Carbonate data in experimental treatments of Emiliania huxleyi, 2011-2012 (E Hux Response to pCO2 project)

Website: https://www.bco-dmo.org/dataset/520651
Data Type: experimental
Version: 2014-07-03

Project
» Planktonic interactions in a changing ocean: Biological responses of Emiliania huxleyi to elevated pCO2 and their effects on microzooplankton (E Hux Response to pCO2)

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Dataset Description

Culturing:

Cultures of *E. huxleyi* Strain CCMP2668, were innoculated at low cell density into media prepared from autoclaved filtered seawater with nutrient amendments based on F/2 medium kit from the National Center for Marine Algae and Microbiota, with a 1:25 reduction in nutrient additions. These were allowed to acclimate for approximately five generations, until cell density neared levels likely to significantly change the pH/pCO2. Daily dilutions of cultures with pre-equilibrated media kept cell density low ($<1x10^5$ cells/ml), ensured cells remained in exponential growth phase and prevented excessive drawdown of nutrients and CO_2. Cell density was determined by flow cytometry (Model) and each flask was diluted with media that was continuously sparged with air containing 400, 750 or 1000 ppm CO2. Air mixtures were created using CO2 free air (Powerex air compressor, and Twin Towers CO2 scrubber) and pure CO2 (Airgas) combined using a system of mass flow controllers (Sierra Instruments) and verified using a non-dispersive infrared CO2 sensor (Licor 820). Cultures were maintained in 1 liter polycarbonate flasks at 15^0 C under a 12/12 light dark cycle. Replicates (n=5) were placed in Plexiglas chambers which were supplied with a flow of the appropriate air mixture for each treatment. Preliminary experiments showed that gas exchange across the air/water surface significantly helped to maintain the target pCO2 in cultures without the mechanical disturbance of bubbling. Sedimentation was minimized by gentle mixing of the cultures by rotation of the bottles twice a day, during sampling and dilution. Cell densities ranged between about 30,000 cells/ml after dilutions to 80,000 cells/ml on the following day. The culture volume that was removed was used for analyses, and replaced with pre-equilibrated media. Cultures were maintained in this fashion for about 8 days. Since the first dilution occurred on day 4 after innoculation, this gives a total of 12 to 14 days in culture at experimental conditions. This experiment was carried out twice, in 2011 and 2012.

SEM:

On days 1 and 8, a few small volume of culture from each replicate was dropped onto SEM stubs and allowed to dry. The stubs were sputter coated with Palladium gold for approximately three minutes (2012) or one minute (2011). Five images of each replicate with three or more cells in each image were taken at 5000x magnification (Smith et al. 2012) using an FEI Quanta 450 Scanning Electron Microscope. To compose each image the field was zoomed out to about 200x magnification to reduce bias in finding clusters of three or more cells. When a cluster of three of more cells was found, the magnification was changed to 5000x magnification and the image was focused and captured.

Parameters including cell size, coccolith size, coccoliths per cell, and percentage of malformed coccoliths were measured from the SEM images. Cell size (control: n= 235, moderate: n= 234, high: n= 200) and coccolith size (control: n= 75, moderate: n= 72, high: n= 67) were calculated using the free hand tool in Image J. To determine the number of coccoliths per cell and the percentage of malformed coccoliths, images were loaded into Windows Photo Viewer (control: n= 1453, moderate: n= 1409, high: n= 1215). Coccoliths malfomation was assessed by the scheme of DeBodt et al, 2010.

Carbonate chemsitry: (THIS DATASET)

pH was measured photometrically using 1 cm cuvettes, m-cresol dye and an Agilent 5480 UV-VIS spectrophotometer (+/- 0.02). Alkalinity was measured by gran titration using a Titrando 888, and 0.1 N HCl titrant, in a temperature controlled titration vessel (+/1 5 ueq/kg). Other parameters were calculated with CO2sys. pCO2 conditions were the same in the two experiments with the exception that the moderate concentration had slightly more elevated pCO2 in 2011 (662 ppm compared to 602 ppm). (When day 1 is included for 2012, there is no significant difference; the difference in the Moderate treatment is present when only day 12 and 14 are included.)

Related dataset: Emiliania huxleyi SEM

Data Processing Description

BCO-DMO Processing notes:

- original data submitted in Excel file 'Malformation_Data_Sheet_2011_2012_with pivot.xlsx'
- added conventional header with dataset name, PI name, version date, reference information
- changed parameter names to be BCO-DMO compatible
- changed date format from day-month (12-Mar) and m/dd/yyyy (7/11/2013) to yyyy-mm-dd (2011-03-28)
- corrected years 2013 to 2011

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Data Files

File
carbonate.csv (Comma Separated Values (.csv), 11.40 KB) MD5:58f19d25ddf7a1ed76cd76fd04e81f5c Primary data file for dataset ID 520651

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Parameters

Parameter	Description	Units

year	year	yyyy
date	date	yyyy-mm-dd
month_local	month; local time	mm
day_local	day of month; local time	dd
yrday_local	local day and decimal time as 326.5 for the 326th day of the year or November 22 at 1200 hours (noon)	unitless
days_expt	number of days since inoculation of the culture	days
treatment	treatment: C=control (400 ppm CO2); M=medium (750 ppm); H=high (1000 ppm)	unitless
sample	sample id: C=control (400 ppm CO2); M=medium (750 ppm); H=high (1000 ppm); A-E=replicate bottles of Emiliania huxleyi culture	unitless
pH	pH: the measure of the acidity or basicity of an aqueous solution	unitless
pCO2	partial pressure of carbon dioxide {pCO2} in the water body by computation from pH and alkalinity	ppm
CO2	concentration of total CO2 in seawater	umol/kg
bicarbonate	concentration of bicarbonate ion ([HCO3]-) in seawater	umol/kg
carbonate	concentration of carbonate ion ([CO3]2-) in seawater	umol/kg
Omega_Ca	the saturation state of seawater with respect to calcite (known as OMEGA_ar) is a measure of the thermodynamic potential for calcite to form or to dissolve and is defined as the product of the concentrations of dissolved calcium and carbonate ions in seawater divided by their product at equilibrium.	unitless
Omega_Ar	the saturation state of seawater with respect to aragonite (known as OMEGA_ar) is a measure of the thermodynamic potential for aragonite to form or to dissolve and is defined as the product of the concentrations of dissolved calcium and carbonate ions in seawater divided by their product at equilibrium.	unitless
TALK	total alkalinity per unit mass of the water body.	umol/kg

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Instruments

Dataset-specific Instrument Name	CO2 Analyzer
Generic Instrument Name	CO2 Analyzer
Dataset-specific Description	Licor 820: a non-dispersive infrared CO2 sensor
Generic Instrument Description	Measures atmospheric carbon dioxide (CO2) concentration.

Dataset-specific Instrument Name	SEM
Generic Instrument Name	Electron Microscope
Dataset-specific Description	FEI Quanta 450 Scanning Electron Microscope
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of reflection and absorption of electrons behaving as waves.

Dataset-specific Instrument Name	Flow Cytometer
Generic Instrument Name	Flow Cytometer
Dataset-specific Description	BD FACS Calibur flow cytometer
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. Description from: http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm

Dataset-specific Instrument Name	MFC
Generic Instrument Name	Mass Flow Controller
Dataset-specific Description	Sierra Instruments
Generic Instrument Description	Mass Flow Controller (MFC) - A device used to measure and control the flow of fluids and gases

Dataset-specific Instrument Name	spectrophotometer
Generic Instrument Name	Spectrophotometer
Dataset-specific Description	Agilent 5480 UV-VIS spectrophotometer (+/- 0.02)
Generic Instrument Description	An instrument used to measure the relative absorption of electromagnetic radiation of different wavelengths in the near infra-red, visible and ultraviolet wavebands by samples.

Dataset-specific Instrument Name	Automatic titrator
Generic Instrument Name	Titrator
Dataset-specific Description	Metrohm 888 Titrando
Generic Instrument Description	Titrators are instruments that incrementally add quantified aliquots of a reagent to a sample until the end-point of a chemical reaction is reached.

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Deployments

Lab_Love

Website	https://www.bco-dmo.org/deployment/521422
Platform	WWU
Start Date	2011-03-23
End Date	2012-07-16
Description	lab experiments

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Project Information

Planktonic interactions in a changing ocean: Biological responses of Emiliania huxleyi to elevated pCO2 and their effects on microzooplankton (E Hux Response to pCO2)

Description from NSF award abstract:
The calcifying Haptophyte *Emiliania huxleyi* appears to be acutely sensitive to the rising concentration of ocean pCO2. Documented responses by *E. huxleyi* to elevated pCO2 include modifications to their calcification rate and cell size, malformation of coccoliths, elevated growth rates, increased organic carbon production, lowering of PIC:POC ratios, and elevated production of the active climate gas DMS. Changes in these parameters are mechanisms known to elicit alterations in grazing behavior by microzooplankton, the oceans dominant grazer functional group. The investigators hypothesize that modifications to the physiology and biochemistry of calcifying and non-calcifying Haptophyte *Emiliania huxleyi* in response to elevated pCO2 will precipitate alterations in microzooplankton grazing dynamics. To test this hypothesis, they will conduct controlled laboratory experiments where several strains of *E. huxleyi* are grown at several CO2 concentrations. After careful characterization of the biochemical and physiological responses of the *E. huxleyi* strains to elevated pCO2, they will provide these strains as food to several ecologically-important microzooplankton and document grazing dynamics. *E. huxleyi* is an ideal organism for the study of phytoplankton and microzooplankton responses to rising anthropogenic CO2, the effects of which in the marine environment are called ocean acidification; *E. huxleyi* is biogeochemically important, is well studied, numerous strains are in culture that exhibit variation in the parameters described above, and they are readily fed upon by ecologically important microzooplankton.

The implications of changes in microzooplankton grazing for carbon cycling, specifically CaCO3 export, DMS production, nutrient regeneration in surface waters, and carbon transfer between trophic levels are profound, as this grazing, to a large degree, regulates all these processes. *E. huxleyi* is a model prey organism because it is one of the most biogeochemically influential global phytoplankton. It forms massive seasonal blooms, contributes significantly to marine inorganic and organic carbon cycles, is a large producer of the climatically active gas DMS, and is a source of organic matter for trophic levels both above and below itself. The planned

controlled study will increase our knowledge of the mechanisms that drive patterns of change between trophic levels, thus providing a wider array of tools necessary to understand the complex nature of ocean acidification field studies, where competing variables can confound precise interpretation.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-0961229

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