Prochlorococcus cell concentrations during the BiG-RAPA expedition (Cruise MV1015) in the Peru Current and Eastern South Pacific Subtropical Gyre between November and December of 2010

Website: https://www.bco-dmo.org/dataset/886299
Data Type: Cruise Results
Version: 1
Version Date: 2023-01-30

Project
» Center for Microbial Oceanography: Research and Education (C-MORE)

Contributors	Affiliation	Role
Chisholm, Sallie W.	Massachusetts Institute of Technology (MIT)	Principal Investigator
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Abstract
These data include Prochlorococcus cell concentrations (total cell densities by flow cytometry and cell densities for specific ecotypes/clades determined by quantitative PCR). Samples were collected during the C-MORE Biogeochemical Gradients Role in Arranging Planktonic Assemblages (BiG-RAPA) expedition (Cruise MV1015) in the Peru Current and Eastern South Pacific Subtropical Gyre between dates 2010-11-19 and 2010-12-10 along a zonal transect from the northern coast of Chile to the island of Rapa Nui. Prochlorococcus is an important primary producer in the oligotrophic South Pacific Gyre and these data facilitate studies examining Prochlorococcus' ecology.

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Coverage

Spatial Extent: **N**:-20.0834 **E**:-70.7997 **S**:-26.249 **W**:-103.961
Temporal Extent: 2010-11-19 - 2010-12-10

Methods & Sampling

These data include Prochlorococcus cell concentrations from samples collected during the C-MORE Biogeochemical Gradients Role in Arranging Planktonic Assemblages (BiG-RAPA) expedition (Cruise MV1015) in

the Peru Current and Eastern South Pacific Subtropical Gyre between dates 2010-11-19 and 2010-12-10 along a zonal transect from the northern coast of Chile to the island of Rapa Nui.

Seawater Collection: For both flow cytometry and qPCR samples, 500 milliliters (mL) of seawater from each depth was collected in a clean HDPE bottle (10% bleach for 20 minutes, followed by thorough rinsing with ship's tap water). Each clean HDPE bottle was rinsed 3 times with seawater from the Niskin bottle before filling.

Flow Cytometry: For each of the 7 stations along the transect, 9 samples were collected across the euphotic zone; from these, water samples were also collected for ecotype qPCR. Depths at each station were chosen based on the location of the deep chlorophyll maximum, the mixed layer depth, and light attenuation profiles. At station 7, samples were also collected from high-resolution deep-chlorophyll maximum casts (63 and 69), and counts for these are included. Fixed samples for flow cytometry were prepared by adding 5 microliters (ul) of 25% glutaraldehyde to 1 mL whole seawater, mixing, and allowing the sample to fix in the dark for 10 minutes, followed by flash freezing in liquid nitrogen. The samples were stored in liquid nitrogen or at -80C for 2-4 months before running flow cytometry. Two or three flow cytometry samples were preserved from each Niskin sampled, but only one of these sampling replicates was run. For each site and depth collected, a sample was run on a BD/Cytopeia Influx flow cytometer with 488 and 457 lasers both illuminating the sample and 2 um fluorescent bead standards mixed in. No stains were used. Detector sensitivity ranges were optimized for Prochlorococcus and held constant throughout all runs. Each sample was thawed in the dark and run twice (flow cytometry technical duplicates). Replicates were run back to back, flushing the lines in between, so that samples weren't left thawed for longer than 1 hour or put through extra freeze-thaw cycles. Values in the table are averages of the pair of technical duplicates. As measurements, these values are good to ~2-3 significant digits (depending on the length of time a sample was run and the number of Prochlorococcus cells actually detected).

Ecotype quantitative PCR: Samples for qPCR were collected and preserved as described (Zinser et al., 2006). 100 milliliters (mL) of seawater was filtered onto 25 millimeter (mm) diameter, 0.2 micrometer (um) pore size, polycarbonate filters (in triplicate) and chased with 3 mL of preservation solution (10 millimolar (mM) Tris pH=8, 100 mM EDTA, 500 mM NaCl). Filters were placed dry inside 2 mL screw cap polypropylene bead beater tubes and stored at -80 degress C until use. DNA from field samples was extracted from filters as previously described (Zinser et al., 2006). Cells were resuspended by adding 650 microliters (uL) of 10 mM Tris pH=8 to the bead beater tube containing the filter and bead beated at maximum speed (~4800 rpm) for 2 minutes. 500 uL of the respuspended cells were transfered to a 1.5 mL centrifuge tube and the cells were heat lysed at 95 degrees C for 15 minutes. DNA samples were then stored at -80 degrees C until analysis.

The qPCR assay was performed as previously described (Ahlgren et al., 2006; Zinser et al., 2006; Malmstrom et al., 2010; Malmstrom et al., 2012) using the same standards and reaction conditions. Technical duplicates of three replicate filters were analyzed for BiG RAPA (i.e. 6 data points per sample). Data quality was assessed using the percent coefficient of variation for the 6 data points. When cell concentrations fell below the detection limit of the assay (indicated by the associated quality flag), cell concentrations were set to the theoretical detection limit of 0.65 cells/mL.

General Issue: In most cases these water samples are not from the same bottles or depths as cruise core measurement casts.

Flow Cytometry: At station 1, the mixed layer samples do not have technical replicates. In the first few attempted flow cytometry runs Prochlorococcus cells were not visible - not because their fluorescence was low (these waters are relatively turbid), but because the cells were scarce. Later, the remainders of these samples were used for single, long slow runs, and we were able to detect a small Prochlorococcus population - these counts are not robust, representing small numbers with background noise, but they round out the depth profile for a rough idea.

Field samples were collected by Jessie Berta-Thompson and Paul Berube, flow cytometry data were collected and analyzed by Jessie Berta-Thompson and Allison Coe, and quantitative PCR data were collected and analyzed by Allison Coe and Paul Berube.

ProChl values were calculated by normalizing chlorophyll fluorescence of the target population to chlorophyll fluorescence of the standard in the following manner: the mean of red fluorescence (680 nm emmission wavelength) for the gated Prochlorococcus was divided by the mean of red fluorescence (680 nm emmission wavelength) for the gated fluorescent bead standard.

ProFSC values were calculated by normalizing forward scatter of the target population to forward scatter of the standard in the following manner: the mean of forward scatter for the gated Prochlorococcus was divided by the mean of forward scatter for the gated fluorescent bead standard.

QUALITY FLAGS column header = variable[QUALITY]
0 = Less than detection limit
1 = Good data
2 = Single measurement - no assessment of measurement variation
3 = >5% difference of techinical duplicates (flow cytometry)
4 = >50 %CV (ecotype qPCR)

Data Processing Description

Prochlorococcus counts were obtained by gating across chlorophyll fluorescence (680/40) vs. forward scatter using FlowJo software. Gates were adjusted by hand separately for each sample, since the cell properties change dramatically across this dataset. Concentrations were calculated using sample run times, sample counts and measured flow rates. For surface/mixed layer samples with higher light exposure and smaller cells, many Prochlorococcus populations in this data set were just above or only partly above the fluorescence noise - these counts represent estimates likely on the low side (gating to avoid noise, faintly fluorescing cells undetectable), and they are not as accurate as estimates for the well-separated deeper populations.

BCO-DMO Processing description:
- Adjusted field/parameter names to comply with BCO-DMO naming conventions
- Added a conventional header with dataset name, PI names, version date
- Rounded columns: "Latitude" and "Longitude" to 3 decimal places (or to the thousandth place)

Data Files

File
prochlorococcus_data.csv(Comma Separated Values (.csv), 12.60 KB)   MD5:7467b8f05eb7390c2a2fcbd451f9c3e4   Primary data file for dataset ID 886299

Related Publications

Ahlgren, N. A., Rocap, G., & Chisholm, S. W. (2006). Measurement of Prochlorococcus ecotypes using real-time polymerase chain reaction reveals different abundances of genotypes with similar light physiologies. Environmental Microbiology, 8(3), 441–454. https://doi.org/10.1111/j.1462-2920.2005.00910.x
Methods

Malmstrom, R. R., Coe, A., Kettler, G. C., Martiny, A. C., Frias-Lopez, J., Zinser, E. R., & Chisholm, S. W. (2010). Temporal dynamics of Prochlorococcus ecotypes in the Atlantic and Pacific oceans. The ISME Journal, 4(10), 1252–1264. doi:10.1038/ismej.2010.60
Methods

Malmstrom, R. R., Rodrigue, S., Huang, K. H., Kelly, L., Kern, S. E., Thompson, A., Roggensack, S., Berube, P. M., Henn, M. R., & Chisholm, S. W. (2012). Ecology of uncultured Prochlorococcus clades revealed through single-cell genomics and biogeographic analysis. The ISME Journal, 7(1), 184–198. https://doi.org/10.1038/ismej.2012.89
Methods

Zinser, E. R., Coe, A., Johnson, Z. I., Martiny, A. C., Fuller, N. J., Scanlan, D. J., & Chisholm, S. W. (2006). Prochlorococcus Ecotype Abundances in the North Atlantic Ocean As Revealed by an Improved Quantitative PCR Method. Applied and Environmental Microbiology, 72(1), 723–732. https://doi.org/10.1128/aem.72.1.723-

732.2006 https://doi.org/10.1128/AEM.72.1.723-732.2006
Methods

Parameters

Parameter	Description	Units
Station	Station ID number	unitless
Cast	CTD cast ID number	unitless
Bottle	Niskin bottle number	unitless
botid_number	Niskin bottle ID in format AABBBCCCDD where AA=cruise; BBB=station; CCC=cast; DD=niskin bottle	unitless
ISO_DateTime_UTC	Sample collection timestamp (UTC) in ISO 8601 format yyyy-mm-ddTHH:MM:SSZ	unitless
Latitude	latitude of study site North (South is negative)	decimal degrees
Longitude	latitude of study site East (West is negative)	decimal degrees
Depth	Sample collection depth relative to sea furface	meters (m)
Prochlorococcus	Prochlorococcus cell concentration based on flow cytometry counts	cells ml-1
Prochlorococcus_quality	Prochlorococcus Quality Flag; 0 = Less than detection limit; 1 = Good data; 2 = Single measurement - no assessment of measurement variation; 3 = >5% difference of techinical duplicates (flow cytometry)	unitless
ProChl	Chlorophyll fluorescence per cell relative to fluorescence of beads: the mean of red fluorescence (680 nm emission wavelength) for the gated Prochlorococcus was divided by the mean of red fluorescence (680 nm emission wavelength) for the gated fluorescent bead standard	unitless
ProFSC	Forward scatter (FSC) per cell relative to forward scatter of beads: the mean of forward scatter for the gated Prochlorococcus was divided by the mean of forward scatter for the gated fluorescent bead standard	unitless
eMIT9312	Prochlorococcus eMIT9312 (HLII clade) cell concentration based on quantitative PCR	cells ml-1

eMIT9312_quality	eMIT9312 Quality Flag; 0 = Less than detection limit; 1 = Good data; 2 = Single measurement - no assessment of measurement variation; 4 = >50 %CV (ecotypes qPCR)	unitless
eMED4	Prochlorococcus eMED4 (HLI clade) cell concentration based on quantitative PCR	cells ml-1
eMED4_quality	eMED4 Quality Flag; 0 = Less than detection limit; 1 = Good data; 2 = Single measurement - no assessment of measurement variation; 4 = >50 %CV (ecotypes qPCR)	unitless
HLIII	Prochlorococcus HLIII (HLIII clade) cell concentration based on quantitative PCR	cells ml-1
HLIII_quality	HLIII Quality Flag; 0 = Less than detection limit; 1= Good data; 2 = Single measurement - no assessment of measurement variation; 4 = >50 %CV (ecotypes qPCR)	unitless
HLIV	Prochlorococcus HLIV (HLIV clade) cell concentration based on quantitative PCR	cells ml-1
HLIV_quality	HLIV Quality Flag; 0 = Less than detection limit; 1= Good data; 2 = Single measurement - no assessment of measurement variation; 4 = >50 %CV (ecotypes qPCR)	unitless
eNATL2A	Prochlorococcus eNATL2A (LLI clade) cell concentration based on quantitative PCR	cells ml-1
eNATL2A_quality	eNATL2A Quality Flag; 0 = Less than detection limit; 1 = Good data; 2 = Single measurement - no assessment of measurement variation; 4 = >50 %CV (ecotypes qPCR)	unitless
eMIT9313	Prochlorococcus eMIT9313 (LLIV clade) cell concentration based on quantitative PCR	cells ml-1
eMIT9313_quality	eMIT9313 Quality Flag; 0 = Less than detection limit; 1 = Good data; 2 = Single measurement - no assessment of measurement variation; 4 = >50 %CV (ecotypes qPCR)	unitless

Instruments

Dataset-specific Instrument Name	Influx Cell Sorter (BD Biosciences and Cytopeia)
Generic Instrument Name	Automated Cell Counter
Generic Instrument Description	An instrument that determines the numbers, types or viability of cells present in a sample.

Dataset-specific Instrument Name	LightCycler 480 System (Roche Applied Science, Indianapolis, Indiana)
Generic Instrument Name	dPCR
Generic Instrument Description	Digital Polymerase Chain Reaction (dPCR)

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Deployments

MV1015

Website	https://www.bco-dmo.org/deployment/58647
Platform	R/V Melville
Report	http://cmore.soest.hawaii.edu/cruises/big_rapa/
Start Date	2010-11-18
End Date	2010-12-14
Description	The South East Pacific (SEP) is characterized by very high nutrient concentrations in the waters adjacent to the Chilean coast, but very low nutrient concentrations (oligotrophic) in the mid- South Pacific Subtropical Gyre (SPSG), near Easter Island. The steep gradient in nutrient concentrations across the region affects the level of marine production, the composition of the microbial community, and the operation of major biogeochemical cycles in ways that are not fully understood. Despite the remarkable diversity of trophic conditions, strong gradients and even some unique singularities, the SEP is still the most sparsely sampled oceanic region of the global ocean from hydrodynamic, biological and biogeochemical points of view. The SPSG is also the most oligotrophic of all sub-tropical gyres. Previous expeditions and remote sensing studies have describes the nutrient and chlorophyll field, but there have been few simultaneous measurements of chemical properties with microbial community structure and function. This expedition is designed to investigate the impact of elemental nutrient (nitrogen, phosphorus, iron, silicon, carbon) ratios on marine productivity and microbial community composition. We propose to sample along a line extending from the Chilean coast near Arica to Easter Island. We will occupy three major "process" stations for up to five days each; a high productivity, near shore station, a mid-cruise station in the nutrient transition zone, and a low productivity, mid-gyre station near Easter Island. In between these stations, we will briefly sample at additional "survey" stations at lower intensity along the cruise track. Cruise information and original data are available from the NSF R2R data catalog. BiG RAPA Home project Web site with additional information

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Project Information

Center for Microbial Oceanography: Research and Education (C-MORE)

Website: http://cmore.soest.hawaii.edu/

Coverage: North Pacific Subtropical Gyre (large region around 22 45 N, 158 W)

Project summary

The **Center for Microbial Oceanography: Research and Education** (C-MORE) is a recently established (August 2006; NSF award: EF-0424599) NSF-sponsored Science and Technology Center designed to facilitate a more comprehensive understanding of the diverse assemblages of microorganisms in the sea, ranging from the genetic basis of marine microbial biogeochemistry including the metabolic regulation and environmental controls of gene expression, to the processes that underpin the fluxes of carbon, related bioelements and energy in the marine environment. Stated holistically, C-MORE's primary mission is: *Linking Genomes to Biomes*.

We believe that the time is right to address several major, long-standing questions in microbial oceanography. Recent advances in the application of molecular techniques have provided an unprecedented view of the structure, diversity and possible function of sea microbes. By combining these and other novel approaches with more well-established techniques in microbiology, oceanography and ecology, it may be possible to develop a meaningful predictive understanding of the ocean with respect to energy transduction, carbon sequestration, bioelement cycling and the probable response of marine ecosystems to global environmental variability and climate change. The strength of C-MORE resides in the synergy created by bringing together experts who traditionally have not worked together and this, in turn, will facilitate the creation and dissemination of new knowledge on the role of marine microbes in global habitability.

The new Center will design and conduct novel research, broker partnerships, increase diversity of human resources, implement education and outreach programs, and utilize comprehensive information about microbial life in the sea. The Center will bring together teams of scientists, educators and community members who otherwise do not have an opportunity to communicate, collaborate or design creative solutions to long-term ecosystem scale problems. The Center's research will be organized around four interconnected themes:

- (Theme I) microbial biodiversity,
- (Theme II) metabolism and C-N-P-energy flow,
- (Theme III) remote and continuous sensing and links to climate variability, and
- (Theme IV) ecosystem modeling, simulation and prediction.

Each theme will have a leader to help coordinate the research programs and to facilitate interactions among the other related themes. The education programs will focus on pre-college curriculum enhancements, in service teacher training and formal undergraduate/graduate and post-doctoral programs to prepare the next generation of microbial oceanographers. The Center will establish and maintain creative outreach programs to help diffuse the new knowledge gained into society at large including policymakers. The Center's activities will be dispersed among five partner institutions:

- Massachusetts Institute of Technology,
- Woods Hole Oceanographic Institution,
- Monterey Bay Aquarium Research Institute,
- University of California at Santa Cruz and
- Oregon State University

and will be coordinated at the University of Hawaii at Manoa.

Related Files:

Strategic plan (PDF file)

Funding

Funding Source	Award
NSF Division of Biological Infrastructure (NSF DBI)	DBI-0424599