Cell size and chemical characteristics of five strains of coccolithophore Emiliania huxleyi (Protist signaling project)

Website: https://www.bco-dmo.org/dataset/684883

Data Type: experimental

Version: 1

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Project

» Environmental stress and signaling based on reactive oxygen species among planktonic protists (Protist signaling)

Contributors	Affiliation	Role
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Abstract

Cell size and chemical characteristics of five strains of coccolithophore Emiliania huxleyi (Protist signaling project)

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Coverage

Spatial Extent: **Lat**:48.5 **Lon**:-122.75

Dataset Description

Cell size and chemical characteristics (H2O2 production, DMSP content) of five strains of coccolithophore *Emiliania huxleyi*, grown in laboratory culture

Acquisition Description

Cultures of Emiliania huxleyi were obtained from the National Center for Marine Algae and Microbiota at Bigelow Laboratories (all CCMP strains), or from Dr. D. Iglesias-Rodriguez at UC Santa Barbara (strain NEZH) and maintained in the Strom laboratory at Shannon Point Marine Center.

Batch cultures were grown in 50-100 ml volumes of seawater (salinity = 30) amended with f/50 nutrients, at a temperature of 15 deg C and an irradiance of 140-300 umol photons m-2 s-1 on a 12L:12D light cycle. Replicate cultures (n=3) were subsampled for chemical measurements at cell densities of 1.4 to 2.7 x 10^5 cells ml-1 (DMSP) and 3.6 to 12.8 x 10^5 cells ml-1 (H2O2). Different chemical and size measurements reported for a given strain were made over the course of several separate experiments.

Dimethylsulfoniopropionate (DMSP) contained within E. huxleyi cells was measured using a Shimadzu GC-14A gas chromatograph and flame photometric detection, following the methods of Wolfe et al. (2002 J. Phycol. 38: 948-960). Cells were captured on 25 mm glass fiber filters (effective pore size 0.7 um) and placed into 3 ml 5N NaOH for hydrolysis. Method was standardized using ultrapure DMSP-Cl (standard range 0.625 to 50 nM; r2 ≥0.998).

Hydrogen peroxide (H2O2) released into the dissolved phase by E. huxleyi was measured using the Amplex Red – horseradish peroxidase method, using a kit from Molecular Probes (now part of Thermo Fisher Scientific) according to kit directions and to Suggett et al. (2008 J Phycol 44: 948-956). Fluorescent reaction product was quantified in a BioTek Synergy M plate reader (565 nm excitation, 585 nm emission). True reagent blanks were obtained by catalase treatment of E. huxleyi culture filtrate (50 U ml-1, 45 min, room temperature) following Shaked et al. (2010 Environ Sci Technol 44: 3238-3244). Method was standardized using ultrapure H2O2 (standard range 0.025 to 0.5 uM; r2 = 0.98)

E. huxleyi cell size was obtained by imaging live cells (n = 23-29) at 1000x magnification on a Leica DM5500 B microscope, and sizing them with associated image analysis software. Calcification (i.e. whether a strain harbored coccoliths) was also confirmed during microscopy. Note that the sample of strain CCMP3266 used for size measurement comprised a mixture of calcifying and non-calcifying cells.

Processing Description

BCO-DMO Data Processing Notes:

-Data compiled into one table from multiple spreadsheets
-Replaced all blank cells with "nd"

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Parameters

Parameter	Description	Units
data_type	Description of the type of data found in the corresponding row.	unitless
CCMP1516	Data for strain CCMP1516	unitless
CCMP3268	Data for strain CCMP3268	unitless
CCMP3266	Data for strain CCMP3266	unitless
CCMP2668	Data for strain CCMP2668	unitless
NEZH	Data for strain NEZH	unitless

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Instruments

Dataset-specific Instrument Name	Shimadzu GC-14A gas chromatograph
Generic Instrument Name	Gas Chromatograph
Dataset-specific Description	Used to measure Dimethylsulfoniopropionate (DMSP)
Generic Instrument Description	Instrument separating gases, volatile substances, or substances dissolved in a volatile solvent by transporting an inert gas through a column packed with a sorbent to a detector for assay. (from SeaDataNet, BODC)

Dataset-specific Instrument Name	Leica DM5500 B microscope
Generic Instrument Name	Microscope-Optical
Dataset-specific Description	Used to determine cell size
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of reflection and absorption of visible light. Includes conventional and inverted instruments. Also called a "light microscope".

Dataset-specific Instrument Name	BioTek Synergy M plate reader
Generic Instrument Name	plate reader
Dataset-specific Description	Used to measure fluorescent reaction
Generic Instrument Description	Plate readers (also known as microplate readers) are laboratory instruments designed to detect biological, chemical or physical events of samples in microtiter plates. They are widely used in research, drug discovery, bioassay validation, quality control and manufacturing processes in the pharmaceutical and biotechnological industry and academic organizations. Sample reactions can be assayed in 6-1536 well format microtiter plates. The most common microplate format used in academic research laboratories or clinical diagnostic laboratories is 96-well (8 by 12 matrix) with a typical reaction volume between 100 and 200 ÂµL per well. Higher density microplates (384- or 1536-well microplates) are typically used for screening applications, when throughput (number of samples per day processed) and assay cost per sample become critical parameters, with a typical assay volume between 5 and 50 ÂµL per well. Common detection modes for microplate assays are absorbance, fluorescence intensity, luminescence, time-resolved fluorescence, and fluorescence polarization. From http://en.wikipedia.org/wiki/Plate_reader, 2014-09-0-23.

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Deployments

Strom_2014	
Website	https://www.bco-dmo.org/deployment/684891
Platform	lab Strom
Start Date	2014-09-01
End Date	2017-08-01
Description	Five strains of coccolithophore Emiliania huxleyi were grown in the lab. Originally collected from the Salish Sea: 48.5, -122.75.

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Project Information

Environmental stress and signaling based on reactive oxygen species among planktonic protists (Protist signaling)

Coverage: Salish Sea: 48.5, -122.75

Description from NSF proposal: This proposal arises from the central premise that the oxidative stress response is an emergent property of phototrophic cellular systems, with implications for nearly every aspect of a phytoplankton cell's life in the upper ocean. Oxidative stress (OS) arises from the uncompensated production of reactive oxygen species (ROS) within a cell, which can occur in response to a myriad of environmental stressors (e.g. nutrient limitation, temperature extremes, toxins, variable light exposure). In addition to the biochemical damage and physiological impairment that OS can cause, the phytoplankton OS response also includes increased net production and extracellular release of ROS, osmolytes, and other compounds that are known or suspected to be potent signals regulating protist behavior. We hypothesize that, through chemical signaling, oxidative stress acts to govern relationships among environmental variability, phytoplankton condition, and protist predation. Our proposed study of these integrated signaling and response processes has three overarching objectives: 1) Create and characterize oxidatively stressed phytoplankton. We will use light stress (variable exposure to visible light and UV) to create oxidatively stressed phytoplankton in the laboratory. Common coastal taxa with contrasting stress responses will be characterized using an array of fluorescent probes, biochemical measurements, and physiological assays. In addition, intracellular production and extracellular release of ROS and the associated chemical signal DMSP will be quantified. Use of Phaeodactylum tricornutum light stress mutants will add an independent means of connecting OS to signal production and predation response. 2) Examine protist predator responses to oxidatively stressed phytoplankton and associated chemical signals. Responses will be investigated by means of manipulation experiments and thorough characterization of associated signal chemistry. Assessment of predator response will be via predation rate measurements and population aggregation/dispersal behaviors in structured columns. 3) Investigate the prevalence of OS, its environmental correlates, and the microzooplankton predation response in the natural waters of a well-characterized local embayment. Application of ROS probes and OS assays to the natural environment and the design of OS manipulation experiments will be informed by the laboratory experiments using local protist species. Our work will help to elucidate some of the multiple ways in which the OS response can affect phytoplankton fitness, contributing information that can be used to characterize the position of key coastal species along an OS response spectrum. Ultimately such information could be used in trait-based conceptual and numerical models in a manner analogous to cell size and other 'master traits'. Our research will also inform the relatively new and exciting field of chemical signaling in planktonic communities, exploring DMSP- and ROS-based signaling between two of the most significant groups in the plankton, the eukaryotic phytoplankton and their protist

predators. Finally, findings will help elucidate the links between environmental stress, phytoplankton response, and predation in planktonic ecosystems. These links relate to central issues in biological oceanography, including the predator-prey interactions that influence bloom demise, and the mechanisms by which protists feed selectively and thereby structure prey communities. The proposed research is a cross-cutting endeavor that unites subjects usually studied in isolation through a novel conceptual framework. Thus the findings have the potential to generate broadly applicable new insights into the ecological and evolutionary regulation of this key trophic link in planktonic food webs.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1434842

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