# Direct measure of phytoplankton cell quotas from field populations sampled from multiple cruises cruises between 2010 and 2016

Website: https://www.bco-dmo.org/dataset/849153

Data Type: Cruise Results

Version: 1

Version Date: 2021-06-02

## **Project**

» Biological Controls on the Ocean C:N:P ratios (Biological C:N:P ratios)

## **Programs**

- » <u>Dimensions of Biodiversity</u> (Dimensions of Biodiversity)
- » Ocean Carbon and Biogeochemistry (OCB)

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

This dataset represents direct measures of phytoplankton cell quotas from field populations, contributing to the project goal of understanding how taxon-specific stoichiometry and growth rates, genomic analyses control the stoichiometry of surface ocean plankton. The field work included several cruises: AE1123 (September/October 2011) and AE1226 (September 2012) both from Bermuda to Puerto Rico, AE1319 (June 2013) from Bermuda to the Labrador Sea, and NH1418 (May 2014) from Hawaii to Tahiti, among others.

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## Coverage

**Spatial Extent:** N:55 **E**:95 **S**:-20.76 **W**:-157 **Temporal Extent:** 2010-10-31 - 2016-04-15

## Methods & Sampling

Phytoplankton sample collection and preparation. Samples for taxon-specific elemental content were collected using 12 liter (L) Niskin bottles, usually from two depths at each station (see supplemental file for map of stations from which samples were collected and cell-sorted to quantify cellular macronutrient content; one within the upper mixed layer (10-20 meters) and one near the deep chlorophyll maximum. Due to timing of the cruises, samples

were collected from a seasonal window limited to summer and early fall. Samples were prepared for sorting by concentrating cells from the ambient sample under darkened conditions using a Memteq High Volume Cell Trap (0.2 micrometer pore size), and then flushed from the Cell Trap using 0.2 micrometer ( $\mu$ m) filtered seawater and a luer lock syringe; roughly 3-4 liters (L) of ambient seawater was concentrated down to ~5 milliliters (mls). Samples were either gravity filtered through the Cell Trap directly from the Niskin bottle or pushed through the cell trap using a peristaltic pump at <50 milliliter per minute (ml min-1). Samples were then kept cool at 4 degrees C if they were going to be sorted live and immediately after collection or fixed with freshly filtered paraformaldehyde (0.5% v/v final concentration) if they were going to be stored for later sorting. Samples were allowed to fix at 4 degrees C for 1-2 hours and then flash frozen in liquid nitrogen and then moved to -80 degrees C for longer term storage.

Cell sorting and elemental analysis. The cell counting and sorting protocols used follow that of Baer et al. (2017). Cell counts and sorts were performed on either a Becton Dickinson Influx or FACSJazz flow cytometer, each utilizing a 200 milliwatt (mW) 488 namometer (nm) laser, with detectors for forward scatter, side scatter, 692 namometer (nm), and 530 namometer (nm), and operated with 8 grams of NaCL kg-1 solution for sheath fluid, which was filtered inline using a 0.22 micrometer (µm) Millipore SterivexTM filter. Instrument alignment was performed with 3.0 micrometer (um) 6-peak rainbow beads, while roughly hourly checks on forward scatter response were performed with 0.53 micrometer (µm) Nile Red beads (Spherotech). Prochlorococcus populations were discriminated based on forward scatter and red fluorescence, and a gate in orange (530 nm) discriminated for Synechococcus. Eukaryotes were all larger autofluorescing cells that did not fit the cyanobacterial gating scheme. For sorting, sort control software was set to "1.0 drop pure" sort mode. In excess of 17 million, 8 million, and 500,000 cells for *Prochlorococcus*, *Synechococcus*, and eukaryotes were sorted for chemical analysis, respectively. Not all phytoplankton groups were sufficiently abundant at each station/depth for practical sorting and subsequent analysis. Post-sort purity tests were run with subsamples of each sorted population; sort purity always exceeded 94%. Sorted populations were collected in polystyrene Falcon tubes (BD Biosciences Inc.) and subsequently filtered on pre-combusted (450 degrees C for 4 hours) GF-75 filters (Ahlstrom; nominal pore size = 0.3 micrometer). To ensure complete capture of all of the cells, the Falcon tubes were rinsed multiple times with 0.2 micrometer (µm) filtered sheath fluid. Filter and sheath fluid blanks were produced each day samples were sorted for subsequent subtraction from the mass of each unknown sample. Following filtration, filters were placed in acid-washed cryovials and frozen (-20 degrees C) until analysis as described below. Average cellular elemental content was determined by dividing the elemental content of the sample (see next section) by the number of sorted cells as determined in a direct post-sort count analysis of the sorted sample.

Particulate nutrients. Particulate organic P (POP) was analyzed using the ash-hydrolysis method, with oxidation efficiency and standard recovery tested with each sample run using an ATP standard solution and a certified phosphate standard (OSIL Phosphate Nutrient Standard Solution). Method precision is 1-2% at 5 nanomoles per kilogram (nmol kg-1). Samples for POC and PON were acid fumed in a desiccator over concentrated HCl prior to analysis. After acid fuming, POC and PON were determined on a Costech 4010 elemental analyzer or a Control Equipment 440 elemental analyzer, depending on the samples. L-glutamic acid (USGS40) was used for standard curve generation and as a check standard (tolerance of <0.1 microgram) approximately every ten samples and at the end of every instrument run. Empty tin capsules (Costech Analytical Technologies) were cleaned with acetone and dried and run as instrument blanks.

Additional methodological details can be found in Lomas et al. 2021 (currently in revision).

## **Data Processing Description**

Data were calculated using standard Microsoft Excel.

## **BCO-DMO Processing Description**

- Converted dates to ISO8601 date format (YYYY-MM-DD);
- Adjusted field/parameter names to comply with BCO-DMO naming conventions;
- Added a conventional header with dataset name, PI names, version date.

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#### **File**

sorted\_cell\_quotas.csv(Comma Separated Values (.csv), 10.71 KB)

MD5:2be6c86d6d27bf679b77c5934c7b56b4

Primary data file for dataset ID 849153

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# **Supplemental Files**

#### File

#### Sorted cell quotas cruise locations map

filename: sorted\_cell\_quotas\_cruise\_locations\_map.pdf (Portable Document Format (.pdf), 560.61 KB)
MD5:225f230981f5963edda812b3ad3f1433

Map of stations from which samples were collected and cell-sorted to quantify cellular macronutrient content.

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### **Related Publications**

Baer, S. E., Lomas, M. W., Terpis, K. X., Mouginot, C., & Martiny, A. C. (2017). Stoichiometry of Prochlorococcus, Synechococcus, and small eukaryotic populations in the western North Atlantic Ocean. Environmental Microbiology, 19(4), 1568–1583. doi:10.1111/1462-2920.13672

Methods

Lomas, M. W., Baer, S. E., Mouginot, C., Terpis, K. X., Lomas, D. A., Altabet, M. A., & Martiny, A. C. (2021). Varying influence of phytoplankton biodiversity and stoichiometric plasticity on bulk particulate stoichiometry across ocean basins. Communications Earth & Environment, 2(1). https://doi.org/10.1038/s43247-021-00212-9

Results

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## **Parameters**

Parameter	Description	Units
Categorical_Region	Ocean region name as defined in Lomas et al. 2021 publication (currently in revision)	unitless
Cruise_ID	Cruise identifier	unitless
Date	Date in format YYYY-MM-DD	unitless
Latitude	Latitude of station sampled	degrees North
Longitude	Longitude of station sampled	degrees West
Station	Station name on the specific cruise	unitless
Depth	Depth of sample collection	meters

Prochlorococcus_POC	Particulate Organic Carbon (POC) per cell of in situ Prochlorococcus population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Prochlorcoccus_PON	Particulate Organic Nitrogen (PNC) per cell of in situ Prochlorococcus population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Prochlorococcus_POP	Particulate Organic Phosphorus (POP) per cell of in situ Prochlorococcus population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Synechococcus_POC	Particulate Organic Carbon (POC) per cell of in situ Synechococcus population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Synechococcus_PON	Particulate Organic Nitrogen (PNC) per cell of in situ Synechococcus population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Synechococcus_POP	Particulate Organic Phosphorus (POP) per cell of in situ Synechococcus population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Eukaryote_POC	Particulate Organic Carbon (POC) per cell of in situ eukaryote population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Eukaryote_PON	Particulate Organic Nitrogen (PNC) per cell of in situ eukaryote population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)
Eukaryote_POP	Particulate Organic Phosphorus (POP) per cell of in situ eukaryote population, derived from flow cytometric sorting and subsequent chemical analysis	femtomole per cell (fmol/cell)

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# Instruments

Dataset-specific Instrument Name	Costech 4010 elemental analyzer, Control Equipment 440 elemental analyzer
Generic Instrument Name	CHN Elemental Analyzer
Dataset-specific Description	Costech 4010 elemental analyzer or a Control Equipment 440 elemental analyzer depending on the sample
Generic Instrument Description	A CHN Elemental Analyzer is used for the determination of carbon, hydrogen, and nitrogen content in organic and other types of materials, including solids, liquids, volatile, and viscous samples.

Dataset- specific Instrument Name	
Generic Instrument Name	Flow Cytometer
Dataset- specific Description	Becton Dickinson Influx or FACSJazz flow cytometer, each utilizing a 200 mW 488 nm laser, with detectors for forward scatter, side scatter, 692 nm, and 530 nm
Generic Instrument Description	Flow cytometers (FC or FCM) are automated instruments that quantitate properties of single cells, one cell at a time. They can measure cell size, cell granularity, the amounts of cell components such as total DNA, newly synthesized DNA, gene expression as the amount messenger RNA for a particular gene, amounts of specific surface receptors, amounts of intracellular proteins, or transient signalling events in living cells. (from: <a href="http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm">http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm</a> )

Dataset- specific Instrument Name	
Generic Instrument Name	Niskin bottle
	A Niskin bottle (a next generation water sampler based on the Nansen bottle) is a cylindrical, non-metallic water collection device with stoppers at both ends. The bottles can be attached individually on a hydrowire or deployed in 12, 24, or 36 bottle Rosette systems mounted on a frame and combined with a CTD. Niskin bottles are used to collect discrete water samples for a range of measurements including pigments, nutrients, plankton, etc.

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# **Deployments**

# AE1319

Website	https://www.bco-dmo.org/deployment/537979
Platform	R/V Atlantic Explorer
Report	http://dmoserv3.whoi.edu/data_docs/Bio_CNP_Ratios/AE1319_Cruise_Report_09182013_reduced2.pdf
Start Date	2013-08-14
End Date	2013-09-11
Description	Cruise for project 'Dimensions of Biodiversity: Biological Controls on the Ocean C:N:P ratios'.

# NH1410

Website	https://www.bco-dmo.org/deployment/628491
Platform	R/V New Horizon
Report	http://dmoserv3.whoi.edu/data_docs/OMZ_SulfurCycling/Cruise_Report_NH1410.pdf
Start Date	2014-05-10
End Date	2014-06-08
Description	Oxygen Minimum Zone Microbial Biogeochemistry Expedition 2 (OMZoMBiE 2) Cruise Track (PDF) Cruise information and original data are available from R2R: <a href="https://www.rvdata.us/search/cruise/NH1410">https://www.rvdata.us/search/cruise/NH1410</a>

## NH1418

Website	https://www.bco-dmo.org/deployment/829909	
Platform	R/V New Horizon	
Start Date	2014-09-19	
End Date	2014-10-07	
Description	For project "Biological Controls on the Ocean C:N:P Ratio".	

## RR1604

Website	https://www.bco-dmo.org/deployment/723194	
Platform	R/V Roger Revelle	
Start Date	2016-03-21	
End Date	2016-04-28	

## **SO243**

Website	https://www.bco-dmo.org/deployment/850839	
Platform	R/V Sonne	
Start Date	2015-10-05	
End Date	2015-10-22	

## **AE1032**

ALIUUL	
Website	https://www.bco-dmo.org/deployment/542529
Platform	R/V Atlantic Explorer
Start Date	2010-10-18
End Date	2010-11-03
Description	Bermuda Atlantic Time-series Study (BATS) Validation stations, Panulirus Hydrographic Stations, Prochlorococcus in the Sargasso Sea

## AE1226

Website	https://www.bco-dmo.org/deployment/852206
Platform	R/V Atlantic Explorer
Start Date	2012-09-26
End Date	2012-10-13

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# **Project Information**

Biological Controls on the Ocean C:N:P ratios (Biological C:N:P ratios)

Coverage: western North Atlantic; 60N to 20N along 66W longitude; 20N to 15S in the tropical Pacific

One of the fundamental patterns of ocean biogeochemistry is the Redfield ratio, linking the stoichiometry of surface plankton with the chemistry of the deep ocean. There is no obvious mechanism for the globally consistent C:N:P ratio of 106:16:1 (Redfield ratio), especially as there is substantial elemental variation among plankton communities in different ocean regions. Thus, knowing how biodiversity regulates the elemental composition of the ocean is important for understanding the ocean and climate as a whole -- now and in the future.

The conceptual hypotheses for this study are as follows: 1. The C:N:P ratio of a cell is constrained by its broad taxonomic group, which determines, for example, whether it has an outer shell, its size, functional metabolism, membrane lipid composition. 2. Within a taxon, there is high genetic diversity. Some of this genetic diversity is potentially laterally transferred, or can be lost within taxa, and confers various functional abilities (organic phosphate assimilation, nitrate assimilation, photoheterotrophy, etc.). Functional diversity provides the cell with further flexibility, such as the ability to respond to varying nutrient supply rates/ratios, and affects a cell's C:N:P ratio within the range specified by the taxon. 3. Given these taxonomic and genetic constraints, a cell is physiologically plastic and modifies how it allocates cellular resources in response to nutrient supply rates/ratios in the environment. 4. The microbial diversity (taxonomic, genetic, and functional) of the surface ocean varies over time and space, driven by many factors in addition to nutrients. The sum of this mixture composes the ecosystem C:N:P, the ratio that Redfield described.

Based on this framework, the CoPIs will make field observations of taxon-specific stoichiometry and growth rates, genomic analyses, and conduct laboratory chemostat experiments to improve understanding of how ocean taxonomic, genetic, and functional biodiversity control the stoichiometry of the surface ocean plankton. Their analyses of these data would lead to a mechanistic understanding of variations in the Redfield ratio, both spatially and temporally.

This study will greatly expand knowledge of the genomic diversity among ocean microbes and how this diversity affects biogeochemistry. The stoichiometry of the ocean's microbes is a parameter that nearly every chemical or biological oceanographer uses, from converting measurements made in one element to another, to estimating regional and global nitrogen budgets. The research also has important implications for the global carbon budget and any changes that might result from climate change.

To understand mechanistically temporal and spatial variability of the plankton C:N:P ratio, biodiversity must be studied not only at the traditional taxonomic level, but at the genetic and functional levels which dictate organism response to their environment. Data will be integrated into a combined ocean ecological, evolutionary, and biogeochemical model, with flexible stoichiometry, including cellular biochemical allocations. Seeding a coupled physical-biological model of the oceans with multiple competing genotypes enables the exploration of ecological and evolutionary patterns of resource acquisition and C:N:P ratios. Developing a more mechanistic examination of the course of ecology and evolution, in which laboratory and field data define tradeoffs between different growth and nutrient acquisition strategies, would estabblish the framework of adaptive dynamics for determining "evolutionarily convergence". Finally, model outcomes will be evaluated against field data.

The field work planned for this project includes several cruises: BV46 (September/October 2011), BV48 (September 2012), a June 2013 cruise from Bermuda to the Labrador Sea, and a cruise from Hawaii to Tahiti (May 2014). Additionally, samples will be be acquired during cruises of opportunity.

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## **Program Information**

Dimensions of Biodiversity (Dimensions of Biodiversity)

Website: <a href="http://www.nsf.gov/funding/pgm\_summ.jsp?pims\_id=503446">http://www.nsf.gov/funding/pgm\_summ.jsp?pims\_id=503446</a>

Coverage: global

(adapted from the NSF Synopsis of Program)

Dimensions of Biodiversity is a program solicitation from the NSF Directorate for Biological Sciences. FY 2010 was year one of the program. [MORE from NSF]

The NSF Dimensions of Biodiversity program seeks to characterize biodiversity on Earth by using integrative, innovative approaches to fill rapidly the most substantial gaps in our understanding. The program will take a broad view of biodiversity, and in its initial phase will focus on the integration of genetic, taxonomic, and functional

dimensions of biodiversity. Project investigators are encouraged to integrate these three dimensions to understand the interactions and feedbacks among them. While this focus complements several core NSF programs, it differs by requiring that multiple dimensions of biodiversity be addressed simultaneously, to understand the roles of biodiversity in critical ecological and evolutionary processes.

## Ocean Carbon and Biogeochemistry (OCB)

Website: <a href="http://us-ocb.org/">http://us-ocb.org/</a>

Coverage: Global

The Ocean Carbon and Biogeochemistry (OCB) program focuses on the ocean's role as a component of the global Earth system, bringing together research in geochemistry, ocean physics, and ecology that inform on and advance our understanding of ocean biogeochemistry. The overall program goals are to promote, plan, and coordinate collaborative, multidisciplinary research opportunities within the U.S. research community and with international partners. Important OCB-related activities currently include: the Ocean Carbon and Climate Change (OCCC) and the North American Carbon Program (NACP); U.S. contributions to IMBER, SOLAS, CARBOOCEAN; and numerous U.S. single-investigator and medium-size research projects funded by U.S. federal agencies including NASA, NOAA, and NSF.

The scientific mission of OCB is to study the evolving role of the ocean in the global carbon cycle, in the face of environmental variability and change through studies of marine biogeochemical cycles and associated ecosystems.

The overarching OCB science themes include improved understanding and prediction of: 1) oceanic uptake and release of atmospheric CO2 and other greenhouse gases and 2) environmental sensitivities of biogeochemical cycles, marine ecosystems, and interactions between the two.

The OCB Research Priorities (updated January 2012) include: ocean acidification; terrestrial/coastal carbon fluxes and exchanges; climate sensitivities of and change in ecosystem structure and associated impacts on biogeochemical cycles; mesopelagic ecological and biogeochemical interactions; benthic-pelagic feedbacks on biogeochemical cycles; ocean carbon uptake and storage; and expanding low-oxygen conditions in the coastal and open oceans.

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## **Funding**

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1046297
NSF Division of Ocean Sciences (NSF OCE)	OCE-1045966

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