

# Counts of Prochlorococcus from on-deck incubations with <sup>13</sup>C-bicarbonate as part of DNA-SIP experiments conducted on Hawaii Ocean Time-series (HOT) cruises, HOT283 and HOT288 in 2016

**Website:** <https://www.bco-dmo.org/dataset/700773>

**Data Type:** Cruise Results, experimental

**Version:** 1

**Version Date:** 2017-05-23

## Project

» [Microbial ecology of coexisting ecotypes: Are all Prochlorococcus equal?](#) (ProEco)

## Program

» [Ocean Time-series Sites](#) (Ocean Time-series)

Contributors	Affiliation	Role
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## Abstract

Counts of Prochlorococcus from on-deck incubations with <sup>13</sup>C-bicarbonate as part of DNA-SIP experiments conducted on Hawaii Ocean Time-series (HOT) cruises, HOT283 and HOT288 in 2016.

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## Coverage

**Temporal Extent:** 2016-04-13 - 2016-11-29

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## Dataset Description

Counts of Prochlorococcus from on-deck incubations with <sup>13</sup>C-bicarbonate as part of DNA-SIP experiments; conducted on Hawaii Ocean Time-series (HOT) cruises 283 and 288.

## Methods & Sampling

Water was sampled from the CTD under trace metal clean conditions into 2 L polycarbonate bottles (acid-washed) and amended with <sup>13</sup>C-bicarbonate. Flow cytometry samples were collected from triplicate bottles at several time points between 0 hours (unlabeled control) and 36 hours after the initiation of incubation. Flow cytometry samples were collected in 1 mL volumes and fixed immediately with 25% TEM-grade glutaraldehyde to a final concentrations of 0.125%. Samples were inverted 10 times to mix, incubated at room temperature in

the dark for 10 minutes, then flash frozen in liquid nitrogen to archive. Samples were stored at -80C until analysis. For analysis, each sample was thawed at room temperature then analyzed by flow cytometry.

Cell counts were determined by flow cytometry using a BD Biosciences Influx high speed cell sorter. A 488 nm laser was used in addition to chlorophyll and phycoerythrin filters, forward scatter relative to 1 um beads, and side scattered light.

## Data Processing Description

The program BD Software was used to collect flow cytometry data and generate distinct files for analysis. The program Flow Jo (version 7.6.5) was used to process flow cytometry files.

BCO-DMO Data Processing:

- modified parameter names to conform with BCO-DMO naming conventions;
- replaced spaces with underscores and removed parentheses (in depth column).

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## Data Files

File
<b>pro_cell_counts.csv</b> (Comma Separated Values (.csv), 1.21 KB) MD5:2d8640b39ffa90e326f95b031f208c70
Primary data file for dataset ID 700773

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## Parameters

Parameter	Description	Units
cruise	Cruise name/identifier	unitless
incubation	Incubation identifier	unitless
depth	Sample depth; DCM = deep chlorophyll maxium	unitless
timepoint	Time point in the incubation	hours
replicate	Replicate identifier	unitless
prochlorococcus	Count of Prochlorococcus cells	cells per milliliter (cells/mL)

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## Instruments

<b>Dataset-specific Instrument Name</b>	CTD
<b>Generic Instrument Name</b>	CTD - profiler
<b>Dataset-specific Description</b>	Water was sampled from the CTD under trace metal clean conditions into 2L polycarbonate bottles (acid-washed) and amended with 13C-bicarbonate.
<b>Generic Instrument Description</b>	The Conductivity, Temperature, Depth (CTD) unit is an integrated instrument package designed to measure the conductivity, temperature, and pressure (depth) of the water column. The instrument is lowered via cable through the water column. It permits scientists to observe the physical properties in real-time via a conducting cable, which is typically connected to a CTD to a deck unit and computer on a ship. The CTD is often configured with additional optional sensors including fluorometers, transmissometers and/or radiometers. It is often combined with a Rosette of water sampling bottles (e.g. Niskin, GO-FLO) for collecting discrete water samples during the cast. This term applies to profiling CTDs. For fixed CTDs, see <a href="https://www.bco-dmo.org/instrument/869934">https://www.bco-dmo.org/instrument/869934</a> .

<b>Dataset-specific Instrument Name</b>	BD Biosciences Influx high speed cell sorter
<b>Generic Instrument Name</b>	Flow Cytometer
<b>Dataset-specific Description</b>	Cell counts were determined by flow cytometry using a BD Biosciences Influx high speed cell sorter.
<b>Generic Instrument Description</b>	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> )

<b>Dataset-specific Instrument Name</b>	
<b>Generic Instrument Name</b>	Trace Metal Bottle
<b>Dataset-specific Description</b>	Water was sampled from the CTD under trace metal clean conditions into 2L polycarbonate bottles (acid-washed) and amended with 13C-bicarbonate.
<b>Generic Instrument Description</b>	Trace metal (TM) clean rosette bottle used for collecting trace metal clean seawater samples.

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## Deployments

SKQ201615S

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/700776">https://www.bco-dmo.org/deployment/700776</a>
<b>Platform</b>	R/V Sikuliaq
<b>Report</b>	<a href="http://dmoserv3.whoi.edu/data_docs/ProEco/cs288.pdf">http://dmoserv3.whoi.edu/data_docs/ProEco/cs288.pdf</a>
<b>Start Date</b>	2016-11-25
<b>End Date</b>	2016-11-29

## KOK1605

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/700778">https://www.bco-dmo.org/deployment/700778</a>
<b>Platform</b>	R/V Ka`imikai-O-Kanaloa
<b>Report</b>	<a href="http://dmoserv3.whoi.edu/data_docs/ProEco/cs283.pdf">http://dmoserv3.whoi.edu/data_docs/ProEco/cs283.pdf</a>
<b>Start Date</b>	2016-04-13
<b>End Date</b>	2016-04-17
<b>Description</b>	Note the cruise report identifies this cruise as KOK16-04. KOK16-04 was the initial cruise ID but it was changed to KOK16-05 after completion of the cruise due to changes in the ship's schedule.

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## Project Information

### Microbial ecology of coexisting ecotypes: Are all Prochlorococcus equal? (ProEco)

**Coverage:** North Pacific Ocean, Station ALOHA

#### *Description from the NSF award abstract:*

Prochlorococcus is a photosynthetic organism that is tremendously abundant in the ocean and influences biogeochemical cycles on global scales. This project aims to link Prochlorococcus community structure to primary productivity in situ. The twelve known Prochlorococcus ecotypes exhibit extensive diversity. It is thought that this diversity allows the Prochlorococcus "collective" to maintain numerical dominance across gradients in light, nutrients, and temperature that accompany changes in depth, season, and latitude. A large gap in our understanding lies in whether we should assess the ecosystem value of Prochlorococcus by its abundance or by its community structure or both. Ecosystem models assign all ecotypes the same role. However, genomic and physiological evidence from cultivated isolates and wild populations suggests tentatively that distinct genotypes may contribute differently to the ecosystem through variation in light and nutrient physiologies and interactions with other microorganisms. The consequences of these molecular-level differences to primary productivity in situ are unknown. This project tests whether absolute abundance, or community structure, determines the contributions of Prochlorococcus to biogeochemical dynamics by measuring the contributions of different ecotypes to primary productivity. The results of this project will inform ecosystem models towards better representation of how shifts in climate and Prochlorococcus diversity will affect global nutrient cycles, trophic cascades, and interactions with other bacteria, viruses, and grazers. The insights and approaches delineated by this work will be generally applicable to the ecology of abundant microbial populations in the open ocean such as pigmented and non-pigmented eukaryotes, heterotrophic bacteria, and other cyanobacterial lineages. A basic understanding of differences between coexisting ecotypes will provide inroads into understanding mechanisms of cooperation, competition, and collaboration among ecotypes in all microbial ecosystems. The investigators will build a teaching module to expose high school students to microbial oceanography, big data, and systems biology through virtual ocean exploration. The primary objective will be to impress upon students the importance of an "invisible forest" of microorganisms in the ocean. Students will examine the distribution patterns of abundant microbial groups in the context of oceanographic data from large publically available databases. High school teachers and student interns, a graduate student, the investigators, and an educational specialist will design, implement, and test the module for classrooms nationwide. This effort will follow a successful education model (Systems Education Experience

- SEE) developed previously.

The investigators will address an overarching hypothesis that *Prochlorococcus* ecotypes vary in their contribution to the ecosystem as primary producers. More specifically, the investigators hypothesize that patterns of cell division and carbon fixation vary between coexisting ecotypes, and these differences are a function of genome content, gene expression, environmental conditions, and community composition. The technical approach will involve two field-based experiments will be applied to three different depths, at the oceanographic Station ALOHA, that differ in *Prochlorococcus* community composition. Experiment 1 will examine whether coexisting ecotypes vary in cell division, using 16S rRNA sequencing to quantify ecotype abundance in G1, S, and G2 cells. Experiment 2 will examine how carbon fixation varies between coexisting ecotypes using RNA-stable isotope probing and 16S rRNA sequencing of RNA enriched in <sup>13</sup>C after incubation with <sup>13</sup>C-bicarbonate. These experiments will be performed with *Prochlorococcus* communities under native in situ conditions and shifts in conditions to mimic light and temperature of other depths. In both experiments, the temporal gene expression of a selected set of carbon fixation and cell division genes will be examined to link gene expression patterns to primary productivity. All data will be related to the oceanographic environment including its physical, chemical, and biological features.

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## Program Information

### Ocean Time-series Sites (Ocean Time-series)

**Coverage:** Bermuda, Cariaco Basin, Hawaii

Program description text taken from Chapter 1: Introduction from the **Global Intercomparability in a Changing Ocean: An International Time-Series Methods Workshop** report published following the workshop held November 28-30, 2012 at the Bermuda Institute of Ocean Sciences. The full report is available from the workshop Web site hosted by US OCB: <http://www.who.edu/website/TS-workshop/home>

Decades of research have demonstrated that the ocean varies across a range of time scales, with anthropogenic forcing contributing an added layer of complexity. In a growing effort to distinguish between natural and human-induced earth system variability, sustained ocean time-series measurements have taken on a renewed importance. Shipboard biogeochemical time-series represent one of the most valuable tools scientists have to characterize and quantify ocean carbon fluxes and biogeochemical processes and their links to changing climate (Karl, 2010; Chavez et al., 2011; Church et al., 2013). They provide the oceanographic community with the long, temporally resolved datasets needed to characterize ocean climate, biogeochemistry, and ecosystem change.

The temporal scale of shifts in marine ecosystem variations in response to climate change are on the order of several decades. The long-term, consistent and comprehensive monitoring programs conducted by time-series sites are essential to understand large-scale atmosphere-ocean interactions that occur on interannual to decadal time scales. Ocean time-series represent one of the most valuable tools scientists have to characterize and quantify ocean carbon fluxes and biogeochemical processes and their links to changing climate.

Launched in the late 1980s, the US JGOFS (Joint Global Ocean Flux Study; <http://usjgofs.who.edu>) research program initiated two time-series measurement programs at Hawaii and Bermuda (HOT and BATS, respectively) to measure key oceanographic measurements in oligotrophic waters. Begun in 1995 as part of the US JGOFS Synthesis and Modeling Project, the CARIACO Ocean Time-Series (formerly known as the Carbon Retention In A Colored Ocean) Program has studied the relationship between surface primary production, physical forcing variables like the wind, and the settling flux of particulate carbon in the Cariaco Basin.

The objective of these time-series effort is to provide well-sampled seasonal resolution of biogeochemical variability at a limited number of ocean observatories, provide support and background measurements for process-oriented research, as well as test and validate observations for biogeochemical models. Since their creation, the BATS, CARIACO and HOT time-series site data have been available for use by a large community of researchers.

Data from those three US funded, ship-based, time-series sites can be accessed at each site directly or by selecting the site name from the Projects section below.

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1646709</a>

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