Polysaccharide hydrolysis rates from mesocosm and bulk water incubations from waters taken aboard the R/V Endeavor in the Western North Atlantic during the research cruise EN683 in May and June 2022

Website: https://www.bco-dmo.org/dataset/985777
Data Type: Cruise Results, experimental
Version: 1
Version Date: 2025-10-02

Project
» Substrate structural complexity and abundance control distinct mechanisms of microbially-driven carbon cycling in the ocean (Substrate complexity and microbes)

Contributors	Affiliation	Role
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Abstract
This dataset includes measurements of polysaccharide hydrolase activities in large volume mesocosm and bulk water incubations. Seawater was collected aboard R/V Endeavor during the research cruise EN683 (2022-05-24 to 2022-06-12). The potential of the seawater microbial community to hydrolyze six high-molecular-weight polysaccharides (arabinogalactan, chondroitin sulfate, fucoidan, laminarin, pullulan, and xylan) was investigated in the DCM and bottom water. Waters for bulk incubation were taken at three sites and various depths. Mesocosm incubations were taken at two sites and two depths. This research tested the hypothesis that the mechanism of polysaccharide processing is related to the cost to a cell of producing the enzymes required for its hydrolysis, and the probability that a cell will receive sufficient return on investment for producing the enzymes. Our conceptual model suggests that external (extracellular) hydrolysis is favored when organic matter is abundant, or when enzyme production costs can be shared (e.g., on particles, in biofilms); selfish uptake (hydrolysis without production of low molecular weight products in the environment) would be a better strategy when high molecular weight (HMW) organic matter is scarce, and particularly when the available HMW organic matter is very complex. Seawater was sampled from four depths at our initial station, and the deep chlorophyll maximum and bottom water from two subsequent stations differing in typical extent of primary productivity. We incubated mesocosms of seawater from the deep chlorophyll maximum and from bottom water from two stations differing in typical extent of primary productivity. Organic matter availability was amended by adding high molecular weight organic matter (dissolved and particulate) from diatoms, or by adding the polysaccharide fucoidan, or by adding the polysaccharide arabinogalactan. Amendments were performed in triplicate mesocosms, while one mesocosm was left unamended for a total of ten mesocosms from each depth and station tested. These data help test this model because they provide information about the activities of various external (extracellular) exo-acting and endo-acting hydrolases, all of which are used to hydrolyze high molecular weight organic matter. Activities of these enzymes differed by station as well as by depth.

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Coverage

Location: Western North Atlantic, stations 21, 22, and 23. Water samples were taken at the depth of the deep chlorophyll maximum (determined via CTD; ca 35m and 152m, respectively) and at the bottom, 4092m and 5305 m, respectively.
Spatial Extent: **N**:42.870093 **E**:-53.873585 **S**:33.7441666 **W**:-75.296666
Temporal Extent: 2022-05-24 - 2022-06-09

Methods & Sampling

Seawater was collected aboard R/V Endeavor during the research cruise EN683 (2022-05-24 to 2022-06-12). Water samples were taken at the depth of the deep chlorophyll maximum (determined via CTD; ca 35m and 152m, respectively) and at the bottom, 4092m and 5305 m, respectively. The following stations were sampled in the Western North Atlantic:

- Station 21 at approximately 33 degrees 44 N, 75 degrees 17 W
- Station 22 at approximately 42 degrees 52 N, 53 degrees 52 W
- Station 23 at approximately 34 degrees 20 N, 69 degrees 49 W

Water was collected via Niskin bottles mounted on a rosette, equipped with a CTD.

Seawater was transferred to 20 L carboys that were rinsed three times with water from the sampling depth and then filled with seawater from a single Niskin bottle, using silicone tubing that had been acid washed then rinsed with distilled water prior to use. From each carboy, water was dispensed into smaller glass containers that were cleaned and pre-rinsed three times with water from the carboy prior to dispensing. This water was used to measure cell counts, bacterial productivity, and the activities of polysaccharide hydrolases, peptidases, and glucosidases. A separate glass Duran bottle was filled with seawater from the carboy and sterilized in an autoclave for 20-30 minutes to serve as a killed control for microbial activity measurements.

For mesocosm (large volume) incubation experiments (referred to as “LV” incubations), seawater was transferred to 20 L carboys that were rinsed three times with water from the sampling depth and then filled with seawater from a single Niskin bottle, using silicone tubing that had been acid washed then rinsed with distilled water prior to use. At stations 22 and 23, carboys were filled from bottom water and deep chlorophyll maximum (DCM) water each, according to the CTD. Triplicate 20L carboys (a total of 9 carboys) were amended with high molecular weight material isolated from the diatom *Thalassiosira*, or the polysaccharide fucoidan, or the polysaccharide arabinogalactan; additional unamended single carboys were used for controls. From each carboy, water was dispensed into smaller glass containers that were cleaned and pre-rinsed three times with water from the carboy prior to dispensing. This water was used to measure the activities of peptidases, and glucosidases. A separate glass Duran bottle was filled with seawater from the carboy and sterilized in an autoclave to serve as a killed control for microbial activity measurements. All mesocosms were incubated in the dark at near in-situ temperatures. Mesocosms were sub-sampled for polysaccharide hydrolase activity measurements at the start of incubation (0 days), and then after at approximately 5d, 10d, 15d, and 25d.

Polysaccharide hydrolase activity was measured at each sub-sampling by filling three 50 mL falcon tubes with mesocosm incubated seawater and one 50 mL falcon tube was filled with autoclaved mesocosm incubated seawater to serve as a killed control, for each substrate. Polysaccharide substrate was added at 3.5 µM monomer-equivalent concentrations, except for fucoidan, which was added at 5 µM concentrations (a higher concentration was necessary for sufficient fluorescence signal). Two 50 mL falcon tubes – one with seawater and one with autoclaved seawater – with no added substrate served as blank controls. Incubations were stored in the dark at as close to in situ temperature as possible.

Subsamples of the incubations were collected at time zero, and at a sequence of subsequent time points. At each time point, 2 mL of seawater was collected from the 50 mL falcon tube using a sterile syringe, filtered through a 0.2 µm pore size syringe filter, and stored frozen until processing.

The hydrolysis of high molecular weight substrate to lower molecular weight hydrolysis products was measured using gel permeation chromatography with fluorescence detection, after the method of Arnosti

(1996, 2003). In short, the subsample was injected onto a series of columns consisting of a 21 cm column of G50 and a 19 cm column of G75 Sephadex gel. The fluorescence of the column effluent was measured at excitation and emission wavelengths of 490 and 530 nm, respectively.

Data Processing Description

Hydrolysis rates were calculated from the change in molecular weight distribution of the substrate over time, as described in detail in Arnosti (2003). Scripts to calculate hydrolysis rates are available in the associated Github repository (Hoarfrost, 2017).

BCO-DMO Processing Description

- Imported "FLA_Rates_EN683_LV_and_Bulk.csv" into the BCO-DMO system
- Replaced periods with underscores in parameter names
- Removed units from parameter names
- Replaced "2022-05-22" with "2022-05-24" in the "date" column, upon submitter request
- Added "Z" to the end of the UTC datetime field
- Exported file as "985777_v1_fla_rates_en683_lv_and_bulk.csv"

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Related Publications

Arnosti, C. (1996). A new method for measuring polysaccharide hydrolysis rates in marine environments. Organic Geochemistry, 25(1-2), 105–115. doi:10.1016/s0146-6380(96)00112-x https://doi.org/10.1016/S0146-6380(96)00112-X
Methods

Arnosti, C. (2003). Fluorescent derivatization of polysaccharides and carbohydrate-containing biopolymers for measurement of enzyme activities in complex media. Journal of Chromatography B, 793(1), 181–191. doi:10.1016/s1570-0232(03)00375-1 https://doi.org/10.1016/S1570-0232(03)00375-1
Methods

Hoarfrost, A., Gawarkiewicz, G., & Arnosti, C. (2017, May 15). Ahoarfrost/Shelf1234: Shelf1234 Initial Release. Zenodo. https://doi.org/10.5281/zenodo.580059
Methods

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Related Datasets

IsRelatedTo

Ghobrial, S., Arnosti, C. (2025) **Bacterial Productivity measurement of bulk seawater and mesocosm experiments taken aboard the R/V Endeavor in the Western North Atlantic during the research cruise EN683 in May and June, 2022.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2025-10-02 http://lod.bco-dmo.org/id/dataset/985783 [view at BCO-DMO]
Relationship Description: Includes data from mesocosm experiments performed using seawater samples collected on EN683.

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Parameters

Parameter	Description	Units
deployment	Cruise ID	unitless
Station	Station number 21, 22, or 23	unitless
latitude	Latitude of sampling site, South is negative	Decimal degrees
longitude	Longitude of sampling site, West is negative	Decimal degrees
date	Date of sample collection	unitless
time	Time of sample collection, US Eastern Time (ET)	unitless
ISO_DateTime_UTC	DateTime of sample collection in ISO format in GMT/UTC	unitless
cast_number	Cast number (refers to cast of CTD/Niskin bottles on cruise)	unitless
depth_actual	Actual depth at which water was collected	m
sample_type	Sample from bulk water (bulk) or Large Volume incubation (LV)	unitless
Incubation_Temp	Temperature of incubation. RT = Room Temperature (~20 C)	degrees Celsius
unamended_amended	Whether high molecular weight organic matter was added or not; U = unamended, F = fucoidan, A = arabinogalactan, T = *Thalassiosira* extract; the following number corresponds to amended incubation replicate	unitless
Sub_sample_day	The amount of incubation time that has elapsed at each timepoint in days	Days
substrate	Polysaccharide used for incubation: ara = arabinogalactan, chn = chondroitin sulfate, fuc = fucoidan, lam = laminarin, pul = pullulan, xyl = xylan	unitless
rate_x	The hydrolysis rate for the kill-control	nmol L-1 hr-1
rate_1	The hydrolysis rate for the first replicate	nmol L-1 hr-1
rate_2	The hydrolysis rate for the second replicate	nmol L-1 hr-1

rate_3	The hydrolysis rate for the third replicate	nmol L-1 hr-1
mean_rate	The average hydrolysis rate for all replicates	nmol L-1 hr-1
sd_rate	The standard deviation of the hydrolysis rates for all replicates	nmol L-1 hr-1
kcrate_x	The kill-corrected hydrolysis rate for the kill-control	nmol L-1 hr-1
kcrate_1	The kill-corrected hydrolysis rate for the first replicate	nmol L-1 hr-1
kcrate_2	The kill-corrected hydrolysis rate for the second replicate	nmol L-1 hr-1
kcrate_3	The kill-corrected hydrolysis rate for the third replicate	nmol L-1 hr-1
mean_kcrate	The average kill-corrected hydrolysis rate for all replicates	nmol L-1 hr-1
sd_kcrate	The standard deviation of the kill-corrected hydrolysis rates for all replicates	nmol L-1 hr-1

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Instruments

Dataset-specific Instrument Name	CTD
Generic Instrument Name	CTD Sea-Bird SBE 911plus
Dataset-specific Description	Water was collected via Niskin bottles mounted on a rosette, equipped with a CTD.
Generic Instrument Description	The Sea-Bird SBE 911 plus is a type of CTD instrument package for continuous measurement of conductivity, temperature and pressure. The SBE 911 plus includes the SBE 9plus Underwater Unit and the SBE 11plus Deck Unit (for real-time readout using conductive wire) for deployment from a vessel. The combination of the SBE 9 plus and SBE 11 plus is called a SBE 911 plus. The SBE 9 plus uses Sea-Bird's standard modular temperature and conductivity sensors (SBE 3 plus and SBE 4). The SBE 9 plus CTD can be configured with up to eight auxiliary sensors to measure other parameters including dissolved oxygen, pH, turbidity, fluorescence, light (PAR), light transmission, etc.). more information from Sea-Bird Electronics

Dataset-specific Instrument Name	HPLC system with Hitachi fluorescence detectors (L-7485, L-2485, Chromaster - 5440)
Generic Instrument Name	High-Performance Liquid Chromatograph
Dataset-specific Description	The hydrolysis of high molecular weight substrate to lower molecular weight hydrolysis products was measured using gel permeation chromatography with fluorescence detection, after the method of Arnosti [1996, 2003]. In short, the subsample was injected onto a series of columns consisting of a 21 cm column of G50 and a 19 cm column of G75 Sephadex gel. The fluorescence of the column effluent was measured at excitation and emission wavelengths of 490 and 530 nm, respectively.
Generic Instrument Description	A High-performance liquid chromatograph (HPLC) is a type of liquid chromatography used to separate compounds that are dissolved in solution. HPLC instruments consist of a reservoir of the mobile phase, a pump, an injector, a separation column, and a detector. Compounds are separated by high pressure pumping of the sample mixture onto a column packed with microspheres coated with the stationary phase. The different components in the mixture pass through the column at different rates due to differences in their partitioning behavior between the mobile liquid phase and the stationary phase.

Dataset-specific Instrument Name	
Generic Instrument Name	Incubator
Dataset-specific Description	Polysaccharide hydrolase activity was measured at each sub-sampling by filling three 50 mL falcon tubes with mesocosm incubated seawater and one 50 mL falcon tube was filled with autoclaved mesocosm incubated seawater to serve as a killed control, for each substrate. Polysaccharide substrate was added at 3.5 µM monomer-equivalent concentrations, except for fucoidan, which was added at 5 µM concentrations (a higher concentration was necessary for sufficient fluorescence signal). Two 50 mL falcon tubes – one with seawater and one with autoclaved seawater – with no added substrate served as blank controls. Incubations were stored in the dark at as close to in situ temperature as possible.
Generic Instrument Description	A device in which environmental conditions (light, photoperiod, temperature, humidity, etc.) can be controlled. Note: we have more specific terms for shipboard incubators (https://www.bco-dmo.org/instrument/629001) and in-situ incubators (https://www.bco-dmo.org/instrument/494).

Dataset-specific Instrument Name	Niskin bottles
Generic Instrument Name	Niskin bottle
Dataset-specific Description	Water was collected via Niskin bottles mounted on a rosette, equipped with a CTD.
Generic Instrument Description	A Niskin bottle (a next generation water sampler based on the Nansen bottle) is a cylindrical, non-metallic water collection device with stoppers at both ends. The bottles can be attached individually on a hydrowire or deployed in 12, 24, or 36 bottle Rosette systems mounted on a frame and combined with a CTD. Niskin bottles are used to collect discrete water samples for a range of measurements including pigments, nutrients, plankton, etc.

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Deployments

EN683

Website	https://www.bco-dmo.org/deployment/956118
Platform	R/V Endeavor
Start Date	2022-05-24
End Date	2022-06-12

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Project Information

Substrate structural complexity and abundance control distinct mechanisms of microbially-driven carbon cycling in the ocean (Substrate complexity and microbes)

Coverage: Western North Atlantic

Substrate Structural Complexity and Abundance Control Distinct Mechanisms of Microbially-Driven Carbon Cycling in the Ocean

Almost half of the organic carbon produced in the ocean is processed by bacteria. Bacteria use extracellular (outside the cell) enzymes to break down large organic molecules to small sizes that can be transported into their cells. It has recently been discovered that bacteria use extracellular enzymes in two ways: 'selfish uptake' and 'external hydrolysis'. External hydrolysis releases low molecular weight products to the environment where they can be used by other organisms. 'Selfish uptake' releases little or no products. This research will determine the extent and location of 'selfish uptake' in ocean waters. This process affects the distribution of organic carbon in the ocean, the flow of small organic molecules to feed a wider range of bacteria, and the composition and dynamics of the bacterial community. Recent results show that 'selfish' bacteria are active in deep ocean waters, where they take up complex polysaccharides (sugars) that are not hydrolyzed externally. These results inspired a new model that links 'selfish uptake' and external hydrolysis to the amount and complexity of the organic matter that is used by bacteria. This project will test the model by describing the polysaccharide fraction of marine organic matter, and studying the relationships between organic matter abundance, structural complexity, and extracellular enzyme use. Graduate and undergraduate students will participate in the project as members of the research team in the field and in the laboratory.

This research will test the hypothesis that the mechanism of polysaccharide processing is related to the cost to a cell of producing the enzymes required for its hydrolysis, and the probability that a cell will receive sufficient return on investment for producing the enzymes. The conceptual model that will be tested suggests that external hydrolysis is favored when organic matter is abundant, or when enzyme production costs can be shared (e.g., on particles, in biofilms); selfish uptake would be a better strategy when high molecular weight (HMW) organic matter is scarce, and particularly when the HMW organic matter is very complex. This study will test this model by characterizing the structure of polysaccharide-containing components of dissolved organic matter (DOM) and particulate organic matter (POM) collected from the ocean, by determining the extent of selfish uptake and rates of external hydrolysis of different polysaccharides by natural microbial communities from the surface and the deep ocean, and by incubation experiments that control for the abundance of polysaccharides of different structural complexity. This project will be carried out in collaboration with colleagues at the Max Planck Institute for Marine Microbiology, whose expertise in carbohydrate chemistry and structural analyses, and in advanced microscopy and analysis of complex microbial communities, are central to the project.

This award reflects NSF's statutory mission and has been deemed worthy of support through evaluation using the Foundation's intellectual merit and broader impacts review criteria.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-2022952

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