

# Abundance of bacteria viruses and chlorophyll containing cells collected from the R/V Oceanus OC1504A in the Oregon/California Coastal Upwelling Zone, between 34-44N and 120-124W during 2015

Website: <https://www.bco-dmo.org/dataset/652223>

Data Type: Cruise Results

Version: 1

Version Date: 2016-07-20

## Project

» [Linking physiological and molecular aspects of diatom silicification in field populations](#)

(Diatom Silicification)

Contributors	Affiliation	Role
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<a href="#">Brzezinski, Mark</a>	University of California-Santa Barbara (UCSB-LifeSci)	Co-Principal Investigator
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## Abstract

Abundance of bacteria viruses and chlorophyll containing cells collected from the R/V Oceanus OC1504A in the Oregon/California Coastal Upwelling Zone, between 34-44N and 120-124W during 2015

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## Coverage

**Spatial Extent:** N:43.65434 E:-120.81017 S:34.55467 W:-124.48169

**Temporal Extent:** 2015-04-20 - 2015-05-01

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## Dataset Description

Enumeration of Bacteria, viruses, and chlorophyll containing particles using flow cytometry.

**For related datasets, click on the project link at the top of the page.**

## Acquisition Description

### ***Environmental Sample Collection***

1. Transfer 1 ml of whole seawater to a 2 ml cryovial.
2. Add 20 ul of 25% glutaraldehyde for a final concentration of 0.5%.
3. Incubate at 4 degrees celsius for 30 min.
4. Flash freeze in liquid N<sub>2</sub> and store at -80 degrees celsius.

### ***Fluorescent DNA staining (for bacterial and viral abundances)***

1. Thaw samples.
2. To 20 ul of sample, add 980 ul 1X TE buffer with SYBR Gold (see recipe below)

3. Heat to 80 degrees celsius for 10 min in the dark
4. Cool at RT for 5 min
5. Analyze via flow cytometry

### ***Analysis (for bacterial and viral abundances)***

Samples are analyzed on Influx Model 209S Mariner flow cytometer using BD Software (BD Biosciences).

1. An initial Forward Scatter (FSC) vs Side Scatter (SSC) configuration is determined using Molecular Probes Flow Cytometry Sub-micron particles size reference kit (Cat#F13839) consisting of 0.02, 0.1, 0.5, 1.0 and 2.0 um fluorescent beads.
2. A gating hierarchy is established using both beads and previously determined virus and bacteria populations as reference (Sybr Gold Fluorescence versus SSC cytogram).
3. Samples are analyzed using a 488 nm laser for excitation and a minimum trigger threshold is established using 542/15 nm (SYBR Gold) emission.

### ***Analysis (for chlorophyll containing cells)***

Samples are analyzed on a BD Accuri C6. Fixed, frozen samples are thawed and analyzed immediately.

1. An initial Forward Scatter (FSC) vs Side Scatter (SSC) configuration is determined using various sized fluorescent beads as reference points (1.0, 2.0, 3.0, 6.0 and 10 um).
2. Gating is established using both beads and previously determined phytoplankton populations as reference (Chlorophyll Fluorescence versus FSC cytogram).
3. Samples are analyzed using 488nm laser for excitation and the default BD Accuri threshold (80,000 RFU) on FSC is used.

### **TE buffer with SYBR Gold recipe**

#### 1X TE (for 100 mls)

1 ml of 1M Tris, pH 8.0

1 ml of 0.5 mM EDTA

98 mls MQ water

Store 4 degrees celsius

#### 1X TE + SYBR Gold (for 10 mls)

1. Filter 10 mls 1 TE buffer, 0.22 um filter
2. 1:20,000 dilution of SYBR Gold (Molecular Probes) stock (0.5 ul stock to 10 mls TE

buffer)

## Processing Description

### DMO notes:

- Changed parameter names to meet BCO-DMO naming conventions
- Added ISO\_DateTime column
- Removed 4 unnecessary lat/lon columns (2 remain).

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## Parameters

Parameter	Description	Units
cast	cast number	unitless
depth	depth of sample collection	meters
date_local	local date of cast; mm-bbb-yy	unitless
time_local	local time of cast; HH:MM	unitless
time_utc	UTC time of cast; HH:MM	unitless
date_utc	UTC date of cast; mm-bbb-yy	unitless
station	consecutive station number	unitless
lat	latitude	decimal degrees
lon	longitude	decimal degrees
bacteria	bacteria-like particle abundance	bacteria per milliliter
virus	virus-like particle abundance (VLP)	VLP per milliliter
chl_total	chlorophyll containing cells	cells per milliliter
ISO_DateTime_UTC	DateTime (UTC) ISO formatted	unitless
cruise_id	The name of the cruise that collected these data.	unitless

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## Instruments

<b>Dataset-specific Instrument Name</b>	Influx Model 209S Mariner Flow Cytometer
<b>Generic Instrument Name</b>	Flow Cytometer
<b>Dataset-specific Description</b>	Samples were analyzed on flow cytometer using BD Software (BD Biosciences). Bacterial and viral abundances were analyzed on this flow cytometer.
<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>	BD Accuri C6
<b>Generic Instrument Name</b>	Flow Cytometer
<b>Dataset-specific Description</b>	Chlorophyll containing cells analyzed on this flow cytometer.
<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> )

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**Instrument Deployments**

## OC1504A

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/560135">https://www.bco-dmo.org/deployment/560135</a>
<b>Platform</b>	R/V Oceanus
<b>Report</b>	<a href="https://musicc2015.wordpress.com">https://musicc2015.wordpress.com</a>
<b>Start Date</b>	2015-04-19
<b>End Date</b>	2015-05-02
<b>Description</b>	Data for the project "Linking physiological and molecular aspects of diatom silicification in field populations" (PIs Kimberlee Thamatrakoln and Mark Brzezinski) were collected on this cruise.

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## Project Information

## **Linking physiological and molecular aspects of diatom silicification in field populations (Diatom Silicification)**

**Coverage:** Oregon/California Coastal Upwelling Zone, between 34-44N and 120-124W

Description from NSF award abstract: Diatoms, unicellular, eukaryotic photoautotrophs, are among the most ecologically successful and functionally diverse organisms in the ocean. In addition to contributing one-fifth of total global primary productivity, diatoms are also the largest group of silicifying organisms in the ocean. Thus, diatoms form a critical link between the carbon and silicon (Si) cycles. The goal of this project is to understand the molecular regulation of silicification processes in natural diatom populations to better understand the processes controlling diatom productivity in the sea. Through culture studies and two research cruises, this research will couple classical measurements of silicon uptake and silica production with molecular and biochemical analyses of Silicification-Related Gene (SiRG) and protein expression. The proposed cruise track off the West Coast of the US will target gradients in Si and iron (Fe) concentrations with the following goals: 1) Characterize the expression pattern of SiRGs, 2) Correlate SiRG expression patterns to Si concentrations, silicon uptake kinetics, and silica production rates, 3) Develop a method to normalize uptake kinetics and silica production to SiRG expression levels as a more accurate measure of diatom activity and growth, 4) Characterize the diel periodicity of silica production and SiRG expression. It is estimated that diatoms process 240 Teramoles of biogenic silica each year and that each molecule of silicon is cycled through a diatom 39 times before being exported to the deep ocean. Decades of oceanographic and field research have provided detailed insight into the dynamics of silicon uptake and silica production in natural populations, but a molecular understanding of the factors that influence silicification processes is required for further understanding the regulation of silicon and carbon fluxes in the ocean. Characterizing the genetic potential for silicification will provide new information on the factors that regulate the distribution of diatoms and influence in situ rates of silicon uptake and silica production. This research is expected to provide significant information about the molecular regulation of silicification in natural populations and the physiological basis of Si limitation in the sea.

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## **Funding**

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1333929</a>
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1334387</a>

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