

## Dissolved zinc in the subarctic North Pacific and Bering Sea: Its distribution, speciation, and importance to primary producers

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[1] The eastern subarctic North Pacific, an area of high nutrients and low chlorophyll, has been studied with respect to the potential for iron to control primary production. The geochemistry of zinc, a critical micronutrient for diatoms, is less well characterized. Total zinc concentrations and zinc speciation were measured in near-surface waters on transects across the subarctic North Pacific and across the Bering Sea. Total dissolved zinc concentrations in the near-surface ranged from 0.10 nmol L<sup>-1</sup> to 1.15 nmol L<sup>-1</sup> with lowest concentrations in the eastern portions of both the North Pacific and Bering Sea. Dissolved zinc speciation was dominated by complexation to strong organic ligands whose concentration ranged from 1.1 to 3.6 nmol L<sup>-1</sup> with conditional stability constants ( $K'_{ZnL/Zn}$ ) ranging from 10<sup>9.3</sup> to 10<sup>11.0</sup>. The importance of zinc to primary producers was evaluated by comparison to phytoplankton pigment concentrations and by performing a shipboard incubation. Zinc concentrations were positively correlated with two pigments that are characteristic of diatoms. At one station in the North Pacific, the addition of 0.75 nmol L<sup>-1</sup> zinc resulted in a doubling of chlorophyll after 4 days.

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### 1. Introduction

[2] Surface macronutrient concentrations (nitrate, phosphorus, silicate) in the subarctic N. Pacific are not depleted by phytoplankton growth [McAllister *et al.*, 1960], leading to its characterization as a high nutrient, low chlorophyll (HNLC) region. Surface concentrations of iron (Fe) in this region are very low (e.g., 0.06 nmol L<sup>-1</sup>) [Martin *et al.*, 1989]. Additions of Fe to both bottle incubations [Martin and Fitzwater, 1988; Coale, 1991; Boyd *et al.*, 1996; Lam *et al.*, 2001; Crawford *et al.*, 2003; Leblanc *et al.*, 2005] and mesoscale ocean patches [Tsuda *et al.*, 2003; Boyd *et al.*, 2004; Marchetti *et al.*, 2006] have demonstrated the ability of Fe to regulate primary production in this region.

[3] Zinc (Zn) concentrations are also very low in the surface waters of the subarctic N. Pacific [Bruland *et al.*, 1978;

Martin *et al.*, 1989; Lohan *et al.*, 2002]. Though Zn is an important micronutrient for phytoplankton, the role of Zn in limiting marine primary production is not as clear as that of Fe. Low Zn concentrations can limit phytoplankton growth in cultures [Anderson *et al.*, 1978; Brand *et al.*, 1983; Morel *et al.*, 1991]. Though similarly low Zn concentrations are found in the open ocean, unlike Fe, Zn additions to bottle experiments generally have had little or no effect on total chlorophyll biomass [Coale, 1991; Coale *et al.*, 1996; Scharek *et al.*, 1997; Gall *et al.*, 2001; Coale *et al.*, 2003; Crawford *et al.*, 2003; Franck *et al.*, 2003; Ellwood, 2004; Lohan *et al.*, 2005; Leblanc *et al.*, 2005], with the exception of Fe-Zn (and Fe-Co) co-limitation observed at the Costa Rica Upwelling Dome [Franck *et al.*, 2003, Table 7; Saito *et al.*, 2005]. The few observations of a primary productivity increase in response to Zn addition is likely due in part to the ability of some phytoplankton to substitute cobalt (Co) and cadmium (Cd) for Zn in enzyme systems [e.g., Lee *et al.*, 1995; Yee and Morel, 1996]. Some phytoplankton can also substitute non-Fe containing proteins [LaRoche *et al.*, 1996] or copper containing proteins [Peers and Price, 2006] to alleviate Fe stress when Fe concentrations are low. The low solubility of Fe at oceanic pH precludes there being a large reservoir of Fe in deep waters in the same way that there is for classic 'nutrient-type' trace metals such as Zn. The reduced deep source of Fe compared to Zn likely contributes to the greater prevalence of observed Fe limitation in the world's oceans.

[4] Co is preferentially utilized over Zn by some phytoplankton species or vice versa [Sunda and Huntsman, 1995;

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Saito *et al.*, 2002; Xu *et al.*, 2007; Saito and Goepfert, 2008], which suggests that the ratio of Zn to Co may influence species composition in the ocean [Sunda and Huntsman, 1995]. While this hypothesis has not been explicitly tested, the ability of Zn to influence species composition has been shown. In an incubation in the eastern subarctic N. Pacific, the addition of Zn and Fe together (+Zn/+Fe) resulted in a higher proportion of phytoplankton in the smaller size fraction (0.2–5  $\mu\text{m}$ ) compared to the addition of Fe alone [Crawford *et al.*, 2003]. The community in the +Zn/+Fe addition had a higher abundance of small diatoms and small flagellates and less coccolithophores and ciliates than the Fe alone addition [Crawford *et al.*, 2003]. Zn may influence not only which taxa dominate but which species within a taxa are most successful. In a bottle incubation experiment in the sub-Antarctic zone near New Zealand, the addition of Zn caused a community shift from a large colonial pennate diatom to a smaller, less-silicified, solitary pennate diatom species [Leblanc *et al.*, 2005].

[5] Zn deficiency can also affect the extent of calcification of *E. huxleyi*, a ubiquitous marine coccolithophore [Schulz *et al.*, 2004]. Under Zn limitation, *E. huxleyi* cells become more heavily calcified due to a slowing of growth rates with no corollary decrease in calcium carbonate ( $\text{CaCO}_3$ ) production rate [Schulz *et al.*, 2004]. This effect, along with the potential of Zn to influence species composition, suggests that Zn may be an important determinant of the organic carbon: $\text{CaCO}_3$  rain ratio. At low Zn:Co, coccolithophores and small cyanobacteria would be expected to dominate over diatoms based on their growth rates at low Zn:Co [Sunda and Huntsman, 1995]. This community shift would decrease the organic carbon: $\text{CaCO}_3$  due to coccolithophores'  $\text{CaCO}_3$  shells. In addition, the coccolithophores growing under low Zn would be more highly calcified than those experiencing replete levels of Zn, further decreasing the organic carbon: $\text{CaCO}_3$  rain ratio.

[6] Culture studies with the synthetic ligand ethylenediaminetetraacetic acid (EDTA) have shown that Zn bioavailability is related to the Zn free ion ( $\text{Zn}^{2+}$ ) concentration rather than the total Zn concentration [e.g., Anderson *et al.*, 1978]. Natural organic ligands in the surface ocean strongly bind Zn [Donat and Bruland, 1990; Bruland, 1989; Ellwood and van den Berg, 2000; Ellwood, 2004; Jakuba *et al.*, 2008]. These ligands dominate the speciation of the total Zn pool, with  $\text{Zn}^{2+}$  typically accounting for 5% or less of the total dissolved Zn. The inventory of Zn binding ligands can be extremely dynamic with production and removal of ligands occurring on timescales of 1 day [Lohan *et al.*, 2005]. Zn speciation measurements of shipboard incubation experiments suggest that the phytoplankton community can survive at lower  $\text{Zn}^{2+}$  than is known to limit some large phytoplankton species in cultures [Ellwood, 2004; Lohan *et al.*, 2005]. However, smaller diatoms such as *Thalassiosira oceanica* are able to grow at  $\text{Zn}^{2+}$  abundances below those that limit larger phytoplankton species [Sunda and Huntsman, 1992, 1995] suggesting that low zinc could cause species composition effects or that diatoms under these conditions can meet their Zn requirement by substituting Co or Cd.

[7] This study examines total dissolved Zn concentrations and Zn speciation across the subarctic N. Pacific and Bering Sea and the potential influence of Zn on phytoplankton growth, species composition and nutrient utilization. The

eastern part of the subarctic N. Pacific, the Gulf of Alaska, is characterized by low levels of chlorophyll and high nutrient concentrations. Fe fertilization experiments have demonstrated the ability of Fe to regulate primary production in this region [e.g., Martin and Fitzwater, 1988; Boyd *et al.*, 2004]. In the western part of the subarctic N. Pacific, there are relatively high rates of atmospheric deposition of Fe [Duce and Tindale, 1991] and a high seasonal export by diatoms [Honjo, 1997], though portions of Western Subarctic Gyre can also experience Fe limitation [Suzuki *et al.*, 2005]. In this area, the east-flowing North Pacific Current originates from the Kuroshio Extension in the central Pacific. Enhanced vertical mixing events associated with variability in the strength of the North Pacific Current and with eddies of the system can bring nutrient-rich waters to the surface [Cummins and Freeland, 2007; Chu and Kuo, 2010] which likely contributes to the observed gradient in Zn concentrations from east to west noted by Fukuda *et al.* [2000].

[8] The Bering Sea is split into a deep western basin whose surface waters are characterized by low summertime primary productivity and a wide continental shelf to the east that is highly productive, particularly at the shelf break with decreasing productivity toward the coast [Springer *et al.*, 1996]. There is evidence that the deep western portion of the Bering Sea is an HNLC limited by Fe [Tyrrell *et al.*, 2005; Aguilar-Islas *et al.*, 2007].

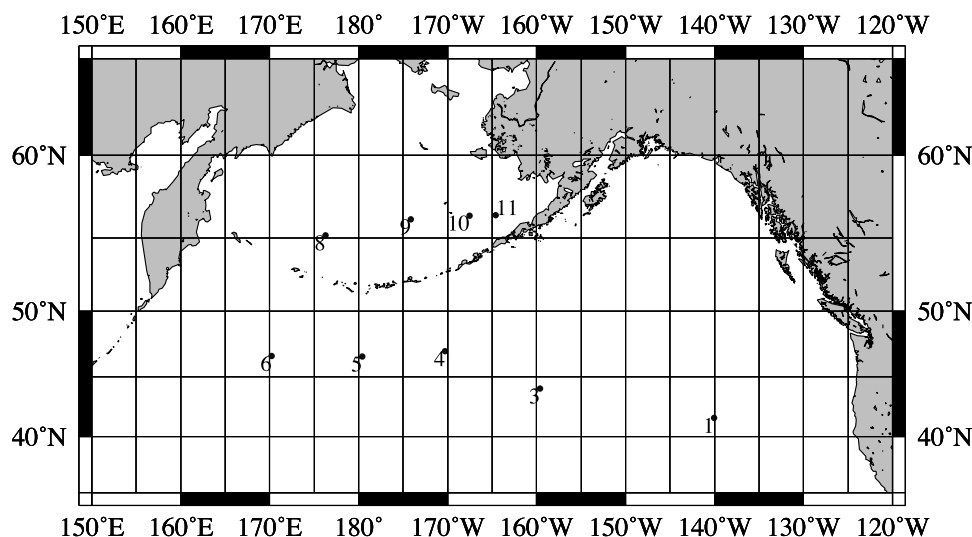
## 2. Methods

### 2.1. Sample Collection and Handling

[9] Samples were collected in the N. Pacific and Bering Sea aboard the R/V Kilo Moana in the summer of 2003 (Figure 1). Water samples were collected using 10 L Teflon-coated Go-Flos (General Oceanics) on a Kevlar hydrowire. Seawater was collected from the Go-Flo bottles by connecting Teflon tubing to both ports and over-pressuring the top port with filtered ultra-high purity nitrogen gas. Seawater passed directly from Teflon tubing through an all plastic sandwich filter rig with acid-cleaned 0.4  $\mu\text{m}$ , 142 mm diameter polycarbonate membrane filters and into rigorously acid-washed (including sequential soaks in warm Citranox detergent, J.T. Baker Instra-analyzed HCl, J.T. Baker Instra-analyzed  $\text{HNO}_3$ , and pH 2 Seastar HCl) low density polyethylene (LDPE) or Teflon bottles. Samples for total dissolved Zn analysis (LDPE bottles) were acidified to approximately pH 2 by the addition of 2 mL HCl (Seastar) per liter of seawater. Samples for Zn speciation analysis (Teflon bottles) were stored refrigerated (4°C) until analysis (typically within 10 days). All manipulation of the samples occurred in a laminar flow bench inside clean laboratories.

### 2.2. Total Dissolved Zn Analysis

[10] Total dissolved zinc ( $\text{Zn}_T$ ) concentrations were measured using isotope dilution and magnesium hydroxide pre-concentration followed by analysis using inductively coupled plasma mass spectrometry (ICP-MS) after Wu and Boyle [1998] and Saito and Schneider [2006]. Centrifuge tubes (15 mL polypropylene, Globe Scientific) were cleaned by soaking in 2N HCl (J.T. Baker instra-analyzed) at 60°C for 48 h followed by rinsing 5 times with pH 2 HCl (J.T. Baker instra-analyzed) and once with pH 2 HCl (Seastar). Finally, tubes were filled to a positive meniscus with pH 2 HCl



**Figure 1.** Map of station locations from the North Pacific and Bering Sea visited aboard the R/V *Kilo Moana* in June–August 2003.

(Seastar) and capped until use. At the time of analysis, tubes were rinsed once with sample and then filled to approximately 13.5 mL (exact volume determined gravimetrically). Samples were then spiked with  $^{66}\text{Zn}$  (98.9% as  $^{66}\text{Zn}$ , Cambridge Isotope Laboratories, Inc.) to an estimated  $^{66}\text{Zn}$ : $^{64}\text{Zn}$  ratio of 9. This ratio minimizes error magnification [Heumann, 1988]:

$$R_{\text{optimum}} = \sqrt{\left(\frac{^{66}\text{Zn}}{^{64}\text{Zn}}\right)_{\text{sample}} \times \left(\frac{^{66}\text{Zn}}{^{64}\text{Zn}}\right)_{\text{spike}}} \quad (1)$$

The added  $^{66}\text{Zn}$  spike was allowed to equilibrate with the samples overnight. The following day, 125  $\mu\text{L}$  of ammonia (Seastar) was added to each tube. After 90 s, the tube was inverted and after an additional 90 s, tubes were centrifuged for 3 min at  $3000 \times g$  (3861 rpm) using a swinging bucket centrifuge (Eppendorf 5810R). The majority of the supernatant was carefully decanted. Tubes were then respun for 3 min to firm pellet and the remaining supernatant was shaken out. Pellets were dissolved on the day of ICP-MS analysis using 0.5–1.5 mL of 5% nitric acid (Seastar).  $\text{Zn}_T$  concentrations were calculated as

$$[\text{Zn}_T] = \frac{f_{64}^{\text{spike}}}{f_{64}^{\text{natural}}} \times [\text{Zn}_{\text{spike}}] \times \frac{\text{spike volume}}{\text{sample volume}} \times \left(\frac{R_{\text{measured}} - R_{\text{spike}}}{R_{\text{natural}} - R_{\text{measured}}}\right) \quad (2)$$

where  $f_{64}$  is the fraction of the abundance of  $^{64}\text{Zn}$  over the abundance of all the Zn isotopes and  $R$  is the ratio of  $^{66}\text{Zn}$ : $^{64}\text{Zn}$ . To measure the procedural blank, 1 mL of low zinc surface seawater was treated the same as samples, and calculations were performed as though it was a 13.5 mL sample (Zn contribution from the 1 mL is considered negligible). The average blank value was  $0.12 \text{ nmol L}^{-1}$  with a detection limit of  $0.09 \text{ nmol L}^{-1}$  (calculated as three times the standard deviation of the blank). The daily procedural blank value was subtracted from measured sample values.

[11] The efficiency of the magnesium hydroxide precipitate at scavenging Zn was tested. One mL aliquots of

acidified seawater ( $n = 3$ ) were equilibrated with the radioisotope  $^{65}\text{Zn}$  (approximately  $0.5 \mu\text{Ci}$ ) for 2 h. Thirty  $\mu\text{L}$  of ammonia was added to each sample and after 1.5 min, the samples were centrifuged for 3 min. The amount of  $^{65}\text{Zn}$  was quantified in the seawater before precipitation, in the precipitate, and in the supernatant using a sodium iodide detector. The percent of  $^{65}\text{Zn}$  that was captured, on average, by the magnesium hydroxide pellet was 96% and the fraction remaining in the supernatant was 2%. The accuracy of the method was evaluated by measuring a NASS-5 seawater standard (National Research Council of Canada). The NASS standard has a certified value of  $1.56 \pm 0.60 \text{ nmol L}^{-1}$  Zn. The value obtained by this method ( $1.00 \pm 0.03 \text{ nmol L}^{-1}$ ) was within the specified range of the certified value.

[12] ICP-MS measurements were made using a Thermo Finnigan ELEMENT2 in medium resolution mode, which was sufficient to resolve  $^{64}\text{Zn}$  from the potential interference peak due to Mg-Ar. Another potential interference at mass 64 is that of  $^{64}\text{Ni}$ , which is a minor isotope of Ni (natural abundance of less than 1%). This interference was not measured explicitly. In many cases, the contribution of the mass 64 signal due to Ni will not be significant relative to  $^{64}\text{Zn}$ , however at the lowest Zn concentrations measured here, the contribution due to Ni may be significant. Total dissolved Ni concentrations were measured in this study using voltammetric methods (see below). The potential influence of the  $^{64}\text{Ni}$  on the  $\text{Zn}_T$  concentrations calculated here is discussed in the Results section.

### 2.3. Total Dissolved Ni Analysis

[13] Total dissolved nickel (Ni) was also measured here as described in Saito *et al.* [2005]. Briefly, a Metrohm 663 hanging mercury drop electrode stand with a Perfluoroalkoxy Teflon (PFA) sample vessel was interfaced with an Ecochemie  $\mu\text{Autolab}$  system and GPES (General Purpose Electrochemical System, Eco Chemie) software. Dimethylglyoxime (DMG) from Aldrich was recrystallized in Milli-Q (Millipore) water in the presence of  $10^{-3}$  ethylenediaminetetraacetic acid (EDTA; Sigma Ultra) to remove impurities, dried, and redissolved in high performance liquid

chromatography (HPLC)-grade methanol. We prepared 1.5 mol L<sup>-1</sup> sodium nitrite (Fluka Puriss) purified by an overnight equilibration with prepared Chelex-100 beads (BioRad). An N-(2-hydroxyethyl)piperazine-N'-(3-propanesulfonic acid) (EPPS) buffer solution (Fisher) was purified by passing through a column with 3 mL of prepared Chelex-100 beads. For total dissolved nickel analyses, 8.50 mL of ultraviolet light irradiated filtered seawater was pipetted into the Teflon sample cup followed by 50  $\mu$ L of 0.5 mol L<sup>-1</sup> EPPS, 20  $\mu$ L 0.1 mol L<sup>-1</sup> DMG, and 1.5 mL of 1.5 mol L<sup>-1</sup> sodium nitrite. The  $\mu$ Autolab protocol involved a 3-min 99.999% N<sub>2</sub> gas purge at 120 kPa, a 90-s deposition time at -0.6 V, a 15-s equilibration period, and a high-speed negative scan from -0.6 to -1.4 V at 10 V s<sup>-1</sup>. A linear sweep waveform was used on drop size three (0.52 mm<sup>2</sup>) and a stirrer speed of five. Total nickel concentrations were determined with three standard additions of 1 nmol L<sup>-1</sup> using a NiCl<sub>2</sub> solution prepared in pH 2 Milli-Q water.

### 2.3. Zn Speciation Determinations

[14] Zn speciation was determined using an anodic stripping voltammetry method [Jakuba *et al.*, 2008] that was adapted from that of Fischer and van den Berg [1999] for the measurement of total lead and cadmium. Measurements were made using a 663 VA stand (Metrohm) consisting of a glassy carbon rotating disc working electrode (2 mm diameter), a glassy carbon rod counter electrode, and a double junction Ag, AgCl, saturated KCl reference electrode. The electrode was interfaced to a PC (IBM Thinkpad) using an IME663 and  $\mu$ Autolab I (Eco Chemie). Before each day of speciation analysis, the glassy carbon rotating disc electrode (RDE) was manually polished with aluminum oxide. Then, a cyclic voltammetry step was performed cycling the voltage between -0.8 V and 0.8 V 50 times. Reagents used included 4-(2-Hydroxyethyl)-1-piperazinepropanesulfonic acid (EPPS), ammonium thiocyanate, and mercuric chloride. The pH of the EPPS buffer was adjusted to 8.1. EPPS and thiocyanate reagents were run through a pre-cleaned chelex column [Price *et al.*, 1989] to remove trace metal contamination. The thiocyanate reagent is added to improve the reproducibility of the mercury film and to ensure full de-plating of the film between samples [Fischer and van den Berg, 1999].

[15] Zn speciation titrations were set up in 15 mL Teflon vials (Savillex). In order to prevent wall loss, Teflon vials were equilibrated with specific concentrations of Zn before use and each vial was always used for the same concentration of Zn. Before a titration, the vials were rinsed with 12 mL of sample. Then, 12 mL of sample was added into each vial, along with 1.7 mmol L<sup>-1</sup> EPPS. For each sample aliquot, the appropriate concentration of Zn was allowed to equilibrate with the sample in the sample vial for 10 min. Then, the sample aliquot was poured into the voltammetric cell and 4.2 mmol L<sup>-1</sup> ammonium thiocyanate and 10.4  $\mu$ mol L<sup>-1</sup> mercuric chloride were added. Each sample aliquot was purged with ultra-high purity nitrogen gas for 5 min to remove oxygen. A conditioning potential of 0.6 V was held for 60 s. The mercury film and Zn' were then deposited at a potential of -1.5 V for 180 s. After a 10 s equilibration time, the voltage was ramped in square wave mode from -1.3 V to 0.6 V. A

frequency of 50 Hz was used and the step potential and modulation amplitude were 4.95 mV and 49.95 mV, respectively. A peak in current was evident at -1.1 V representing the concentration of inorganic Zn species (Zn'). The total ligand concentration ([L<sub>T</sub>]) and conditional stability constant (K'<sub>cond,Zn'</sub>) with respect to Zn' were calculated by performing a linearization of the titration data [Ružič, 1982; van den Berg, 1982], where [Zn']/[ZnL] is plotted versus [Zn']. The equation for the resulting line is

$$\frac{[Zn']}{[ZnL]} = \frac{[Zn']}{[L_T]} + \frac{1}{(K'_{cond,Zn'} \times [L_T])} \quad (3)$$

Hence, [L<sub>T</sub>] can be calculated as  $\frac{1}{slope}$  and K'<sub>cond,Zn'</sub> as  $\frac{1}{([L_T] \times y\text{-intercept})}$ . The Zn<sup>2+</sup> concentrations in Table 1 were calculated based on the relationships:

$$K'_{cond,Zn'} = \frac{[ZnL]}{[Zn'][L']} \quad (4)$$

$$[Zn'] = \alpha_{Zn} \times [Zn^{2+}] \quad (5)$$

[Zn'] was calculated from equation 3, assuming that [L]<sub>T</sub> - [Zn]<sub>T</sub> = [L'] and that [ZnL] = [L]<sub>T</sub> - [L']. A value of 2.2 was used for  $\alpha_{Zn}$  [Turner *et al.*, 1981].

### 2.4. Shipboard Incubation

[16] Trace metal clean water was collected with an air-driven Teflon pump that pumped water from approximately 15 m depth directly into an acid-washed HDPE carboy, housed in a trace-metal free bubble constructed of a HEPA filter and plastic sheeting. Acid-washed polycarbonate bottles were filled with unfiltered seawater from the carboy. A time zero sample for chlorophyll *a* was also collected from the carboy. Additions were made to duplicate polycarbonate bottles as follows: control (no addition), +Fe (2.5 nmol L<sup>-1</sup> FeCl<sub>3</sub>), +Zn (0.75 nmol L<sup>-1</sup> ZnCl<sub>2</sub>), +Zn/+Fe (0.75 nmol L<sup>-1</sup> ZnCl<sub>2</sub>, 2.5 nmol L<sup>-1</sup> FeCl<sub>3</sub>). Bottles were tightly capped and placed in an on-deck water bath supplied with flowing seawater for temperature control. Sunlight was attenuated with bluegel shading (Roscolux 65: Daylight Blue, Stage Lighting Store) to mimic 15 m irradiances. On days 2 and 4, one bottle was removed from the incubator and sampled for chlorophyll. In order to avoid potential contamination by Zn, which is a highly contamination prone element, bottles were sacrificed upon sampling. Samples for analysis of dissolved nutrients from select bottles were also collected, filtered, and frozen.

### 2.5. Chlorophyll, Nutrients, and HPLC Pigments

[17] For chlorophyll *a* analysis, seawater was passed through a GF/F filter. Filters were extracted in 90% acetone overnight at -20°C. Chlorophyll *a* (Chla) concentrations were corrected for the interference of phaeophytin by the addition of acid. All samples for nutrient analysis were filtered through the trace metal filtration rig described above into acid-cleaned polypropylene tubes and stored frozen until analysis. Analysis was performed by the Ocean Data Center (ODC) at the Scripps Institution of Oceanography for nitrate, nitrite, ammonium, silicic acid, and phosphate. In addition, phosphate analyses were run on board at select

**Table 1.** Dissolved Zn, SRP, and Ni Data<sup>a</sup>

Station	Dates Occupied	Latitude	Longitude	Depth (m)	Zn <sub>T</sub> (nmol L <sup>-1</sup> )	Ni <sub>T</sub> (nmol L <sup>-1</sup> )	SRP (μmol L <sup>-1</sup> )	Chla (μg L <sup>-1</sup> )	L <sub>T</sub> (nmol L <sup>-1</sup> )	logK' <sub>ZnL/Zn'</sub>	Zn <sup>2+</sup> (nmol L <sup>-1</sup> )											
1	24 Jun–28 Jun 2003	41°06'N	140°06'W	15	0.10 (0.02) <sup>b</sup>	3.3	0.29	0.14	1.1	10.0	0.005											
				50	0.35 (0.01)	3.6	<b>0.33</b>	0.99														
				75	0.10 (0.02)		<b>0.48</b>	0.27	1.6	10.0	0.003											
				150	1.99 (0.02)	3.5																
				300	2.80 (0.10)																	
				500	6.82 (0.03)	8.3																
3	4 Jul–5 Jul 2003	44°02'N	159°59'W	20	0.20 (0.03) <sup>b</sup>	4.2	<b>1.10</b>	0.37	1.8	10.5	0.002											
				4	7 Jul–8 Jul 2003	47°01'N	170°30'W	13	0.56 (0.10)	4.0	1.34	0.72										
								26	0.71 (0.06)	5.6	1.35	1.01	2.2	10.5	0.007							
								40	0.89 (0.08)	8.0	1.47	0.41	2.3	11.0	0.003							
								60	1.24 (0.11)	5.9	<b>1.63</b> <sup>c</sup>	0.16										
								100	1.51 (0.01)	6.6	1.57	0.03	2.6	10.1	0.048							
150	5.10 (0.07)	6.1	<b>2.08</b>					0.01														
5	12 Jul–13 Jul 2003	46°59'N	179°58'W	25	0.65 (0.02)	6.2	1.40	0.81	2.3	10.5	0.006											
				6	15 Jul 2003	46°60'N	170°20'E	20	0.78 (0.11)	7.5	1.40	0.56	2.5	9.8	0.033							
								40	1.66 (0.09)	6.4	1.45		2.7	10.1	0.059							
								50	2.39 (0.29)		<b>1.64</b>	0.65										
								80	2.73 (0.01)	5.9	<b>1.74</b> <sup>c</sup>	0.13										
								150	5.26 (0.02)	7.5	2.33	0.02										
300	8.52 (0.02)	10.9	3.13																			
8	21 Jul–22 Jul 2003	55°18'N	176°31'E	20	0.89 (0.01)	8.2	1.46	0.86	3.6	9.3	0.071											
				9	24 Jul 2003	56°19'N	174°15'W	20	1.15 (0.02)													
								10	26 Jul 2003	56°43'N	167°50'W	14	0.42 (0.05)		<b>0.31</b>	0.63						
												11	27 Jul 2003	56°45'N	164°60'W	16	0.29 (0.02) <sup>b</sup>	5.1	<b>0.24</b>	1.21		
																1000	9.76 (0.10)		2.23 <sup>c</sup>			
																1500	9.58 (0.27)		3.03			
3000	12.27 (0.19) <sup>d</sup>		2.77 <sup>c</sup>																			

<sup>a</sup>Values in parentheses represent the standard deviation of duplicate or triplicate analyses for Zn<sub>T</sub>. The majority of the SRP values were measured from sub-samples collected from Go-Flo bottles. Where sub-samples from Go-Flo bottles were unavailable, values are reported from samples collected from Niskin bottles and measured on-board (bold values).

<sup>b</sup>Potentially high level of Ni interference, see section 3 for details.

<sup>c</sup>Samples where the SRP values measured from the Go-Flo sub-samples did not match expected oceanographic depth trend. When available these values were replaced by SRP measured on-board from Niskin bottles. See section 3 and Figure 4a.

<sup>d</sup>Contamination suspected, see section 3 for details.

stations using the molybdate blue method [Murphy and Riley, 1962]. In general, analyses from ODC are used here, except on several occasions where frozen nutrient values were not oceanographically consistent due to preservation problems [Maher and Woo, 1998, and references therein] and shipboard analyses were substituted. Besides these few outliers, shipboard and frozen samples compared well. Samples were collected for phytoplankton pigment analysis from Niskin bottles on a CTD rosette frame. Analysis of HPLC pigments was performed after Bidigare [1991] and Bidigare and Trees [2000].

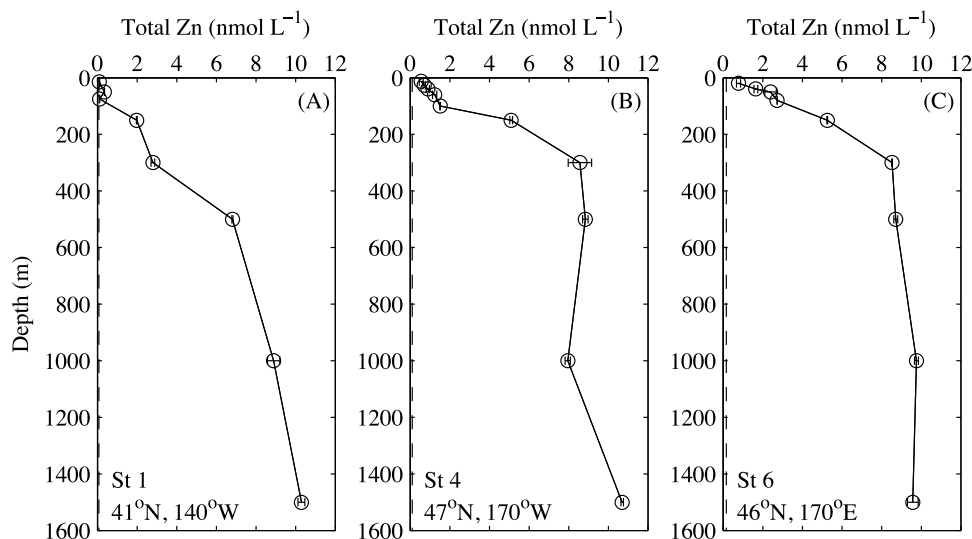
### 3. Results

#### 3.1. Total Dissolved Zn

[18] Zn<sub>T</sub> was measured in the near-surface at 5 stations in the N. Pacific and 4 stations in the Bering Sea. Zn<sub>T</sub> concentrations in the near-surface ranged from 0.10 to 1.15 nmol L<sup>-1</sup> (Table 1). Concentrations increased from

east to west in the North Pacific, in agreement with reports of integrated Zn<sub>T</sub> in the upper 100 m of this region [Fukuda et al., 2000]. The Zn<sub>T</sub> value at Station 1 (0.10 nmol L<sup>-1</sup>) is within the range reported for the near-surface in that vicinity (0.06–0.25 nmol L<sup>-1</sup>) [Martin et al., 1989]. During the SEEDS Fe enrichment experiment, which was performed further west of Station 6 (48.5°N, 165°W), near-surface Zn<sub>T</sub> concentrations were even higher than those observed at Station 6 (1.18–2.35 nmol L<sup>-1</sup>) [Kinugasa et al., 2005], which may indicate that the east–west trend of increasing Zn<sub>T</sub> continues beyond our study region.

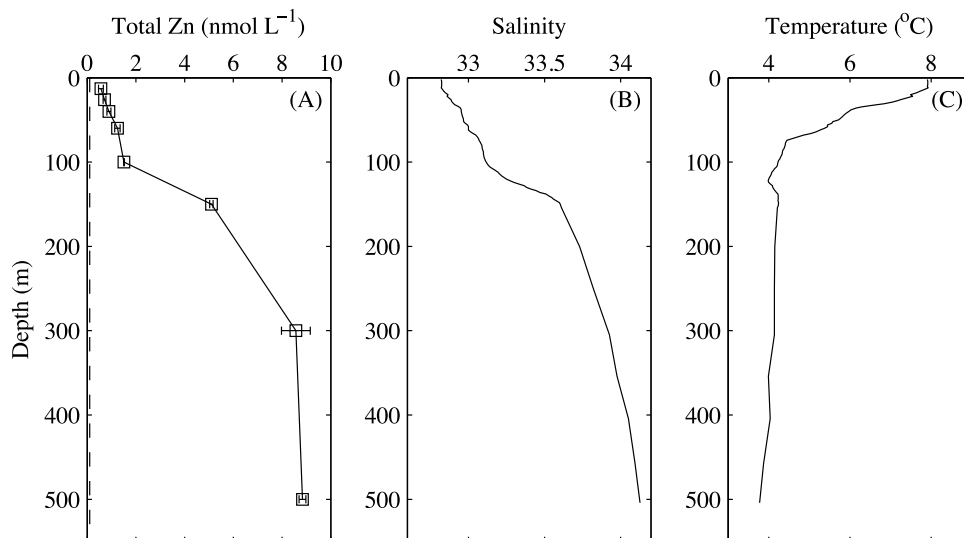
[19] Near-surface Zn<sub>T</sub> concentrations in the Bering Sea were high (~1 nmol L<sup>-1</sup>) in the deep western portion of the basin and low in the eastern waters over the Bering shelf. Previous studies of Zn in the Bering Sea have shown elevated Zn<sub>T</sub> concentrations at depth (3.5 nmol L<sup>-1</sup> at 30 m and 20 nmol L<sup>-1</sup> at 3000 m) in the western Bering Sea [Fujishima et al., 2001], but low surface Zn<sub>T</sub> concentrations (<0.41 nmol L<sup>-1</sup>) [Fujishima et al., 2001; Leblanc et al.,



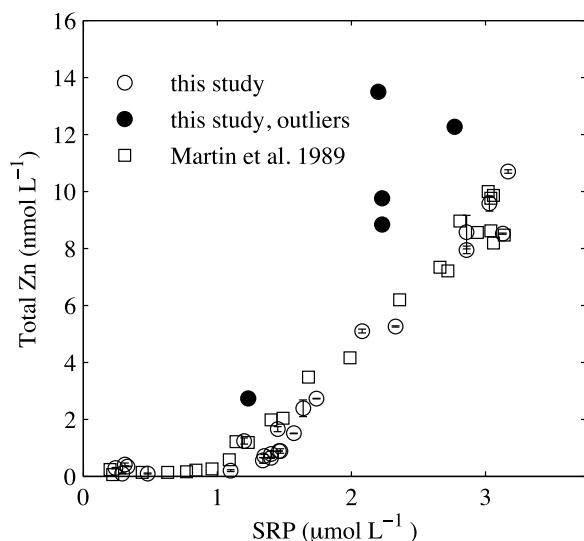
**Figure 2.** Depth profiles down to 1500 m of total dissolved Zn concentrations from (a) stations 1, (b) 4, and (c) 6. Error bars represent the standard deviation of duplicate or triplicate analyses. Dashed line represents the detection limit.

2005]. At Station 8, the mixed layer was only 15 m, and the SRP concentration at 20 m was  $1.56 \mu\text{mol L}^{-1}$  compared with only  $1.25 \mu\text{mol L}^{-1}$  at the surface. Thus, the high  $\text{Zn}_T$  at Station 8 is likely a result of sampling below the mixed layer. The mixed layer at Station 9 is about 18–20 m and the near-surface sample from this station was taken from 20 m. Unfortunately, nutrient data is not available for Station 9 to confirm that the high  $\text{Zn}_T$  value was collected from a depth of active nutrient remineralization. The western Bering Sea can experience high dust inputs from Asian deserts, which may contribute to relatively high Zn concentrations in this area [Boyd *et al.*, 1998; Nishioka *et al.*, 2003]. Lower  $\text{Zn}_T$  was observed in the eastern Bering Sea on the Bering Shelf where *Chla* concentrations were high, possibly reflecting biological uptake.

[20] As mentioned in the Methods section, the possible interference of  $^{64}\text{Ni}$  contributing to our calculated  $\text{Zn}_T$  concentrations was not measured explicitly. The  $\text{Ni}_T$  concentrations in those near-surface samples measured ranged from 3.3 to  $8.2 \text{ nmol L}^{-1}$  (Table 1). While this  $\text{Ni}_T$  technique has somewhat variable precision due to Ni's slow kinetic properties and the small mercury drop surface of the Metrohm 663 hanging drop mercury electrode, the surface and deep water values reported here were generally consistent to that reported by Bruland [1980] for the Central North Pacific. These Ni concentrations could cause an overestimation of our  $\text{Zn}_T$  values that was generally less than 10% of  $\text{Zn}_T$  values; however at the lowest  $\text{Zn}_T$  values the potential for the Ni interference is more significant. For example, in the near-surface at Station 1, the  $^{64}\text{Ni}$  concentration was



**Figure 3.** Comparison of (a) total dissolved Zn concentrations with (b) salinity and (c) temperature profiles in the upper 500 m at Station 4. Dotted line and error bars in Figure 3a as in Figure 2.



**Figure 4a.** Total dissolved Zn concentrations versus soluble reactive phosphorus (SRP) concentrations from all stations. Open squares represent data from VERTEX stations T-5 and T-6, which are in close proximity to Station 1 [Martin *et al.*, 1989]. Both open and closed circles are data from this study. The closed circles represent data where the SRP values are suspected outliers. Zinc error bars as in Figure 2.

30 pmol L<sup>-1</sup>, which could account for approximately 30% of the Z<sub>nT</sub> measured at this depth. The three samples where the <sup>64</sup>Ni could account for over 10% of the Z<sub>nT</sub> concentration are noted in Table 1. The potential contribution of Ni<sub>T</sub> to the deeper Z<sub>nT</sub> values was generally very low (average possible contribution of less than 3%).

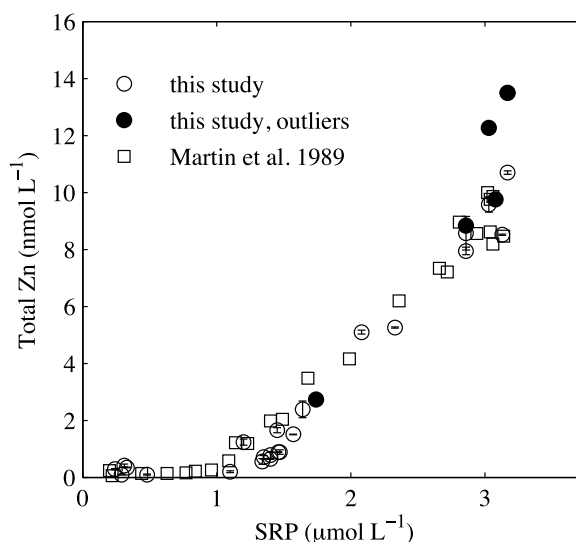
[21] Profiles of 8–11 depths were collected at three stations in the N. Pacific (Figure 2). Zinc profiles exhibited nutrient-like behavior, in agreement with previous studies in the region [Bruland *et al.*, 1978; Bruland, 1980; Martin *et al.*, 1989; Bruland *et al.*, 1994; Lohan *et al.*, 2002]. At the majority of stations, the near-surface samples collected were close to the base of the surface mixed layer (e.g., Station 4, Figure 3). In the N. Pacific, the seasonal thermocline extended down to roughly 40–150 m. In the Bering Sea, the thermocline occurred over shallower depths (25–50 m).

[22] A plot of Z<sub>nT</sub> versus soluble reactive phosphorus (SRP), shows good agreement between this study and Martin *et al.* [1989] from the N. Pacific in the vicinity of Station 1 (Figure 4a), with a few exceptions. At high SRP and Zn, four data points fall above the trend (Station 4, 500 m; Station 4, 3000 m; Station 6, 1000 m; Station 6, 3000 m). They occur at depths in the profile (1000–3000 m) where SRP concentrations are expected to remain relatively constant with depth, yet the SRP in these samples is significantly lower than the two depths surrounding it or the depth above it in the case of the 3000 m points. SRP was measured on board the ship and on frozen samples for this depth. The shipboard value of 1.7 μmol L<sup>-1</sup> SRP is significantly higher than the frozen sample of 1.2 μmol L<sup>-1</sup> and is a value that matches the trend. The anomalously low SRP values suggest that in some cases SRP may have been lost during storage before analysis by ODC, a potential problem of sample storage [Maher and Woo, 1998, and references therein]. If

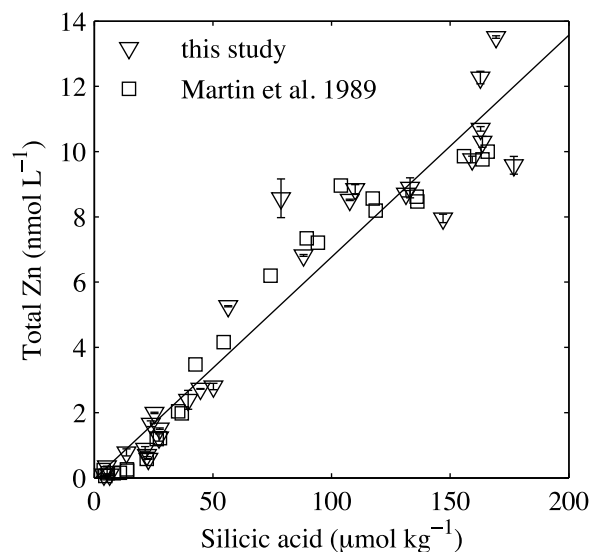
the SRP values for these four data points are replaced with the average value for the two surrounding depths or the value above it in the case of 3000 m, the two shallower samples (Station 4, 500 m; Station 6, 1000 m) fall back in line with the expected trend (Figure 4b). The final two data points (Station 4, 3000 m; Station 6, 3000 m) fall closer to the line but still seem to have higher than expected Z<sub>nT</sub> concentrations. The 3000 m samples were collected with Go-Flo bottles attached to the ship's CTD rosette frame, so they may have been contaminated. A final point that seems to fall somewhat off the trend is that at 1.2 μmol L<sup>-1</sup> SRP and 2.7 nmol L<sup>-1</sup> Z<sub>nT</sub> (Station 6, 80 m). Once again, when the ODC SRP value is replaced with the shipboard SRP value, the data point conforms with the trend.

[23] A tight correlation was observed between silicic acid (Si) and Z<sub>nT</sub> concentrations (Figure 5, R<sup>2</sup> = 0.93). The highest Zn values again fall above the trendline—the likely contamination of these samples has already been noted. The Zn/Si relationship is generally linear, but data from both this study and Martin *et al.* [1989] trend slightly above the linear relationship in the mid-values.

[24] Zinc concentrations reached maximum concentrations in deep waters of 10–10.5 nmol L<sup>-1</sup> (excluding the presumably contaminated 3000 m data) in agreement with Martin *et al.* [1989] who reported similar concentrations at 1500 m for VERTEX stations T-5 thru T-8 and Fujishima *et al.* [2001] who found deep water Z<sub>nT</sub> concentrations across the subarctic N. Pacific to be 10–11 nmol L<sup>-1</sup>. In the central North Pacific, between 20–30°N, Z<sub>nT</sub> concentrations reached maximums of only about 9.5 nmol L<sup>-1</sup> [Bruland *et al.*, 1978; Bruland, 1980; Bruland *et al.*, 1994]. The higher deep Zn values in the subarctic N. Pacific relative to the central N. Pacific are consistent with the increasing time for remineralized Zn to accumulate in deep waters as



**Figure 4b.** Total dissolved Zn concentrations versus soluble reactive phosphorus (SRP) concentrations from all stations. Suspect SRP values (closed circles) have been replaced with average values from surrounding depths or with SRP values measured at sea (see section 3 for more detail). Zinc error bars as in Figure 2.



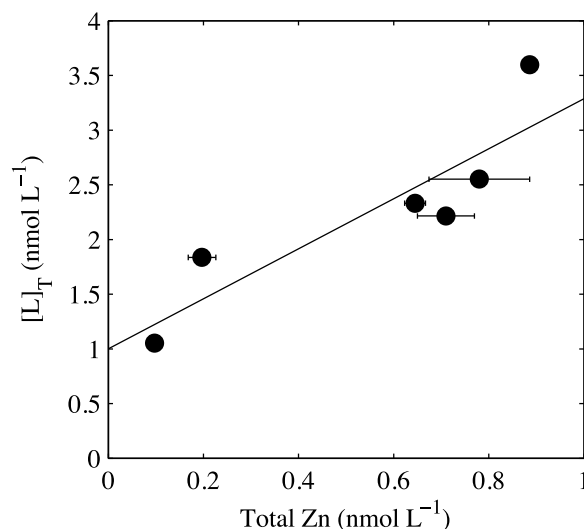
**Figure 5.** Total dissolved Zn concentrations from stations 1, 4, and 6 compared to silicic acid taken from the WOCE archive from stations close to these stations (triangles). Squares are data taken from VERTEX stations T-5 and T-6, which are in close proximity to Station 1 [Martin *et al.*, 1989]. Line is a least squares fit line for the  $Zn_T$  concentrations from this study and the WOCE silicic acid data ( $R^2 = 0.93$ ). Zn error bars as in Figure 2.

they move northward. At Station 4,  $Zn_T$  concentrations from the same profile were also measured using isotope dilution, magnesium hydroxide pre-concentration coupled with an anion exchange resin, and the results from that method showed excellent agreement with this study [John, 2007].

### 3.2. Zn Speciation

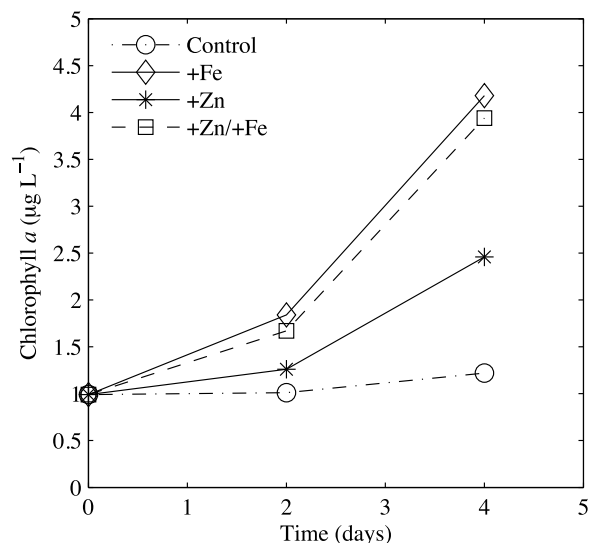
[25] Zn speciation titrations were performed on near-surface samples for stations 1–8. The concentration of Zn binding ligands ( $L_T$ ) ranged from 1.1 to 3.6  $nmol L^{-1}$  in the near-surface with an average concentration of 2.3  $nmol L^{-1}$  (Table 1). The concentration of  $L_T$  followed a general trend of increasing concentrations from east to west in the N. Pacific, similar to  $Zn_T$  concentrations (Figure 6). The highest concentration of  $L_T$  was observed at Station 8 in the western Bering Sea. The  $K'_{cond,Zn}$  of the natural organic ligands ranged from  $10^{9.3}$  to  $10^{11.0}$  with an average value of  $10^{10.2}$  (Table 1). The resultant  $Zn^{2+}$  concentrations ranged from 2–71  $pmol L^{-1}$ , which represented 3% on average of the total Zn concentration. Where multiple depths of a profile were analyzed for speciation, the Zn binding ligands had similar concentrations and conditional stability constants at each depth (Table 1). Free  $Zn^{2+}$  concentrations in the near-surface were very low at the eastern North Pacific stations (2–7  $pmol L^{-1}$ ), similar to previous studies in this area [Bruland, 1989; Donat and Bruland, 1990; Lohan *et al.*, 2005]. The  $Zn^{2+}$  concentrations at the westernmost station in the North Pacific and in the Bering Sea were much higher at 33 and 71  $pmol L^{-1}$ , respectively.

[26] The lowest  $Zn^{2+}$  concentrations were observed in the eastern portion of the subarctic N. Pacific where the majority of previous studies have focused. Ligand concentrations of



**Figure 6.** Concentration of Zn binding ligands ( $L_T$ ) in the near surface shown in relation to the total dissolved Zn concentration. Line is a least squares fit line ( $R^2 = 0.79$ ,  $p < 0.05$ ).

1.0–2.2  $nmol L^{-1}$  that had  $K'_{cond,Zn}$ 's of  $10^{10.6}$ – $10^{11.2}$  were observed in the central and northeast North Pacific by these workers. However, the ligand concentrations were higher and the  $K'_{cond,Zn}$ 's lower here than those reported by Bruland [1989]. The differences cancel out when determining free  $Zn^{2+}$ , hence the agreement.  $K'_{cond,Zn}$  and  $[L_T]$  observed in this study are more similar to those in subarctic waters near New Zealand where the  $[L_T]$  ranged from



**Figure 7.** Results from a bottle incubation experiment examining the influence of Zn and Fe on phytoplankton biomass. The incubation was performed at Station 5 in the North Pacific. Chlorophyll *a* concentrations are shown over the 4 days of the experiment. One bottle was sacrificed at each time point. Additions were: Control (no addition), +Fe (2.5  $nmol L^{-1}$   $FeCl_3$ ), +Zn (0.75  $nmol L^{-1}$   $ZnCl_2$ ), +Zn/+Fe (0.75  $nmol L^{-1}$   $ZnCl_2$ , 2.5  $nmol L^{-1}$   $FeCl_3$ ).



**Table 2.** Time Final (Day 4) Nutrient Concentrations and Nutrient Drawdown Ratios From the Shipboard Incubation Performed at Station 5 in the North Pacific

Treatment	NO <sub>3</sub> ( $\mu\text{mol L}^{-1}$ )	SRP ( $\mu\text{mol L}^{-1}$ )	Si ( $\mu\text{mol L}^{-1}$ )	$\Delta\text{Si}/\Delta\text{NO}_3$	$\Delta\text{Si}/\Delta\text{SRP}$
Time Zero	14.3	1.3	14.5		
Control	13.0	1.2	12.4	1.62	21.0
+Fe	7.4	0.8	9.2	0.77	10.6
+Zn/Fe	7.9	0.9	9.5	0.78	12.5
+Zn	10.6	1.0	11.4	0.84	10.3

1.3–2.5 nmol L<sup>-1</sup> and  $K'_{\text{cond,Zn}}$  was between 10<sup>9.7</sup> and 10<sup>10.4</sup> [Ellwood, 2004].

[27] Culture studies have shown that *E. huxleyi* can exude Zn binding ligands [Vasconcelos et al., 2002]. In this study, though the concentrations of L<sub>T</sub> followed a trend similar to Chla, there was not a significant relationship between Chla and [L<sub>T</sub>] ( $R^2 = 0.51$ ,  $p > 0.1$ ).

[28] The [L<sub>T</sub>] at Station 8 in the Bering Sea (3.6 nmol L<sup>-1</sup>) is the highest reported value of Zn binding ligands in the open ocean measured by voltammetric methods to the authors' knowledge. Ligand concentrations as high as 6 nmol L<sup>-1</sup> have been reported in bottle incubations after the addition of Zn [Lohan et al., 2005], and much higher concentrations (>20 nmol L<sup>-1</sup>) have been observed in coastal waters and phytoplankton cultures [van den Berg, 1985; Vasconcelos et al., 2002]. Station 8 had the highest Zn<sub>T</sub> concentrations of any station where Zn speciation measurements were performed (0.89 nmol L<sup>-1</sup>). The pool of Zn binding ligands is dynamic and can change on short timescales in response to the addition of Zn [Lohan et al., 2005]. The high L<sub>T</sub> concentrations may reflect a phytoplankton community that is well adapted to synthesizing additional ligands in response to atmospheric dust, which can be deposited to the Bering Sea from Asian deserts [Duce and Tindale, 1991].

### 3.3. Shipboard Incubations

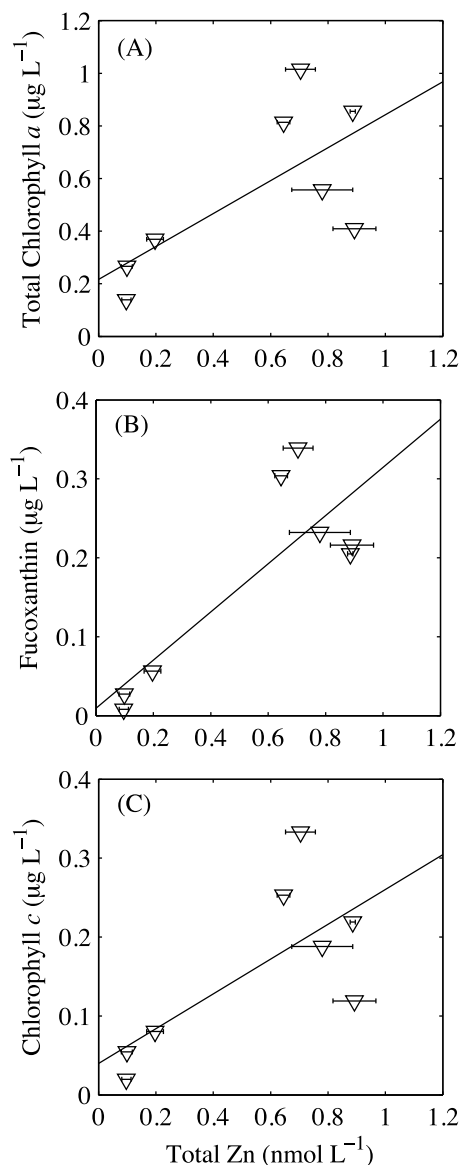
[29] Results from the shipboard incubation that began at Station 5 are presented (Figure 7). At time zero of the incubation, total Chla concentrations were 0.99  $\mu\text{g L}^{-1}$ . The dominant phytoplankton pigments at Station 5 were hexanoyloxyfucoxanthin, fucoxanthin, chlorophyll *c*, and chlorophyll *b*. This pigment signature is consistent with an initial phytoplankton community dominated mainly by

diatoms, prymnesiophytes, and green algae [Mackey et al., 1998]. The near-surface Zn<sub>T</sub> concentration at Station 5 was 0.65 nmol L<sup>-1</sup>. Chlorophyll *a* values remained constant in the control treatment over 4 days (Figure 7). In the +Zn treatment, Chla concentrations increased over the control by 25% on day 2 and reached twice the value in the control on day 4. An even more pronounced effect was seen in the treatments where Fe was added (+Fe, +Zn/+Fe). In these treatments, Chla increased by between 60 and 80% over the control on day 2 and reached over 3 times the Chla values observed in the control on day 4. At the end of the experiment, nutrients were most depleted in the treatments where Fe was added. Nutrient concentrations in the +Zn treatment were intermediate between those of the control and the Fe additions. The drawdown of nutrients in the incubation bottles matched the Chla trend (Table 2). Incubation experiments that included Zn additions were performed at 7 other stations. No increase in response to Zn addition was observed at any other station (Table 3). Previous shipboard incubations in the N. Pacific have shown little effect on total Chla concentration due to the addition of Zn [Coale, 1991; Crawford et al., 2003] or no effect [Lohan et al., 2005; Leblanc et al., 2005]. There is no obvious explanation for the anomalous result at Station 5. Near-surface Zn<sub>T</sub> concentrations at Station 5 were relatively high, and the concentration of Zn<sup>2+</sup> was not particularly low at 7 pmol L<sup>-1</sup>. The pigment distribution at Station 5 was very similar to that at Station 6, where Zn addition had no effect. Fe contamination is a possible explanation for the positive result to Zn addition at this site. However, no growth was observed in the control bottles or in a treatment of 500 pmol L<sup>-1</sup> added Co (M. Saito et al., unpublished data, 2003). Thus it seems unlikely that contamination would have randomly occurred in the two +Zn bottles that

**Table 3.** Summary Table of Incubation Experiments Performed in the North Pacific and Bering Sea During the Summer of 2003<sup>a</sup>

Station	Location	+Zn	+DIP	+N	+Zn/N	+Fe	+Zn/Fe
1	41.60°N, 140.06°W	–	–	+	+		
3	44.02°N, 159.59°W	–	–	–	–		
4	47.01°N, 170.30°W	–	–	–	–		
5	46.59°N, 179.58°W	+				+	+
6	46.60°N, 170.20°E	–				+	+
Station	Location	+Zn	+DIP	+Zn/DIP	+Fe/DIP	+Fe	+Zn/Fe
8	55.18°N, 176.31°E	–				+	+
10	56.43°N, 167.50°W	–	–	–	–	–	–
11	56.45°N, 164.60°W	–	–	–	–	–	–

<sup>a</sup>Stations 1–6 are the subarctic North Pacific; stations 8–10 are the Bering Sea. An + indicates that a treatment resulted in an increase in chlorophyll *a* above a no-addition control, whereas a – indicates no increase observed. Where spaces are empty, the treatment was not performed at that station. +Fe additions were performed by Saito et al. (unpublished data, 2003) at stations 1, 3 and 4. A chlorophyll *a* increase due to Fe addition was observed at stations 3 and 4 but not at station 1.



**Figure 8.** Comparison of total Zn concentrations and several phytoplankton pigments (as determined by HPLC). (a) The apparent positive relationship between total chlorophyll *a* and Zn is not significant ( $R^2 = 0.49$ ,  $p > 0.05$ ); however, there is a significant positive correlation between Zn and the relative contribution of two pigments found in diatoms: (b) fucoxanthin ( $R^2 = 0.70$ ,  $p < 0.01$ ) and (c) chlorophyll *c* ( $R^2 = 0.50$ ,  $p < 0.05$ ). Error bars as in Figure 2. Lines are least squares fit lines.

were sacrificed at successive time points and not in any of the control or +Co bottles.

### 3.4. Chlorophyll, Nutrients, and HPLC Pigments

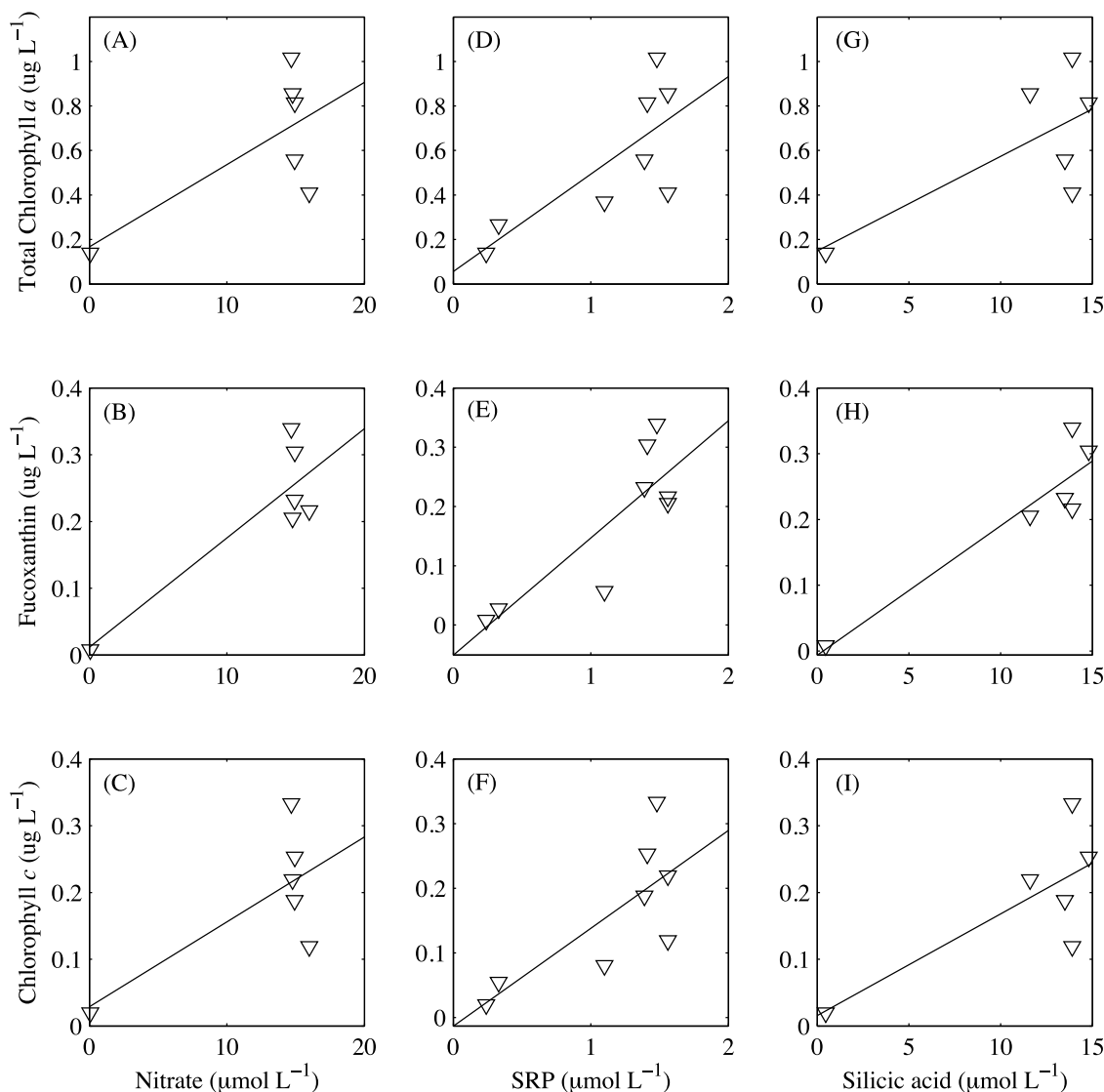
[30] In the N. Pacific, near-surface SRP was lowest in the east and increased significantly between Station 1 and Station 3 and then remained relatively constant (Table 1). In the Bering Sea, SRP was highest in the west and decreased significantly on the Bering shelf. There were twelve samples where SRP was measured both by the Popp group at sea and from frozen samples by the ODC. When the SRP values

from both analysts are plotted against one another, the vast majority of the samples fall within 10% of the 1:1 line (data not shown). Two exceptions are the samples from Station 4, 60 m and Station 6, 80 m. In both cases, the values measured by the ODC ( $1.20 \mu\text{mol L}^{-1}$ ,  $1.23 \mu\text{mol L}^{-1}$ ) were significantly lower than those measured at sea ( $1.63 \mu\text{mol L}^{-1}$ ,  $1.74 \mu\text{mol L}^{-1}$ ). The ODC SRP values for these two samples were also lower than would be expected based on the SRP concentrations from the shallower depths of the same profile. For both these samples, the values measured at sea are used in Table 1 and in Figures 4 and 9.

[31] Nitrate plus nitrite values in the near-surface were also very low at Station 1 ( $<0.1 \mu\text{mol L}^{-1}$ ) and much higher and more consistent over the western half of the N. Pacific transect ( $\sim 15 \mu\text{mol L}^{-1}$ ). Silicic acid followed the same trend (Station 1:  $0.5 \mu\text{mol L}^{-1}$ ; Sts. 4–6:  $14 \mu\text{mol L}^{-1}$ ).

[32] Total Chl *a* concentrations in the near-surface were lowest in the eastern N. Pacific (Table 1). Chlorophyll *a* increased moving west from Station 1 to Station 5. Moderate Chl *a* values were observed for stations 6 through 10 in the western N. Pacific and Bering Sea, and the highest Chl *a* concentration encountered was at Station 11 on the Bering Shelf. The most abundant phytoplankton marker pigments (i.e., non-Chl *a*) were hexanoyloxyfucoxanthin, fucoxanthin, chlorophyll *c*, and chlorophyll *b*. The pigment signatures agree with previous examinations of phytoplankton communities containing prymnesiophytes, diatoms, pelagophytes, and green algae. In the northeast subarctic Pacific at Ocean Station Papa ( $50^\circ\text{N}$ ,  $145^\circ\text{W}$ ), the phytoplankton community is typically dominated by small prymnesiophytes (such as *E. huxleyi*), pelagophytes, small diatoms, and cyanobacteria, with larger diatoms and other large phytoplankton contributing 10–30% of the total Chl *a* [e.g., Boyd and Harrison, 1999; Thibault et al., 1999]. The waters of the northwest subarctic Pacific often host phytoplankton communities with abundant prasinophytes and a higher relative abundance of diatoms than in the eastern subarctic North Pacific [e.g., Suzuki et al., 2002; Obayashi et al., 2001]. Hexanoyloxyfucoxanthin was the most abundant marker pigment at Sts. 1, 2, 3, and 10, while fucoxanthin was dominant at Sts. 4, 5, 6, 8, and 11. This pattern meshes with the above studies and indicates that at the eastern North Pacific stations the community was likely dominated by prymnesiophytes, while at the western North Pacific stations, diatoms were more likely to dominate. The higher concentrations of fucoxanthin at the western North Pacific stations is consistent with the proliferation of diatoms in waters containing relatively high levels of macro- and micronutrients.

[33] Phytoplankton pigment concentrations were compared to Zn concentrations from those samples in the upper thermocline where Zn speciation data were available. This criterion excludes data from St 6, 40 m and St 4, 100 m, where significant mixing with deep waters is evidenced by the more than doubling of the total Zn concentrations relative to mixed layer values. There was not a significant relationship ( $p > 0.05$ ) between  $\text{Zn}_T$  concentration and total Chl *a* (Figure 8a). However, for two major pigments found in diatoms, fucoxanthin ( $p < 0.01$ ) and chlorophyll *c* ( $p < 0.05$ ), positive relationships were observed with  $\text{Zn}_T$  (Figures 8b and 8c), which may indicate the relative importance of Zn



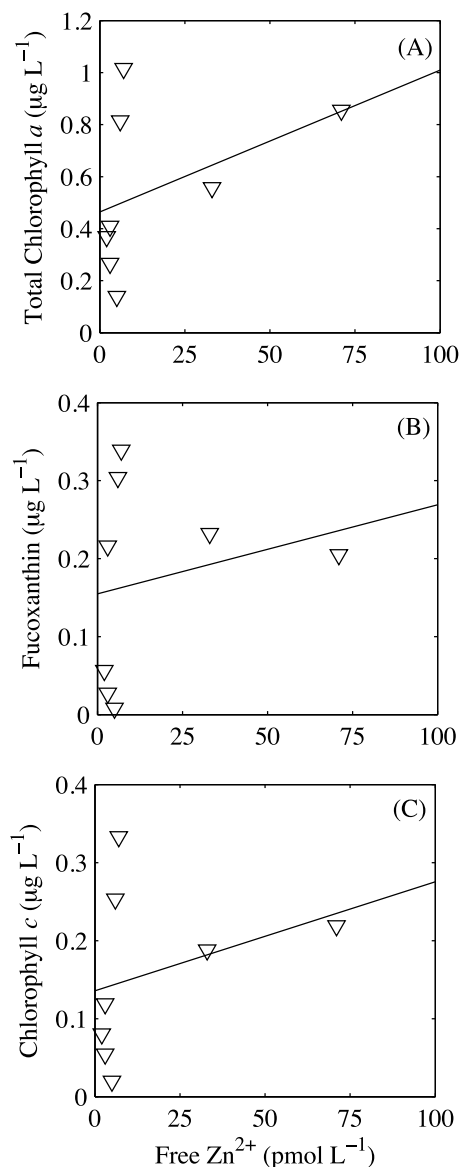
**Figure 9.** Comparison of total  $\text{NO}_3$ , SRP, and silicic acid concentrations with several phytoplankton pigments (as determined by HPLC). There are not significant correlations between the concentration of  $\text{NO}_3$  and the concentration of (a) total chlorophyll  $a$  ( $R^2 = 0.49$ ,  $p > 0.1$ ) or (c) chlorophyll  $c$  ( $R^2 = 0.51$ ,  $p > 0.1$ ). There was a significant correlation between the concentration of  $\text{NO}_3$  and the concentration of (b) fucoxanthin ( $R^2 = 0.76$ ,  $p < 0.05$ ). There are significant correlations between the concentration of SRP and the concentration of (d) total chlorophyll  $a$  ( $R^2 = 0.58$ ,  $p < 0.05$ ), (e) fucoxanthin ( $R^2 = 0.72$ ,  $p < 0.01$ ), and (f) chlorophyll  $c$  ( $R^2 = 0.58$ ,  $p < 0.05$ ). Similar to  $\text{NO}_3$ , there are not significant correlations between the concentration of silicic acid and the concentration of (g) total chlorophyll  $a$  ( $R^2 = 0.51$ ,  $p > 0.1$ ) or (i) chlorophyll  $c$  ( $R^2 = 0.58$ ,  $p > 0.05$ ). There was a strong correlation between the concentration of silicic acid and the concentration of (h) fucoxanthin ( $R^2 = 0.86$ ,  $p < 0.01$ ). Lines are least squares fit lines.

to this fraction of the phytoplankton community. Diatoms have high Zn quotas relative to other taxa [Sunda and Huntsman, 1995]. The relationship observed here is consistent with Zn being an essential micronutrient for diatoms.

[34] Significant correlations ( $p < 0.01$ ) were also observed between both SRP and silicic acid and fucoxanthin (Figure 9) but nitrate was not as strongly correlated. When pigment concentrations were compared to the free  $\text{Zn}^{2+}$  concentration rather than total Zn concentration, no significant correlations were observed (Figure 10). Instead, aside from two high points, most of the data were below

$10 \text{ pmol L}^{-1}$ , regardless of pigment concentration, suggesting that at very low concentrations, free  $\text{Zn}^{2+}$  is relatively invariant with biology within the capabilities of this methodology.

[35] Two minor pigments, alloxanthin and chlorophyllide  $a$ , also exhibited positive correlations with  $\text{Zn}_T$  (data not shown). Alloxanthin is a marker for cryptomonads and was found in very low quantities (average relative contribution to total Chla  $\sim 1\%$ ). Chlorophyllide  $a$  is a degradation product of Chla and was observed in somewhat higher concentrations (average relative contribution to Chla  $< 9\%$ ). Degradation of



**Figure 10.** Comparison of free  $\text{Zn}^{2+}$  concentrations and several phytoplankton pigments (as determined by HPLC). There are not significant correlations between (a) the concentration of  $\text{Zn}^{2+}$  and the concentration of total chlorophyll *a* ( $R^2 = 0.05$ ,  $p > 0.1$ ) and the relative contribution of two pigments found in diatoms: (b) fucoxanthin ( $R^2 = 0.05$ ,  $p > 0.1$ ) and (c) chlorophyll *c* ( $R^2 = 0.10$ ,  $p > 0.1$ ). Lines are least squares fit line.

Chl*a* to chlorophyllide *a* can occur during copepod grazing and during the filtration of some diatoms [Klein and Sournia, 1987, and references therein]. These relationships are based on limited data, and interpreting pigment data can be difficult because organisms contain multiple pigments and the pigment composition of field organisms does not accurately match those of cultured organisms.

#### 4. Discussion

[36] A prominent feature of the data presented here is the strong gradient in zinc concentration and zinc speciation in

surface waters on the east–west transect. Increasing zinc concentrations toward the west were matched by increasing ligand concentrations, leading to relatively little variability in free  $\text{Zn}^{2+}$  in the mixed layer until the westernmost station (Station 6).  $\text{Zn}_T$  at Station 6 is higher in surface waters, and free  $\text{Zn}^{2+}$  increases to values seen only in the subsurface further east. Together, these results show an emerging picture of a large region in the central and eastern N Pacific where mixed layer free  $\text{Zn}^{2+}$  is less than  $10 \text{ pmol L}^{-1}$ . This region is bounded by waters underlying the mixed layer, where  $\text{Zn}_T$  and free  $\text{Zn}^{2+}$  are much higher, and by nutrient-rich surface waters in the west. Increasing Zn levels toward the west are associated with an increase in biomass and diatoms, as indicated by Chl*a* and fucoxanthin data. These increasing zinc abundances may be one of the factors, with the increased macronutrients, that contributes to this trend toward higher primary productivity in the west. Establishing a causal relationship between Zn and biological activity is premature, since Zn additions to bottle incubations resulted in a positive response at only one station.

[37] Free  $\text{Zn}^{2+}$  is presumed to be the biologically available form [Sunda and Huntsman, 1992, 1995] so the observation that total dissolved Zn has a stronger correlation with pigment parameters is surprising. However, for acquisition of some micronutrients like Fe and Co, some phytoplankton appear to be capable of accessing the organically complexed forms [Maldonado and Price, 1999; Saito et al., 2002], hence free ion concentrations may be less relevant than total concentrations for some species because of these high affinity uptake systems that render organically complexed forms biologically available. Correlations here also reflect the fact that  $\text{Zn}_T$  can be measured much more precisely than the scarcer free  $\text{Zn}^{2+}$ , making it difficult to look for correlations between the latter and other parameters.

[38] In spite of these limitations, our free  $\text{Zn}^{2+}$  data enable us to compare field data with culture studies, which are typically carried out using metal ion buffers like EDTA, where effects (e.g., specific growth rates, cellular Zn quota) are reported as a function of free  $\text{Zn}^{2+}$ . Comparison of our free  $\text{Zn}^{2+}$  data with phytoplankton culture studies suggest that Zn is a limiting micronutrient for some phytoplankton, in particular larger diatoms [Sunda and Huntsman, 1995], and is low enough to induce biochemical responses such as substitution of Co for Zn [Yee and Morel, 1996; Xu et al., 2007; Saito et al., 2008].

[39] Sunda and Huntsman [1992, 1995] demonstrated a robust relationship between free  $\text{Zn}^{2+}$  and Zn:C in phytoplankton cultures, building on earlier studies [Sunda and Guillard, 1976; Anderson et al., 1978]. Using a Redfieldian-type analysis, they estimated Zn:P ratios in phytoplankton biomass through analysis of the dissolved Zn:P data in the upper 1000 m of the North Pacific reported in Martin et al. [1989]. While Zn shows the strongest correlation with Si over the entire water column, the strong correlation with P in the upper water column indicates a close coupling of Zn and nutrient remineralization. Therefore, Sunda and Huntsman [1992] argue that these dissolved ratios presumably reflect the Zn:P of remineralized phytoplankton biomass, and using their data for Zn:C in culture, they estimated the corresponding free  $\text{Zn}^{2+}$  concentration required to produce a sufficiently large Zn:P ratio. Their calculations suggest that the Zn:P ratio from the nutricline

could only arise from diatoms growing with a free zinc abundance of  $\sim 1000$  pmol L<sup>-1</sup>. The estimated “nutriline value” for free Zn<sup>2+</sup> is an order of magnitude higher than the highest value reported here and in Lohan *et al.* [2002]. The only source of nutrient-rich, high Zn water to lead to such conditions is substantially deeper than 100 m, yet the mixed layer does not penetrate that depth in this region, even during the winter [Anderson *et al.*, 1969]. Therefore, the Zn must come from elsewhere. One possibility is that episodic deep mixing events, perhaps within boundary regions, lead to conditions with elevated Zn. A major shortcoming of all Zn studies, including ours, is that they do not examine the dynamics of Zn injection and drawdown during bloom periods. For instance, it is possible that deep mixing events preceded the SEEDS experiment in the western North Pacific, when surface Zn was extremely high [Kinugasa *et al.*, 2005].

[40] The Zn:P ratio calculated from data between 50 to 150 m is much smaller, and yields a smaller cell quota and free Zn<sup>2+</sup> abundance of 0.8 to 2 pmol L<sup>-1</sup>. The free Zn<sup>2+</sup> abundances estimated from the shallow dissolved Zn:P ratios are quite similar to that measured in this study in the low picomolar range (Table 1), suggesting that much less bioavailable Zn is in the photic zone and which also is within the range of what would affect phytoplankton species composition [Sunda and Huntsman, 1995].

[41] In summary, data from a variety of sources, including culture work and measurements of remineralized Zn:P inferred from dissolved Zn and phosphate abundances, provide a mechanism for Zn cycling that is consistent with the free ion model. A principal, if accidental role of organic ligands is to act as a natural Zn buffer. Complexation decreases the rate of “luxury” uptake or non-biological scavenging under conditions of high particle scavenging, leaving a residual concentration available for organisms with high affinity transport systems. From Sunda and Huntsman’s work, this would appear to include small diatoms, but not larger ones. Zn limitation will probably induce community shifts to species or biochemistries with lower Zn requirements before total Zn is completely removed. Thus organic complexation provides a useful natural service to organisms with a small, but nonzero Zn requirement.

[42] The inter-replacement of Co and Zn as micronutrients has been shown in phytoplankton cultures [Saito *et al.*, 2008, and references therein]. Therefore, Co distributions should also be considered in evaluating the biological implications of our data, particularly for station 5, the only location where we had evidence for Zn limitation. A strong depletion in near-surface Co concentrations was observed at Station 5, with some of the lowest Co values observed on this transect ( $\sim 30$  pmol L<sup>-1</sup>; M. Saito, manuscript in preparation, 2012). The low concentration of Co is consistent with the biochemical substitution of Co for Zn under low Zn conditions and with the observation that Co draw down in the North Pacific typically occurs when Zn concentrations are depleted relative to SRP concentrations [Sunda and Huntsman, 1995].

[43] Much of the region we surveyed exhibits Fe limitation to varying degrees [Martin and Fitzwater, 1988; Kinugasa *et al.*, 2005]. The relationship between Fe and Zn limitation has been studied previously in cultures and deck-board incubations. In cultures of the diatom *T. weissflogii*,

Fe and Zn deficiency resulted in cells with higher Si contents, and Zn-stressed cells had a significantly higher Si:NO<sub>3</sub> than Zn replete diatoms [De La Rocha *et al.*, 2000]. Franck *et al.* [2003] performed a series of incubation experiments in Fe-limited upwelling regions and observed a decrease in the Si:NO<sub>3</sub> utilization ratio when Fe or Fe and Zn together were added, due to a greater stimulation of NO<sub>3</sub> uptake versus Si uptake. In contrast to the Fe addition results, the Si:NO<sub>3</sub> utilization ratio increased when Zn was added as a result of either a decline in NO<sub>3</sub> uptake (Costa Rica and Point Conception, CA stations) or enhanced Si uptake (Big Sur, CA) [Franck *et al.*, 2003]. In this study, a similar decrease in the Si:NO<sub>3</sub> utilization ratio was observed in all the metal treatments relative to the control (Table 2), which is consistent with previous Fe addition experiments in the subarctic North Pacific [Takeda, 1998]. The mechanism for the alteration of the nutrient ratios was not explored; however, our results support previous observations that both Zn and Fe limitation may influence a phytoplankton cell’s ability to compete for Si [De La Rocha *et al.*, 2000; Franck *et al.*, 2003].

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## References

- Aguilar-Islas, A. M., M. P. Hurst, K. N. Buck, B. Sohst, G. J. Smith, M. C. Lohan, and K. W. Bruland (2007), Micro- and macronutrients in the south-eastern Bering Sea: Insight into iron-replete and iron-depleted regimes, *Prog. Oceanogr.*, *73*, 99–126, doi:10.1016/j.pcean.2006.12.002.
- Anderson, G. C., T. R. Parsons, and K. Stephens (1969), Nitrate distribution in the subarctic Northeast Pacific Ocean, *Deep Sea Res. Oceanogr. Abstr.*, *16*, 329–334.
- Anderson, M. A., F. M. M. Morel, and R. R. L. Guillard (1978), Growth limitation of a coastal diatom by low zinc ion activity, *Nature*, *276*, 70–71, doi:10.1038/276070a0.
- Bidigare, R. R. (1991), Analysis of algal chlorophylls and carotenoids, in *Marine particles: Analysis and Characterization*, *Geophys. Monogr. Ser.*, vol. 63, edited by D. C. Hurd and D. W. Spencer, pp. 119–123, AGU, Washington, D. C., doi:10.1029/GM063p0119.
- Bidigare, R. R., and C. C. Trees (2000), HPLC phytoplankton pigments: Sampling, laboratory methods, and quality assurance procedures, in *Ocean Optics Protocols For Satellite Ocean Color Sensor Validation, Revision 2*, *NASA Tech. Memo. 2000–209966*, edited by J. Mueller and G. Fargion, pp. 154–161, NASA Goddard Space Flight Cent., Greenbelt, Md.
- Boyd, P., and P. J. Harrison (1999), Phytoplankton dynamics in the NE subarctic Pacific, *Deep Sea Res., Part II*, *46*, 2405–2432, doi:10.1016/S0967-0645(99)00069-7.
- Boyd, P. W., D. L. Muggli, D. E. Varela, R. H. Goldblatt, R. Chretien, K. J. Orians, and P. J. Harrison (1996), In vitro iron enrichment experiments in the NE subarctic Pacific, *Mar. Ecol. Prog. Ser.*, *136*, 179–193, doi:10.3354/meps136179.
- Boyd, P. W., C. S. Wong, J. Merrill, F. Whitney, J. Snow, P. J. Harrison, and J. Gower (1998), Atmospheric iron supply and enhanced vertical carbon flux in the NE subarctic Pacific: Is there a connection?, *Global Biogeochem. Cycles*, *12*, 429–441, doi:10.1029/98GB00745.
- Boyd, P. W., et al. (2004), The decline and fate of an iron-induced subarctic phytoplankton bloom, *Nature*, *428*, 549–553, doi:10.1038/nature02437.
- Brand, L. E., W. G. Sunda, and R. R. L. Guillard (1983), Limitation of marine phytoplankton reproductive rates by Zn, Mn, and Fe, *Limnol. Oceanogr.*, *28*, 1182–1198, doi:10.4319/lo.1983.28.6.1182.
- Bruland, K. W. (1980), Oceanographic distributions of cadmium, zinc, nickel, and copper in the North Pacific, *Earth Planet. Sci. Lett.*, *47*, 176–198, doi:10.1016/0012-821X(80)90035-7.

- Bruland, K. W. (1989), Complexation of zinc by natural organic ligands in the central North Pacific, *Limnol. Oceanogr.*, *34*, 269–285, doi:10.4319/lo.1989.34.2.0269.
- Bruland, K. W., G. A. Knauer, and J. H. Martin (1978), Zinc in north-east Pacific water, *Nature*, *271*, 741–743, doi:10.1038/271741a0.
- Bruland, K. W., K. J. Orians, and J. P. Cowen (1994), Reactive trace metals in the stratified central North Pacific, *Geochim. Cosmochim. Acta*, *58*, 3171–3182, doi:10.1016/0016-7037(94)90044-2.
- Chu, P. C., and Y. Kuo (2010), Nutrient pumping/advection by propagating Rossby Waves in the Kuroshio Extension, *Deep Sea Res., Part II*, *57*, 1809–1819, doi:10.1016/j.dsr2.2010.04.007.
- Coale, K. H. (1991), Effects of iron, manganese, copper, and zinc enrichments on productivity and biomass in the subarctic Pacific, *Limnol. Oceanogr.*, *36*, 1851–1864, doi:10.4319/lo.1991.36.8.1851.
- Coale, K. H., et al. (1996), A massive phytoplankton bloom induced by an ecosystem-scale iron fertilization experiment in the equatorial Pacific Ocean, *Nature*, *383*, 495–501, doi:10.1038/383495a0.
- Coale, K. H., X. J. Wang, S. J. Tanner, and K. S. Johnson (2003), Phytoplankton growth and biological response to iron and zinc addition in the Ross Sea and Antarctic Circumpolar Current along 170 degrees W, *Deep Sea Res., Part II*, *50*, 635–653, doi:10.1016/S0967-0645(02)00588-X.
- Crawford, D. W., et al. (2003), Influence of zinc and iron enrichments on phytoplankton growth in the Northeastern subarctic Pacific, *Limnol. Oceanogr.*, *48*, 1583–1600, doi:10.4319/lo.2003.48.4.1583.
- Cummins, P. F., and H. J. Freeland (2007), Variability of the North Pacific Current and its bifurcation, *Prog. Oceanogr.*, *75*, 253–265, doi:10.1016/j.pocan.2007.08.006.
- De La Rocha, C. L., D. A. Hutchins, M. A. Brzezinski, and Y. Zhang (2000), Effects of iron and zinc deficiency on elemental composition and silica production by diatoms, *Mar. Ecol. Prog. Ser.*, *195*, 71–79, doi:10.3354/meps195071.
- Donat, J. R., and K. W. Bruland (1990), A comparison of two voltammetric techniques for determining zinc speciation in Northeast Pacific Ocean waters, *Mar. Chem.*, *28*, 301–323, doi:10.1016/0304-4203(90)90050-M.
- Duce, R. A., and N. W. Tindale (1991), Atmospheric transport of iron and its deposition in the ocean, *Limnol. Oceanogr.*, *36*(8), 1715–1726, doi:10.4319/lo.1991.36.8.1715.
- Ellwood, M. J. (2004), Zinc and cadmium speciation in subantarctic waters east of New Zealand, *Mar. Chem.*, *87*, 37–58, doi:10.1016/j.marchem.2004.01.005.
- Ellwood, M. J., and C. M. G. van den Berg (2000), Zinc speciation in the Northeastern Atlantic Ocean, *Mar. Chem.*, *68*, 295–306, doi:10.1016/S0304-4203(99)00085-7.
- Fischer, E., and C. M. G. van den Berg (1999), Anodic stripping voltammetry of lead and cadmium using a mercury film electrode and thiocyanate, *Anal. Chim. Acta*, *385*, 273–280, doi:10.1016/S0003-2670(98)00582-0.
- Franck, V. M., K. W. Bruland, D. A. Hutchins, and M. A. Brzezinski (2003), Iron and zinc effects on silicic acid and nitrate uptake kinetics in three high-nutrient, low-chlorophyll (HNLC) regions, *Mar. Ecol. Prog. Ser.*, *252*, 15–33, doi:10.3354/meps252015.
- Fujishima, Y., K. Ueda, M. Maruo, E. Nakayama, C. Tokutome, H. Hasegawa, M. Matsui, and Y. Sohrin (2001), Distribution of trace bioelements in the subarctic North Pacific Ocean and the Bering Sea (the R/V Hakuho Maru cruise KH-97-2), *J. Oceanogr.*, *57*, 261–273, doi:10.1023/A:1012426411228.
- Fukuda, R., Y. Sohrin, N. Saotome, H. Fukuda, T. Nagata, and I. Koike (2000), East–west gradient in ectoenzyme activities in the subarctic Pacific: Possible regulation by zinc, *Limnol. Oceanogr.*, *45*, 930–939, doi:10.4319/lo.2000.45.4.0930.
- Gall, M. P., R. Strzpek, M. Maldonado, and P. W. Boyd (2001), Phytoplankton processes. Part 2: Rates of primary production and factors controlling algal growth during the Southern Ocean Iron Release Experiment (SOIREE), *Deep Sea Res., Part II*, *48*, 2571–2590, doi:10.1016/S0967-0645(01)00009-1.
- Heumann, K. G. (1988), Isotope dilution mass spectrometry, in *Inorganic Mass Spectrometry*, *Chem. Anal. Ser.*, vol. 95, edited by F. Adams, R. Gijbels, and R. Van Grieken, pp. 301–375, John Wiley, New York.
- Honjo, S. (1997), The Northwestern Pacific Ocean, a crucial ocean region to understand global change: Rationale for new international collaborative investigations, in *Biogeochemical Processes in the North Pacific*, edited by S. Tsunogai, pp. 233–248, Jpn. Mar. Sci. Found., Tokyo.
- Jakuba, R. W., J. W. Moffett, and M. A. Saito (2008), Use of a modified, high-sensitivity, anodic stripping voltammetry method for determination of zinc speciation in the North Atlantic Ocean, *Anal. Chim. Acta*, *614*(2), 143–152, doi:10.1016/j.aca.2008.03.006.
- John, S. G. (2007), The marine biogeochemistry of zinc isotopes, PhD thesis, MIT-WHOI Joint Prog., Woods Hole, Mass.
- Kinugasa, M., T. Ishita, Y. Sohrin, K. Okamura, S. Takeda, J. Nishioka, and A. Tsuda (2005), Dynamics of trace metals during the subarctic Pacific iron experiment for ecosystem dynamics study (SEEDS2001), *Prog. Oceanogr.*, *64*, 129–147, doi:10.1016/j.pocan.2005.02.005.
- Klein, B., and A. Sourmia (1987), A daily study of the diatom spring bloom at Roscoff (France) in 1985. II. Phytoplankton pigment composition studied by HPLC analysis, *Mar. Ecol. Prog. Ser.*, *37*, 265–275, doi:10.3354/meps037265.
- Lam, P. J., P. D. Tortell, and F. M. M. Morel (2001), Differential effects of iron additions on organic and inorganic carbon production by phytoplankton, *Limnol. Oceanogr.*, *46*, 1199–1202, doi:10.4319/lo.2001.46.5.1199.
- LaRoche, J., P. W. Boyd, R. M. L. McKay, and R. J. Geider (1996), Flavodoxin as a *in situ* marker for iron stress in phytoplankton, *Nature*, *382*, 802–805, doi:10.1038/382802a0.
- Leblanc, K., C. E. Hare, P. W. Boyd, K. W. Bruland, B. Sohst, S. Pickmere, M. C. Lohan, K. Buck, M. Ellwood, and D. A. Hutchins (2005), Fe and Zn effects on the Si cycle and diatom community structure in two contrasting high and low-silicate HNLC areas, *Deep Sea Res., Part I*, *52*, 1842–1864, doi:10.1016/j.dsr.2005.06.005.
- Lee, J. G., S. B. Roberts, and F. M. M. Morel (1995), Cadmium: A nutrient for the marine diatom, *Thalassiosira weissflogii*, *Limnol. Oceanogr.*, *40*, 1056–1063, doi:10.4319/lo.1995.40.6.1056.
- Lohan, M. C., P. J. Statham, and D. W. Crawford (2002), Total dissolved zinc in the upper water column of the subarctic North East Pacific, *Deep Sea Res., Part II*, *49*, 5793–5808, doi:10.1016/S0967-0645(02)00215-1.
- Lohan, M. C., D. W. Crawford, D. A. Purdie, and P. J. Statham (2005), Iron and zinc enrichments in the northeastern subarctic Pacific: Ligand production and zinc availability in response to phytoplankton growth, *Limnol. Oceanogr.*, *50*, 1427–1437, doi:10.4319/lo.2005.50.5.1427.
- Mackey, D. J., H. W. Higgins, M. D. Mackey, and D. Holdsworth (1998), Algal class abundances in the western equatorial Pacific: Estimation from HPLC measurements of chloroplast pigments using CHEMTAX, *Deep Sea Res., Part I*, *45*, 1441–1468, doi:10.1016/S0967-0637(98)00025-9.
- Maher, W., and L. Woo (1998), Procedures for the storage and digestion of natural waters for the determination of filterable reactive phosphorus, total filterable phosphorus and total phosphorus, *Anal. Chim. Acta*, *375*, 5–47, doi:10.1016/S0003-2670(98)00274-8.
- Maldonado, M. T., and N. M. Price (1999), Utilization of iron bound to strong organic ligands by plankton communities in the subarctic Pacific Ocean, *Deep Sea Res., Part II*, *46*, 2447–2473, doi:10.1016/S0967-0645(99)00071-5.
- Marchetti, A., N. D. Sherry, H. Kiyosawa, A. Tsuda, and P. J. Harrison (2006), Phytoplankton processes during a mesoscale iron enrichment in the NE subarctic Pacific: Part I—Biomass and assemblage, *Deep Sea Res., Part II*, *53*, 2095–2113, doi:10.1016/j.dsr2.2006.05.038.
- Martin, J. H., and S. E. Fitzwater (1988), Iron deficiency limits phytoplankton growth in the north-east Pacific subarctic, *Nature*, *331*, 341–343, doi:10.1038/331341a0.
- Martin, J. H., R. M. Gordon, S. Fitzwater, and W. W. Broenkow (1989), VERTEX: Phytoplankton/iron studies in the Gulf of Alaska, *Deep Sea Res.*, *36*, 649–680, doi:10.1016/0198-0149(89)90144-1.
- McAllister, C. D., T. R. Parsons, and J. D. H. Strickland (1960), Primary productivity and fertility at station “P” in the north-east Pacific Ocean, *J. Cons., Cons. Int. Explor. Mer.*, *25*, 240–259, doi:10.1093/icesjms/25.3.240.
- Morel, F. M. M., R. J. M. Hudson, and N. M. Price (1991), Limitation of productivity by trace-metals in the sea, *Limnol. Oceanogr.*, *36*, 1742–1755, doi:10.4319/lo.1991.36.8.1742.
- Murphy, J., and J. P. Riley (1962), A modified single solution method for the determination of phosphate in natural waters, *Anal. Chim. Acta*, *27*, 31–36, doi:10.1016/S0003-2670(00)88444-5.
- Nishioka, J., S. Takeda, I. Kudo, D. Tsumune, T. Yoshimura, K. Kuma, and A. Tsuda (2003), Size-fractionated iron distributions and iron-limiting processes in the subarctic NW Pacific, *Geophys. Res. Lett.*, *30*(14), 1730, doi:10.1029/2002GL016853.
- Obayashi, Y., E. Tanoue, K. Suzuki, N. Handa, Y. Nojiri, and C. S. Wong (2001), Spatial and temporal variabilities of phytoplankton community structure in the northern North Pacific as determined by phytoplankton pigments, *Deep Sea Res., Part I*, *48*, 439–469, doi:10.1016/S0967-0637(00)00036-4.
- Peers, G., and N. M. Price (2006), Copper-containing plastocyanin used for electron transport by an oceanic diatom, *Nature*, *441*(7091), 341–344, doi:10.1038/nature04630.
- Price, N. M., G. I. Harrison, J. G. Hering, R. J. Hudson, P. M. V. Nirel, B. Palenik, and F. M. M. Morel (1989), Preparation and chemistry of the artificial algal culture medium Aquil, *Biol. Oceanogr.*, *6*, 443–461.
- Ružić, I. (1982), Theoretical aspects of the direct titration of natural waters and its information yield for trace metal speciation, *Anal. Chim. Acta*, *140*, 99–113, doi:10.1016/S0003-2670(01)95456-X.

- Saito, M. A., and T. J. Goepfert (2008), Zinc-cobalt colimitation of *Phaeocystis antarctica*, *Limnol. Oceanogr.*, 53(1), 266–275, doi:10.4319/lo.2008.53.1.0266.
- Saito, M. A., and D. L. Schneider (2006), Examination of precipitation chemistry and improvements in precision using the Mg(OH)<sub>2</sub> preconcentration ICP-MS method for high-throughput analysis of open ocean Fe and Mn in seawater, *Anal. Chim. Acta*, 565, 222–233, doi:10.1016/j.aca.2006.02.028.
- Saito, M. A., J. W. Moffett, S. W. Chisholm, and J. B. Waterbury (2002), Cobalt limitation and uptake in *Prochlorococcus*, *Limnol. Oceanogr.*, 47, 1629–1636, doi:10.4319/lo.2002.47.6.1629.
- Saito, M. A., G. Rocab, and J. W. Moffett (2005), Production of cobalt binding ligands in a *Synechococcus* feature at the Costa Rica upwelling dome, *Limnol. Oceanogr.*, 50, 279–290, doi:10.4319/lo.2005.50.1.0279.
- Saito, M. A., T. J. Goepfert, and J. T. Ritt (2008), Some thoughts on the concept of colimitation: Three definitions and the importance of bioavailability, *Limnol. Oceanogr.*, 53(1), 276–290, doi:10.4319/lo.2008.53.1.0276.
- Scharek, R., M. A. Van Leeuwe, and H. J. W. Debaar (1997), Responses of Southern Ocean phytoplankton to the addition of trace metals, *Deep Sea Res., Part II*, 44, 209–227, doi:10.1016/S0967-0645(96)00074-4.
- Schulz, K. G., I. Zondervan, L. J. A. Gerringa, K. R. Timmermans, M. J. W. Veldhuis, and U. Riebesell (2004), Effect of trace metal availability on coccolithophorid calcification, *Nature*, 430, 673–676, doi:10.1038/nature02631.
- Springer, A. M., C. P. McRoy, and M. V. Flint (1996), The Bering Sea Green Belt: Shelf-edge processes and ecosystem production, *Fish. Oceanogr.*, 5, 205–223, doi:10.1111/j.1365-2419.1996.tb00118.x.
- Sunda, W. G., and R. R. L. Guillard (1976), The relationship between cupric ion activity and the toxicity of copper to phytoplankton, *J. Mar. Res.*, 34, 511–529.
- Sunda, W. G., and S. A. Huntsman (1992), Feedback interactions between zinc and phytoplankton in seawater, *Limnol. Oceanogr.*, 37, 25–40, doi:10.4319/lo.1992.37.1.0025.
- Sunda, W. G., and S. A. Huntsman (1995), Cobalt and zinc inter-replacement in marine phytoplankton: Biological and geochemical implications, *Limnol. Oceanogr.*, 40, 1404–1417, doi:10.4319/lo.1995.40.8.1404.
- Suzuki, K., C. Minami, H. Liu, and T. Saino (2002), Temporal and spatial patterns of chemotaxonomic algal pigments in the subarctic Pacific and the Bering Sea during the early summer of 1999, *Deep Sea Res., Part II*, 49, 5685–5704, doi:10.1016/S0967-0645(02)00218-7.
- Suzuki, K., A. Hinuma, H. Saito, H. Kiyosawa, H. Liu, T. Saino, and A. Tsuda (2005), Responses of phytoplankton and heterotrophic bacteria in the northwest subarctic Pacific to in situ iron fertilization as estimated by HPLC pigment analysis and flow cytometry, *Prog. Oceanogr.*, 64, 167–187, doi:10.1016/j.pocean.2005.02.007.
- Takeda, S. (1998), Influence of iron availability on nutrient consumption ratio of diatoms in oceanic waters, *Nature*, 393, 774–777, doi:10.1038/31674.
- Thibault, D., S. Roy, C. S. Wong, and J. K. Bishop (1999), The downward flux of biogenic material in the NE subarctic Pacific: Importance of algal sinking and mesozooplankton herbivory, *Deep Sea Res., Part II*, 46, 2669–2697, doi:10.1016/S0967-0645(99)00080-6.
- Tsuda, A., et al. (2003), A mesoscale iron enrichment in the western subarctic Pacific induces a large centric diatom bloom, *Science*, 300, 958–961, doi:10.1126/science.1082000.
- Turner, D. R., M. Whitfield, and A. G. Dickson (1981), The equilibrium speciation of dissolved components in freshwater and seawater at 25°C and 1 atm pressure, *Geochim. Cosmochim. Acta*, 45, 855–881, doi:10.1016/0016-7037(81)90115-0.
- Tyrrell, T., A. Merico, J. J. Wanick, C. S. Wong, N. Metzl, and F. Whitney (2005), Effect of seafloor depth on phytoplankton blooms in high-nitrate, low-chlorophyll (HNLC) regions, *J. Geophys. Res.*, 110, G02007, doi:10.1029/2005JG000041.
- van den Berg, C. M. G. (1982), Determination of copper complexation with natural organic ligands in seawater by equilibration with MnO<sub>2</sub> I. theory, *Mar. Chem.*, 11, 307–322, doi:10.1016/0304-4203(82)90028-7.
- van den Berg, C. M. G. (1985), Determination of the zinc complexing capacity in seawater by cathodic stripping voltammetry of zinc APDC complex-ions, *Mar. Chem.*, 16, 121–130, doi:10.1016/0304-4203(85)90017-9.
- Vasconcelos, M. T. S. D., M. F. C. Leal, and C. M. G. van den Berg (2002), Influence of the nature of the exudates released by different marine algae on the growth, trace metal uptake and exudation of *Emiliania huxleyi* in natural seawater, *Mar. Chem.*, 77, 187–210, doi:10.1016/S0304-4203(01)00087-1.
- Wu, J., and E. A. Boyle (1998), Determination of iron in seawater by high-resolution isotope dilution inductively couple plasma mass spectrometry after Mg(OH)<sub>2</sub> coprecipitation, *Anal. Chim. Acta*, 367, 183–191, doi:10.1016/S0003-2670(98)00145-7.
- Xu, Y., D. Tang, Y. Shaked, and F. M. M. Morel (2007), Zinc, cadmium, and cobalt interreplacement and relative use efficiencies in the coccolithophore *Emiliania huxleyi*, *Limnol. Oceanogr.*, 52, 2294–2305, doi:10.4319/lo.2007.52.5.2294.
- Yee, D., and F. M. M. Morel (1996), In vivo substitution of zinc by cobalt in carbonic anhydrase of a marine diatom, *Limnol. Oceanogr.*, 41, 573–577, doi:10.4319/lo.1996.41.3.0573.