assay yielded quantitatively comparable PON estimates for coastal waters. Finally, in a comparison with the GF/F filter, the higher trapping efficiency of the 0.2 µm TF-200 filter resulted in a 4-fold increase in PON estimates.

METHODS

Reagents and preparation of glassware

Reagent grade chemicals were used for all analyses. Aqueous solutions were prepared with deionized distilled water (DDW) obtained from a Millipore Milli-Q system. Pyrex and Kimax screw cap test tubes (50 ml), used with the wet-oxidation method, were baked at 500 °C for 6 h and treated with dimethyldichlorosilane (Sigma 3879). The glass test tubes, centrifuge tubes, and sample vials were acid washed and DDW rinsed between experiments. The 7-ml glass digestion vials used in the protein assay were washed in a dilute NaOH solution (0.1 M), rinsed with DDW, and combusted at 500 °C overnight.

Persulfate analysis

The persulfate oxidation method used was a modification of the semiautomatic method described by Raimbault and Slawyk (1991). The oxidizing reagent was prepared with recrystallized potassium persulfate, $K_2S_2O_8$ (Baker 3239). Potassium persulfate (160 g) was dissolved in 1 L of DDW at 70-80 °C, and then recrystallized by cooling to ~0 °C. The crystals were collected and then dried at 45 °C. The working oxidation reagent was prepared daily by dissolving 5 g recrystallized persulfate in 15 ml of 3.75 N NaOH and bringing the volume to 100 ml with DDW (pH >13). The solution was stirred for 1 h to assure complete dissolution, and then filtered through a GF/F filter to remove any particulate material.

The recrystallizing and reagent filtering steps reduce the variability and intensity of the reagent blank signal by at least a factor of two.

The Tris buffer solution was prepared daily by dissolving 15.8 g Trizma-HCL (Sigma 3253) in 100 ml of DDW (1 M, pH 3.2) (Raimbault and Slawyk, 1991). Although the Tris buffer is reportedly stable for long periods of time, a significant increase in the reagent blank was found after more than 1 d storage. The reagents used in NO_3^- + NO_2^- analysis on the Technicon AutoAnalyzer II (AAII) were prepared according to Atlas et al., 1971. Protein standards were prepared using bovine serum albumin (BSA, Pierce 23209) which has a protein concentration of 2 mg/ml and is ~16% N by weight.

The primary BSA standard (2 mg/ml) was diluted 1:9 with DDW. Aliquots of the secondary standard (0.2 mg/ml) were used to achieve final concentrations of 0, 7.7, 15.3, 23.0, and 38.0 µM N for analysis on the AAII. The protein standards and filter samples were placed in 50-ml test tubes with 10 ml of DDW and 5 ml of persulfate reagent. The test tubes were tightly sealed and digested for 4 h at 120-125 °C and >15 PSI.

After digestion, samples were cooled, mixed well, and 3.5 ml aliquots were transferred into 14-ml glass centrifuge tubes. The alkaline samples (pH >12) were buffered with 0.5 ml of 1 M Tris buffer to achieve a pH between 8 - 8.5 and 3.0 ml of DDW was added. The buffered samples were centrifuged for 2 min at 2000 rpm to remove filter debris, and the supernatant was transferred from each to a 20-ml glass sample vial. Total N was determined by analysis of NO_3^- + NO_2^-.

BCA protein assay

The reagents for the BCA protein assay (Pierce 23235) consist of an alkaline solution containing bicinchoninic acid and a 4% $CuSO_4$ solution. The two solutions were mixed 50:1 [alkaline:4% $CuSO_4$] to produce the BCA working reagent, which was prepared daily. Protein standards for this assay were also prepared using BSA (2 mg/ml), and a secondary standard (diluted 1:9, 0.2 mg/ml) was used to prepare the range of standard concentrations; 0, 3.2, 4.8, 6.4, and 8.0 µg N.

The filter samples and standards were placed in 7-ml vials, and 2 ml of DDW was added to the samples, while an appropriate volume of DDW was added to the standards to achieve a volume of 2 ml. After adding 2 ml of the BCA working reagent, the samples were mixed, capped or covered with foil and placed in a 60 °C water bath for 30 min. After color development the samples were cooled, thoroughly mixed and sample absorbance was measured at 562 nm. The absorbances of the standards were read before and after reading the sample absorbances to check for changes in the extent of color development. During the time required to complete the analysis (~15 min for 30 samples), no significant change was detected.

Comparison of glass fiber and membrane filter

A variety of 0.2 µm membrane filters (47 mm) were evaluated for use with the persulfate oxidation and the protein assay (Table I.1). Whatman GF/F glass fiber filters (47 mm) were used as the primary comparison. This filter is commonly used in determining PON and POC because of its low N and C background and fast flow

TABLE II.1 Types of 0.2 μm, 47 mm membrane filters analyzed.

Filter brand name	Membrane composition	Manufacturer
FP Vericel	fluoropolymer	Gelman
Nylaflo	nylon	Gelman
PCTE (plain)	polycarbonate	Poretics
PVPF (PVP-free)	polycarbonate, without wetting agent polyvinylpyrrolidone	Poretics
Supor	polysulfone	Gelman
TF (PTFE)	polytetrafluorethylene (teflon)	Gelman

rate. Tests were run to evaluate the filters for background nitrogen and protein levels, chemical interference, and adsorption of dissolved/colloidal organic nitrogen.

Prior to use, GF/F filters were precombusted at 500 °C for 2 h. The membrane filters, with the exception of the FP-200 filters, were used directly as supplied from the manufacturer without treatment. Both untreated and 95% EtOH washed FP-200 filters were analyzed. In the filtration experiments, both untreated and handled filters were used as blanks. The handled blanks were processed in the same manner as sample filters, barring actual filtration of a sample. The TF-200 filters are hydrophobic and were wetted with 95% EtOH (1 ml) just before filtration.

Double filter and multiple volume experiments

Fresh seawater was obtained from Hatfield Marine Science Center, Newport, OR. The samples were stored at 15 °C for ~6 h before filtration. In the double filter experiment, 100 ml of sample was filtered through a stack consisting of two membrane filters separated by a GF/F filter to maintain high flow rates ($n = 4$ for TF-200 and $n = 3$ for PVP-free), and 500 ml was filtered through the GF/F filter stack ($n = 3$). The filters were analyzed using the BCA protein assay.

In the multiple volume experiment, 50, 100, and 200 ml samples were filtered through membrane filters. PON concentrations were determined using both the persulfate oxidation method and the BCA protein assay. Concurrently, retention of PON on GF/F filters was determined by filtering 500 ml of sample. The pressure applied during vacuum filtration was < 200 mm Hg.

Sample storage

The effects of sample storage were evaluated by comparing the PON estimates of fresh and stored replicate GF/F and TF-200 samples. Fresh samples were run within 2 h of filtration. The stored samples were either dried at 45 °C or frozen at -23 °C for 24 h. In a second experiment, samples were frozen for a range of time periods (1 d to 2 mo) to determine the effect of extended storage. Filters samples were placed in vacutainers before storage.

RESULTS

Filter blanks

Persulfate analysis

The GF/F, TF-200, and PVP-free filter blanks were low (0.021 - 0.040 µmol N, Table II.2) and were not significantly different from one another (Student's *t* test, *p*-value > 0.10). These three filters had significantly lower blanks than the plain Poretic and FP-200 filters (*p*-value < 0.001), which had N blank values more than five times greater than the GF/F filter blank. Supor and Nylaflo filters had background N levels that were much greater than the highest standard used in our calibrations (1.14 µmol N). Because of their high N content, Supor and Nylaflo filters were not included in subsequent tests or comparisons.

BCA protein assay

Only GF/F and TF-200 filters had low blanks (0.065 and 0.026 µmol N respectively, Table II.3). The protein N blanks for the GF/F and TF-200 filters were not significantly different from each other (*p*-value > 0.10). The untreated FP-200 filters had high protein N blanks (0.655 µmol N), but an EtOH rinse reduced the blanks by 50%. The protein N blanks for the Poretic (plain and PVP-free) and EtOH washed FP-200 filters were, on average, 5 times greater than the blanks for the GF/F and TF-200 filters.

TABLE II.2 Nitrogen background comparison for 47 mm filters analyzed by the persulfate method.

Filter	µmol N / filter	SD	n
GF/F	0.040	0.016	5
TF-200	0.021	0.007	5
PVP-free Poretic	0.025	0.016	4
Plain Poretic	0.167	0.021	5
FP-200 (untreated)	0.261	0.021	5
Supor-200	> 1.14		
Nylaflo	> 1.14		

TABLE II.3 Nitrogen background comparison for 47 mm filters analyzed using the BCA protein assay.

Filter	µmol N / filter	SD	n
GF/F	0.065	0.015	3
TF-200	0.026	0.015	11
PVP-free Poretic	0.115	0.017	36
Plain Poretic	0.246	0.010	8
FP-200	0.655	0.110	5
(EtOH washed)	0.297	0.082	7

Chemical interference

Persulfate analysis

To assess chemical interference, the PON results for a series of protein standards with and without filters were compared. Persulfate oxidation of protein standards gave linear results up to 40 µM N and, for these three experiments, resulted in a mean slope of 1.12 ± 0.04 and intercept of 0.04 ± 0.01 µmol N (Fig. II.1a). The standard curves with GF/F and TF-200 filters were not significantly different from one another (intercepts, p-value > 0.30; slope, p-value > 0.20) and had very good linear fits (r^2 > 0.99). Their slopes were similar to the standard curves without filters, indicating minimal chemical interference by the filters (filter/standard slopes ratios of ~1, Table II.4).

The low intercept for the FP-200 filter suggests a low blank value, but the intercept of the standard curve with filters was lower than the intercept without filters, suggesting chemical interference. The slopes of the standard curves for both types of the Poretic filters were approximately half that of the standards without filters, indicating significant chemical interference. The variability associated with the intercepts and slopes of these three filters was quite high.

BCA protein assay

The chemical interference results for the BCA protein assay were similar to those discussed above. The protein assay gave linear results over the range of protein standards, up to 50 µg protein (or ~0.6 µmol N, Fig. II.1b). None of the 5 filters showed significant chemical interference, as the slopes of the standard curves with filters were similar to those without filters (filter/standard slope ratios range

TABLE II.4 Standard curves with and without filters analyzed by the persulfate method over the course of three experiments.

	Intercept (µmol N)	SE	Slope	SE	Filter slope / Standard slope	r^2	n
Standards without filters							
Expt. 1	0.053	0.023	1.165	0.018		0.998	10
Expt. 2	0.034	0.030	1.090	0.024		0.996	10
Expt. 3	0.030	0.042	1.094	0.034		0.993	10
Standards with filters							
GF/F (expt. 1)	0.078	0.032	1.147	0.037	0.98	0.997	5
TF-200 (expt. 3)	0.041	0.030	1.060	0.022	0.97	0.996	11
FP-200 * (expt. 3)	-0.006	0.099	0.974	0.075	0.89	0.950	11
Plain (expt. 1)	0.218	0.227	0.509	0.259	0.44	0.564	5
PVP-free (expt. 2)	0.016	0.083	0.508	0.067	0.47	0.879	10

* untreated filters (FP-200 EtOH wash resulted in colored digest that interfered with analysis.)

TABLE II.5 Standard curves with and without filters analyzed using the BCA protein assay over the course of two experiments. Intercept values presented in absorbance units (abs.) at 562 nm.

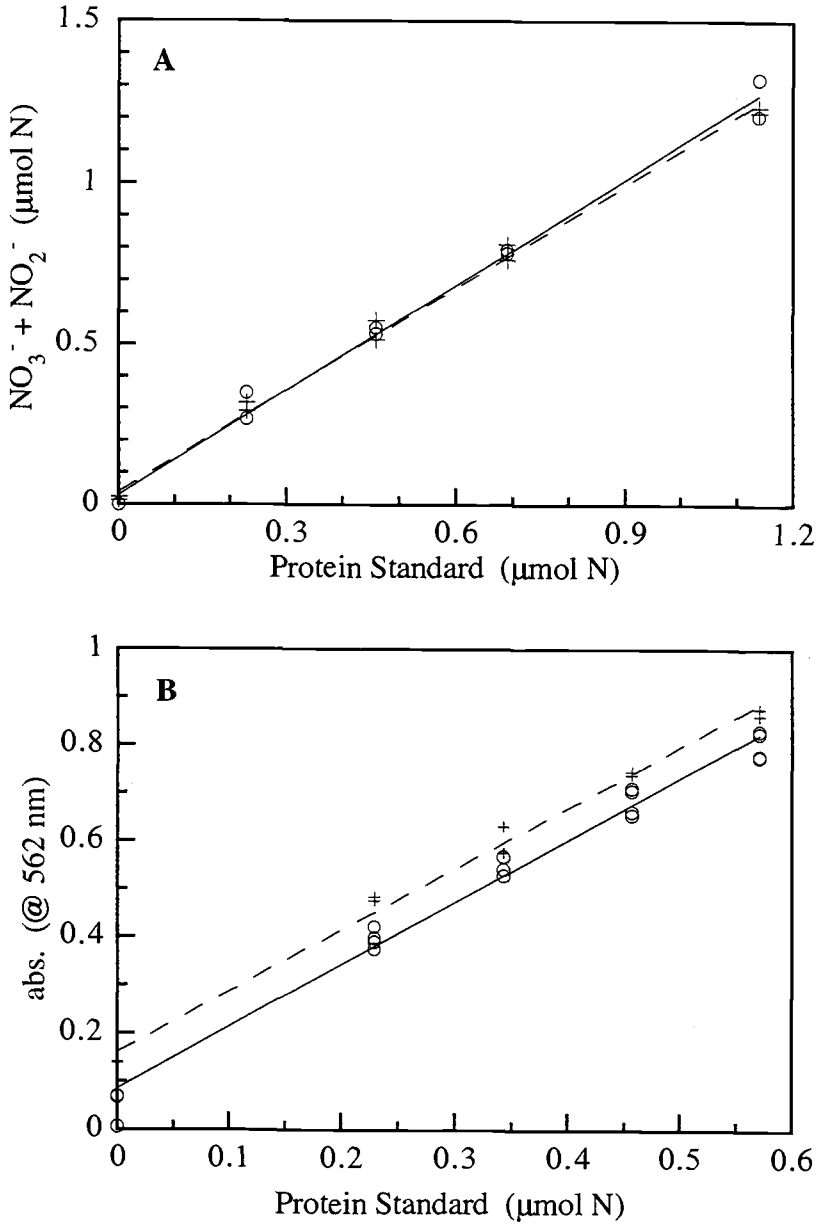
	Intercept (abs.)	SE	Slope	SE	Filter slope / Standard slope	r^2	n
Standards without filters							
Expt. 1	0.071	0.020	1.067	0.019		0.993	24
Expt. 2	0.086	0.027	1.290	0.030		0.991	19
Standards with filters							
GF/F (expt. 1)	0.171	0.043	0.949	0.058	0.89	0.964	12
Plain (expt. 1)	0.339	0.015	1.018	0.014	0.95	0.996	24
PVP-free (expt. 1)	0.217	0.030	0.942	0.024	0.88	0.985	24
FP-200 * (expt. 2)	0.337	0.035	1.100	0.057	0.85	0.979	10
TF-200 (expt. 2)	0.161	0.025	1.270	0.040	0.98	0.992	10

* EtOH washed

Figure II.1 A) Protein standards with (+) and without (o) TF-200 filters analyzed by the persulfate method. Dashed and solid regression lines represent the standards with (y-intercept = 0.041, slope = 1.060) and without TF-200 filters (y-intercept = 0.030, slope = 1.094). The output of the Technicon AA II has been converted into µmol N by calibration with a series of undigested NO_3^- standards.

B) Protein standards with (+) and without (o) TF-200 filters analyzed using the BCA protein assay. Dashed and solid regression lines represent the standards with (y-intercept = 0.161, slope = 1.270) and without TF-200 filters (y-intercept = 0.086, slope = 1.290).

Figure II.1


A
1.5
1
0.5
0
0
0.3
0.6
0.9
1.2
$NO_3^- + NO_2^-$ (µmol N)
Protein Standard (µmol N)
B
1
0.8
0.6
0.4
0.2
0
0
0.1
0.2
0.3
0.4
0.5
0.6
abs. (@ 562 nm)
Protein Standard (µmol N)

from 0.89 to 0.98, Table II.5). The GF/F and TF-200 filters had significantly lower absorbance blanks than the other three filters (p-value < 0.001 for each) and were not significantly different from one another (p-value > 0.40).

Double filter and multiple volume experiments

These experiments were performed to evaluate the degree of N adsorption of the various filters. The double filter experiments were interpreted by assuming that the top filter yields a PON estimate based on trapped particles plus retained (adsorbed or trapped) dissolved organic nitrogen. The nitrogen content of the bottom filter is then presumed to be a quantitative measure of the adsorbed dissolved material. DON is simply defined here as the <0.2 μm organic N fraction, and no attempt was made to characterize it further into truly dissolved or colloidal (<0.2 μm particulates) groupings. The data for the multiple volume experiments were interpreted based on the quantitative relationship between N measured and sample volume filtered.

In the double filter experiments, the N content of the bottom filters for both the GF/F and TF-200 filters was low and similar to handled filter blanks. The bottom GF/F filters (0.015 ± 0.003 μmol N, mean ± SD) accounted for 7.9% of the N content of top filters (0.190 ± 0.051 μmol N), while the bottom TF-200 filters (0.017 ± 0.005 μmol N) accounted for 14.3% of the N content of top filters (0.119 ± 0.040 μmol N). As the absolute value of N in bottom filters was small and similar to the handled blanks, subtraction of the handled blank from measured PON for single filter measurements should provide a good correction for filter adsorption of

DON by GF/F and TF-200 filters. In contrast, bottom PVP-free filters retained 0.57 ± 0.035 µmol N while top filters retained 0.70 ± 0.011 µmol N. Although the difference between the top and bottom filter gave a similar estimate of PON for both the PVP-free and TF-200 filters, 81% of the measured N (i.e. not corrected for background adsorption) for PVP-free filters appeared to result from adsorption of DON.

The results from the multiple volume experiment for the TF-200 filters indicated a direct proportionality between N measured and volume filtered (Fig. II.2). The intercepts of the regression lines in Fig. II.2 were 0.017 and 0.024 µmol N for the persulfate method and the protein assay, respectively. The direct proportionality between N measured and volume filtered, in combination with the low intercepts, further indicates that adsorption is minimal and that the N retained is primarily due to particulates. In contrast, the results for the PVP-free filters indicated a strong propensity for N adsorption. The increases in PON, resulting from a doubling of the sample volume, were quite small (average < 15%, Fig. II.2), and the intercept of the regression was 0.39 µmol N. This high level of N adsorption makes it nearly impossible to distinguish the PON signal from that of the PON + adsorbed DON for the PVP-free filters.

Sample storage

The first storage experiment compared two methods of sample storage, drying and freezing. The results indicated the TF-200 filters were not amenable to being oven dried. The heat caused the polypropylene support mesh of the TF-200 filters to

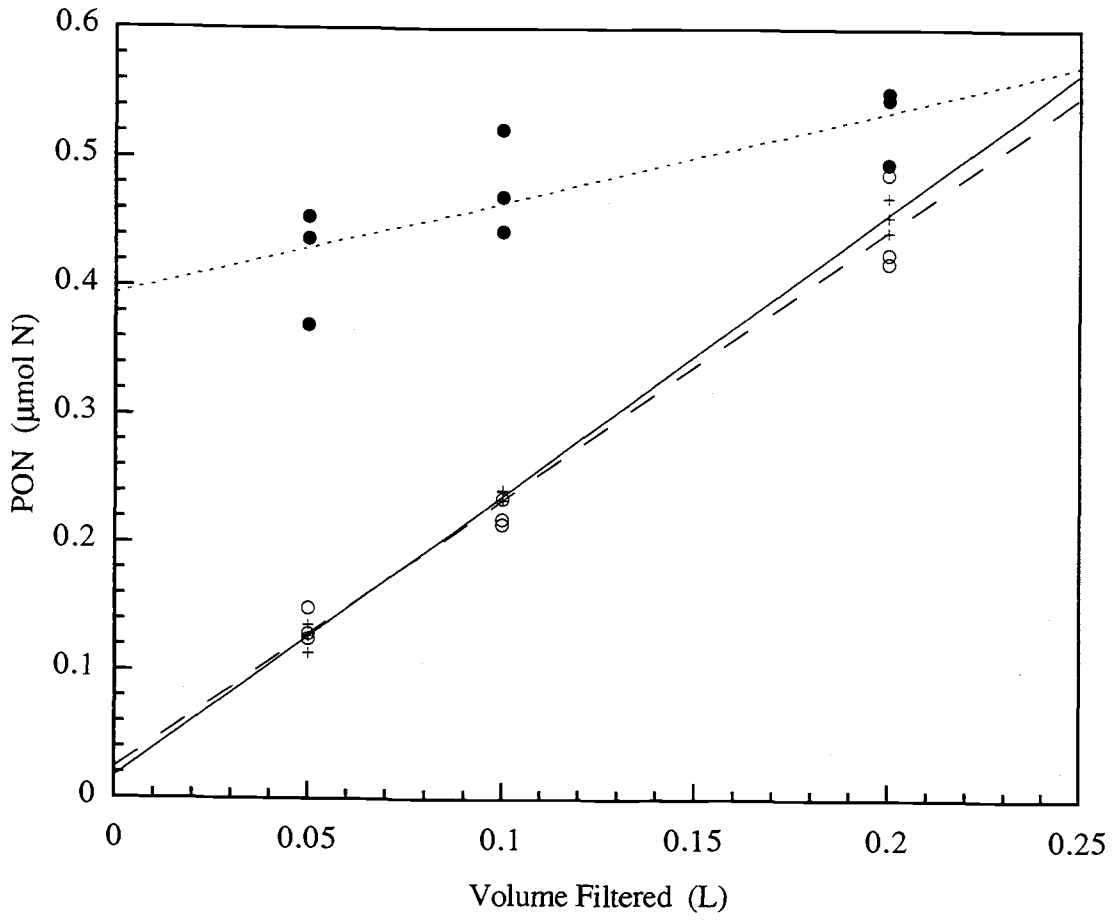


Figure II.2 Multiple volume filtration experiment results. TF-200 filters analyzed using both persulfate (○) and BCA (+) assays (solid, *y*-intercept = 0.017, slope = 2.20; and dashed, *y*-intercept = 0.024, slope = 2.09, respectively). PVP-free filters analyzed by the BCA protein assay (●; dotted, *y*-intercept = 0.39, slope = 0.71).

contract, forming isolated pockets of sample. This resulted in low and variable PON estimates (data not shown).

An experiment comparing freshly filtered samples with samples that had been frozen for 1 d to 2 mo was conducted to determine the effect of frozen storage. The persulfate PON estimates were only minimally affected by frozen storage (data not shown). The change in GF/F PON estimates was negligible, < 5% for 2 mo of storage, and the TF-200 estimates decreased by 13% over that time span. The high variability in the protein assay results prevented drawing any conclusions about storage effects. The ease of the BCA protein assay, however, negates the need to store samples. Further experiments are being conducted to more clearly characterize the storage effects.

Comparison of PON and protein assays

This study compared PON estimates determined by the persulfate and protein methods using TF-200 filters for samples from the multiple volume experiments (Fig. II.3). The low volume requirement for the TF-200 filters is clearly illustrated by the good agreement of PON concentrations for sample volumes ranging from 50 to 200 ml. There was also excellent agreement between the two assays at each of the three sample volumes. Moreover, the results indicated a 4-fold difference between PON concentrations for 0.2 µm TF-200 and 0.7 µm GF/F filters, demonstrating a higher PON retention efficiency for the TF-200 filters.

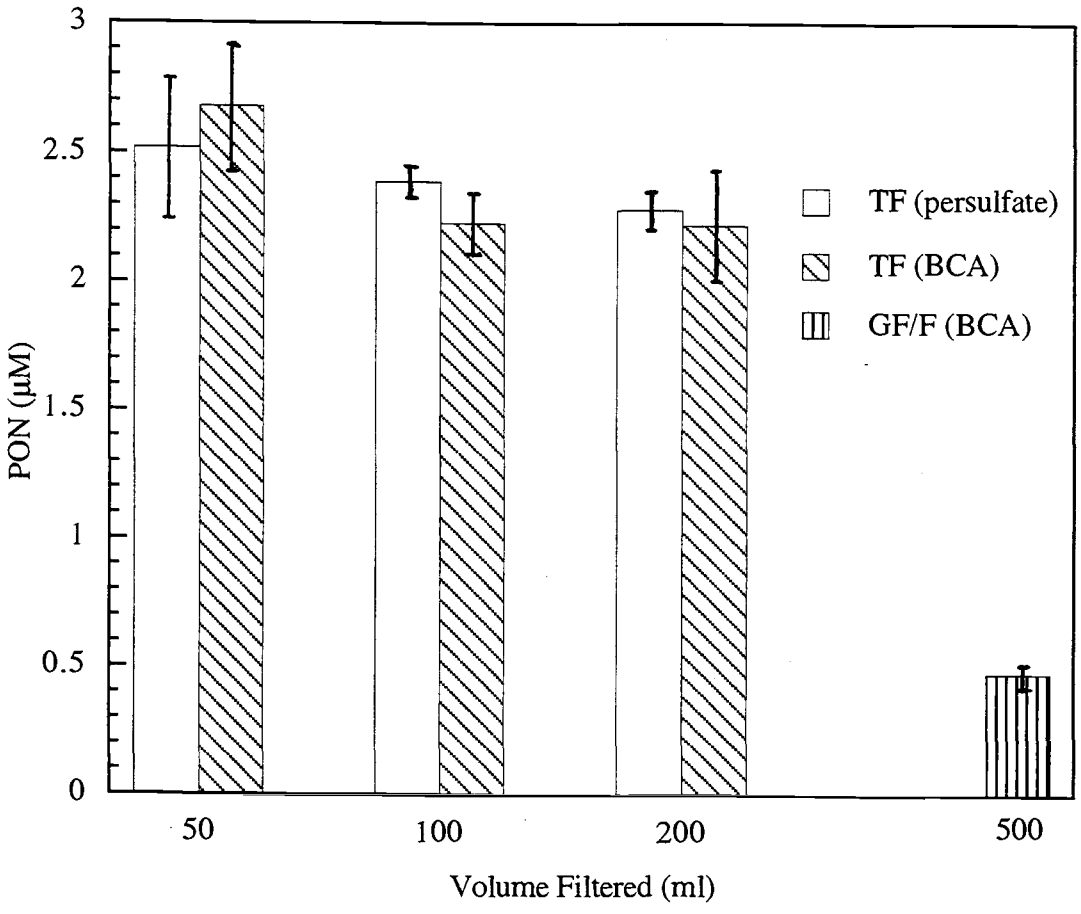


Figure II.3 Sample µM N measured plotted against volume filtered for TF-200 and GF/F filters analyzed using both the persulfate method and the BCA protein assay. The error bars represent the standard error (n = 3).

Filtration rates

Fast filtration rates are one of the primary reasons that GF/F filters have gained prominence in particulate measurements. Typical flow rates for GF/F filters are about 1 L/min in oligotrophic waters and 100 ml/min in productive oceanic and coastal waters for vacuum pressures < 200 mm Hg. Flow rates for the TF-200 filters were slower, 10 to 20 ml/min for coastal waters. However, the actual filtration times were dependent upon the volume filtered. The higher trapping efficiency and low N blank of the TF-200 filters allow sample volumes to be < 20% of that needed for PON analysis using GF/F filters. This easily compensated for the slower flow rates and resulted in comparable filtration times, 1 to 10 minutes for GF/F filters and 5 to 10 minutes for TF-200 filters.

DISCUSSION

Filter characteristics

Inorganic filters are most often used for analysis of particulate organic material. Two types of 0.2 µm inorganic filters are available and have been examined during previous experiments -- silver and aluminum oxide (Anopore). Problems were encountered with both types of inorganic filters. As reported by Gordon and Sutcliffe (1974), silver filters had high blanks even after combustion. Moveover, silver filters failed to retain more than 50% of bacteria in seawater samples (D. Kirchman, pers. comm., 1991). Anopore filters have low C and N background after combustion (Altabet, 1990), but could not be used with the persulfate oxidation as described here. The aluminum oxide caused a large drop in the pH during digestion and resulted in incomplete oxidation of the samples. It is possible that the background of silver filters could be reduced by a chemical wash procedure, but the failure of these filters to retain bacteria diminishes their suitability for trapping submicron PON. Similarly, it might be possible to modify the persulfate oxidation to eliminate the drop in pH for aluminum oxide filters. However, preliminary studies (Wheeler unpub.) suggested that the aluminum oxide filters have a high adsorption capacity for dissolved proteins which makes them unsuitable for submicron PON analysis.

This study investigated the use of organic (membrane) filters as an alternative for trapping submicron PON. The filter evaluation showed that 5 of the 6 membrane filters had high N blanks and/or interfered with the chemical analysis. The TF-200

filter was the only filter tested that was applicable to the persulfate method without a pre-filtration cleaning treatment. The N blank of the TF-200 filter was comparable to the GF/F filter (0.021 and 0.040 μmol N respectively) and there was no indication of chemical interference with either filter. Moreover, the teflon filters required much smaller sample volumes than those needed with the GF/F filters. The smaller sample volume alleviated the problem of slower flow rates, and filtration times were generally 5-10 min.

The adsorption of N by filters was evaluated in the multiple volume and double filter experiments. Both approaches have been applied in the investigation of C and N adsorption previously (Menzel, 1966; Menzel, 1967; Banoub and Williams, 1972; Sharp, 1974; Gordon and Sutcliffe, 1974; Feely, 1975). Characterization of the adsorbed material has been incomplete. The contributions of dissolved (Menzel, 1966; Menzel, 1967), colloidal (Sharp, 1974), and small particles (Bishop and Edmond, 1976) to measured PON have been discussed, but the relative importance of each has not been determined. The results in this study indicate that adsorbed N contributes minimally to the PON estimates for both TF-200 and GF/F filters. Apparent DON adsorption on Poretic PVP-free filters, however, contributed 81% of the measured PON + DON concentration. The low N background and low N adsorption characteristics of the TF-200 filters match the characteristics of the GF/F filters and make them highly suitable for analysis of submicron PON.

Comparison of persulfate oxidation and BCA assay for PON

The BCA protein assay yielded PON estimates that were quantitatively comparable to the persulfate wet-oxidation method for the coastal water studied. The BCA assay has the benefits of both being a simple technique and providing real-time PON estimates. However, the variable response of different proteins to this analysis (Smith et al., 1985) could pose serious problems for standard calibration. This problem and the BCA versus persulfate PON comparisons are addressed in more detail in chapter III.

Significance of submicron PON

It is now clear that an abundance of living and non-living submicron particles are present in seawater. Autotrophic picoplankton (Li et al., 1983; Herbland et al., 1985; Peña et al., 1990; Li et al., 1992; Dickson and Wheeler, 1993), heterotrophic bacteria (Fuhrman et al., 1989; Cho and Azam, 1990) and viral particles (Bergh et al., 1989; Proctor and Fuhrman, 1990) are plentiful and a major component of living biomass in oligotrophic waters. Despite the recognized importance of picoplankton, techniques for direct analysis of their contribution to POC and PON have not been developed. Bacteria that pass through GF/F filters can be enumerated and, by using biomass conversion factors (Lee and Fuhrman, 1987), the missed bacterial C and N can be estimated. This missed bacterial POC and PON is then added to the measured POC and PON on GF/F filters to estimate total POC and PON (Cho and Azam, 1988). Using this technique, Cho and Azam conclude that heterotrophic bacteria account for 40% of POC in the Pacific. However, analysis of particle

abundance in the 0.38-1.0 μm size range indicates that bacteria account for less than 5% of these particles (Koike et al., 1990). Obviously, accurate determination of the contribution of bacteria (and other picoplankton) to POC and PON will require direct estimates of POC and PON of the appropriate size classes. The method for PON analysis on 0.2 μm filters outlined in this chapter will allow a direct estimate of PON which includes the picoplankton size class of particles.

PON has historically been considered a minor component of the total N pool. Estimates of the contribution of PON to total N in open ocean waters ranges from ~10% in nutrient depleted surface waters to ≪1% for nutrient rich waters below the surface mixed layer (Sharp, 1983). These figures are based on PON estimates derived using current standard protocols (i.e. filtration with GF/F filters). The preliminary results of this study indicate a four-fold increase in PON estimates using 0.2 μm TF-200 filters in comparison with GF/F filters. These results suggest that PON in the 0.2 - 0.7 μm size class is a significant component of the nitrogen pool and that a comprehensive study of this 'new' PON pool is necessary for detailed nitrogen budgets.

The persulfate oxidation method described here can be used to determine >0.2 μm PON on TF-200, teflon filters. In combination with other biomass measures, the persulfate determination of >0.2 μm PON will allow a more accurate comparison of picoplankton standing stocks to total PON concentrations. Moreover, preliminary estimates indicate that inclusion of submicron particulates results in a four-fold increase in measured PON concentrations. If this result is found to be

generally valid, then submicron PON is a major component of both PON and total nitrogen (particulate plus dissolved) and needs to be considered more carefully in studies of biogeochemical fluxes.

Chapter III

PARTICULATE AND DISSOLVED ORGANIC NITROGEN IN THE CENTRAL EQUATORIAL PACIFIC

One of the most notable features of the Equatorial Pacific is the 'cold tongue' of surface seawater stretching from the coast of South America to the central equatorial region (Wyrtki, 1981; Bryden and Brady, 1985). The high nitrate concentrations associated with this cool water have been described and used in a wide range of studies; determining the structure of meridional flow (Wyrtki and Kilonsky, 1984), estimating new production in the equatorial Pacific (Chavez and Barber, 1987) and tracking the passage of instability waves (Feely et al., 1993). In contrast, we have only a cursory knowledge about the distributions of dissolved and particulate organic nitrogen in this region. Particulate nitrogen distributions have been reported recently for equatorial transects at 135°W (Peña et al., 1991) and 150°W (Eppley et al., 1992). Thomas et al. (1971) measured dissolved organic nitrogen in the eastern tropical Pacific by UV-oxidation, but the concentrations and distribution of DON in this region have not been re-examined since the recent debate over the adequacy of older analytical methods (Suzuki et al., 1985; Hedges and Farrington, 1993). A comprehensive evaluation of both inorganic and organic N-

pools is important to achieve an understanding of the transport and regeneration of nitrogen in the equatorial Pacific (Sarmiento et al., 1988).

The central equatorial Pacific has persistently high nutrient concentrations and lower than expected chlorophyll levels (HNLC; Cullen, 1991). As in other HNLC regions (i.e. the subarctic Pacific and Southern Ocean), the phytoplankton biomass of the central equatorial Pacific is dominated by picoplankton (Li et al., 1983; Chavez, 1989; Peña et al., 1990). In addition, small (< 1 μm) particles have been shown to account for a majority of the PON measured in the equatorial Pacific (Peña et al., 1991; Eppley et al., 1992). Despite the recognized importance of picoplankton and submicron particulate material in oceanic regions, collection of particulates for biomass measurements are still commonly made using glass fiber filters (Whatman GF/F) with a nominal pore size of 0.7 μm (Feely et al., 1991). These filters are easily combusted to remove background organic carbon and nitrogen and have fast flow rates. However, in oceanic waters, GF/F filters fail to retain 50% of bacteria (Lee and Fuhrman, 1987; Kirchman et al., 1989), 40-74% of the Chl *a* (Phinney and Yentsch, 1985; Dickson and Wheeler, 1993) and 30-40% of POC and PON (Altabet, 1990). Clearly, better quantification of the standing stock of living and detrital particulate material in oceanic regions requires modification of current protocols for particle collection.

Resolution of discrepancies in mass balance comparisons, for both short term experimental work and large scale models of nitrogen fluxes, requires better information on the concentration and fluxes of DON and PON. Recent work in the central equatorial Pacific suggests that net NO_3^- depletion in incubation bottles

greatly exceeds the accumulation of nitrogen in PON (Eppley and Renger, 1992). As suggested by the authors, the discrepancy could result from unmeasured fluxes to DON and submicron PON. On a larger scale, mass balance of the fluxes of dissolved and particulate nitrogen in ocean circulation models requires the postulation of a significant but presently unmeasured flux of DON (Toggweiler, 1989; Najjar et al., 1992). Definitive tests of the potential importance of PON and submicron DON have been hampered by the lack of a suitable method for direct measurement of submicron PON and a small and questionable data set on the oceanic distribution of DON.

The use of 0.2 μm filters should result in more accurate estimates of picoplankton biomass and PON (Li et al., 1983; Fuhrman et al., 1989). The need for direct measurement of submicron PON has led to the development of the method outlined in Chapter II using 0.2 μm teflon, TF-200 filters. A modified persulfate oxidation which gives results comparable to CHN analysis (Raimbault and Slawyk, 1991) was used in the analysis of PON .

DON concentrations have been measured by persulfate oxidation (D'Elia et al., 1977; Solórzano and Sharp, 1980), UV-oxidation (Armstrong et al., 1966; Thomas et al., 1971), dry combustion (Gordon and Sutcliffe, 1973), high-temperature combustion (Walsh, 1989) and most recently by a high-temperature Pt-catalyzed oxidation (Suzuki et al., 1985). Although the controversy about the accuracy of the various analytical methods is not yet resolved, the older methods do seem to provide reliable results (Hansell and Ward, 1993). For our work in the central equatorial Pacific, we chose to use the persulfate oxidation technique for DON analysis. Maita

and Yanada (1990) reported good agreement between DON measurements made using persulfate oxidation and high-temperature catalytic oxidation methods. Our primary goal was to obtain sufficient data on the vertical distribution of DON along a series of north-south transects to determine if any significant variations occur. The documentation of any such variations will be a useful starting point for the development and testing of hypotheses regarding the source and fate of DON in this oceanic region.

In this study chlorophyll, DON, NO_3^-, NH_4^+, and both submicron and GF/F PON were measured in the equatorial Pacific along N-S transects at 140°, 125°, 110° and 95°W and an equatorial time series station at 140°W. The transect cruises provided information on the spatial distributions of these measurements, while the time series allowed for an investigation of temporal variability. The comparison of >0.2 μm and >0.7 μm PON estimates showed PON to be underestimated by the glass fiber filters. PON estimates in the mixed layer were ~1.6 times higher when measured using 0.2 μm filters compared with glass fiber filters. Data from the time series suggested that temporal variations in submicron PON may be the result of biological interactions.

METHODS

Sampling was conducted during the fall of 1992 on NOAA transect cruises aboard the R/V Discoverer (DI-92-03 & 04) and the NSF-EqPac time series station aboard the R/V Thompson (TT012). Twenty-three stations were sampled during transects along 95°, 110°, 125° and 140° W from 10°N to 10°S (Fig. III.1). Station positions and sampling times are listed in Table III.1. CTD casts were made to the 1,000 m depth using a 24-bottle rosette system equipped with 10 liter Niskin bottles (for stations 68 to 74 a 12-bottle CTD/rosette was used). Standard bottle depths were chosen prior to the cruise and our sampling concentrated on bottle depths in the upper 150 m. Time series samples were taken on the equator at 140°W between October 4 and October 20. Sampling was conducted during productivity casts to 120 m using Go-Flo bottles (General Oceanics).

PON Sample Handling and Filtration

One liter samples were taken from 12 depths (18 during high-resolution profiles) for the determination of PON, DON and NO_3^-+NO_2^-. Duplicate 10 ml sub-samples for DON and NO_3^-+NO_2^- measurements were immediately transferred to 30 ml teflon vials, frozen at -20 °C and analyzed upon return to OSU. Duplicate 100 ml aliquots were filtered through 25 mm 0.2 µm TF filters and a single 500 ml aliquot was filtered through a 25 mm GF/F filter to estimate PON for each sample. The GF/F filters were combusted at 500 °C prior to the cruise and the hydrophobic TF filters were wetted with 95% EtOH (1 ml) just before filtration. At half the

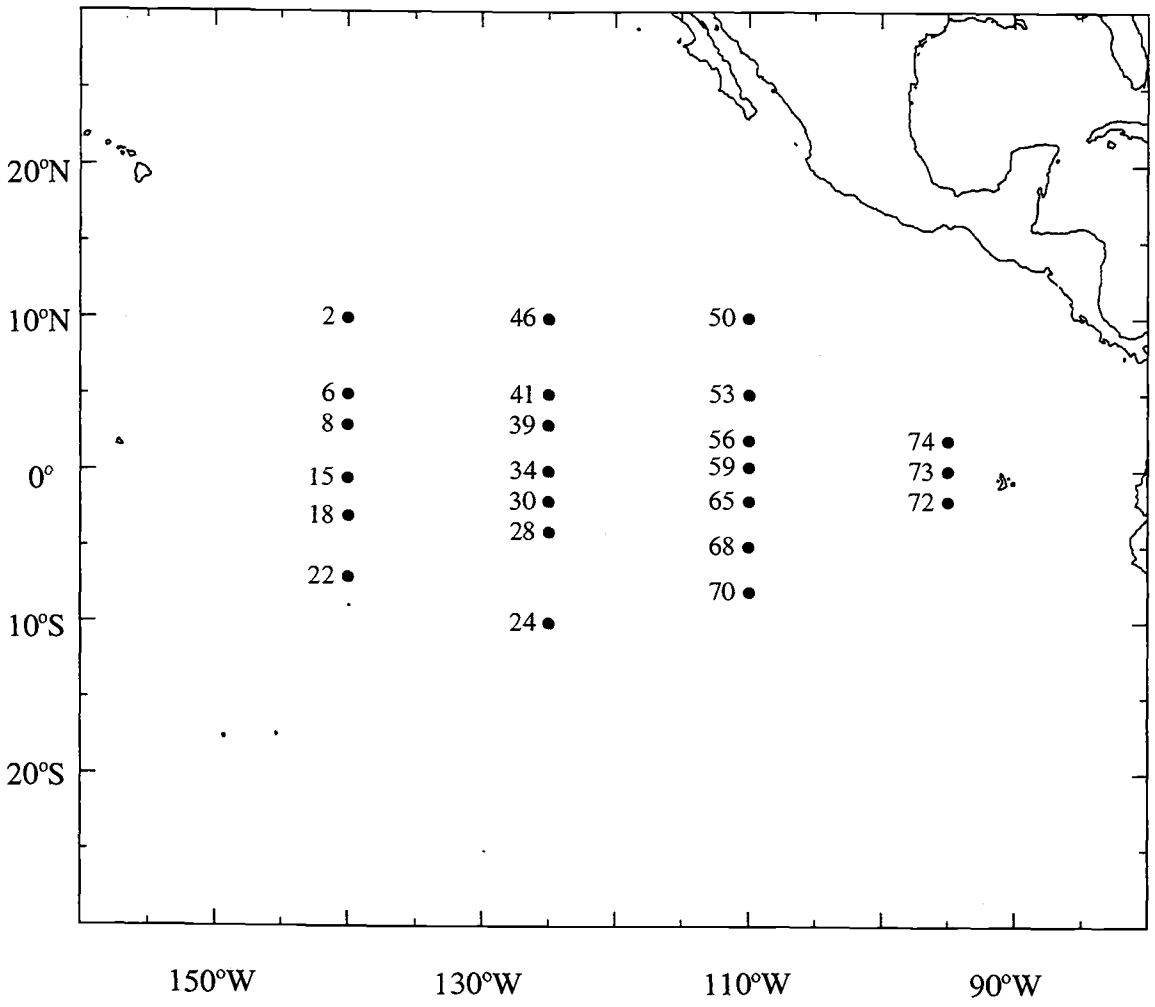


Figure III.1 Stations sampled aboard the R/V Discoverer on the NOAA EPOCS/CO_2/US JGOFS cruises DI-92-03 and DI-92-04 the fall of 1992.

TABLE III.1 Time and location of sampling stations.

Station #	Date	Local Time	Latitude	Longitude
2	9-09-92	18:17	10 00.38 N	139 59.45 W
6	9-11-92	06:42	5 00.39 N	140 03.33 W
8	9-12-92	10:53	2 59.35 N	140 07.21 W
15	9-14-92	23:42	0 29.73 S	139 59.92 W
18	9-16-92	02:47	2 59.88 S	140 00.10 W
22	9-17-92	09:20	7 00.05 S	140 00.16 W
24	9-20-92	12:40	10 00.05 S	125 00.03 W
28	9-23-92	10:34	4 00.11 S	125 00.00 W
30	9-24-92	01:06	1 59.98 S	125 00.03 W
34	9-26-92	06:55	0 01.35 S	124 57.66 W
39	9-28-92	15:52	2 59.86 N	125 00.26 W
41	9-29-92	18:11	5 03.12 N	125 00.90 W
46	10-01-92	18:37	9 59.28 N	124 59.36 W
50	10-16-92	22:15	10 01.08 N	109 56.85 W
53	11-01-92	10:47	4 58.14 N	109 54.98 W
56	11-02-92	06:43	2 06.75 N	110 06.80 W
59	11-03-92	19:28	0 15.26 N	109 59.98 W
65	11-06-92	05:31	2 05.60 S	109 54.10 W
68	11-08-92 deep	05:54	4 59.86 S	110 02.13 W
	shallow	07:45	4 59.95 S	110 01.72 W
70	11-09-92 deep	04:29	8 00.05 S	109 59.96 W
	shallow	06:19	7 59.68 S	110 00.66 W
72	11-13-92	15:10	2 00.00 S	95 00.00 W
73	11-14-92	17:35	0 00.86 S	95 03.34 W
74	11-15-92	14:29	1 57.28 N	94 09.05 W

stations, duplicate TF samples were also taken to estimate PON using the bicinchoninic acid (BCA) protein assay (Smith et al., 1985) as described in Chapter II. The pressure applied during vacuum filtration was < 200 mm Hg. Sample filters were placed in vacutainers and frozen at -20 °C. All protein samples were analyzed aboard the R/V Discoverer.

PON comparisons were also made on the NSF-EqPac time series cruise TT012. Water samples were taken at depths of 0, 10, 20, 35, 49, 64, 82 and 120 m during 8 Go-Flo casts over a period of 2 weeks. The filtration protocol was the same as outlined above. Nutrients, including NH_4^+, were analyzed using standard protocols on a continuous-flow analyzer.

Two strategies for estimating filter blanks were undertaken -- handled and refiltered blanks. Handled blanks were processed in the same manner as sample filters, without the actual filtration of a sample. The refiltered blanks were prepared by passing the filtrate from a previous sample through a second filter. The refiltered blanks had a high degree of unexplained variation, and I suspect this was due to contamination of the filtration apparatus used to retain the filtrate. The suitability of the handled filters as blanks for this method was discussed in Chapter II.

Chlorophyll a Measurements

Chlorophyll *a* was determined on CTD casts at Stations 2 and 8 to compare the Chl *a* measurements of GF/F and 0.2 µm Nuclepore filters. Chl *a* concentrations were measured using a Turner Designs fluorometer after extraction in 90% acetone at -20 °C in the dark for 24 h. The fluorometer was calibrated prior to the cruise using

a pure Chl *a* standard (Sigma), and a coproporphyrin standard (Sigma) was used to standardize the fluorometer during the cruise. Chl *a* data sets at the 140°W and 110°W transects have been provided by R. Barber and F. Chavez, respectively. These data were used to evaluate how the PON/Chl ratio changed with latitude.

Sample Analysis

The PON samples were analyzed using the persulfate oxidation method detailed in Chapter II. The method is a modification of the semiautomatic method described by Raimbault and Slawyk (1991). Persulfate digests were analyzed using standard techniques for $NO_3^-+NO_2^-$ on a Technicon Autoanalyzer II (Atlas et al., 1971). The BCA protein assay was used to estimate PON at half the stations. In direct comparisons with the persulfate method, the BCA protein assay gave consistently lower results (in the mixed layer, persulfate PON/ BCA PON = 2.04 ± 0.45). When no other PON data was available, PON was estimated from the BCA data and the estimated values are appropriately marked in the tables.

Persulfate oxidation (Solórzano and Sharp, 1980 as modified by Raimbault and Slawyk, 1991) was also used to digest DON samples. $NO_3^-+NO_2^-$ was determined in parallel undigested samples, and DON was calculated as total dissolved nitrogen (TDN) minus $NO_3^-+NO_2^-$. The digestion efficiency for the persulfate oxidations was estimated by comparing the slope of digested protein, bovine serum albumin (BSA) and digested NO_3^- standard curves. The digestion efficiencies (81.3 ± 4.8%, n=3) were used to correct the calculated DON values.

Deionized distilled water (Millipore Milli-Q system) was used in determining the reagent blank. In comparisons with HPLC grade water (Baker Analyzed), there was no significant difference in the reagent blanks (Student's t test, p=0.20).

RESULTS

The four transects encompassed at least two distinct regions of the equatorial Pacific. Barber and Chavez (1991) suggest that the Galapagos region should be considered separately from the central and eastern equatorial Pacific. They attribute the high primary production and Chl *a* standing stocks in the region to the west of the Galapagos (~90° to 93°W) to inputs of Fe and subsurface nutrients from the Galapagos platform. Changes in physical circulation patterns caused by the shoaling of the Equatorial Undercurrent near 95°W (Bryden and Brady, 1985) may also contribute to the characteristically high biomass and productivity measurements for this region. The other three transects showed consistent patterns in the distribution of nutrients and biomass, and therefore were grouped together for the presentation and discussion of results. The data for 95°W are reported here (Tables III.3 & 4 and Appendices), but are not included in the discussion of our results.

Particulate Nitrogen

PON was measured at 22 of 23 sampling stations. Of those measurements, particulate protein estimates of PON were used at 3 stations for the TF filters and 5 stations for the GF/F filters. Average PON concentrations in the mixed layer ranged from 0.9 to 1.6 µM for >0.2 µm PON and from 0.4 to 1.2 µM for >0.7 µm PON (Table III.2). Along 125° and 140°W, there were local minima in >0.7 µm PON at the equator (0.77 and 0.64 µM, respectively) and maxima within 3° to the

TABLE III.2 Average >0.2 µm and >0.7 µm PON concentrations in the mixed layer for each station. (n = # of sample depths)

Lat	Long	>0.2 µm PON Mean (µM)	>0.2 µm PON SD (µM)	>0.7 µm PON Mean (µM)	>0.7 µm PON SD (µM)	>0.2 µm PON / >0.7 µm PON	n
5°N	140°W	1.37	0.04	0.78	0.05	1.76	3
3°N	140°W	*1.10	0.06	*0.83	0.11	1.33	3
0° 30'S	140°W	1.20	0.13	0.64	0.00	1.89	2
3°S	140°W	1.09	0.09	0.88	0.07	1.24	2
7°S	140°W	*1.42	0.16	*0.67	0.11	2.12	2
10°N	125°W	1.06	0.03	0.74	0.13	1.43	2
5°N	125°W	1.33	0.08	0.90	0.01	1.47	2
3°N	125°W	1.27	0.00	0.90	0.02	1.41	2
0°N	125°W	1.14	0.16	0.77	0.17	1.48	3
2°S	125°W	1.21	0.02	0.90	0.07	1.34	2
4°S	125°W	1.25	0.04	0.64	0.09	1.96	2
10°S	125°W	0.90	0.01	0.62	0.05	1.45	2
10°N	110°W	*0.87	0.11	*0.38	0.06	2.30	3
5°N	110°W	1.21	0.10	0.55	0.08	2.20	5
2°N	110°W	1.04	0.13	0.61	0.08	1.71	5
0° 15'N	110°W	1.42	0.12	*0.81	0.10	1.75	5
2°S	110°W	1.55	0.12	1.16	0.06	1.33	5
5°S	110°W	1.21	0.03	0.89	0.02	1.36	2
8°S	110°W	1.35	0.06	0.94	0.09	1.44	5
						mean 1.63	
						SD 0.32	
2°N	95°W	1.60	0.05	*0.77	0.02	2.09	3
0°	95°W	1.89	0.81	1.62	0.76	1.17	3
2°S	95°W	1.48	0.29	1.06	0.31	1.39	3

* indicates PON values were estimated from BCA protein assay results.

north and south (0.88 ± 0.03 µM, n=4). Average concentrations of >0.2 µm PON were generally higher away from the equator.

The difference between >0.2 µm and >0.7 µm PON estimates was compared by calculating the ratio of >0.2 µm/>0.7 µm PON (Table III.2). On average, the TF PON estimates were 1.63 ± 0.32 times higher than the coincident GF/F estimates in the mixed layer. The uniform vertical distribution of PON in the mixed layer was evident in the low standard deviations (0.00 to 0.17 µM; Table III.2) of the mean TF and GF/F PON estimates. The mean coefficients of variance for PON estimates were 6.5% and 10.0%, respectively, in the mixed layer for each station. There were no apparent east-west trends in PON concentrations.

Representative PON profiles are shown in Figures III.2a & 2b. Submicron PON was calculated as the difference between total PON (measured on 0.2 µm TF filters) and >0.7 µm PON. The vertical distribution of PON was similar both near (15'N) and away (5°N) from the equator. The vertical distributions of the >0.7 µm, >0.2 µm and submicron PON were similar, with mixed layer maxima and characteristic decreases between 50 to 100 m. A significant portion of the PON gradient was driven by the submicron PON (0.2 to 0.7 µm) not retained by the GF/F filters.

A composite profile of submicron PON was generated using all the data from 110°W to 140°W (Table III.3 and Fig. III.3). Error bars in the composite profile are large and presumably reflect both spatial and temporal variability inherent in such a large data set. Nonetheless, the composite profile clearly shows three distinct regions

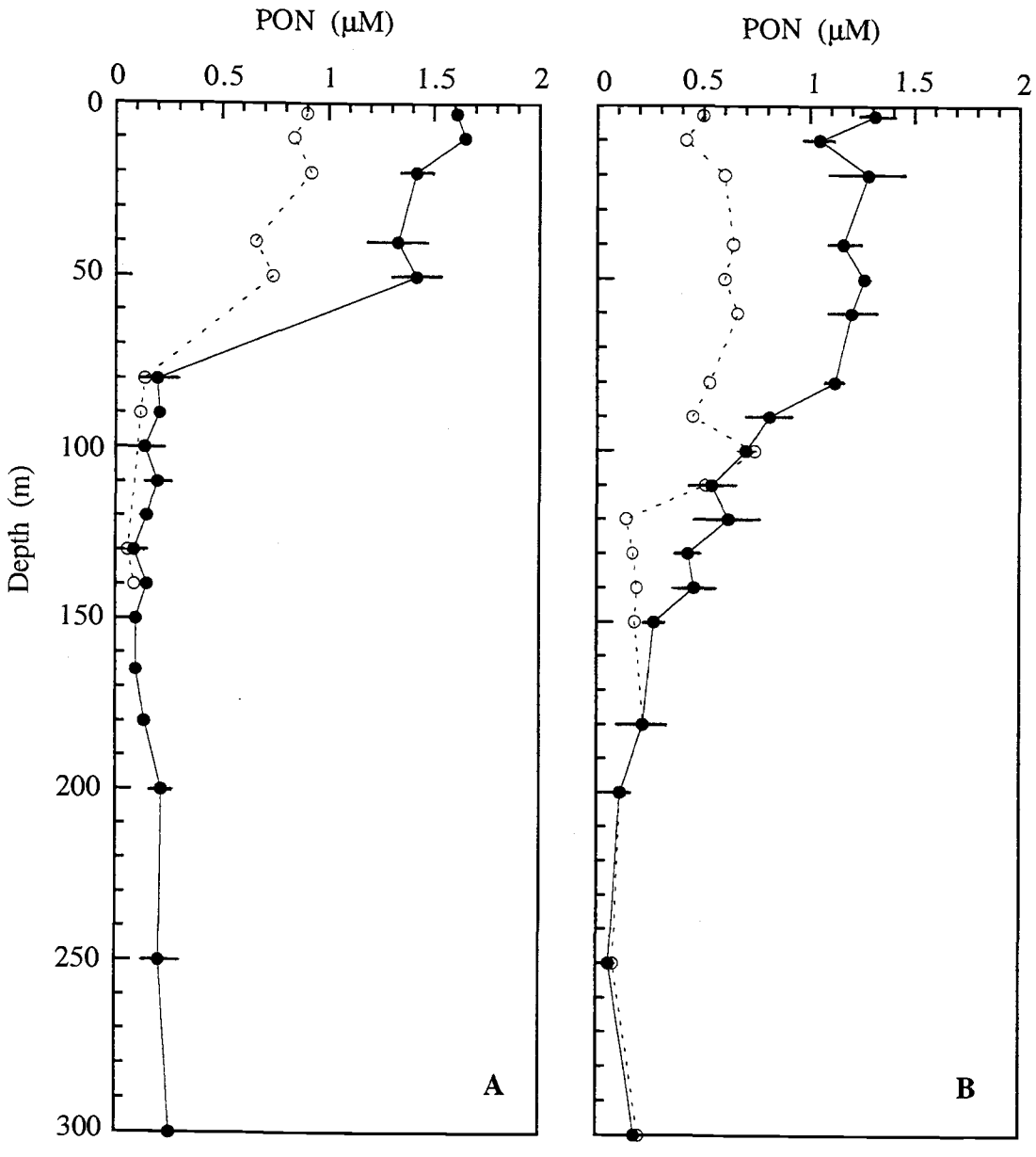


Figure III.2 Representative depth profiles of >0.2 µm (●) and >0.7 µm (○) PON estimates. A) 0° 15'N 110°W B) 5°N 110°W.

TABLE III.3 Data from composite of PON depth profiles at 110°, 125° and 140°W transects. Results for the statistical comparison of the mean values for each of the two types of filters are shown in the last column.

Depth (m)	>0.2 µm PON Mean (µM)	>0.2 µm PON SD (µM)	>0.7 µm PON Mean (µM)	>0.7 µm PON SD (µM)	Submicron PON* (µM)	n	
3	1.22	0.21	0.77	0.22	0.45	15	**
10	1.28	0.20	0.83	0.24	0.45	9	**
20	1.24	0.23	0.73	0.24	0.51	9	**
30	1.26	0.25	0.83	0.17	0.44	8	**
40	1.25	0.14	0.84	0.18	0.42	8	**
50	1.15	0.24	0.65	0.19	0.50	7	**
60	1.00	0.24	0.66	0.20	0.35	14	**
80	0.66	0.37	0.48	0.22	0.18	13	ns
90	0.50	0.30	0.37	0.23	0.13	9	ns
100	0.45	0.12	0.35	0.17	0.11	11	ns
110	0.32	0.17	0.28	0.15	0.04	5	ns
120	0.26	0.11	0.22	0.08	0.03	12	ns
130	0.27	0.17	0.22	0.14	0.05	11	ns
140	0.25	0.11	0.18	0.04	0.07	7	ns
150	0.24	0.13	0.21	0.06	0.03	11	ns
180	0.22	0.09	0.18	0.08	0.03	10	ns
200	0.16	0.07	0.16	0.05	0.00	14	ns
250	0.22	0.12	0.20	0.09	0.02	12	ns
300	0.20	0.06	0.17	0.08	0.03	11	ns

* calculated as (>0.2 µm PON) - (>0.7 µm PON)
** indicates significant difference ($p < 0.01$)
ns indicated no significant difference ($p > 0.10$)

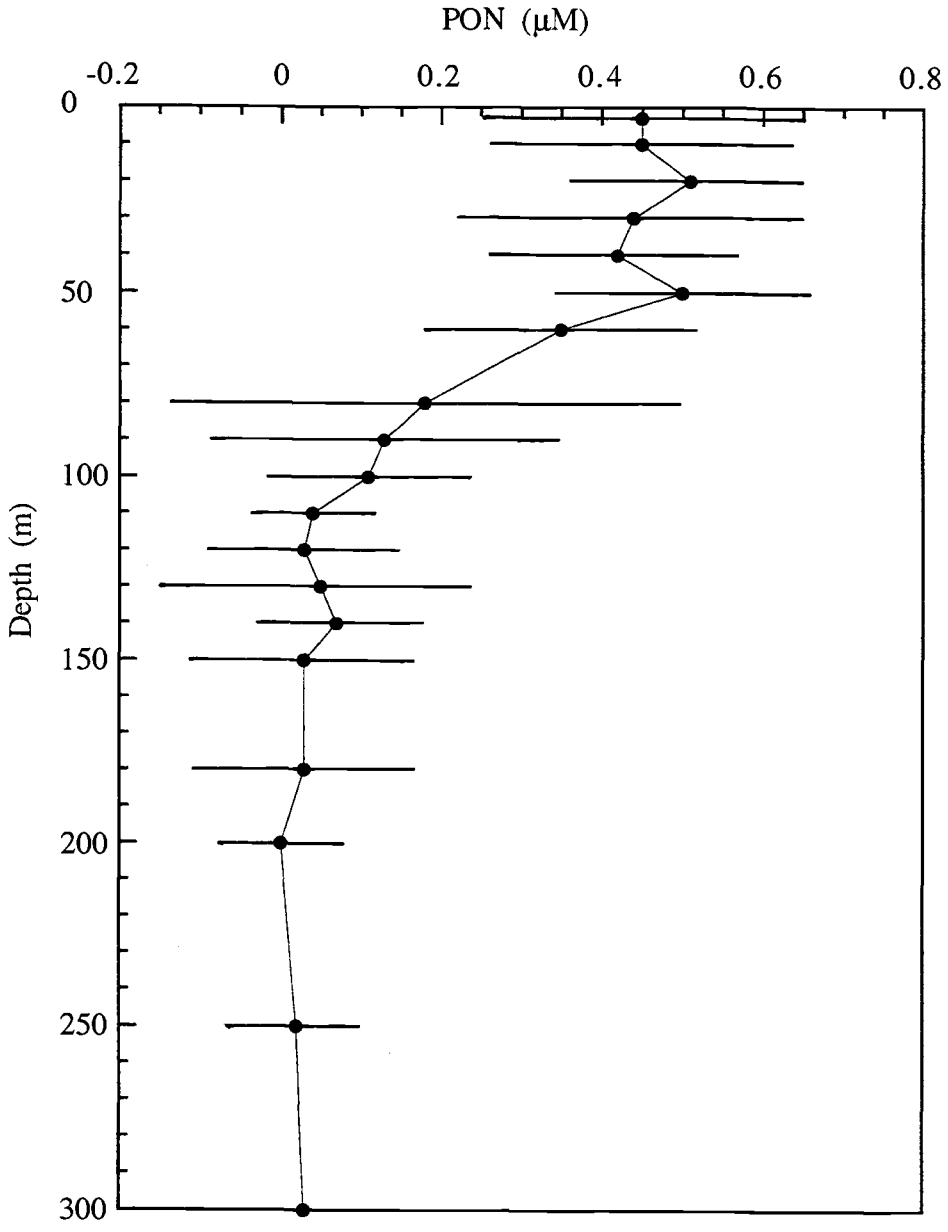


Figure III.3 Composite depth profile of submicron PON (>0.2 µm - >0.7 µm PON). Error bars represent one standard deviation (see Table III.3 for n).

in the vertical distribution of submicron PON. Maximum concentrations (0.46 ± 0.17 µm, n=56) were found in the mixed layer (0-50 m). A sharp decrease in concentration is apparent between 50 and 100 m, while concentrations were very low and relatively constant (0.03 ± 0.02 µm, n=93) below 100 m. The gradient of submicron PON in the intermediate layer (50-100 m) is -6.89×10^{-3} µM m^{-1} with $r^2 = 0.968$.

Chlorophyll a and PON/Chl ratios

Comparisons of >0.2 µm and >0.7 µm Chl *a* were made at two stations. The Chl *a* profiles for Station 8 (3°N 140°W; Fig. III.4a) were similar to the PON profiles for the region. In the mixed layer, a greater than 2-fold increase in Chl *a* was found when comparing 0.2 µm Nuclepore and GF/F filters (mean = 2.2 ± 0.3, n=5). At Station 2 (10°N 140°W) there was a subsurface Chl *a* maximum at 90 m and the 0.2 µm Nuclepore Chl *a* estimates were almost twice that of the GF/F filters (Fig. III.4b; mean = 1.73 ± 0.26, n=16).

Chl *a* data from 140°W (R. Barber) and 110°W (F. Chavez) were used in calculating PON/Chl ratios for each of these transects. Since the data along these two transects varied by only ~10% and others have found no systematic east-west gradients in Chl *a* concentrations (Murray et al. 1992), the Chl *a* data from 140°W was also used to calculate PON/Chl ratios at 125°W. There were minima in the PON/Chl ratios at the equator for each of the transects (47.1 ± 4.3, n=3) and the ratio increased steadily poleward (Fig. III.5). These trends and values were relatively

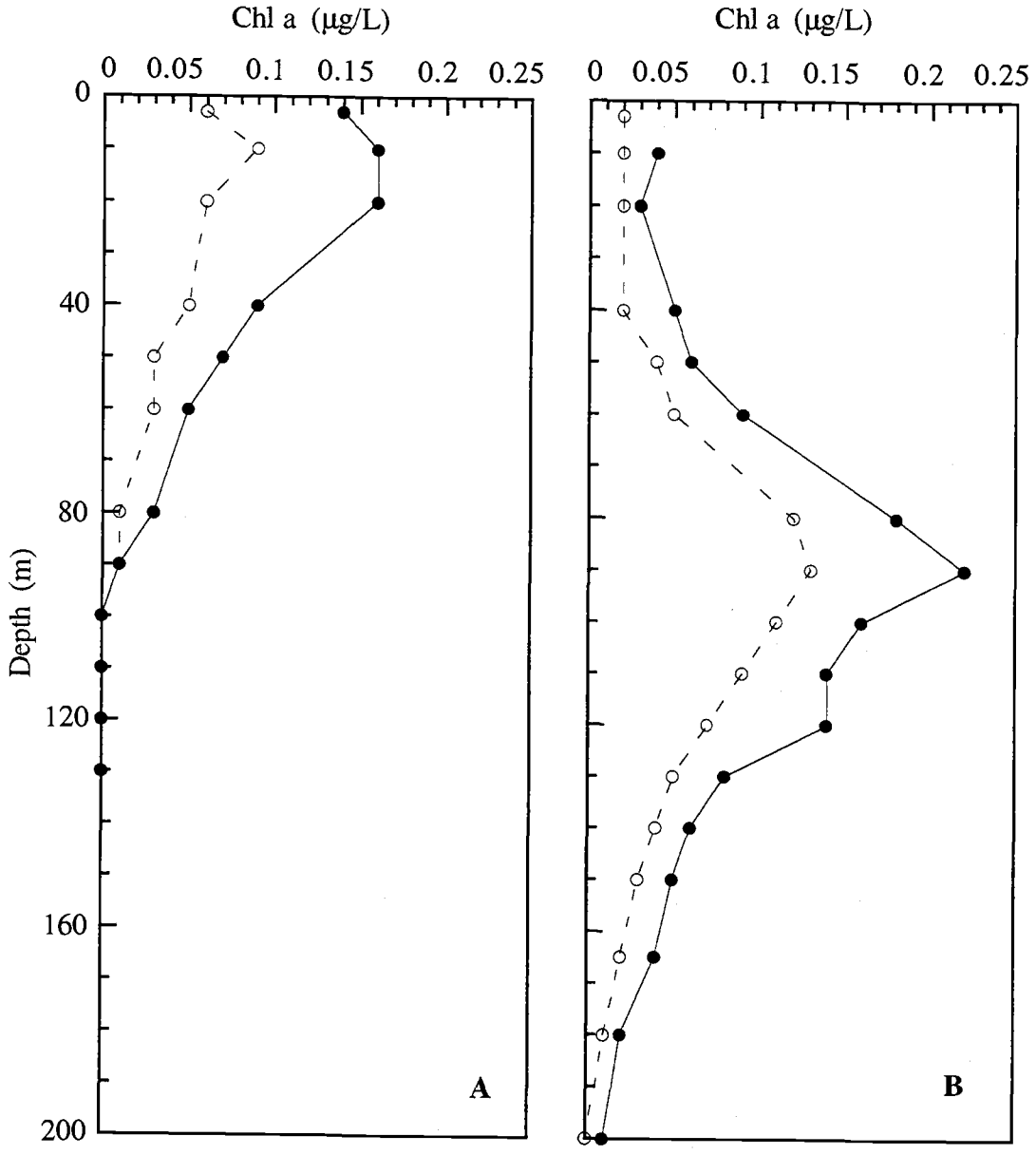


Figure III.4 Chlorophyll *a* profiles comparing measurements made using 0.2 µm Nuclepore (●) and GF/F (○) filters. A) 3°N 140°W B) 10°N 140°W.

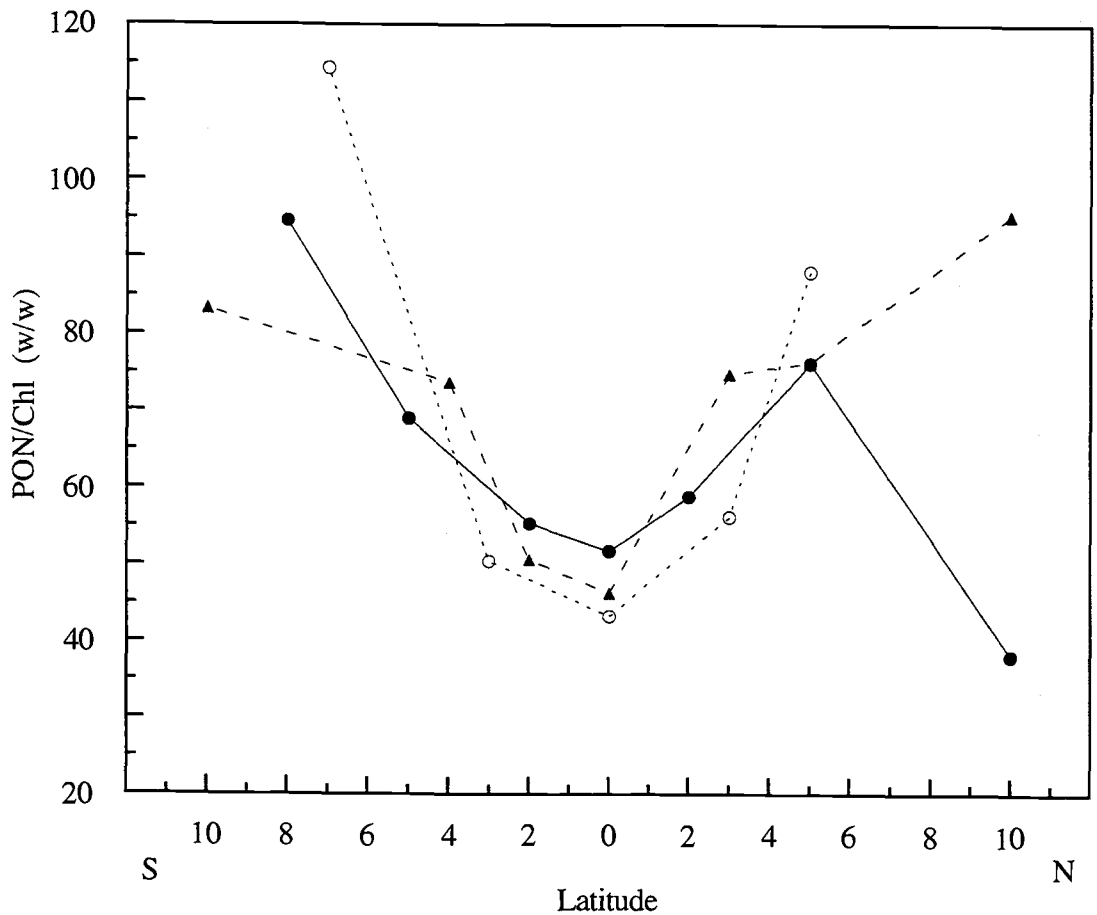


Figure III.5 Ratio of integrated PON and Chl *a* for transects at 110°W (●), 125°W (▲) and 140°W (○). Data integrated over the euphotic zone using the trapezoidal method (Press et al., 1989). Chl *a* data courtesy of R. Barber and F. Chavez.

consistent over the three transects. PON/Chl ratios in Fig. III.5 were calculated using >0.2 μm PON and GF/F Chl *a* measurements. The trends were similar, but values lower, when GF/F PON estimates were used (mean of 27.7 ± 4.9 at the equator, n=3). We suggest that the value of the >0.2 μm PON/>0.2 μm Chl and GF/F PON/Chl ratios are comparable based on the 2-fold difference found between 0.2 μm and GF/F Chl *a* measurements.

Dissolved Nitrogen

Dissolved organic and inorganic N were measured at all 23 stations and the concentrations averaged over the mixed layer are presented in Table III.4. For each transect, the NO_3^-+NO_2^- concentrations were highest (7.3 to 10.6 μM) at the equator and decreased abruptly to the north and gradually to the south. We found an eastward increase in NO_3^-+NO_2^- concentrations at the equator and a general increase south of the equator, but no east-west trend was evident north of the equator. There were no systematic east-west variations in DON, but the minimum along each transect was centered on the equator (see below). DON concentrations in the mixed layer ranged from 6.7 to 10.6 μM (Table III.4). DON was the major constituent of total dissolved N (TDN) in the mixed layer of this region (station means = 67% ± 17%, n=20). The importance of DON in the mixed layer is illustrated by the representative profiles of DON, NO_3^-+NO_2^- and TDN from both high and low nitrate surface waters (Fig. III.6a & 6b). Below the nitracline, however, DON accounts for less than 20% of TDN (see Appendix A). The profiles also suggested an increase of DON in the mixed layer to the north of the equator.

TABLE III.4 Comparison of total dissolved nitrogen (TDN), $NO_3^-+NO_2^-$ (N+N) and dissolved organic nitrogen (DON) concentrations averaged over the mixed layer.

Lat	Long	TDN Mean (µM)	TDN SD (µM)	N+N Mean (µM)	N+N SD (µM)	DON Mean (µM)	DON SD (µM)	%DON in TDN
10°N	140°W	8.04	0.94	0.33	0.06	7.71	0.90	95.9
5°N	140°W	13.50	0.13	4.02	0.51	9.48	0.64	70.2
3°N	140°W	13.31	1.26	3.86	1.44	9.45	0.22	71.0
0°30'S	140°W	14.69	0.64	7.27	0.75	7.42	1.39	50.5
3°S	140°W	15.20	1.35	5.80	0.15	9.40	1.20	61.8
7°S	140°W	14.19	0.30	6.46	0.07	7.73	0.37	54.5
10°N	125°W	15.83	4.39	6.33	5.53	9.50	1.14	60.0
5°N	125°W	10.50	0.68	0.64	0.02	9.86	0.70	93.9
3°N	125°W	12.86	2.39	3.02	0.53	9.85	1.86	76.6
0°	125°W	18.88	2.63	9.91	1.17	8.97	3.80	47.5
2°S	125°W	14.98	0.93	6.96	0.03	8.02	0.90	53.5
4°S	125°W	13.73	1.35	4.17	0.03	9.56	1.32	69.7
10°S	125°W	11.45	0.56	3.00	0.03	8.46	0.53	73.9
10°N	110°W	11.22	3.13	1.79	1.82	9.42	1.32	84.0
5°N	110°W	10.94	4.82	0.36	0.16	10.58	4.74	96.7
2°N	110°W	10.74	3.18	1.22	1.42	9.51	3.13	88.6
0°15'N	110°W	17.24	0.89	10.58	0.55	6.66	0.62	38.6
2°S	110°W	17.81	3.38	8.32	0.18	9.49	3.26	53.3
5°S	110°W	16.89	0.69	9.12	0.08	7.78	0.61	46.0
8°S	110°W	19.07	3.41	8.77	0.13	10.30	3.37	54.0
							mean	67.0
							SD	17.4
2°N	95°W	8.09	1.06	0.27	0.39	7.82	1.13	96.6
0°	95°W	23.85	8.26	12.97	3.34	10.89	5.08	45.6
2°S	95°W	28.13	3.93	15.66	4.85	12.47	6.67	44.3

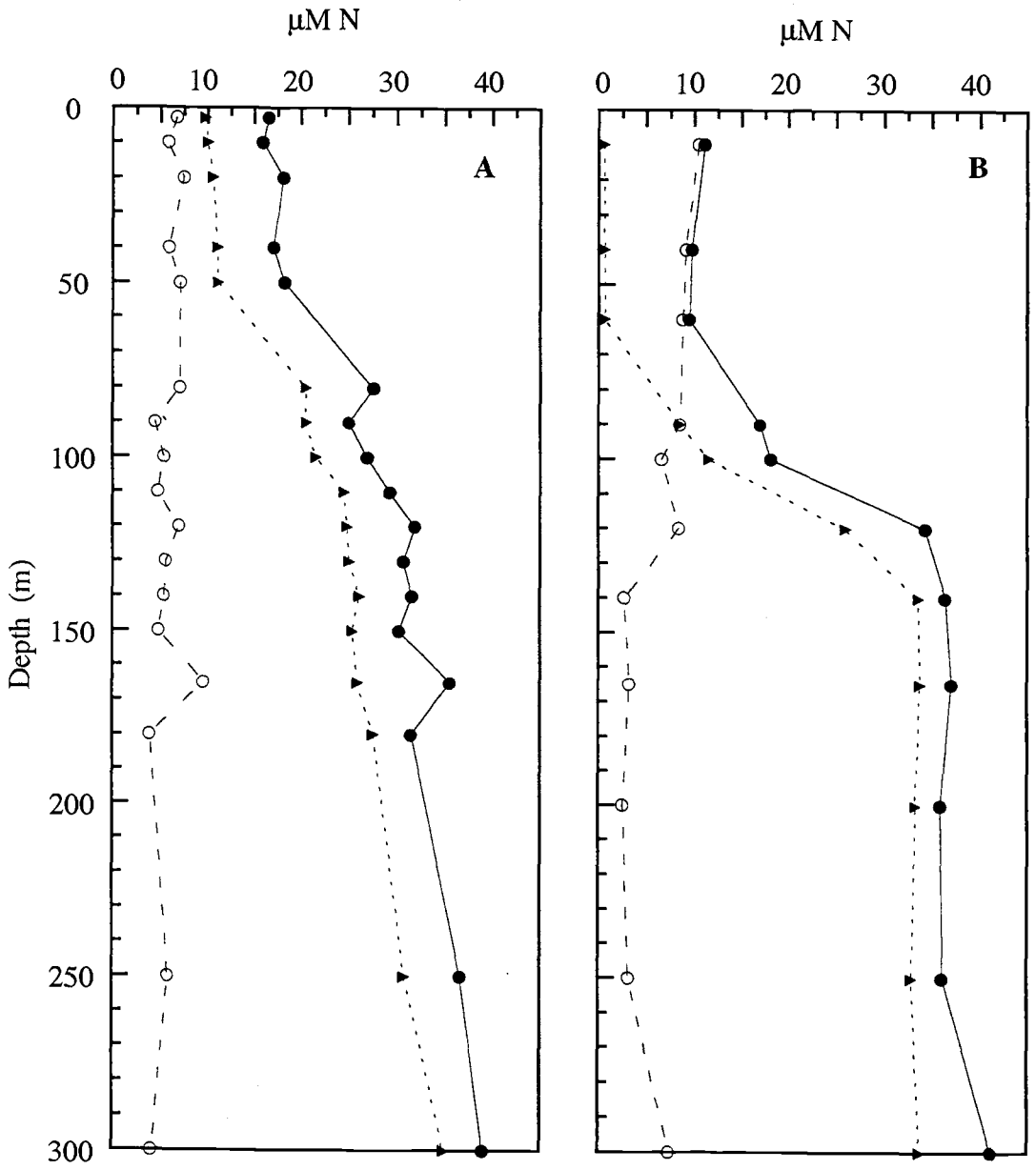


Figure III.6 Representative depth profiles of DON (○), NO_3^-+NO_2^- (▲) and total dissolved nitrogen (●). A) 0° 15'N 110°W B) 5°N 125°W.

The northward increase was emphasized further when the latitudinal distribution of mean DON concentrations in the euphotic zone was examined (Fig. III.7). A DON minimum of 6.8 ± 0.3 μM (n=3) was found at the equator. Mean DON concentrations increased to 11.7 ± 2.1 μM at 5°N along the three transects. Within three degrees of the equator DON increased to 9.8 ± 0.8 μM (n=3) to the north and to 9.2 ± 0.8 μM (n=3) to the south. The poleward increase in DON coincided with a sharp decrease in surface NO_3^- (>4.5 μM) to the north and a modest decrease in surface NO_3^- (<2.0 μM) to the south of the equator.

Equatorial Time Series

The time series sampling coincided with the passage of an instability wave that was evidenced by abrupt changes in the nutrient concentrations (Fig. III.8a). There was a marked increase in NO_3^- from 5.8 to 7.2 μM and a coincident decrease in NH_4^+ from 0.11 to 0.03 μM. The changes in nutrients were followed by changes in Chl *a* and PON (Fig. III.8b). The Chl *a* concentrations increased 80% reaching a maximum of 0.46 μg/L and there was a distinct increase in >0.7 μm PON from 0.65 to 1.00 μM with a corresponding decrease in submicron PON from 0.65 to 0.35 μM. After Oct. 12, there was a significant decrease in NO_3^- of ~2.0 μM, which corresponded to a 3-fold increase in NH_4^+ to 0.13 μM, a return to initial Chl *a* concentrations and a doubling of submicron PON to 0.80 μM. The increase in submicron PON accounted for ~20% of the nitrogen removed from the NO_3^- pool.

We were unable to measure DON during the time series, but speculate below on the possibility that changes in DON could account for a significant portion of the net decrease in NO_3^-.

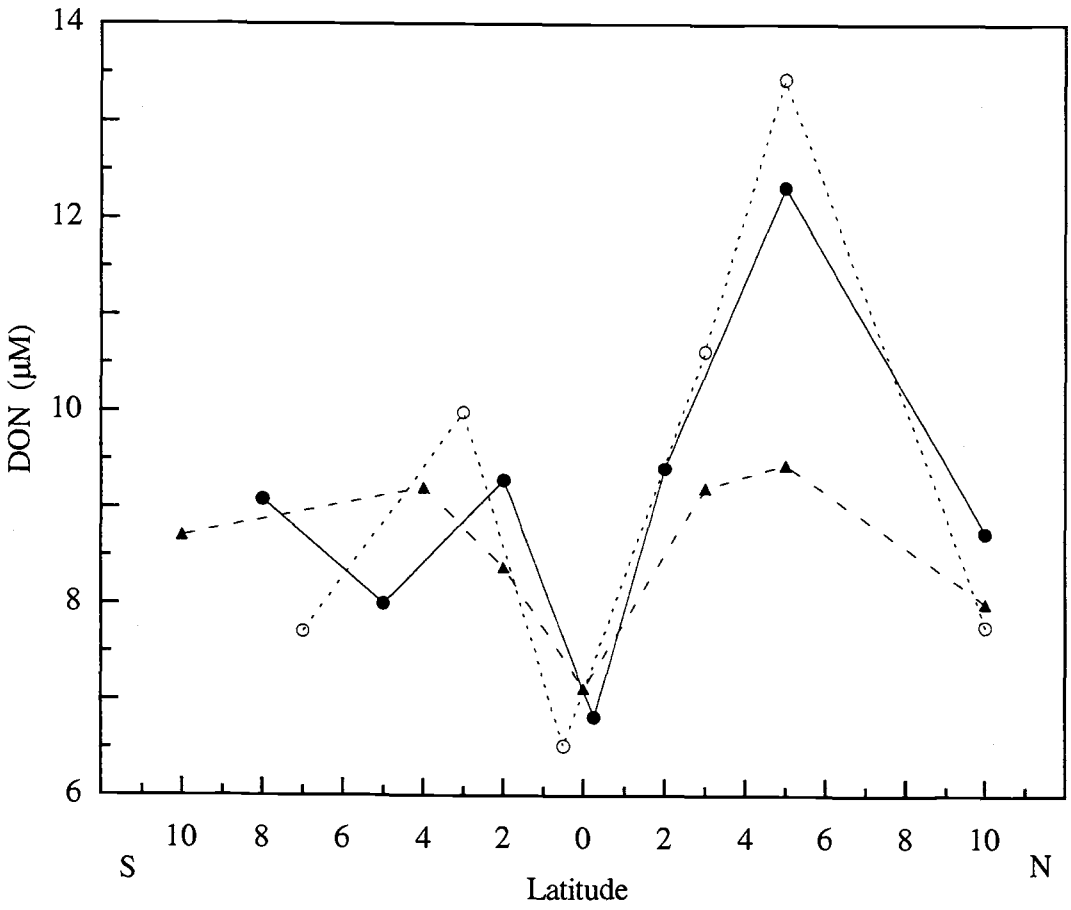


Figure III.7 Distribution of DON averaged over the euphotic zone for transects at 110°W (●), 125°W (▲) and 140°W (○).

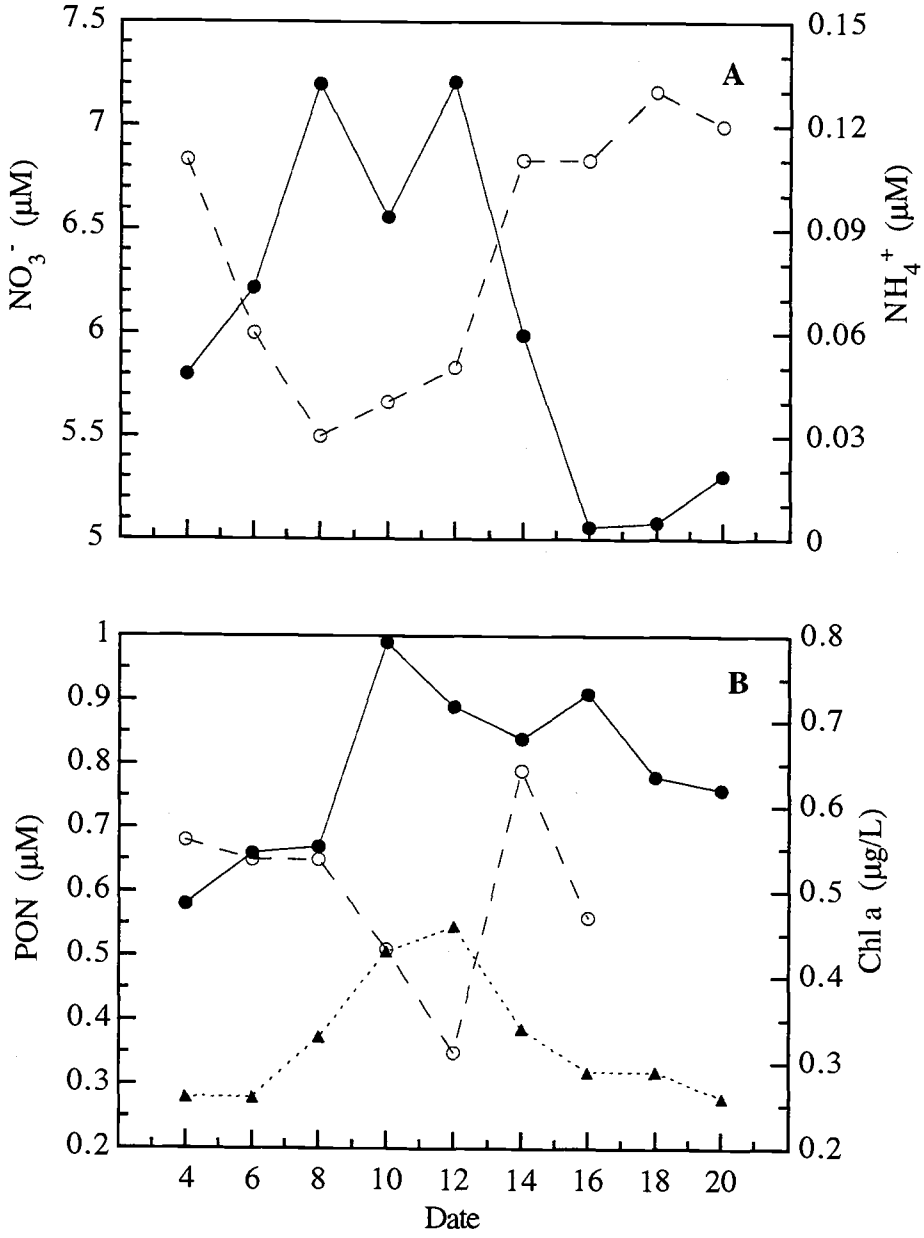


Figure III.8 Time series measurements at 0° 140°W during US JGOFS cruise TT012 from October 4-20, 1992. A) NO_3^- (●) and NH_4^+ (○) B) submicron PON (○), >0.7 µm PON (●) and Chl *a* (▲).

DISCUSSION

In general, our estimates of PON using GF/F filters were comparable to the values determined previously by Eppley et al. (1992) and Peña et al. (1991). However, there were differences in the latitudinal trends. During March (Eppley et al., 1992) and April (Peña et al., 1991) of 1988, maximum PON values were observed at the equator. Our study showed local minima at the equator for 140°W and 125°W transects and maxima 2°-3° to the north and south. The differences may result from a combination of seasonal (higher upwelling intensity) and temporal (occurrence of instability waves) variations associated with our fall sampling.

The >0.2 μm PON estimates were ~1.6 times higher than the GF/F estimates for the mixed layer, while there were no significant differences between the two measurements below 60 m. The submicron PON pool makes up a significant portion of PON in the mixed layer and may play a critical role in the regeneration and transport of nitrogen. The sharp gradients (Fig. III.2a & 2b and III.3) and the negligible sinking rates of submicron PON should result in strong diffusive fluxes of this material out of the mixed layer. Low concentrations of submicron PON below 80 to 100 m would suggest intense degradation and/or recycling of this material in the lower euphotic zone. These findings support the hypothesis of Peña et al. (1991) that the increase in the C/N ratio of POM in the lower euphotic zone results from the faster turnover of PON than of POC.

The submicron PON that passes through GF/F filters is not limited to detrital and bacterial PON. Comparisons of 0.2 μm Nuclepore and GF/F filters indicated that

half of the Chl *a* also passed through the GF/F filters. These findings could have important implications for productivity measurements. Use of glass fiber filters for ^{14}C productivity measurements in oceanic waters could result in incomplete recovery of phytoplankton (Phinney and Yentsch, 1985; Dickson and Wheeler, 1993) and an underestimate of total primary production. Although the inefficient trapping of Chl *a* by glass fiber filters has been noted in the literature, the extension of filter comparisons to ^{14}C productivity measurements has not been addressed. The potential significance of inefficient trapping of biogenic particles has been discussed, however, for the measurement of rates of nitrogen assimilation by plankton. Eppley and Renger (1992) report a 7-fold difference in the net rate of NO_3^- depletion compared with ^{15}N-measured rates of NO_3^- assimilation into PON. Review of a larger data set for similar comparisons suggests that 1.0 to 2.1-fold differences between NO_3^- depletion and ^{15}N assimilation are more common (cited in Table 4 of Eppley and Renger, 1992). Based on the assumption that heterotrophic bacteria are the active component of submicron PON, Eppley and Renger argue that inefficient trapping of submicron PON would account for, at most, a 2-fold difference in rates. However, as both phytoplankton and bacteria are important components of submicron PON, use of glass fiber filters to determine rates of production (^{14}C and ^{15}N) could lead to a >2-fold underestimate of rates. Resolution of the discrepancy between the two types of rate measurements will require more precise determination of the rates of nutrient depletion, as well as direct estimates of primary production and nutrient assimilation by phytoplankton and bacteria which can pass through glass fiber filters.

PON is comprised of phytoplankton, heterotrophic protists, bacteria and detrital material. Examination of PON/Chl ratios provides a useful indication of the relative importance of autotrophic biomass (Kokkinakis and Wheeler, 1987; Dortch and Packard, 1989). Low ratios are typical of waters dominated by phytoplankton, whereas high ratios result from a predominance of heterotrophic organisms and detrital material. For our equatorial Pacific transects, the lowest PON/Chl ratios were found at the equator and the ratios increased steadily to the north and to the south. Our results for PON/Chl for material collected on glass fiber filters are similar to those reported by Peña et al. (1991). Our PON/Chl ratios calculated with >0.2 μm PON and GF/F Chl *a* are higher, as should be expected. For oceanic regions, the ratio of >0.2 μm PON/>0.2 μm Chl would be the most appropriate value to use. Unfortunately, the database for such a comparison is rather small. Our equatorial results, however, suggest the inclusion of the submicron component for both PON and Chl *a* results in PON/Chl ratios that are similar to the data reported for glass fiber filters. The consistent increase in PON/Chl ratios away from the equator for both our data and Peña et al. (1991) suggests a steady increase in the relative contribution of heterotrophic and/or detrital nitrogen to PON.

A similar latitudinal pattern was evident in the distribution of DON. A DON minimum of 6.8 μM was found at the equator with an increase of 4.9 μM to the north and 2.4 μM to the south. These concentrations and poleward increases in DON compare well with earlier work. Thomas et al. (1971) reported a mean concentration of 6.9 ± 3.4 μM DON in the eastern equatorial Pacific and 8.2 ± 3.4 μM DON in the surrounding nutrient depleted waters. Although the accuracy of DON measurements

remains to be confirmed, the consistency between our data set and Thomas et al. (1971) does suggest that the relative distributions are real. The nutrient rich, recently upwelled waters have lower concentrations of DON and the dissolved organic pool increases with decreasing NO_3^- to the north and south. The large increases in DON that were found to the north (Fig. III.7) corresponded to sharp decreases in surface NO_3^- of >4.5 µM and suggests that the DON pool serves as a 'sink' or reservoir for upwelled NO_3^-. This is also implied by DON depth profiles, which show decreases of 2-3 µM in DON that are inversely correlated with NO_3^-. The large poleward increases in DON suggest that in the equatorial region meridional advection may be as important as vertical diffusion in the removal of N, as DON, from the equatorial region.

The studies of temporal variations in biological parameters for the equatorial Pacific have concentrated on the El Niño/La Niña cycles and seasonal variations. Less information is available on biological responses to short term variability, such as instability waves. Instability waves were first observed in satellite images of sea surface temperature (SST) by Legeckis (1977). These waves are caused by shear within the region's zonal circulation patterns and are most intense during the boreal fall, when the westward flow of the Southern Equatorial Current is strongest (Philander, 1978). The instability is manifested as N-S shifts in meridional flow which move cool water north and warm water south (Halpern et al., 1988), resulting in the SST signature of the instability waves. Coincident fluctuations in nutrients and chlorophyll have also been reported (Chavez et al., 1990; Carr et al., 1992; Feely et al., 1993).

During the NSF time series cruise, we successfully sampled chemical and biological parameters in the upper water column during the passage of an instability wave. The most apparent features associated with the shift from warm to cool water were significant increases in nutrients and biomass. Similar changes were noted in earlier work by Chavez et al. (1990), who also measured significant increases in primary productivity and nanoplankton biomass (mainly pennate diatoms). Although it is tempting to propose a cause and effect relationship between increased NO_3^- and production of larger phytoplankton, Chavez et al. (1990) report that the increase in total phytoplankton biomass is accompanied by increases in all major taxa. Nitrate levels in the equatorial Pacific are well above levels known to limit phytoplankton growth (Goldman and Glibert, 1983), and the simultaneous increases in all the major taxa of phytoplankton argues against regulation of community structure by competition for nitrate.

Factors regulating phytoplankton standing stocks and primary production in the equatorial Pacific and other high-nutrient, low-chlorophyll (HNLC) waters have been debated intensely since 1988. The two major hypotheses are Fe-limitation of nitrate use (Martin and Fitzwater, 1988) and control of phytoplankton stocks by zooplankton grazing (Miller et al., 1991). Recent work in the equatorial Pacific indicates that the dominant phytoplankton are well-adapted to light and nutrient conditions and are growing rapidly (Cullen et al., 1992). Fast growth rates, but constant standing stocks are indicative of the proximate control of primary production by grazing (Cullen, 1991). Nonetheless, Fe-addition experiments clearly indicate a stimulatory effect on diatom abundance and production (Price et al., 1991).

As argued by Cullen (1991) and Miller et al. (1991), both factors seem to play a role simultaneously in HNLC regions. The dominant pico- and nanoplankton are growing rapidly but fail to accumulate due to close coupling with micrograzers, while diatoms appear to be Fe-limited and only accumulate when iron supplies are enhanced.

Our time series results combined with results from other studies in HNLC regions are consistent with the following scenario. The recently upwelled (nitrate-rich) water may contain elevated levels of iron (Martin and Gordon, 1988). This iron input could stimulate diatom growth and result in the observed increase in Chl *a* standing stocks and production (Chavez et al., 1990). The most recently upwelled water is also low in Chl *a* and NH_4^+. As production of the pico- and nanoplankton increases, grazing activity of the micrograzers also increases. This results in the establishment of a grazing dominated food web, with high growth rates but constant biomass. In such systems, NH_4^+ concentrations are relatively high and NH_4^+, rather than NO_3^-, is the major nitrogenous nutrient for phytoplankton (Wheeler and Kokkinakis, 1990).

Our observations of changes in submicron PON, during the passage of the instability wave, are also consistent with an important role for grazing in the equatorial Pacific. Submicron PON appears to be inversely correlated with NO_3^- concentrations and directly correlated with NH_4^+ concentrations. The shifts observed in submicron PON could reflect changes in growth rates and/or grazing rates within the microbial community. Changes in submicron PON during the time series, as well as the observation of low concentrations at depth, suggest that this material is produced and degraded rapidly in the upper portions of the water column.

Early models of nitrogen fluxes for oceanic regions were primarily two dimensional; i.e. nitrate enters the euphotic zone from depth (either by diffusion or upwelling), is assimilated into PON and then sinks out of the euphotic zone as PON. Nitrate influx and PON sinking flux do appear to be closely balanced in oceanic gyres (Eppley and Peterson, 1979). These two-dimensional models are inappropriate for the equatorial Pacific. Physical processes result in both meridional and zonal transport of water in the central equatorial Pacific. Moreover, as we show here, two pools of organic nitrogen (submicron PON and DON) have significant concentrations in the upper water column. Our observations of large spatial variations in both pools of organic nitrogen suggest that each is an important and dynamic component of the equatorial nitrogen cycle.

Primary production in the central equatorial Pacific is fueled by nitrate-rich water which is upwelled from depth near the equator and then flows poleward producing a band of nutrient-rich water which extends about 5° meridionally on either side of the equator. Our results show that as NO_3^- in this water is utilized there are simultaneous increases in PON and DON. These changes, as well as the vertical distribution of PON and DON, support the conclusion that the organic forms of nitrogen are produced and degraded in the upper 100 m of the water column. Full elucidation of the equatorial N cycle will require more detailed studies of the mechanism and rate of cycling of these pools of organic nitrogen.

Chapter IV

SUMMARY

The main goal of this thesis was the development and application of a method that directly measures >0.2 μm PON . Previous work had shown silver and aluminum oxide inorganic filters to be incompatible with persulfate oxidation. This led to the examination of membrane filters, which due to a high C content are not suitable for dry-combustion techniques, but are amendable to persulfate oxidation. Evaluations of the 0.2 μm membrane filters indicated that 5 of the 6 filter types had high N blanks and/or interfered with the chemical analysis. The TF-200, teflon filter was the only membrane filter tested that was suitable for the persulfate method. The TF-200 and glass fiber filters had comparable filter N blanks, comparable filtration times and showed negligible chemical interference.

Field work in the central equatorial Pacific indicated an increase in PON estimates using TF-200 filters. The 0.2 μm filters retained ~1.6 times more PON than the GF/F filters in the mixed layer. The concentrations of the submicron PON (0.2 to 0.7 μm) made up a significant portion of the total PON, were relatively constant in the mixed layer and decreased rapidly in the lower euphotic zone. In general, PON increased away from the equator and this correlated with a relative increase in detrital and/or heterotrophic N. These findings suggest that significant

amounts of organic nitrogen are removed from the mixed layer by downward diffusive fluxes and from the point of upwelling by meridional advection of detrital PON.

Our results also suggest that a significant amount of organic nitrogen is laterally advected away from the equator as DON. Meridional increases in DON were comparable and coincident with decreases in surface NO_3^- and both exhibited sharper gradients to the north of the equator. Depth profiles showed a significant decrease in DON concentrations below the mixed layer and support the notion that the diffusive flux and degradation of DON are important mechanisms in the equatorial Pacific. The vertical change in DON was similar to the horizontal gradient to the south, but smaller than the gradient to the north. Meridional advection of DON may be as important as vertical DON fluxes for the removal of N from the upwelling region.

Temporal variations in submicron and >0.7 μm PON, nutrients and Chl *a* were observed during the time series experiments. Large increases in NO_3^- and decreases in NH_4^+ signified the passage of an instability wave. Submicron PON appeared to be correlated with NH_4^+ concentrations and both increased over time in the recently upwelled water. These increases, in conjunction with a decrease in Chl *a*, could reflect changes within the microbial community. The increases in submicron PON and NH_4^+ accounted for ~25% of the nitrogen from coincident decreases in NO_3^-. Our transect results suggest that this 'missing' nitrogen may have been converted to DON.

Application of the persulfate oxidation method for measurement of submicron PON and dissolved organic nitrogen in the equatorial Pacific has shown both of these pools to be important nitrogenous components in the mixed layer. Concentrations of both pools decrease with depth suggesting a significant degradation and downward diffusive flux. Even higher poleward gradients were measured for both pools in the mixed layer. The spatial distribution of PON and DON, in conjunction with measurements of inorganic nitrogen and Chl *a*, indicates that lateral fluxes of organic nitrogen are comparable to the vertical fluxes. Temporal variations in the same parameters offer further evidence for the role of phytoplankton and zooplankton in the transformation of recently upwelled water. The observations made here should serve as a foundation for future work that includes more detailed measurements of particulate and dissolved organic nitrogen and elucidation of their significance in the biogeochemical fluxes of nitrogen.

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APPENDICES

Appendix 1 Primary data collected on the NOAA EPOCS/CO_2/US JGOFS cruises DI-92-03 and DI-92-04 aboard the R/V Discoverer during the fall of 1992 (local date and time are given).

* = PON sample estimated from BCA protein N data

Station 2		Lat: 10 0.38N		Long: 139 59.45W		Date:9-9-92	Time:18:17
			PON		Protein N		
CTD Bottle	Pres.	Depth (m)	TF (µM)	GF/F (µM)	TF (µM)	GF/F (µM)	DON (µM)	N+N (µM)
24	5.0	3					6.93	0.24
22	20.4	20					8.97	0.38
20	52.9	50					7.22	0.37
18	82.6	80					7.64	0.47
17	94.6	90					7.79	17.86
15	111.7	110					5.51	28.00
13	132.3	130					4.09	31.84
11	152.0	150					7.53	32.52
9	184.3	180					3.61	35.28
8	201.0	200					3.72	35.63
7	253.1	250					2.76	35.90
6	303.0	300					3.79	36.65

Station 6		Lat: 5 0.39N		Long: 140 3.33W		Date:9-11-92	Time:06:42
			PON		Protein N		
CTD Bottle	Pres.	Depth (m)	TF (µM)	GF/F (µM)	TF (µM)	GF/F (µM)	DON (µM)	N+N (µM)
24	5.6	3	1.37	0.72			10.33	3.35
22	22.4	20	1.32	0.79	0.87		9.33	4.12
20	52.4	50	1.43	0.83	0.77		8.79	4.58
18	83.6	80	1.37	0.72			31.56	4.90
17	93.2	90	0.85	0.47			10.05	7.23
15	113.3	110	0.47	0.32	0.20		8.05	8.87
13	131.8	130	*0.66	0.38	0.33		7.78	9.43
11	153.6	150		0.24			6.48	10.54
9	182.8	180	0.24	0.17	0.12		5.49	26.32
8	202.7	200	0.28	0.15	0.17		4.25	32.81
7	252.5	250	0.39	0.40			4.34	35.68
6	304.7	300	0.25	0.26	0.24		3.62	36.75

Station 8 Lat: 2 59.35N Long: 140 7.21W Date:9-12-92 Time:10:53

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	2.9	3	*1.16	*0.98	0.58	0.49	9.75	2.02
22	22.6	20	*1.04	*0.76	0.52	0.38	9.28	4.03
20	53.6	50		*0.74		0.37	9.31	5.54
18	82.6	80	*0.58	*0.34	0.29	0.17	16.03	7.91
17	94.5	90		*0.40		0.20	7.60	8.47
15	112.2	110		*0.09		0.09	7.63	9.15
13	133.4	130	*0.15	*0.15	0.15	0.15	4.94	17.96
11	153.4	150		*0.17		0.17	9.96	26.45
9	182.8	180		*0.09	<0.05	0.09	1.92	32.05
8	202.8	200				<0.05	3.66	30.81
7	252.7	250		*0.08		0.08	0.16	31.81
6	302.1	300		*0.20		0.20	3.46	35.97

Station 15 Lat: 0 29.73S Long: 139 59.45W Date:9-14-92 Time:23:42

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	5.5	3	1.07	0.64	0.41		8.80	6.52
21	32.9	30	1.33	0.63			6.03	8.02
19	63.7	60	*0.46	0.20	0.23		6.08	11.97
18	81.4	80	0.25	0.21	0.21		5.22	13.81
17	103.5	100	0.49	0.06	0.20		5.62	14.85
16	123.3	120	*0.13	0.08	0.13		3.44	15.48
15	132.6	130	*0.12	0.06	0.12		3.67	17.00
14	144.6	140	0.34				4.88	18.04
12	177.4	175	*0.10	0.06	0.10		4.69	23.78
11	202.8	200		0.08			3.44	26.34
9	261.4	260	0.37	0.29	0.21		3.47	32.70
8	292.9	290	0.18	0.08	0.18		1.26	35.21

Station 18 Lat: 2 59.88S Long: 140 0.10W Date:9-16-92 Time:02:47

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	6.2	3	1.18	0.95			10.60	5.95
21	32.7	30	1.00	0.81			8.20	5.65
19	62.2	60	1.03	0.93			8.38	6.26
18	83.3	80	0.45	0.96			18.08	6.79
17	102.1	100	0.59	0.47			8.78	7.07
16	122.2	120		0.28			7.66	8.03
15	133.0	130		0.25			7.03	7.77
13	153.0	150	0.54	0.28			5.05	20.83
12	178.1	175		0.21			3.71	31.63
11	202.4	200		0.22			4.82	32.62
9	263.3	260		0.24			10.19	32.57
8	293.8	290		0.19			21.12	34.39

Station 22 Lat: 7 0.04S Long: 140 0.16W Date:9-17-92 Time:09:20

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	4.9	3	*1.26	*0.56	0.63	0.28	8.10	6.39
21	33.5	30	*1.58	*0.78	0.79	0.39	7.36	6.53
19	63.4	60	*0.96	*0.38	0.48	0.19	7.76	7.40
18	83.9	80	*0.74	*0.38	0.37	0.19	7.90	7.61
17	102.3	100	*0.52		0.26	<0.05	5.98	7.02
16	121.9	120	*0.18		0.18	<0.05	18.08	7.13
15	133.6	130	*0.18		0.18	<0.05	5.98	7.41
13	151.1	150	*0.20		0.20	<0.05	4.04	13.68
12	178.8	175				<0.05	3.74	20.92
11	201.7	200	*0.20		0.20	<0.05	2.63	27.05
9	261.8	260	*0.23		0.23	<0.05	3.68	34.52
8	292.7	290	*0.33	*0.06	0.33	0.06	2.28	36.79

Station 24 Lat: 10 0.05S Long: 125 0.03W Date:9-20-92 Time:12:40

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	12.5	10	0.89	0.57			7.93	2.96
21	42.6	40	0.91	0.67			8.99	3.03
19	83.0	80	0.84	0.62			9.19	2.21
18	100.9	100		0.44			9.43	1.84
17	122.7	120	0.32	0.29			7.79	2.63
16	132.5	130	0.11	0.29			9.10	3.13
15	143.5	140	0.24				6.27	3.19
13	177.1	175	0.10	0.19			7.78	3.80
12	204.6	200		0.17			10.09	6.62
11	233.5	230		0.16			4.47	11.13
9	292.0	290		0.21			5.14	28.95
8	333.7	330		0.14			3.60	33.03

Station 28 Lat: 4 0.11S Long: 125 0.00W Date:9-23-92 Time:10:34

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	6.0	3	1.29	0.54			8.24	4.13
21	31.9	30	1.20	0.73	0.51		10.88	4.20
19	63.1	60	1.35	0.75			9.05	3.86
18	81.8	80	*0.98	0.54	0.49		6.46	9.98
17	102.8	100	0.44	0.40	0.28		4.67	19.27
16	122.7	120	0.38	0.25			4.51	29.00
15	131.7	130	0.37	0.20	0.24		3.56	31.72
13	153.4	150	0.39	0.22	0.21		4.06	32.10
12	178.1	175	0.29	0.22	0.16		4.17	32.59
11	203.5	200	0.12	0.18			4.13	32.73
9	262.8	260	0.19	0.24	0.22		3.17	35.24
8	292.1	290	0.26	0.17	0.19		3.76	33.93

Station 30 Lat: 1 59.98S Long: 125 0.03W Date:9-24-92 Time:01:06

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	13.1	10	1.23	0.97			7.12	6.93
21	41.8	40	1.19	0.83			8.92	6.99
19	62.0	60	0.80	0.57			9.03	15.19
17	122.7	120	0.34	0.33			4.39	27.56
16	133.2	130	0.27	0.25			3.13	27.70
15	141.0	140	0.36	0.22			5.52	28.48
13	177.5	175	0.13	0.21			3.01	30.95
12	202.3	200		0.22			2.81	32.20
11	232.5	230	0.11	0.19			2.95	33.38
9	292.3	290	0.13	0.21			3.72	34.12
8	331.0	330		0.14			3.00	34.80

Station 34 Lat: 0 1.35S Long: 124 57.66W Date: 9-26-92 Time:06:55

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	5.3	3	1.34	1.01			14.26	8.29
22	22.7	20	0.95	0.63	0.67		7.17	10.39
20	53.0	50	1.13	0.67	0.78		5.48	11.04
18	82.1	80	0.18	0.29	0.33		4.79	15.27
17	92.2	90	0.10	0.24	0.18		5.79	15.82
15	112.7	110	*0.20	0.24	0.20		7.82	17.31
13	132.8	130	0.09	0.55			7.11	18.55
11	152.3	150	*0.15	0.21	0.15		4.64	22.81
9	183.0	180	*0.27	0.22	0.27		3.20	24.46
8	201.2	200	0.06	0.16	0.13		4.83	26.53
7	252.6	250	*0.19	0.19	0.19		2.98	34.30
6	303.1	300		0.14			6.76	36.77

Station 39 Lat: 2 59.86N Long: 125 0.26W Date: 9-28-92 Time: 15:52

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	13.2	10	1.27	0.88			7.99	2.48
22	42.8	40	1.26	0.92			11.70	3.55
20	63.2	60	1.17	0.89			8.62	4.08
18	92.6	90	0.74	0.58			7.18	11.73
17	102.4	100		0.40			6.19	19.57
15	123.6	120	0.36	0.32			7.88	25.72
13	141.8	140		0.18			5.11	29.15
11	168.5	165	0.16	0.36			5.09	30.32
9	202.1	200	0.24	0.31			4.26	32.29
8	253.4	250	0.18	0.25			4.34	33.86
7	302.9	300	0.13	0.31			4.98	31.21
6	401.6	400	0.17	0.20			3.54	37.28

Station 41 Lat: 5 3.12N Long: 125 0.90W Date: 9-29-92 Time: 18:11

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	13.1	10	1.25	0.89	0.72		10.56	0.62
22	41.4	40	1.40	0.91	0.71		9.17	0.66
20	62.2	60	1.11	0.75	0.47		8.88	0.69
18	91.2	90	0.70	0.79			8.54	8.53
17	102.0	100	0.44	0.37	0.20		6.62	11.56
15	121.2	120	0.23	0.29	0.27		8.43	25.96
13	143.4	140	0.23	0.21	0.17		2.77	33.69
11	166.7	165	0.15	0.21	0.11		3.25	33.83
9	203.1	200	0.09	0.11			2.56	33.35
8	252.0	250	0.08	0.14	0.07		3.20	32.94
7	304.5	300	0.19		0.21		7.43	33.80
6	401.8	400	0.22	0.49	0.23		6.32	37.79

Station 46 Lat: 9 59.28N Long: 124 59.36W Date: 10-1-92 Time:18:37

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	13.3	10	1.02	0.61			10.65	0.79
22	53.6	50	1.09	0.87			8.36	11.86
20	62.2	60	0.85	0.62			6.33	27.25
18	91.2	90	0.17	0.28			3.92	34.26
17	101.5	100	0.31	0.26			4.50	34.88
15	123.0	120	0.11	0.26			5.89	35.43
13	142.8	140		0.17			4.48	35.71
11	165.6	165	0.11	0.18			5.18	35.78
9	199.8	200	0.23	0.20			3.05	36.74
8	251.5	250	0.41	0.27			6.07	36.47
7	301.7	300	0.17	0.26			7.71	35.51

Station 50 Lat: 10 1.08N Long: 109 56.85W Date: 10-16-92 Time:22:15

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	6.7	3	*0.76	*0.46	0.38	0.23	8.41	0.12
22	23.1	20	*1.02	*0.32	0.51	0.16	8.57	0.94
20	53.0	50	*0.84	*0.36	0.42	0.18	11.28	4.32
18	83.5	80		*0.18		0.09	4.18	28.37
17	93.1	90	*0.54	*0.10	0.27	0.05	5.41	28.78
15	113.8	110	*0.19	*0.12	0.19	0.12	6.88	29.32
13	133.2	130	*0.19		0.19	<0.05	5.41	31.66
11	153.2	150		*0.08		0.08	5.26	31.96
9	183.5	180		*0.16		0.16	8.77	33.54
8	203.6	200				<0.05	11.22	33.81
7	253.5	250		*0.22		0.22	4.44	34.83
6	303.4	300		*0.21		0.21	4.13	34.20

Station 53 Lat: 4 58.14N Long: 109 54.98W Date: 11-1-92 Time:10:47

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
24	3.0	3	1.31	0.50	0.57		6.81	0.22
23	12.6	10	1.05	0.42	0.87		8.08	0.15
22	23.3	20	1.28	0.60	0.72		7.02	0.39
21	42.6	40	1.16	0.64	0.56		19.43	0.42
20	54.0	50	1.26	0.60	0.47		11.54	0.62
19	61.9	60	1.20	0.66	0.53		18.04	0.72
18	81.9	80	1.12	0.53	0.68		9.09	1.15
17	93.1	90	0.81	0.45	0.48		8.22	2.94
16	101.8	100	0.70	0.74	0.41		17.40	12.27
15	111.4	110	0.54	0.51	0.28		5.93	17.87
14	122.5	120	*0.34	0.14	0.34		8.40	19.92
13	131.1	130	0.43	0.17	0.31		6.15	20.51
12	143.8	140	0.46	0.19	0.17		24.57	26.91
11	151.4	150	0.27	0.18	0.32		4.91	31.47
9	180.5	180	0.22	0.22	0.25		8.25	30.74
8	203.5	200	0.11	0.11	0.32		12.46	30.66
7	254.1	250	0.06	0.08	0.41		5.16	32.34
6	304.1	300	0.18	0.20	0.33		5.52	34.85

Station 56 Lat: 2 6.75N Long: 110 6.80W Date: 11-2-92 Time:06:43

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
23	5.8	3	1.04	0.70			8.50	0.47
22	12.9	10	1.23	0.63			8.24	0.46
21	22.5	20	1.11	0.59			7.61	0.50
20	44.1	40	0.97	0.67			15.72	0.91
19	51.6	50	0.86	0.46			7.50	4.02
18	61.2	60	0.80	0.47			9.33	4.53
17	81.3	80	0.54	0.47			6.29	17.69
16	91.3	90	0.34	0.26				21.49
15	102.1	100	0.31	0.23			6.76	23.94
14	112.8	110	0.19	0.22			4.61	24.32
13	122.8	120	0.18	0.15			4.42	24.66
12	133.3	130	0.18	0.17			5.38	24.49
11	142.5	140	0.19	0.19			5.36	24.73
9	168.0	165	0.32	0.15			4.81	26.18
8	182.6	180	0.27	0.14			8.48	26.94
7	202.1	200	0.11	0.10			4.23	28.49
6	252.5	250	0.16	0.18			5.37	31.15

Station 59		Lat:	0 15.26N		Long:	109 59.98W	Date:	11-3-92 Time:19:28	
			PON			Protein N			
CTD Bottle	Pres.	Depth (m)	TF (µM)	GF/F (µM)		TF (µM)	GF/F (µM)	DON (µM)	N+N (µM)
23	6.1	3	*1.30	*0.90		0.65	0.45	6.75	9.80
22	12.4	10	1.65	*0.84		0.69	0.42	5.91	10.11
21	23.2	20	1.42	*0.92		0.67	0.46	7.51	10.65
20	42.8	40	1.33	*0.66		0.64	0.33	6.00	11.15
19	52.9	50	1.42	*0.74		0.47	0.37	7.13	11.19
18	83.3	80	0.20	*0.14		0.23	0.07	7.09	20.55
17	90.6	90	0.21	*0.12		0.17	0.06	4.48	20.62
16	102.6	100	0.14			0.25	<0.05	5.39	21.58
15	112.5	110	0.20			0.22	<0.05	4.74	24.56
14	121.8	120	0.15			0.13	<0.05	7.03	24.89
13	132.0	130	0.09	*0.06		0.22	0.06	5.61	25.09
12	142.6	140	0.15	*0.09		0.21	0.09	5.44	26.18
11	152.0	150	0.10			0.27	<0.05	4.84	25.43
10	168.5	165	0.10			0.23	<0.05	9.65	25.96
9	183.1	180	0.14			0.18	<0.05	3.93	27.61
8	202.9	200	*0.22			0.22	<0.05	11.52	27.65
7	251.2	250	0.21			0.25	<0.05	5.87	30.82
6	301.5	300	*0.26			0.26	<0.05	4.12	34.95

Station 65		Lat:	2 5.60S		Long:	109 54.10W	Date:	11-6-92 Time:05:31	
			PON			Protein N			
CTD Bottle	Pres.	Depth (m)	TF (µM)	GF/F (µM)		TF (µM)	GF/F (µM)	DON (µM)	N+N (µM)
24	3.3	3	1.60	1.06				6.67	8.00
23	9.0	10	1.48	1.23				10.26	8.27
22	20.5	20	1.69	1.19				8.26	8.41
21	28.7	30	1.61	1.15				6.80	8.37
20	39.2	40	1.35	1.17				15.47	8.54
19	58.9	60	1.12	0.83				8.20	10.51
18	79.9	80	0.76	0.65				6.63	22.39
17	100.0	100	0.41	0.31				5.56	24.70
16	118.7	120	0.22	0.18				8.64	24.98
15	129.5	130	0.23	0.16				4.34	25.53
14	151.3	150	0.25	0.23				8.29	25.49
13	174.0	175	0.35	0.13				4.25	27.22
12	198.3	200	0.20	0.11				8.65	30.15
11	230.6	230	0.17	0.13				6.61	31.25
10	260.2	260	0.21	0.10				9.48	32.53
9	289.8	290	0.17	0.09				5.85	32.77
8	329.6	330	0.36	0.08				7.45	33.94
7	380.3	380	0.32	0.12				12.74	36.74

Station 68

deep >175	Lat:	4 59.86S	Long:	110 2.13W	Date:11-8-92	Time:05:54	
shallow	Lat:	4 59.95S	Long:	110 1.72W	Date:11-8-92	Time:07:45	

CTD Bottle	Pres.	Depth (m)	PON TF (μM)	PON GF/F (μM)	Protein N TF (μM)	Protein N GF/F (μM)	DON (μM)	N+N (μM)
24		3	1.24	0.91	0.63		7.17	9.03
21		30	1.18	0.87	0.54		8.38	9.20
19		60	0.98	0.74	0.49		9.27	7.33
18		80	0.44	0.50	0.43		5.08	16.01
17		100	*0.44	0.36	0.22		4.73	20.32
16		120	0.27	0.14	0.16		8.53	28.68
14		140	*0.17	0.18	0.17		5.47	31.23
12	175.4	175	0.06	0.35	0.21		2.80	31.20
11	200.9	200	0.14	0.19	0.16		4.90	30.91
10	230.0	230		0.13	0.04		4.68	31.29
9	259.0	260	0.09	0.11	0.15		3.27	29.92
8	287.8	290	0.19	0.12	0.18		6.86	31.97

Station 70

deep >175	Lat:	8 0.05S	Long:	109 59.96W	Date:11-9-92	Time:04:29	
shallow	Lat:	7 59.68S	Long:	110 0.16W	Date:11-9-92	Time:06:19	

CTD Bottle	Pres.	Depth (m)	PON TF (μM)	PON GF/F (μM)	Protein N TF (μM)	Protein N GF/F (μM)	DON (μM)	N+N (μM)
12	2.5	3	1.47	1.06			7.36	8.91
11	9.1	10	1.32	0.99			11.92	8.62
10	20.0	20	1.32	0.80			6.94	8.62
9	28.8	30	1.29	0.96			16.06	8.89
8	39.7	40	1.37	0.90			9.24	8.80
7	59.0	60	1.37	0.76			6.55	8.73
6	79.7	80	0.94	0.45			7.84	4.42
5	102.9	100	0.54	0.31			5.23	7.75
4	116.8	120	0.41	0.26			11.48	15.25
3	129.2	130	0.33	0.23			4.16	16.67
2	140.2	140	0.22	0.18			5.85	23.76
1	149.5	150	0.22	0.12			6.03	26.47
12	172.0	175	0.23	0.12			12.81	29.93
11	201.2	200	0.18	0.17			4.97	29.64
10	228.0	230	0.24	0.13			12.64	32.08
9	257.4	260	0.31	0.17				32.97
8	287.5	290	0.22	0.15			5.50	33.63
7	332.2	330	0.21	0.14			9.07	34.56

Station 72	Lat: 2S	Long: 95W	Date: 11-13-92	Time:15:10

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
12		3	1.65	1.31	0.57		21.50	11.49
11		20	1.71	1.24	0.64		10.33	13.04
10		40	1.07	0.63	0.42		5.58	22.46
9		60	0.55	0.46	0.22		5.98	23.97
8		80	0.44	0.22	0.18		8.52	23.06
7		100	0.41	0.20	0.22		11.06	26.09
5		150	0.26	0.16	0.13		3.97	25.25
4		200	0.28	0.15	0.16		9.15	27.32
3		330	0.20	0.09	0.19		7.33	34.15

Station 73	Lat: 0 00.86S	Long: 95 03.34W	Date: 11-14-92	Time:17:35

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
12		3	2.91	2.68	0.99		7.75	9.39
11		20	1.82	1.24	0.77		6.86	12.08
10		40	0.94	0.93	0.46		18.06	17.43
9		60	0.31	0.30	0.14		4.99	16.97
8		80	0.28	0.22	0.23		5.93	18.48
7		100	0.20	0.23	0.14		5.25	19.78
6		120	0.29	0.15	0.10		6.54	20.53
5		150	0.18	0.15	0.06		4.77	22.28
4		200	*0.12	0.15	0.12		4.75	25.45
3		330	0.21	0.17	0.19		13.87	35.91

Station 74	Lat: 1 57.28N	Long: 94 09.05W	Date: 11-15-92	Time:14:29

CTD Bottle	Pres.	Depth (m)	PON TF (µM)	PON GF/F (µM)	Protein N TF (µM)	Protein N GF/F (µM)	DON (µM)	N+N (µM)
12		3	1.67	*0.78	0.72	0.39	9.38	0.00
11		20	1.54	*0.74	0.72	0.37	6.78	0.00
10		40	1.60	*0.78	0.71	0.39	7.28	0.82
9		60	1.27	*0.68	0.70	0.34	6.06	13.28
8		80	0.56	*0.30	0.20	0.15	5.18	21.64
7		100	0.26	*0.14	0.24	0.07	6.36	21.00
6		120	0.34		0.22	<0.05	4.71	23.29
5		150	0.15		0.29	<0.05	17.56	23.95
4		200	0.10		0.15	<0.05	6.13	24.18
3		330	0.08	*0.19	0.17	0.19	6.22	34.29

Appendix 2 Summary of chlorophyll data from NOAA EPOCS/CO_2/US JGOFS cruises DI-92-03 and DI-92-04 the fall of 1992. Measured using GF/F filters except where 0.2 µm Nucleopore filters are noted. (local time and date)

A) 140°W transect

Station: 2 Latitude: 10 00.38N Date: 9/9/92 Time: 18:17			Station: 6 Latitude: 5 00.39N Date: 9/11/92 Time: 07:20		Station: 8 Latitude: 2 59.35N Date: 9/12/92 Time: 10:53		
Depth	GF/F µg/L	0.2 µm µg/L	Depth	µg/L	Depth	GF/F µg/L	0.2µm µg/L
3	0.02	---	0	0.16	3	0.06	0.14
10	0.02	0.04	8	0.16	10	0.09	0.16
20	0.02	0.03	17	0.18	20	0.06	0.16
40	0.02	0.05	27	0.16	40	0.05	0.09
50	0.04	0.06	42	0.20	50	0.03	0.07
60	0.05	0.09	65	0.17	60	0.03	0.05
80	0.12	0.18			80	0.01	0.03
90	0.13	0.22			90	0.01	0.01
100	0.11	0.16			100	0.00	0.00
110	0.09	0.14			110	0.00	0.00
120	0.07	0.14			120	0.00	0.00
130	0.05	0.08			130	0.00	0.00
140	0.04	0.06					
150	0.03	0.05					
165	0.02	0.04					
180	0.01	0.02					
200	0.00	0.01					

A) 140°W continued

Station: 10 Latitude: 1 00.50N Date: 9/13/92 Time: 04:50		Station: 15 Latitude: 0 30.00S Date: 9/15/92 Time: 01:00	
Depth	µg/L	Depth	µg/L
0	0.05	0	0.21
7	0.07	10	0.11
15	0.07	13	0.11
23	0.07	21	0.11
37	0.09	34	0.15
57	0.10	52	0.16

B) 125°W transect

Station: 46 Latitude: 9 57.36N Date: 10/1/92 Time: 22:18		Station: 41 Latitude: 5 03.36N Date: 9/29/92 Time: 19:30		Station: 38 Lat: 2 01.78N Date: 9/28/92 Time: 07:51	
Depth	µg/L	Depth	µg/L	Depth	µg/L
0	0.10	0	0.20	0	0.15
8	0.12	8	0.17	8	0.15
18	0.14	18	0.21	17	0.14
29	0.16	29	0.17	27	0.15
46	0.19	46	0.22	42	0.17
70	0.03	70	0.25	65	0.31

B) 125°W continued

Station: 34 Latitude: 0 01.69N Date: 9/26/92 Time: 08:24		Station: 30 Latitude: 1 59.56S Date: 9/24/92 Time: 07:15	
Depth	µg/L	Depth	µg/L
0	0.40	0	0.15
6	0.37	8	0.17
13	0.40	17	0.17
21	0.37	27	0.17
34	0.40	42	0.18
52	0.49	65	0.27

C) 110°W transect

Station: 51 Latitude: 8 00.02N Date: 10/31/92 Time: 06:30		Station: 53 Latitude: 5N Date: 11/1/92 Time: 09:50		Station: 56 Latitude: 2N Date: 11/2/92 Time: 05:55	
Depth	µg/L	Depth	µg/L	Depth	µg/L
0	0.10	11	0.12	0	0.15
38	0.08	20	0.17	9	0.16
63	0.08	31	0.16	15	0.17
80	0.13	49	0.16	23	0.16
92	0.19	76	0.17	37	0.18
108	0.06			57	0.16

C) 110°W continued

Station: 60 Latitude: 0 09.00N Date: 11/4/92 Time: 08:00		Station: 65 Latitude: 2S Date: 11/6/92 Time: 08:58		Station: 67 Latitude: 4S Date: 11/7/92 Time: 10:05	
Depth	µg/L	Depth	µg/L	Depth	µg/L
0	0.21	0	0.16	0	0.12
8	0.17	10	0.18	9	0.16
14	0.25	17	0.18	15	0.13
22	0.21	27	0.20	23	0.13
35	0.30	42	0.32	37	0.12
54	0.24	65	0.23	57	0.17

Station: 68 Latitude: 5S Date: 11/8/92 Time: 06:51		Station: 70 Latitude: 8S Date: 11/9/92 Time: 05:30	
Depth	µg/L	Depth	µg/L
0	0.11	0	0.06
9	0.11	9	0.07
15	0.10	16	0.07
23	0.13	25	0.07
37	0.14	39	0.07
57	0.15	60	0.09

D) 95°W transect

Station: 74 Latitude: 2N Date: 11/15/92 Time: 06:50		Station: 73 Latitude: 0 Date: 11/14/92 Time: 06:36		Station: 72 Latitude: 2S Date: 11/13/92 Time: 06:39	
Depth	µg/L	Depth	µg/L	Depth	µg/L
0	0.19	0	0.28	0	0.12
11	0.17	8	0.29	6	0.15
19	0.20	14	0.21	13	0.16
30	0.21	22	0.35	21	0.17
48	0.27	35	0.46	34	0.26
73	0.32	54	0.27	52	0.32