Hydrolytic enzyme activities during CDOM monoculture experiment with Coscinodiscus

Website: https://www.bco-dmo.org/dataset/748445
Data Type: experimental
Version: 1
Version Date: 2018-10-17

Project
» Collaborative Research: Planktonic Sources of Chromophoric Dissolved Organic Matter in Seawater (PlankDOM)

Contributors	Affiliation	Role
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Abstract
This dataset is from a laboratory experiment. Four phytoplankton cultures and their associated bacterial communities were incubated in replicate roller bottles (1.9 L) over 3-6 weeks under laboratory conditions. Bacterial dynamics in the culture bottles were measured and correlated with geochemical parameters to determine the role of bacterial activities on the formation of CDOM in the cultures (Kinsey et al., 2018, see below). The data include fluorescence and bacterial enzyme activity during CDOM Coscinodiscus monoculture experiments. Growth stages were initial and exponential.

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Coverage

Temporal Extent: 2016-06-16 - 2016-08-27

Dataset Description

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The data include fluorescence and bacterial enzyme activity during CDOM Coscinodiscus monoculture experiments. Growth stages were initial and exponential.

Methods & Sampling

Hydrolytic enzyme activities were determined using L-leucine-4-methylcoumarinyl-7-amide (MCA) hydrochloride, 4-methylumbelliferyl α-D-glucopyranoside, and 4-methylumbelliferone (MUF) β-D-

glucopyranoside (Sigma-Aldrich) as substrate proxies for leucine-aminopeptidase, α-glucosidase, and β-glucosidase activities, respectively. For each bottle and substrate proxy, 196 µL of unfiltered experimental or control water was added in duplicate to a pure-grade black 96-well plate (Brand Life Sciences) containing a single substrate proxy at saturation levels (final concentration 200 µM). Fluorescence (excitation 370 nm, emission 440 nm) was measured in a Tecan Infinite 200 Pro microplate reader immediately following the addition of the substrate and several more times over 7-20 h. The well plates were incubated in the dark at in situ temperature. MUF and MCA standard solutions prepared in seawater were used to determine hydrolysis rates. Killed controls (boiled sample water) and ultrapure water samples showed little change over the incubations.

Data Processing Description

BCO-DMO Processing Notes:
- added conventional header with dataset name, PI name, version date
- modified parameter names to conform with BCO-DMO naming conventions
- converted Excel file tables to a flat file and combined a-glu, b-glu, and leu substrate treatments.

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Data Files

File	
coscinodiscus_enzyme.csv	(Comma Separated Values (.csv), 6.61 KB) MD5:ef6801a2e176a2e2030245bdd79518c7
Primary data file for dataset ID 748445	

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Parameters

Parameter	Description	Units
substrate	substrate for measuring enzyme activity: a-glu = 4-methylumbelliferyl a-D-glucopyranoside; b-glu = 4-methylumbelliferone (MUF) ß-D-glucopyranoside; leu = L-leucine-4-methylcoumarinyl-7-amide	unitless
sample	sample identifier denoted as growth stage (days from start) replicate id	unitless
fluor_t0	fluorescence intensity at time 0	relative fluorescence units
fluor_t1	fluorescence intensity at time 1	relative fluorescence units
fluor_t2	fluorescence intensity at time 2	relative fluorescence units
fluor_t3	fluorescence intensity at time 3	relative fluorescence units
time_t0	time since start of experiment; time point 0	hours
time_t1	time elapsed from start of experiment; time point 1	hours
time_t2	time elapsed from start of experiment; time point 2	hours
time_t3	time elapsed from start of experiment; time point 3	hours
enz_activity_t0	enzymatic activity at time 0	nanoMol/hour
enz_activity_t1	enzymatic activity at time 1	nanoMol/hour
enz_activity_t2	enzymatic activity at time 2	nanoMol/hour
enz_activity_t3	enzymatic activity at time 3	nanoMol/hour
SLOPE	the slope of the graph of fluorescence intensity vs substrate concentration	unitless
RSQR	the square of the correlation coefficient of fluorescence intensity vs substrate concentration	unitless

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Instruments

Dataset-specific Instrument Name	FACSCalibur flow cytometer (Becton-Dickson)
Generic Instrument Name	Flow Cytometer
Dataset-specific Description	Used to make cell counts.
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	Tecan Infinite 200 Pro microplate reader
Generic Instrument Name	plate reader
Dataset-specific Description	Used to measure fluorescence from which hydrolysis rates were calculated.
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 uL 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 µL 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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Project Information

Collaborative Research: Planktonic Sources of Chromophoric Dissolved Organic Matter in Seawater (PlankDOM)

Coverage: Northern Atlantic Ocean, 34.65 N, 69.63 W

NSF abstract:

Chromophoric dissolved organic matter (CDOM) is a small but important fraction of the marine carbon pool that interacts with solar radiation and thus affects many photochemical and biological processes in the ocean. Despite its importance, the chemical basis for the formation of oceanic CDOM remains unclear. CDOM may be formed from two possible sources: 1) heterotrophic bacterial transformations of primary productivity (plankton-derived), or 2) terrestrially-derived. This project will examine the role of phytoplankton as a source of CDOM in the ocean by utilizing a powerful, new technique to measure particulate organic matter absorbance and fluorescence, discrete chemical measurements of probable precursors to planktonic CDOM, and enzymatic assays. Results of this research will provide new insights into the origin and production of planktonic CDOM and its transformation by heterotrophic bacteria. This research on CDOM will be shared broadly through a module at a North Carolina Aquarium, and streaming live feeds of shipboard activities to elementary school classrooms.

Terrestrial and oceanic dissolved organic matter (DOM) differ in their chemical composition. Laboratory and open-ocean observations suggest that bacterial transformation of phytoplankton DOM produces humic-like CDOM signals that are visually similar to those in terrestrial CDOM. However, prior studies of oceanic CDOM using absorbance and fluorescence fit an electronic interaction (EI) model of intramolecular charge transfer (CT) reactions between donor and acceptor molecules common to partially-oxidized terrestrial molecules found in humic substances. This project will test the hypothesis that phytoplankton and bacteria provide a source of donors and acceptors that are microbially-transformed and linked, enabling CT contacts between them and creating oceanic CDOM. To address this, researchers will systematically study phytoplankton growth, including marine snow formation. A new technique for measuring base-extracted POM (BEPOM) absorbance and fluorescence will be used to incorporate planktonic CDOM results into the EI model, and supplemented with measurements of its probable chemical precursors. These experiments will improve understanding of how the

production of CDOM in the ocean is linked to the optics and chemistry of planktonic CDOM formation. Determining the time course and extent of phytoplankton POM and DOM transformation by heterotrophic bacteria during the same phytoplankton growth experiments will provide an in-depth understanding as to how bacterial transformation of marine snow-associated planktonic organic matter drives CDOM production throughout the ocean.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1459406

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