# Dissolved macronutrient concentrations from incubation experiments performed on RVIB Nathaniel B. Palmer cruise NBP 16-08 from September to October 2016

Website: https://www.bco-dmo.org/dataset/743072

Data Type: Cruise Results

Version: 1

Version Date: 2018-07-31

#### **Project**

» <u>Collaborative Research: Investigating Iron-binding Ligands in Southern Ocean Diatom Communities: The Role of Diatom-Bacteria Associations</u> (Diatom Bacteria Ligands)

Contributors	Affiliation	Role
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#### **Abstract**

Dissolved macronutrient concentrations from incubation experiments performed on RVIB Nathaniel B. Palmer cruise NBP 16-08 from September to October 2016.

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## Coverage

**Spatial Extent**: N:-62.332 **E**:-59.565 **S**:-62.46 **W**:-64.647

Temporal Extent: 2016-09-13 - 2016-10-09

#### **Dataset Description**

NBP16-08 incubation dissolved macronutrient concentrations.

#### Methods & Sampling

#### Sampling and analytical procedures:

**Incubation 1** was setup from unfiltered seawater collected aboard the R/V/I/B Nathanial B. Palmer using a SeaBird GEOTRACES style SBE32 rosette system deployed on a conducting Kevlar line with

OceanTestEquipment, Inc. X-Niskin samplers modified for trace element sampling. Seawater was collected from three consecutive casts to 25-35 m depth at -62.332N, -64.647E and the seawater from each cast was homogenized in three acid-cleaned and Milli-Q (>18.2 M $\Omega$  cm) conditioned 50-L polypropylene carboys, then distributed into acid-cleaned and Milli-Q conditioned 4-L polycarbonate incubation bottles that were rinsed three times with the seawater prior to filling. Once filled, incubation bottles were spiked with their treatments in laminar flow hoods in a shipboard trace metal bubble, sealed with the caps wrapped in parafilm and placed in a 2  $^{\circ}$ C temperature-controlled lit incubator van with 24 hour blue light (Hopkinson et al. 2007). Dark treatment bottles were placed in the same van but wrapped in heavy-duty black construction garbage bags.

**Incubation 2** was setup from unfiltered seawater collected aboard the R/V/I/B Nathanial B. Palmer using a SeaBird GEOTRACES style SBE32 rosette system deployed on a conducting Kevlar line with OceanTestEquipment, Inc. X-Niskin samplers modified for trace element sampling. Seawater was collected from two consecutive casts to 25-35 m depth at -62.46N, -59.565E and the seawater from each cast was homogenized in three acid-cleaned and Milli-Q (>18.2 M $\Omega$  cm) conditioned 50-L polypropylene carboys, then distributed into acid-cleaned and Milli-Q conditioned 4-L polycarbonate incubation bottles that were rinsed three times with the seawater prior to filling. Once filled, incubation bottles were spiked with their treatments in laminar flow hoods in a shipboard trace metal bubble, sealed with the caps wrapped in parafilm and placed in a 2  $^{\circ}$ C temperature-controlled lit incubator van with 24 hour blue light (Hopkinson et al. 2007). Dark treatment bottles were placed in the same van but wrapped in heavy-duty black construction garbage bags.

Incubation 3 was setup from a combination of filtered and unfiltered seawater collected aboard the R/V/I/B Nathanial B. Palmer using a SeaBird GEOTRACES style SBE32 rosette system deployed on a conducting Keylar line with OceanTestEquipment, Inc. X-Niskin samplers modified for trace element sampling. Seawater was collected from one cast to 25-35 m depth at -62.46N, -59.565E, the location of Incubation 2, filtered through a 0.2 um Acropak membrane capsule filter (Pall) and stored in an acid-cleaned and Milli-Q (>18.2 M $\Omega$  cm) conditioned 50-L polypropylene carboy. Additional unfiltered seawater was collected from one cast to 25-35 m depth at -62.332N, -64.647E, the location of Incubation 1, and homogenized in two acid-cleaned and Milli-Q  $(>18.2 \text{ M}\Omega \text{ cm})$  conditioned 50-L polypropylene carboys. The unfiltered water from the Incubation 1 station was distributed into acid-cleaned and Milli-Q conditioned 4-L polycarbonate incubation bottles that were rinsed three times with the seawater prior to filling and used for one treatment of Incubation 3 (Q in the light, V in the dark). The filtered seawater from the Incubation 2 station was mixed in a 50:50 ratio with the unfiltered water from the Incubation 1 station in acid-cleaned and Milli-Q conditioned 4-L polycarbonate incubation bottles that were rinsed three times with the seawater prior to filling and used for the second treatment in Incubation 3 (R in the light. W in the dark). Once filled, incubation bottles were sealed with the caps wrapped in parafilm and placed in a 2  $^{\circ}$ C temperature-controlled lit incubator van with 24 hour blue light (Hopkinson et al. 2007). Dark treatment bottles were placed in the same van but wrapped in heavy-duty black construction garbage bags.

For all incubations, samples for macronutrients were filtered through sequential 5 um and 0.4 um acid-cleaned polycarbonate track etched filters (Whatman Nuclepore) on Teflon filtration rigs (Savillex) and the filtrate collected in 50 mL Falcon tubes that had been rinsed with distilled water (DIW), soaked overnight in 10% hydrocholoric acid (HCl, Fisher, Trace Metal Grade), rinsed three times with DIW again, dried and rinsed three times with sample prior to filling. Samples were analyzed shipboard, typically within 24 hours, for nitrate+nitrite, phosphate, silicate, and occasionally nitrite. Until analyzed shipboard, samples were stored sealed at 4 °C in the dark and, following analyses, samples were frozen at -20 °C and shipped back to the University of South Florida for laboratory-based analyses of nitrite and ammonium, and in some cases again for nitrate+nitrite, phosphate and silicate.

Analytical methodology was based on established methods (Parsons 1984; Gordon et al. 1993) as described for the Lachat 8500 QuickChem in the Lachat QuickChem methods manuals and for the Technicon AAII in the CARIACO methods manual. All labware were either glass or high density polyethylene and were cleaned by an initial distilled water (DIW) rinse, followed by an overnight 10% hydrochloric acid (Fisher, Trace Metal Grade) soak, then rinsed three times with DIW and three times with solvent, analyte or sample prior to fill. Dedicated glassware was used for reagents and standards to avoid cross contamination issues. All reagents were made in high purity Milli-Q (>18 M $\Omega$  cm) water.

An artificial seawater matrix was used as the carrier for the analyses on the Lachat QuickChem 8500 at sea and on the Technicon AAII in the lab. 4L batches of artificial seawater were made by dissolving pre-weighed salts (128.50 g NaCl, 28.50 g MgSO4\*7H2O, 0.6714 g NaHCO3) in Milli-Q to match Southern Ocean seawater salinity and adjusted to match Southern Ocean seawater pH with 10% hydrochloric acid.

Five-point standard curves were analyzed in duplicate at the beginning and end of each run with duplicate reagent blanks, and quality control checks every seventh sample. Quality control check consisted of either an international reference sample or a quality control sample. The certified nutrient reference samples used, lots CC and CD, were purchased from Kanso Technos in Osaka, Japan. The quality control sample was made from a

20-L homogenized surface Southern Ocean filtered and autoclaved seawater sample. The midpoint standard from the calibration curve was also analyzed every fourteenth sample to check for drift during the runs.

Detection limits for all five parameters on the two instruments used were determined from three times the standard deviation of replicate artificial seawater blanks (n>6). On the Lachat QuickChem 8500, limits of detection were 0.01 uM for nitrate+nitrite, 0.02 uM for phosphate, 0.03 uM for silicate, 0.05 uM for nitrate+nitrite and 0.5 uM for ammonium. On the Technicon AAII, limits of detection were 0.06 uM for nitrate+nitrite, 0.02 uM for phosphate, 0.2 uM for silicate, 0.01 uM for nitrite, and 0.05 uM for ammonium.

Sample analyses for macronutrients were performed by William Abbott (USF).

Gordon, L.I., Jennings, J., J.C., Ross, A.A. and Krest, J.M. 1993. A suggested protocol for continuous flow automated analysis of seawater nutrients (phosphate, nitrate, nitrite and silicic acid) in the WOCE Hydrographic Program and the Joint Global Ocean Fluxes Study, Methods Manual WHPO 91-1. WOCE Hydrographic Program Office.

Hopkinson, B. M., B. G. Mitchell, R. A. Reynolds, H. Wang, K. E. Selph, C. I. Measures, C. D. Hewes, O. Holm-Hansen, and K. A. Barbeau. 2007. Iron limitation across chlorophyll gradients in the southern Drake Passage: Phytoplankton responses to iron addition and photosynthetic indicators of iron stress. Limnology and Oceanography 52: 2540-2554.

Parsons, T.R., Maita, Y. and Lalli, C.M. 1984. A manual of chemical and biological methods for seawater analysis. Pergammon Press, Oxford, 173 pp.

#### **Data Processing Description**

Data were processed using Omnion 4.2 software.

#### **BCO-DMO Processing:**

- changed date format from m/dd/yyyy to yyyy-mm-dd;
- replaced % with "pcnt" in param names;
- replaced spaces with underscores in TREATMENT column;
- combined 3 separate files into one dataset.

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#### **Data Files**

#### File

Nutrients\_Inc.csv(Comma Separated Values (.csv), 37.32 KB)

MD5:a2500c338d1192b72c46b30ca23362f0

Primary data file for dataset ID 743072

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#### **Related Publications**

Gordon, L. I., J. C. Jennings, JR, A. A. Ross, and J. M. Krest. (1994). A suggested protocol for continuous flow analysis of seawater nutrients (phosphate, nitrate, nitrite, and silicic acid) in the WOCE Hydrographic Program and the Joint Global Ocean Fluxes Study. WHP Office Report 91-1. Revision 1, Nov. 1994. WOCE Hydrographic Program Office, Woods Hole, MA.

Related Research

Hopkinson, B. M., Mitchell, B. G., Reynolds, R. A., Wang, H., Selph, K. E., Measures, C. I., ... Barbeau, K. A. (2007). Iron limitation across chlorophyll gradients in the southern Drake Passage: Phytoplankton responses to iron addition and photosynthetic indicators of iron stress. Limnology and Oceanography, 52(6), 2540–2554. doi:10.4319/lo.2007.52.6.2540

Methods

Parsons, T. R., Y. Maita, and C. M. Lalli. "A Manual of Chemical and Biological Methods of Seawater Analysis", Pergamon Press (1984). ISBN: <u>9780080302874</u> *Methods* 

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# **Parameters**

Parameter	Description	Units
INCUBATION	Incubation identifier	unitless
DATE	GMT date when incubation sample was pulled from the incubation bottle for filtering, in format yyyy-mm-dd	
DAY	Day of incubation when sample was collected. Days start from 0 for the day the incubation was setup.	unitless
ID	Sample identifier for incubation bottle and treatment that sample was collected from.	unitless
TREATMENT	Incubation treatment identifier. Treatments A-F were exposed to light, treatments G-L were kept in the dark. Treatments were as follows: A and $G=+0$ , control; B and $H=+1$ nM 57FeCl3; C and $I=+4$ nM Fe 57FeCl3; D and $J=+10$ nM Fe 57FeCl3; E and $K=+600$ pM Vitamin B12; F and $L=+4$ nM 57FeCl3 and $+600$ pM Vitamin B12. The R-A, R-B, R-C, R-D, R-E, and R-F bottles were the same light treatments as A, B, C, D, E, and F, respectively, in replicate R-labeled 4-L incubation bottles.	
BTLNBR	Incubation bottle number. Each 4-L incubation bottle was assigned a unique number from 1-99 across all shipboard incubations.	unitless
NO3_NO2	Concentration of dissolved nitrate+nitrite. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value. Noted as "bdl" when result was below detection limit of instrument(s) used.	micromoles per liter (uM)
NO3_NO2_STDEV	Standard deviation of replicate nitrate+nitrite concentration measurements. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	micromoles per liter (uM)
NO3_NO2_pcnt_RSD	Percent relative standard deviation of replicate nitrate+nitrite concentration measurements. Calculated as NO3_NO2_STDEV divided by NO3_NO2 and multiplied by 100. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless (percent)
NO3_NO2_INSTR	Notes which instrument was used for nitrate+nitrite measurements. $1 = \text{Lachat } 8500 \text{ QuickChem}$ , $2 = \text{Technicon AAII}$ , $3 = \text{Both Lachat } 8500 \text{ QuickChem}$ and Technicon AAII. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless
PO4	Concentration of dissolved reactive phosphate. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value. Noted as "bdl" when result was below detection limit of instrument(s) used.	micromoles per liter (uM)
PO4_STDEV	Standard deviation of replicate phosphate concentration measurements.  Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	
PO4_pcnt_RSD	Percent relative standard deviation of replicate phosphate concentration measurements. Calculated as PO4_STDEV divided by PO4 and multiplied by 100. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless (percent)

PO4_INSTR	Notes which instrument was used for phosphate measurements. 1 = Lachat 8500 QuickChem, 2 = Technicon AAII, 3 = Both Lachat 8500 QuickChem and Technicon AAII. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless
SiO4	Concentration of dissolved silicate. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value. Noted as "bdl" when result was below detection limit of instrument(s) used.	micromoles per liter (uM)
SiO4_STDEV	Standard deviation of replicate silicate concentration measurements. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	micromoles per liter (uM)
SiO4_pcnt_RSD	Percent relative standard deviation of replicate silicate concentration measurements. Calculated as SiO4_STDEV divided by SiO4 and multiplied by 100. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless (percent)
SiO4_INSTR	Notes which instrument was used for silicate measurements. $1 = \text{Lachat}$ 8500 QuickChem, $2 = \text{Technicon AAII}$ , $3 = \text{Both Lachat}$ 8500 QuickChem and Technicon AAII. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless
NO2	Concentration of dissolved nitrite. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value. Noted as "bdl" when result was below detection limit of instrument(s) used.	micromoles per liter (uM)
NO2_STDEV	Standard deviation of replicate nitrite concentration measurements. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	micromoles per liter (uM)
NO2_pcnt_RSD	Percent relative standard deviation of replicate nitrite concentration measurements. Calculated as NO2_STDEV divided by NO2 and multiplied by 100. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless (percent)
NO2_INSTR	Notes which instrument was used for nitrite measurements. $1 = \text{Lachat}$ 8500 QuickChem, $2 = \text{Technicon AAII}$ , $3 = \text{Both Lachat 8500 QuickChem}$ and Technicon AAII. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless
NH4	Concentration of dissolved ammonium. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value. Noted as "bdl" when result was below detection limit of instrument(s) used.	micromoles per liter (uM)
NH4_STDEV	Standard deviation of replicate ammonium concentration measurements. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	micromoles per liter (uM)
NH4_pcnt_RSD	Percent relative standard deviation of replicate ammonium concentration measurements. Calculated as NH4_STDEV divided by NH4 and multiplied by 100. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless (percent)
NH4_INSTR	Notes which instrument was used for ammonium measurements. 1 = Lachat 8500 QuickChem, 2 = Technicon AAII, 3 = Both Lachat 8500 QuickChem and Technicon AAII. Noted as "nda" when no data available for this sample, no sample analyzed, or obviously erroneous data value.	unitless

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## Instruments

Dataset- specific Instrument Name	Lachat QuickChem 8500
Generic Instrument Name	Flow Injection Analyzer
Dataset- specific Description	Lachat QuickChem 8500 series 2, 4 channel analyzer
	An instrument that performs flow injection analysis. Flow injection analysis (FIA) is an approach to chemical analysis that is accomplished by injecting a plug of sample into a flowing carrier stream. FIA is an automated method in which a sample is injected into a continuous flow of a carrier solution that mixes with other continuously flowing solutions before reaching a detector. Precision is dramatically increased when FIA is used instead of manual injections and as a result very specific FIA systems have been developed for a wide array of analytical techniques.

Dataset- specific Instrument Name	Technicon AAII
Generic Instrument Name	Technicon AutoAnalyzer II
Dataset- specific Description	Technicon AAII
Generic Instrument Description	A rapid flow analyzer that may be used to measure nutrient concentrations in seawater. It is a continuous segmented flow instrument consisting of a sampler, peristaltic pump, analytical cartridge, heating bath, and colorimeter. See more information about this instrument from the manufacturer.

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# **Deployments**

#### **NBP1608**

Website	https://www.bco-dmo.org/deployment/742174	
Platform	RVIB Nathaniel B. Palmer	
Start Date	2016-09-07	
End Date	2016-10-14	

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# **Project Information**

Collaborative Research: Investigating Iron-binding Ligands in Southern Ocean Diatom Communities: The Role of Diatom-Bacteria Associations (Diatom\_Bacteria\_Ligands)

Coverage: Southern Ocean, Western Antarctic Peninsula 60-65 S, 63 W

This project focuses on an important group of photosynthetic algae in the Southern Ocean (SO), diatoms, and

the roles associated bacterial communities play in modulating their growth. Diatom growth fuels the SO food web and balances atmospheric carbon dioxide by sequestering the carbon used for growth to the deep ocean on long time scales as cells sink below the surface. The diatom growth is limited by the available iron in the seawater, most of which is not freely available to the diatoms but instead is tightly bound to other compounds. The nature of these compounds and how phytoplankton acquire iron from them is critical to understanding productivity in this region and globally. The investigators will conduct experiments to characterize the relationship between diatoms, their associated bacteria, and iron in open ocean and inshore waters. Experiments will involve supplying nutrients at varying nutrient ratios to natural phytoplankton assemblages to determine how diatoms and their associated bacteria respond to different conditions. This will provide valuable data that can be used by climate and food web modelers and it will help us better understand the relationship between iron, a key nutrient in the ocean, and the organisms at the base of the food web that use iron for photosynthetic growth and carbon uptake. The project will also further the NSF goals of training new generations of scientists and of making scientific discoveries available to the general public. The project supports early career senior investigators and the training of graduate and undergraduate students as well as outreach activities with middle school Girl Scouts in Rhode Island, inner city middle and high school age girls in Virginia, and middle school girls in Florida.

The project combines trace metal biogeochemistry, phytoplankton cultivation, and molecular biology to address questions regarding the production of iron-binding compounds and the role of diatom-bacterial interactions in this iron-limited region. Iron is an essential micronutrient for marine phytoplankton. Phytoplankton growth in the SO is limited by a lack of sufficient iron, with important consequences for carbon cycling and climate in this high latitude regime. Some of the major outstanding questions in iron biogeochemistry relate to the organic compounds that bind >99.9% of dissolved iron in surface oceans. The investigators' prior research in this region suggests that production of strong iron-binding compounds in the SO is linked to diatom blooms in waters with high nitrate to iron ratios. The sources of these compounds are unknown but the investigators hypothesize that they may be from bacteria, which are known to produce such compounds for their own use. The project will test three hypotheses concerning the production of these iron-binding compounds, limitations on the biological availability of iron even if present in high concentrations, and the roles of diatom-associated bacteria in these processes. Results from this project will provide fundamental information about the biogeochemical trigger, and biological sources and function, of natural strong iron-binding compound production in the SO, where iron plays a critical role in phytoplankton productivity, carbon cycling, and climate regulation.

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## **Funding**

Funding Source	Award
NSF Office of Polar Programs (formerly NSF PLR) (NSF OPP)	OPP-1443483
NSF Office of Polar Programs (formerly NSF PLR) (NSF OPP)	OPP-1443474
NSF Office of Polar Programs (formerly NSF PLR) (NSF OPP)	OPP-1443646

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