3A: Removal of organic carbon by natural bacterioplankton communities as a function of pCO2 from laboratory experiments between 2012 and 2016

Website: https://www.bco-dmo.org/dataset/472032 Data Type: experimental Version: 1 Version Date: 2016-12-05

Project

» <u>Will high CO2 conditions affect production, partitioning and fate of organic matter?</u> (OA - Effects of High CO2)

Programs

» <u>Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES): Ocean Acidification</u> (formerly CRI-OA) (SEES-OA)

» Ocean Carbon and Biogeochemistry (OCB)

Contributors	Affiliation	Role
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Abstract

Factors that affect the removal of organic carbon by heterotrophic bacterioplankton can impact the rate and magnitude of organic carbon loss in the ocean through the conversion of a portion of consumed organic carbon to CO2. Through enhanced rates of consumption, surface bacterioplankton communities can also reduce the amount of dissolved organic carbon (DOC) available for export from the surface ocean. The present study investigated the direct effects of elevated pCO2 on bacterioplankton removal of several forms of DOC ranging from glucose to complex phytoplankton exudate and lysate, and naturally occurring DOC. Elevated pCO2 (1000 – 1500 ppm) enhanced both the rate and magnitude of organic carbon removal by bacterioplankton communities compared to low (pre-industrial and ambient) pCO2 (250 – ~400 ppm). The increased removal was largely due to enhanced respiration, rather than enhanced production of bacterioplankton biomass.

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Coverage

Spatial Extent: N:34.407 E:-64.6353 S:-17.45 W:-149.8727

Dataset Description

This dataset includes results of laboratory experiments which measured dissolved organic carbon (DOC) usage by natural bacteria in seawater at different pCO2 levels. Included in this dataset are; bacterial abundance, total organic carbon (TOC), what DOC was added to the experiment, target pCO2 level. The experiments were conducted between 2012 and 2016 during the R/V Kilo Moana cruise KM1416, at the Bermuda Institute for Ocean Sciences (BIOS), and the University of Santa Barbara.

Methods & Sampling

TOC measurements:

The procedures used to set up each experiment (inoculum filtration and dilution with 0.2 um filtrate) removed the majority of particulate organic carbon such that changes in bacterioplankton carbon production and DOC removal were mainly a function of the growth of the inoculum. Ideally, samples collected for organic carbon would be filtered in order to directly assess DOC removal separate from bacterioplankton carbon production over the course of the incubations. However, sample handling during filtration can result in contamination that obscures changes in DOC on the scale of a few micro-molar C. To avoid contamination, seawater samples from the incubation experiments were not filtered. Therefore, measured values of organic carbon include both DOC and bacterioplankton carbon and are considered total organic carbon (TOC).

TOC samples were collected into 60 mL high-density polyethylene bottles (Sargasso Sea and South Pacific Subtropical Gyre) or in combusted 40 mL glass EPA vials with Teflon coated silicone septa (Santa Barbara Channel). All TOC samples were frozen at -20 C until analysis. Samples were analyzed via high temperature combustion method on a modified Shimadzu TOC-V or Shimadzu TOC-L using the standardization and referencing approaches described in Carlson et al. 2010.

Bacterioplankton abundance measurement – Samples for bacterioplankton abundance were analyzed by epifluorescence microscopy with 0, 6-diamidino -2-phenyl dihydrochloride (5ug/mL, DAPI, SIGMA-Aldrich, St. Louis, MO, USA) according to Porter and Feig 1980, or by Flow Cytometry (FCM) on an LSR II with SYBR Green I according to Nelson et al. 2011. See Parsons et al. 2014 and Nelson et al. 2011 regarding sample preparation and instrument settings for epifluorescence microscopy and FCM analyses, respectively. DAPI direct counts and FCM analysis enumerate total prokaryotic abundance. We were not able to differentiate between bacterial and archaeal domains and refer to the combined cell densities as bacterioplankton abundance (Glockner et al. 1999).

Water sources:

Experiment OA11 was conducted on board a research cruise R/V Kilo Moana KM1416. The Sargasso Sea experiments were conducted at the Bermuda Institute for Ocean Sciences (BIOS) with water was collected via the R/V Atlantic Explorer. The Santa Barbara Channel experiments were conducted with water collected near-shore via a pier near the UCSB campus.

Experimental design:

At all three study sites, experiments consisted of 0.2 um-filtered (0.2 um GSWP, Millipore, Billerica, MA) seawater or 0.2 um-filtered phytoplankton exudate that was inoculated with natural bacterial communities. The inoculum of natural bacterial communities consisted of either unfiltered whole seawater (Sargasso Sea and South Pacific Subtropical Gyre experiments) or 1.2 um filtrate (Santa Barbara Channel experiments; 1.2 um RAWP, Millipore, Billerica, MA). Particulate organic carbon concentration in oligotrophic gyres is low (1-3 umol L-1) so to avoid filtration artifacts such as reduced bacterial production (unpublished data) and contamination of DOC due to handling, the inoculum was not pre-filtered for the experiments conducted in oligotrophic waters. Because particulate organic carbon concentration can be much greater in coastal upwelling systems it was necessary to remove large particles and organisms from the inoculum. Inoculum was added at 25 – 30% of final volume, effectively diluting grazer concentrations and grazing pressure. All filters were pre-rinsed with ~2 L of deionized distilled water and sample water prior to use in order to remove organic contaminants from the filters.

Four types of DOC treatments were used and are described in the data as "doc_additions":

1. None: unamended seawater, which provided naturally occurring DOC.

2. CNP: Naturally occurring DOC amended with glucose (~10 uM C) plus NH4 Cl (1uM) and K2HPO4 (0.1uM) (CNP)

3. Species name + " exudate": phytoplankton exudate

4. Species name + " lysate": naturally occurring DOC amended with phytoplankton lysate (~10 uM C L-1; labeled by phytoplankton species used).

The various treatments were generated by inoculating the 0.2 um pre-filtered seawater or exudate with the microbial community; this solution was then divided into two polycarbonate (PC) containers to adjust pCO2. pCO2 levels were adjusted via chemical additions (Sargasso Sea experiment) or by bubbling with CO2-mixed air (Santa Barbara Channel and South Pacific Subtropical Gyre experiments). Adjusted seawater incubations were then transferred into new PC carboys and CNP or lysate was added, if appropriate. A very small volume of lysate (1.2 mL to 11.5 L of experimental volume) or CNP (12 mL to 10 L of experimental water for the Sargasso Sea experiment; 0.28 mL to 10 L of experimental volume for the Santa Barbara Channel experiment) was added to minimize perturbing the carbonate chemistry. All experiments were conducted in duplicate, at in situ temperatures, and in the dark to eliminate photoautotrophic production. All PC bottles had been acid-washed (5 % or 10 % HCL) and rinsed with deionized distilled water and sample water before use.

Data Processing Description

Experiment refers to the experiment name; sites refer to the Sargasso Sea, the Santa Barbara Channel (SBC) and the South Pacific Subtropical Gyre (SPSG); bacterial abundance; standard error and standard deviation. Toc refers to measurements of total organic carbon, for which the units are uM C.

DMO Processing Notes:

* New data version 28 Nov 2016 replaces previous data version from 21 Nov 2013. This version includes more experimental runs. Data parameter names vary between the two data versions. This version also added lat/lon locations for sample sites.

- * New data version 5 Nov 2016 which includes updated data for experiment O15.
- * added a conventional header with dataset name, PI name, version date
- * modified parameter names to conform with BCO-DMO naming conventions
- * Data values of "None" replaced with "nd" meaning no data.
- * Date format changed from mm.dd.yyyy to ISO date format yyyy-mm-dd
- * More exact lat/lon value of 34.4070,-119.8433 for SBC, supplied by Anna James.

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Data Files

File BactDOC.csv(Comma Separated Values (.csv), 32.72 KB) MD5:05cb5324e7e5f7295db371b62a9d51d3

Primary data file for dataset ID 472032

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Parameters

Parameter	Description	Units
experiment	Experiment identifier	unitless
site	Site the water for the experiment came from	unitless
latitude	Latitude where water samples were collected; north is positive.	decimal degrees
longitude	Longitude where water samples were collected; west is negative.	decimal degrees
bottle_number	Bottle identifier	unitless
doc_addition	Dissolved organic carbon additions. See Aquisition Description section for an explaination of values.	unitless
target_pCO2	Target pCO2 level	parts per million (ppm)
time_point	Time point identifier in experiment	unitless
time_days	Elapsed time since start of experiment in days	unitless
date	Date of experiment in format YYYY-MM-DD	unitless
bact_abun_x10e6_avg	Bacterial abundance multiplied by 10^6	cells per milliliter
bact_abun_x10e6_stderr	Standard error of bacterial abundance multiplied by 10^6	cells per milliliter
bact_abun_x10e6_stdev	Standard deviation Bacterial abundance multiplied by 10^6	cells per milliliter
toc_avg	Total organic carbon	micromoles per liter (uM)
toc_stderr	Standard error of total organic carbon	micromoles per liter (uM)
toc_stdev	Standard deviation of total organic carbon	micromoles per liter (uM)

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Instruments

Dataset- specific Instrument Name	Flow Cytometry (FCM)
Generic Instrument Name	Flow Cytometer
Dataset- specific Description	Flow Cytometry (FCM) on an LSR II with SYBR Green I according to Nelson et al. 2011.
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. (from: http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm)

Dataset- specific Instrument Name	Epifluorescence microscopy
Generic Instrument Name	Fluorescence Microscope
Dataset- specific Description	Samples for bacterioplankton abundance were analyzed by epifluorescence microscopy with 0, 6-diamidino -2-phenyl dihydrochloride (5µg mL-1, DAPI, SIGMA-Aldrich, St. Louis, MO, USA) according to Porter and Feig 1980.
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of fluorescence and phosphorescence instead of, or in addition to, reflection and absorption of visible light. Includes conventional and inverted instruments.

Dataset- specific Instrument Name	modified Shimazdu TOC-L
Generic Instrument Name	Shimadzu TOC-L Analyzer
Dataset- specific Description	Samples were analyzed via high-temperature combustion method on a modified Shimadzu TOC-V or Shimadzu TOC-L using the standardization and referencing approaches described in Carlson et al. 2010.
Generic Instrument Description	A Shimadzu TOC-L Analyzer measures DOC by high temperature combustion method. Developed by Shimadzu, the 680 degree C combustion catalytic oxidation method is now used worldwide. One of its most important features is the capacity to efficiently oxidize hard-to- decompose organic compounds, including insoluble and macromolecular organic compounds. The 680 degree C combustion catalytic oxidation method has been adopted for the TOC-L series. <u>http://www.shimadzu.com/an/toc/lab/toc-l2.html</u>

Dataset- specific Instrument Name	modified Shimadzu TOC-L
Generic Instrument Name	Shimadzu TOC-V Analyzer
Dataset- specific Description	Samples were analyzed via high-temperature combustion method on a modified Shimadzu TOC-V or Shimadzu TOC-L using the standardization and referencing approaches described in Carlson et al. 2010.
Generic Instrument Description	A Shimadzu TOC-V Analyzer measures DOC by high temperature combustion method.

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Deployments

KM1416

Website	https://www.bco-dmo.org/deployment/665488
Platform	R/V Kilo Moana
Start Date	2014-07-19
End Date	2014-08-08
Description	Processing Description Water used for OA11 Experiments

lab_UCSB_MSI_Passow

Website	https://www.bco-dmo.org/deployment/58780
Platform	UCSB MSI Passow
Report	http://www.msi.ucsb.edu/people/research-scientists/uta-passow
Start Date	2009-09-01
End Date	2016-01-22
Description	Results form a series of controlled laboratory experiments investigating the effect of altered carbonate system chemistry on the abiotic formation of TEP

lab_UCSB_MSI_Passow

Website	https://www.bco-dmo.org/deployment/58780
Platform	UCSB MSI Passow
Report	http://www.msi.ucsb.edu/people/research-scientists/uta-passow
Start Date	2009-09-01
End Date	2016-01-22
Description	Results form a series of controlled laboratory experiments investigating the effect of altered carbonate system chemistry on the abiotic formation of TEP

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Project Information

Will high CO2 conditions affect production, partitioning and fate of organic matter? (OA - Effects of High CO2)

Website: http://www.msi.ucsb.edu/people/research-scientists/uta-passow

Coverage: Passow Lab, Marine Science Institute, University of California Santa Barbara

From the NSF Award Abstract

Coastal waters are already experiencing episodic exposure to carbonate conditions that were not expected until the end of the century making understanding the response to these episodic events as important as understanding the long-term mean response. Among the most striking examples are those associated with coastal upwelling along the west coast of the US, where the pH of surface waters may drop to 7.6 and pCO2 can reach 1100 uatm. Upwelling systems are responsible for a significant fraction of global carbon export making them prime targets for investigations on how ocean acidification is already affecting the biological pump today. In this study, researchers at the University of California at Santa Barbara will investigate the potential effects of ocean acidification on the strength of the biological pump under the transient increases in CO2 experienced due to upwelling. Increases in CO2 are expected to alter the path and processing of carbon through marine food webs thereby strengthening the biological pump. Increases in inorganic carbon without proportional increases in nutrients result in carbon over-consumption by phytoplankton. How carbon over-consumption affects the strength of the biological pump will depend on the fate of the extra carbon that is either incorporated into phytoplankton cells forming particulate organic matter (POM), or is excreted as dissolved organic matter (DOM). Results from mesocosm experiments demonstrate that the mechanisms controlling the partitioning of fixed carbon between the particulate and dissolved phases, and the processing of those materials, are obscured when both processes operate simultaneously under natural or semi-natural conditions. Here, POM and DOM production and the heterotrophic processing of these materials will be separated experimentally across a range of CO2 concentrations by conducting basic laboratory culture experiments. In this way the mechanisms whereby elevated CO2 alters the flow of carbon along these paths can be elucidated and better understood for use in mechanistic forecasting models.

Broader Impacts- The need to understand the effects of ocean acidification for the future of society is clear. In addition to research education, both formal and informal, will be important for informing the public. Within this project 1-2 graduate students and 2-3 minority students will be recruited as interns from the CAMP program (California Alliance for Minority Participation). Within the 'Ocean to Classrooms' program run by outreach personnel from UCSB's Marine Science Institute an educational unit for K-12 students will be developed. Advice and support is also given to the Education Coordinator of NOAA, Channel Islands National Marine Sanctuary for the development of an education unit on ocean acidification.

PUBLICATIONS PRODUCED AS A RESULT OF THIS RESEARCH

Arnosti C, Grossart H-P, Muehling M, Joint I, Passow U. "Dynamics of extracellular enzyme activities in seawater under changed atmsopheric pCO2: A mesocosm investigation.," Aquatic Microbial Ecology, v.64, 2011, p. 285.

Passow U. "The Abiotic Formation of TEP under Ocean Acidification Scenarios.," Marine Chemistry, v.128-129, 2011, p. 72.

Passow, Uta; Carlson, Craig A.. "The biological pump in a high CO2 world," MARINE ECOLOGY PROGRESS SERIES, v.470, 2012, p. 249-271.

Gaerdes, Astrid; Ramaye, Yannic; Grossart, Hans-Peter; Passow, Uta; Ullrich, Matthias S.. "Effects of Marinobacter adhaerens HP15 on polymer exudation by Thalassiosira weissflogii at different N:P ratios," MARINE ECOLOGY PROGRESS SERIES, v.461, 2012, p. 1-14.

Philip Boyd, Tatiana Rynearson, Evelyn Armstrong, Feixue Fu, Kendra Hayashi, Zhangi Hu, David Hutchins, Raphe Kudela, Elena Litchman, Margaret Mulholland, Uta Passow, Robert Strzepek, Kerry Whittaker, Elizabeth Yu, Mridul Thomas. "Marine Phytoplankton Temperature versus Growth Responses from Polar to Tropical Waters - Outcome of a Scientific Community-Wide Study," PLOS One 8, v.8, 2013, p. e63091.

Arnosti, C., B. M. Fuchs, R. Amann, and U. Passow. "Contrasting extracellular enzyme activities of particleassociated bacteria from distinct provinces of the North Atlantic Ocean," Frontiers in Microbiology, v.3, 2012, p. 1.

Koch, B.P., Kattner, G., Witt, M., Passow, U., 2014. Molecular insights into the microbial formation of marine dissolved organic matter: recalcitrant or labile? Biogeosciences Discuss. 11 (2), 3065-3111.

Taucher, J., Brzezinski, M., Carlson, C., James, A., Jones, J., Passow, U., Riebesell, U., submitted. Effects of warming and elevated pCO2 on carbon uptake and partitioning of the marine diatoms Thalassiosira weissflogii and Dactyliosolen fragilissimus. Limnology and Oceanography

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Program Information

Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES): Ocean Acidification (formerly CRI-OA) (SEES-OA)

Website: <u>https://www.nsf.gov/funding/pgm_summ.jsp?pims_id=503477</u>

Coverage: global

NSF Climate Research Investment (CRI) activities that were initiated in 2010 are now included under Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES). SEES is a portfolio of activities that highlights NSF's unique role in helping society address the challenge(s) of achieving sustainability. Detailed information about the SEES program is available from NSF (<u>https://www.nsf.gov/funding/pgm_summ.jsp?</u> <u>pims_id=504707</u>).

In recognition of the need for basic research concerning the nature, extent and impact of ocean acidification on oceanic environments in the past, present and future, the goal of the SEES: OA program is to understand (a) the chemistry and physical chemistry of ocean acidification; (b) how ocean acidification interacts with processes at the organismal level; and (c) how the earth system history informs our understanding of the effects of ocean acidification on the present day and future ocean.

Solicitations issued under this program:

<u>NSF 10-530</u>, FY 2010-FY2011 <u>NSF 12-500</u>, FY 2012 <u>NSF 12-600</u>, FY 2013 <u>NSF 13-586</u>, FY 2014 NSF 13-586 was the final solicitation that will be released for this program.

PI Meetings:

<u>1st U.S. Ocean Acidification PI Meeting</u>(March 22-24, 2011, Woods Hole, MA) <u>2nd U.S. Ocean Acidification PI Meeting</u>(Sept. 18-20, 2013, Washington, DC) 3rd U.S. Ocean Acidification PI Meeting (June 9-11, 2015, Woods Hole, MA – Tentative)

NSF media releases for the Ocean Acidification Program:

Press Release 10-186 NSF Awards Grants to Study Effects of Ocean Acidification

Discovery Blue Mussels "Hang On" Along Rocky Shores: For How Long?

<u>Discovery nsf.gov - National Science Foundation (NSF) Discoveries - Trouble in Paradise: Ocean Acidification</u> <u>This Way Comes - US National Science Foundation (NSF)</u>

<u>Press Release 12-179 nsf.gov - National Science Foundation (NSF) News - Ocean Acidification: Finding New</u> <u>Answers Through National Science Foundation Research Grants - US National Science Foundation (NSF)</u>

Press Release 13-102 World Oceans Month Brings Mixed News for Oysters

<u>Press Release 13-108 nsf.gov - National Science Foundation (NSF) News - Natural Underwater Springs Show</u> <u>How Coral Reefs Respond to Ocean Acidification - US National Science Foundation (NSF)</u>

<u>Press Release 13-148 Ocean acidification: Making new discoveries through National Science Foundation</u> <u>research grants</u>

<u>Press Release 13-148 - Video nsf.gov - News - Video - NSF Ocean Sciences Division Director David Conover</u> answers questions about ocean acidification. - US National Science Foundation (NSF)

<u>Press Release 14-010 nsf.gov - National Science Foundation (NSF) News - Palau's coral reefs surprisingly</u> resistant to ocean acidification - US National Science Foundation (NSF)

<u>Press Release 14-116 nsf.gov - National Science Foundation (NSF) News - Ocean Acidification: NSF awards</u> \$11.4 million in new grants to study effects on marine ecosystems - US National Science Foundation (NSF)

Ocean Carbon and Biogeochemistry (OCB)

Website: http://us-ocb.org/

Coverage: Global

The Ocean Carbon and Biogeochemistry (OCB) program focuses on the ocean's role as a component of the global Earth system, bringing together research in geochemistry, ocean physics, and ecology that inform on and advance our understanding of ocean biogeochemistry. The overall program goals are to promote, plan, and coordinate collaborative, multidisciplinary research opportunities within the U.S. research community and with international partners. Important OCB-related activities currently include: the Ocean Carbon and Climate Change (OCCC) and the North American Carbon Program (NACP); U.S. contributions to IMBER, SOLAS, CARBOOCEAN; and numerous U.S. single-investigator and medium-size research projects funded by U.S. federal agencies including NASA, NOAA, and NSF.

The scientific mission of OCB is to study the evolving role of the ocean in the global carbon cycle, in the face of environmental variability and change through studies of marine biogeochemical cycles and associated ecosystems.

The overarching OCB science themes include improved understanding and prediction of: 1) oceanic uptake and release of atmospheric CO2 and other greenhouse gases and 2) environmental sensitivities of biogeochemical cycles, marine ecosystems, and interactions between the two.

The OCB Research Priorities (updated January 2012) include: ocean acidification; terrestrial/coastal carbon fluxes and exchanges; climate sensitivities of and change in ecosystem structure and associated impacts on biogeochemical cycles; mesopelagic ecological and biogeochemical interactions; benthic-pelagic feedbacks on biogeochemical cycles; ocean carbon uptake and storage; and expanding low-oxygen conditions in the coastal and open oceans.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	<u>OCE-1041038</u>

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