Suspended particulate nitrogen $\delta^{15}N$ versus nitrate utilization: observations in Monterey Bay, CA

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Abstract

Over a four-year period the $\delta^{15}N$ values of particulate nitrogen, PN, suspended in surface waters at several sites in Monterey Bay were observed to vary between 1.3 and 7.6‰, a significant portion of the known oceanic $\delta^{15}N_{PN}$ range. $\delta^{15}N_{PN}$ generally increased as [NO$_3^-$] declined, in keeping with the hypothesis that $\delta^{15}N_{PN}$ rises with increased NO$_3^-$ utilization. Previously reported measurements conducted elsewhere in the Pacific and Indian oceans generally match this trend. The distinctly non-linear relationship between $\delta^{15}N_{PN}$ and ln[NO$_3^-$] is consistent with a closed-system, Rayleigh fractionation model where: NO$_3^-$ is the reactant, PN is the accumulating end product, initial [NO$_3^-$] = 30 µM, initial $\delta^{15}N_{NO_3}$ = 7‰, and biological fractionation, $\epsilon$ = 9‰. There is, however, considerable scatter in the data about this trend ($r^2 = 0.37$), with the uncertainty (± 1 standard deviation) of empirically estimating nitrate utilization from $\delta^{15}N_{PN}$ of ± 4.3 µM. Also, the $\epsilon$ required to fit the model to observed $\delta^{15}N_{PN}$ variations produces $\delta^{15}N_{NO_3}$ at intermediate and low [NO$_3^-$] that is much higher than has thus far been observed in the ocean. One way to rectify this discrepancy would be if $\epsilon$ declined as [NO$_3^-$] decreased. Lack of significant linear correlation between $\delta^{15}N_{PN}$ and phytoplankton growth rate, cell size, and 1/[NO$_3^-$] argues against $\delta$ being influenced by limitations imposed by diffusive NO$_3^-$ transport. However, logarithmic functions of these factors can explain up to 50% of the observed $\delta^{15}N_{PN}$ variability, suggesting that active NO$_3^-$ transport or species-specific effects may be influencing $\epsilon$ and hence $\delta^{15}N_{PN}$ in Monterey Bay.

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1. Introduction

The $^{15}$N/$^{14}$N in various marine inorganic and organic nitrogen pools has been shown to vary significantly over a range of spatial and temporal scales (reviews by Owens, 1987; Wada and Hattori, 1991, Altabet, 1996). While it is clear that such variations must largely reflect the isotopically selective processing of nitrogen by biota, the use of $^{15}$N/$^{14}$N measurements in field samples to discern or quantify the operation of specific N pathways can be problematic. In the case of $\delta ^{15}$N measurements of marine particulate nitrogen (PN, here presumed to be largely if not entirely particulate organic N), these concerns include the potential complexity of nitrogen sources for PN formation as well as often undescribed and/or variable fractionations involved with N uptake by biota and subsequent N excretion/loss. Nevertheless, significant correlations between $\delta ^{15}$N$_{PN}$ and [NO$_3^-$ ] have been observed and appear to be generally consistent with a model where organic nitrogen is formed through $^{14}$N-selective uptake of NO$_3^-$ in a closed system (review by Altabet, 1996). $\delta ^{15}$N$_{PN}$ subsequently has been used as a relative index of nutrient utilization, notably in conjunction with paleo $\delta ^{15}$N$_{PN}$ reconstructions from the sedimentary record (e.g., François et al., 1992; Calvert et al., 1992; Altabet and François, 1994 a, b; Farrell et al., 1995; Ganeshram et al., 1995; François et al., 1997).

Fundamental to the use of $\delta ^{15}$N$_{PN}$ as a proxy for nutrient utilization is the veracity with which present-day observations match those expected by such a model. There are numerous reasons to anticipate that $\delta ^{15}$N$_{PN}$ will not behave as predicted by the above closed-system scenario, and therefore will not be a faithful recorder of NO$_3^-$ utilization over broad spatial and temporal scales. The potential difficulties in employing such a model include the possibility that NO$_3^-$ may not be the only or even the primary source for new nitrogen production in surface waters. Imported N$_2$, NH$_4^+$, and dissolved organic N utilization, each with potentially distinct $\delta ^{15}$N signatures may significantly influence resident $\delta ^{15}$N$_{PN}$ (e.g. Cifuentes et al., 1988; Minagawa and Wada, 1986; Montoya et al., 1991; Carpenter et al., 1997). Secondly, the isotopic fractionation imparted during the biological incorporation of any one of these N substrates into PN may vary with substrate concentrations as well with algal species, physiology, and growth rate (e.g. Wada and Hattori, 1978; Wada, 1980; Montoya and McCarthy, 1995; Pennock et al., 1996; Waser et al., 1998). Strictly speaking, PN should be viewed as an intermediate product, not as end product, because it is subject to “downstream” metabolism, diageneis, and trophic transfer, each with potentially significant isotopic consequences to the residual PN (e.g. Wada, 1980; Minagawa and Wada, 1984; Macko and Estep, 1984; Libes and Deuser, 1988; Checkley and Miller, 1989). Also, PN sampled via bulk filtration is usually a complex mixture of autotrophic and heterotrophic biota and detrital material where trophic and metabolic $^{15}$N fractionations likely have altered the $\delta ^{15}$N$_{PN}$ over that representative of the organic nitrogen initially formed by phytoplankton (e.g. Wada, 1980; Rau et al., 1990).

Despite these complexities in using any single-isotope fractionation model to interpret $\delta ^{15}$N$_{PN}$, this latter parameter has been observed to be generally higher in low-[NO$_3^-$] waters, broadly consistent with the idea that $\delta ^{15}$N$_{PN}$ increases with the
degree to which surface ocean NO$_3^-$ is consumed (e.g. Wada, 1980; Goering et al., 1990; François et al., 1992; Altabet and François, 1994a, b; Altabet, 1996). An ongoing, long-term study of Monterey Bay oceanography by the Monterey Bay Aquarium Research Institute (MBARI) allowed us to further investigate $\delta^{15}$N$_{PN}$ variations in one area over many seasons, and in concert with other biological and chemical measurements by MBARI.

The Bay is characterized by large spatial and temporal changes in nutrient chemistry and phytoplankton productivity as a result of seasonally varying, wind-driven upwelling (Skogsberg, 1936; Bolin and Abbott, 1963; Breaker and Broenkow, 1994; Chavez, 1995, 1996; Pilskañ et al., 1996). During the winter, oceanographic conditions are relatively uniform in both space and time, with low levels of nitrate ($\sim 1$ $\mu$M), chlorophyll ($\sim 1$ $\mu$g/l), and primary productivity ($< 500$ mg C/m$^2$) (Chavez, 1996). During the upwelling period (spring–summer), variability increases as a result of episodic upwelling and downwelling events. Maximum levels of nitrate are close to 30 $\mu$M, and blooms of phytoplankton (primarily diatoms) usually develop that can reach concentrations approaching 20 $\mu$g chlorophyll/l and with primary productivity rates in excess of 4 g C/m$^2$ d$^{-1}$ (Chavez, 1996). During late summer and occasionally during the upwelling period, wind reversals can bring oligotrophic waters from offshore close to the coast (Rosenfeld et al., 1994). Sea-surface temperatures range from around 9°C during upwelling to $> 16$°C in late summer and even higher during El Niño conditions. The region therefore presents an excellent opportunity to investigate $\delta^{15}$N$_{PN}$ variations and their causes in a highly dynamic yet readily accessible oceanographic setting.

2. Methods

Suspended PN was collected by filtering 1 l of surface seawater through a precombusted Whatman GFF filter (nominal pore size = 0.7 $\mu$m). Water for filtering was obtained aboard R/V Pt Lobos by pumping surface waters from nearshore (36°47.8'N, 121°50.8'W), a mid-bay (36°44.7'N, 122°01.2'W), and an outer bay (36°41.9'N, 122°23.9'W) sites at approximately monthly intervals beginning in July 1993. The filters were stored frozen prior to analysis.

Upon return to the laboratory, all filters were prepared and analyzed for total nitrogen elemental and isotopic abundances as described by Rau et al. (1990). By convention, the $^{15}$N/$^{14}$N of each sample is reported as the relative per mil (‰) difference between the sample ratio and the ratio of a standard. That is:

$$\delta^{15}\text{N} = \frac{R_{\text{sample}}/R_{\text{standard}} - 1}{R_{\text{standard}}} \times 1000(\%\text{oo})$$

where, $R = ^{15}$N/$^{14}$N and “standard” = air N$_2$, respectively. The analytical precision (1 standard deviation) of these measurements was approximately 0.2‰.

Measurements made in tandem with the sampling for isotopic analysis included [NO$_3^-$] by conventional wet chemistry (Sakamoto et al., 1990) and phytoplankton biomass, growth rate, size, and taxonomic composition following the procedures described in Chavez et al. (1991).
3. Results and discussion

$\delta^{15}$N$_{PN}$ ranged from 1.3 to 7.6‰ over the 4 years studied. This covers a large portion of the oceanic $\delta^{15}$N$_{PN}$ range thus far reported (roughly −5 to +15‰, Wada and Hattori, 1991), and attests to the N dynamics in this region and the associated factors that influence $\delta^{15}$N$_{PN}$ here. There is a negative relationship between $\delta^{15}$N$_{PN}$ and $[\text{NO}_3^-]$, and this relationship generally overlaps that observed in several other locations in the temperate and subpolar N. Pacific, Indian, and Southern oceans (Fig. 1). This compiled trend also fits ($r^2 = 0.37$) that anticipated (e.g. Mariotti et al., 1981) if PN represents the accumulating product in a closed system where the initial nitrate concentration, $[\text{NO}_3^-]_i = 30 \mu$M, initial $\delta^{15}$N$_{\text{NO}_3^-}$, $\delta^{15}$N$_{\text{NO}_3^-}$, = 7‰, and the kinetic isotope fractionation, $\varepsilon$, associated with PN production is 9‰ (Fig. 1). The general curvilinear relationship between observed $\delta^{15}$N$_{PN}$ and ln$[\text{NO}_3^-]$ is not characteristic of the behavior of the instantaneous product, IP, in a closed system model (Fig. 1). However, the overlap between observed $\delta^{15}$N$_{PN}$ and modeled $\delta^{15}$N$_{IP}$ in high $[\text{NO}_3^-]$ waters, and the scatter of observed $\delta^{15}$N$_{PN}$ above the modeled $\delta^{15}$N$_{AP}$ trend (Fig. 1) could reflect a varying contribution of IP to the PN analyzed. Variations

Fig. 1. Surface water $\delta^{15}$N$_{PN}$ vs $[\text{NO}_3^-]$ as observed in: Monterey Bay (open triangle, squares, circles, crosses; this study), the N. Pacific Ocean (solid triangles; Wada, 1980; Saino and Hattori, 1985; Goering et al., 1990), and the Indian/Southern Ocean (dots; Altabet and François, 1994a). $[\text{NO}_3^-]$ values < 0.01 µM are assigned a value of 0.01. Lines denote the response of accumulated product $\delta^{15}$N($\delta^{15}$N$_{AP}$), instantaneous product $\delta^{15}$N($\delta^{15}$N$_{IP}$), and NO$_3^-$ $\delta^{15}$N ($\delta^{15}$N$_{\text{NO}_3^-}$) in a closed system where initial $[\text{NO}_3^-] = 30 \mu$M, initial $\delta^{15}$N$_{\text{NO}_3^-}$, = 7‰, and kinetic N isotope fractionation, $\varepsilon$, = 9‰. The sensitivities of the modeled $\delta^{15}$N$_{PN}$ to changes in these parameter values are explored in Fig. 2. See text for further discussion.
in $\delta^{15}N_{PN}$ could therefore be sensitive to the relative importance of IP to the total suspended PN pool, and thus an indicator of the rapidity with which PN is removed from the surface waters via sinking or diagenesis (e.g., Voss et al., 1996).

In March of 1997 Altabet et al. (submitted) found deep water $\delta^{15}N_{NO_3}$ to be 7–8‰ (and $[NO_3^{-}] \approx 30 \mu M$) in this region, similar to the $\delta^{15}N_{NO_3}$ and $[NO_3^{-}]$ values inferred above from the Rayleigh model fit to data (Fig. 1). Subsurface $\delta^{15}N_{NO_3}$ elsewhere in the NE Pacific has ranged from 5 to 8‰ when denitrification was not significant (Cline and Kaplan, 1975; Liu and Kaplan, 1989; Altabet and François, 1994b). The presence of heterotrophic biomass in the PN analyzed could have elevated the $\delta^{15}N_{PN}$ asymptote over that anticipated for the primary accumulated product in such a system, and variability in the size and trophic makeup of this heterotrophic component may also contribute to the scatter in observed $\delta^{15}N_{PN}$ about its mean trend with $[NO_3^{-}]$ (Fig. 1). Also, sediment resuspension (Washburn et al., 1993; Silver et al., 1998) could have influenced the $\delta^{15}N_{PN}$ measured. Yet, we found no significant correlation between $\delta^{15}N_{PN}$ and either the contemporaneously measured C/N ($r^2 = 0.024, N = 122$) or the % heterotroph component (Chavez and Buck, unpublished data) of the total plankton analyzed ($r^2 = 0.104, N = 75$).

A value of 9‰ for the biological fractionation factor, $\epsilon$, is within the range observed experimentally in algae (Wada and Hattori, 1978; Montoya and McCarthy, 1995; Pennock et al., 1996; Waser et al., 1998), but is somewhat higher than most values estimated empirically from field measurements (e.g., Wada, 1980; Goering et al., 1990; Horrigan et al., 1990; Altabet et al., 1991; Sigman et al., 1997; Altabet et al., submitted). However, the higher 9‰ $\epsilon$ value inferred here is primarily required to accomodate the low $\delta^{15}N_{PN}$ found elsewhere in the Pacific or Indian Oceans (Fig. 1). A value of approximately 6‰ appears to be more characteristic of the Monterey Bay data (Fig. 1). Based on vertical profiles of Monterey Bay $\delta^{15}N_{NO_3}$ and $[NO_3^{-}]$ in March of 1997, Altabet et al. (submitted) infer $\epsilon$ values of about 5‰. However, lack of $\delta^{15}N_{NO_3}$ measurements contemporaneous with our $\delta^{15}N_{PN}$ does not allow us to more directly determine $\epsilon$ and its variability during the study. Even then, with f ratios ranging as low as 0.1 in Monterey Bay (Kudela, 1995) it is evident that referencing such isotope fractionation to $NO_3^-$ alone ignores isotope affects imparted on PN by non-nitrate N assimilation.

3.1. Other Model Uncertainties

The significant scatter of $\delta^{15}N_{PN}$ about the modeled $\delta^{15}N_{AP}$ trend could reflect spatial or temporal variations in the preceding $[NO_3^{-}]$, $\delta^{15}N_{NO_3}$, and $\epsilon$ values relevant to Monterey Bay. Using the above parameter settings as a base case, these values are separately varied in Fig. 2 to ascertain their potential contribution to the observed $\delta^{15}N_{PN}$ variability. For example, varying $[NO_3^{-}]$ from 10 to 50 µM increases $\delta^{15}N_{AP}$ at a given $[NO_3^{-}]$ and does so with increasing sensitivity at higher $[NO_3^{-}]$ (Fig. 2a). There is a negative relationship between $\epsilon$ and $\delta^{15}N_{AP}$ whose sensitivity is also higher at elevated $[NO_3^{-}]$ (Fig. 2e). $\delta^{15}N_{AP}$ at a given $[NO_3^{-}]$ also increases with $\delta^{15}N_{NO_3}$, but in contrast to the preceding factors, the sensitivity of $\delta^{15}N_{PN}$ response to $\delta^{15}N_{NO_3}$ slightly decreases with increasing $[NO_3^{-}]$ (Fig. 2c). The
presence of some significantly lower $\delta^{15}N_{PN}$ values at very low $[\text{NO}_3^-]$ (e.g. Fig. 2a) has been ascribed to the utilization of nitrogen sources other than nitrate, especially nitrogen fixation (e.g. Wada, 1980; Minagawa and Wada, 1986; Carpenter et al., 1997), or isotopic fractionation associated with N recycling and/or sedimentary N loss from the euphotic zone (e.g. Saino and Hattori, 1985; Altabet, 1988; Voss et al., 1996; Altabet, 1996).

Whatever the cause of the $\delta^{15}N_{PN}$ scatter about its trend with $[\text{NO}_3^-]$, the resulting uncertainty of empirically estimating $\text{NO}_3^-$ loss from $\delta^{15}N_{PN}$ amounts to an estimation error ($\varepsilon$) of $\pm 4.3 \, \mu\text{M}$ in Monterey Bay. A 95% confidence interval for such estimate would then be roughly $\pm 8.5 \, \mu\text{M}$. If $[\text{NO}_3^-]$ is specified to be $30 \, \mu\text{M}$, then the equivalent uncertainty of estimating the proportion of $\text{NO}_3^-$ utilized amounts to $\pm 28\%$. The uncertainty in knowing $[\text{NO}_3^-]$ and $\delta^{15}N_{NO_3}$ for any point in time adds to this estimation error. We point out that the complex circulation and hydrography of the region (e.g. Brecker and Broenkow, 1994; Rosenfeld et al., 1994) means that upwelling source waters are variable, and thus $[\text{NO}_3^-]$ and $\delta^{15}N_{NO_3}$ are likely not constant. Less confidence in applying such a model to reconstruct paleo N utilization from sedimentary $\delta^{15}N_{PN}$ might be expected due to uncertainties in the magnitude and variability of diagenetic PN isotope effects and the potential for seasonally selective preservation of surface water $\delta^{15}N_{PN}$ in sediments. For example, Altabet et al. (submitted) found sinking PN $\delta^{15}N$ in Monterey Bay to seasonally vary from 6 to 9.5%oo on average higher and less variable than the suspended surface water $\delta^{15}N_{PN}$ reported here.

There is an apparent incongruity, however, in the observed behavior of $\delta^{15}N_{PN}$ and $\delta^{15}N_{NO_3}$ and that predicted by the above closed-system model. Specifically, the $\delta^{15}N_{NO_3}$ response simulated by the preceding base model is usually significantly higher and more sensitive to changes in $[\text{NO}_3^-]$ than has thus far been observed (Fig. 2b, d and f). $\delta^{15}N_{NO_3}$ values $>40\%$ are required by the base model to generate appropriate $\delta^{15}N_{NP}$ vs $[\text{NO}_3^-]$ trends at low and moderate $[\text{NO}_3^-]$, while surface water $\delta^{15}N_{NO_3}$ of this magnitude has never been seen in the N. Pacific (Fig. 2b, d and f) or elsewhere (Altabet, 1996; Voss et al., 1996; Sigman et al., 1997; Altabet et al., submitted). While it is possible to roughly fit the modeled $\delta^{15}N_{NO_3}$ trend to that observed (e.g., Fig. 2f, line $\varepsilon = 3\%_o$) this requires a substantial lowering of $\varepsilon$ and thus an unrealistically high $\delta^{15}N_{PN}$ at high $[\text{NO}_3^-]$ (e.g., Fig. 2e, line $\varepsilon = 3\%_o$). It is impossible to approximate satisfactorily both observed $\delta^{15}N_{PN}$ and $\delta^{15}N_{NO_3}$ trends with the closed system model with any given fixed parameterization. Certainly, future tests of the relevance of the closed system model will require measurement of both

Fig. 2. Observed $\delta^{15}N_{PN}$ and $\delta^{15}N_{NO_3}$ variations with $[\text{NO}_3^-]$ in comparison to those modeled by a closed system with various initial $[\text{NO}_3^-]$ ($[\text{NO}_3^-]$), initial $\delta^{15}N_{NO_3}$ ($\delta^{15}N_{NO_3}$), and kinetic fractionation ($\varepsilon$) values as indicated. Solid line denotes initial model fit as shown in Fig. 1. Observed $\delta^{15}N_{PN}$ as in Figure 1. Observed $\delta^{15}N_{NO_3}$ as reported for the north central Pacific Ocean (triangles; Cline and Kaplan, 1975), S. California Coastal waters (dots; Liu, 1979), and the subarctic Pacific Ocean (squares; Altabet and François, 1994b). Parentheses denote potential analytical uncertainties in $\delta^{15}N_{NO_3}$ at low $[\text{NO}_3^-]$ (Cline and Kaplan, 1975; Liu, 1979).
δ¹⁵Nₚₙ and δ¹⁵N_NO₃ over a wide range of [NO₃⁻]. Presently, there are few δ¹⁵N_NO₃ analyses available, and the few reported at low [NO₃⁻] are inconsistent (Fig. 2b, d and f), possibly due to analytical artifacts in low-[NO₃⁻] samples (Cline and Kaplan, 1975; Liu, 1979).

3.2. Potential Variations in ε

One way of preventing significant δ¹⁵N_NO₃ elevation in such a model while matching observed δ¹⁵Nₚₙ trends with [NO₃⁻] would be to remove the assumption that biological isotope fractionation is constant. Specifically, ε could decline with decreasing [NO₃⁻] in such a way that the modeled δ¹⁵Nₚₙ and δ¹⁵N_NO₃ trends would better fit those observed. Indeed, ε has been found in algal cultures to be quite variable, affected by algal species, growth rate, and physiology in addition to [NO₃⁻] (Wada and Hattori 1978; Montoya and McCarthy, 1995; Pennock et al., 1996; Waser et al., 1998). After finding a significant negative relationship between ¹⁵N fractionation and algal growth rate, Wada and Hattori (1978) proposed that ε should vary directly with the ratio of intracellular to ambient [NO₃⁻]. This is analogous to the effect of the ratio of intracellular to ambient [CO₂aq] on the δ¹³C in plants (Farquhar et al., 1982) as later applied to plankton (e.g., Rau et al., 1992) in a diffusively transported, CO₂aq-based system. Similarly, if intracellular [NO₃⁻] as well as δ¹⁵N_NO₃ are held constant, then δ¹⁵Nₚₙ should linearly increase as 1/[NO₃⁻] increases (as [NO₃⁻] declines). In keeping with this idea we observe that Monterey Bay δ¹⁵Nₚₙ generally increases with 1/[NO₃⁻], although in a manner that is not obviously linear (Fig. 3a).

With regard to intracellular [NO₃⁻], it has been shown for intracellular [CO₂aq] that under diffusive transport this quantity should be negatively related to phytoplankton cell growth rate and cell size (Goericke et al., 1994, Rau et al., 1996, 1997). Thus, at constant δ¹³C_CO₂aq, phytoplankton δ¹³C should increase with cell size/[CO₂aq] and growth rate/[CO₂aq], with such effects having been observed in algal cultures (Fry and Wainwright, 1991; Laws et al., 1995). Are there analogous relationships between δ¹⁵Nₚₙ, [NO₃⁻], and growth rate that are consistent with that expected in a NO₃⁻ diffusive transport scheme?

In addition to [NO₃⁻] measurements, both phytoplankton community growth rate and mean cell size at the Monterey Bay sampling sites were usually determined concurrently (F. Chavez and K. Buck, unpublished data) with δ¹⁵Nₚₙ during 1993–1995. With these data δ¹⁵Nₚₙ is observed to increase with both cell size/[NO₃⁻] and growth rate/[NO₃⁻] (Fig. 3b and c). We hasten to add, however, that the principal contributor to these trends is 1/[NO₃⁻]: neither cell size (r² = 0.116, N = 68) nor especially growth rate (r² = 0.005, N = 65) alone is significantly correlated with δ¹⁵Nₚₙ. This lack of correlation argues against ¹⁵N fractionation that is affected by the limitations of diffusive NO₃⁻ transport as envisioned by Wada and Hattori (1978). It is also noted that the δ¹⁵Nₚₙ trends with [NO₃⁻]-normalized growth rate and cell size are highly non-linear (Fig. 3), a feature that again is not anticipated in a diffusional setting if δ¹⁵N_NO₃ is held constant. Non-linearity, however, has been encountered experimentally in phytoplankton δ¹³C response to growth
Fig. 3. Monterey Bay $\delta^{15}$N$_{PN}$ vs: (a) $1/[\text{NO}_3^-]$, (b) phytoplankton growth rate $/[\text{NO}_3^-]$, and (c) mean spherical diameter of phytoplankton cells $/[\text{NO}_3^-]$ (c). Symbols as in Fig. 1. Cell size and growth rate data were not available for 1996.
rate/[CO₂\text{aq}]), and was attributed to the effects of non-diffusive transport at low [CO₂\text{aq}] (Laws et al., 1997).

3.3. Active NO₃⁻ transport and species effects

As a negatively charged molecule, non-diffusive, active NO₃⁻ transport across cell membranes is indeed relevant (Wheeler, 1983; Montoya, 1994). After failing to find a significant growth rate effect on algal ε in nitrate-limited algal cultures, Montoya and McCarthy (1995) argued that ¹⁵N fractionation associated with active NO₃⁻ transport across the cell membrane rather than intracellular fractionation could be the primary determinant of the overall ε expressed by the cell. They suggested that variations in light regime and/or species-specific effects cause variation in the isotope fractionation associated with active transport, possibly reconciling the lack of growth rate effects between their nitrate-limited cultures and the large effects seen in Wada and Hattori's (1978) light-limited experiments.

With regard to species effects, it is possible that the δ¹⁵N_{PN} increase seen with increasing cell size (Fig. 3c) could reflect a decline in ¹⁴N selectivity that is intrinsic to larger sized algal species. For example reduced ¹³C fractionation has been observed in larger diatoms (Fry and Wainright, 1991), and in the case of ¹⁵N fractionation, large inter-taxa effects have been reported (e.g. Wada, 1980; Montoya and McCarthy, 1995). We do note a positive though non-statistically significant relationship between centric diatom biomass and δ¹⁵N_{PN} in our data set (Fig. 4), possibly suggesting that certain species within this group may fractionate N isotopes less than other taxa. However, in

![Fig. 4. δ¹⁵N_{PN} versus centric diatom carbon concentration in Monterey Bay 1993–1995.](image)
the preceding discussions the lack of (i) parallel $\delta^{15}N_{\text{NO}_3}$ measurements in Monterey Bay, and (ii) assurance that NO$_3^-$ was the dominant N substrate do not allow us to effectively address $^{15}$N fractionation and thus to factor out $\delta^{15}N_{\text{NO}_3}$ variations from the observed changes in $\delta^{15}N_{\text{PN}}$.

4. Conclusions

A general, multi-year $\delta^{15}N_{\text{PN}}$ trend with [NO$_3^-$] was observed in Monterey Bay that is consistent with: (1) trends seen elsewhere in the N. Pacific, Indian, and Southern oceans, and (2) a model of $\delta^{15}N_{\text{PN}}$, where PN represents accumulating product in a nitrate-based, closed biological system. There is, however, considerable scatter in the observed relationship, and hence the uncertainties in using $\delta^{15}N_{\text{PN}}$ alone as a proxy for NO$_3^-$ utilization even in one location appear to be substantial. Also, the high rate at which $\delta^{15}N_{\text{NO}_3}$ increases with declining [NO$_3^-$] in such a model appears to be inconsistent with observations elsewhere. Factors that might reconcile such differences between modeled and observed $\delta^{15}N_{\text{NO}_3}$ behavior include allowing biological isotope fractionation to vary. Lack of correlation between $\delta^{15}N_{\text{PN}}$ and growth rate or cell size argues against diffusional NO$_3^-$ transport as a factor affecting $^{15}$N fractionation. However, significant $\delta^{15}N_{\text{PN}}$ correlations with highly nonlinear functions of cell size, growth rate, and 1/[NO$_3^-$] suggest that variations in species composition or active NO$_3^-$ transport might be affecting $\delta^{15}$N in this region. Parallel measurements of $\delta^{15}N$ and $\delta^{15}N_{\text{NO}_3}$ especially on how [NO$_3^-$] are needed to more effectively test these ideas.

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