North Atlantic subseafloor sediment viable microbe numbers/metabolisms from cruise KN223 on R/V Knorr in the North and West Atlantic Ocean in November 2014

Website: https://www.bco-dmo.org/dataset/686389
Data Type: Cruise Results
Version: 1
Version Date: 2017-03-28

Project
» Defining the interplay between oxygen, organic carbon, and metabolism in subseafloor sediment communities (Subseafloor metabolisms)

Program
» Center for Dark Energy Biosphere Investigations (C-DEBI)

Contributors	Affiliation	Role
Boyd, Eric S.	Montana State University	Principal Investigator
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Abstract
North Atlantic subseafloor sediment viable microbe numbers/metabolisms from cruise KN223 on R/V Knorr in the North and West Atlantic Ocean in November 2014.

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Coverage

Spatial Extent: **N**:29.6767 **E**:-50.6203 **S**:14.4 **W**:-58.3283
Temporal Extent: 2014-11-03 - 2014-11-21

Dataset Description

North Atlantic subseafloor sediment viable microbe numbers/metabolisms from cruise KN223 on R/V Knorr in the North and West Atlantic Ocean in November 2014.

Methods & Sampling

All samples used in this work were collected as part of the North Atlantic long-coring expedition in Oct.-Dec. 2014 (R/V Knorr, Cruise KN223); this project focuses on sediments from 4 sites (2, 3, 11, 12) exhibiting variations in the depth to which oxygen penetrates. The sediment subsamples were collected from long piston cores or shorter gravity cores. While oxygen penetrates through the full long core depth at sites 11 and 12, oxygen was consumed in the sediment column at site 3 and especially at site 2. All samples were collected anaerobically in order to perform on-board culture enrichments via the most probable number (MPN) method. Sediments were placed in sterile serum vials, capped with butyl rubber stoppers and flushed with N2 for 2 min and maintained at 4 degrees C for immediate shipboard MPN inoculation work (this dataset). Parallel samples were similarly collected from these and additional core sections and maintained at 4 degrees C for later determination of microbial production rates (see microbial production dataset).

Twenty ml of anaerobic saline media was added to the 30 ml sediment within each serum vial and mixed to create a sediment slurry. MPN assays were initiated on-board and were designed to quantify the abundance of viable microbial cells with specified metabolisms. Hungate tubes with synthetic marine base salts media were amended with various combinations of electron donors (acetate, peptone) and acceptors (oxygen, nitrate, and manganese(IV) oxide):

Electron donor/acceptor	O_2	NO_3^-	Mn^{4+}
acetate	Not tested	200 µM / 58 mM	200 µM / 50 mM
peptone	20 mg L^{-1} / 21%(air)	20 mg L^{-1} / 58 mM	20 mg L^{-1} / 50 mM

Saline basal medium was composed of (g/L): 0.2 NH4Cl, 30 NaCl, 2.8 MgCl2, 0.33 KCl, 0.3 CaCl2, 0.3 KH2PO4, 0.01 NaBr, 0.015 H3BO3, 0.02 SrCl2, 0.02 KI, 0.02 FeCl3, 0.0075 MnSO4, 0.0045 Na2WO4.2H2O, 0.003 NiCl2, 0.02 CoSO4, 0.0015 ZnSO4, 0.002 CuSO4, and 0.0015 Na2MoO4. pH was adjusted to 6.2.

In the case of anaerobic MPN assays, Hungate tubes and their contents were boiled and purged with high purity N2 for 30 min. Aerobic assay tubes were prepared in air and were not purged. MPN assays were inoculated with 1 ml of sediment slurry and were diluted using 10-fold dilutions. MPN assays were incubated for 6-12 months at room temperature and were then assayed for activity/growth. Activity in MPN assays was evaluated by determining the headspace concentration of CO2 using an infrared gas detector. Additionally, colorimetric approaches specific to each anaerobic metabolism were used: the azo dye method (Strickland and Parsons 1968, Bull Fish Res Board Can 167:71) to detect the reduction of nitrate to nitrite and the T(4-CP)P method (Madison et al. 2011, Talanta, 84:374) to quantify production of Mn(II) from Mn(IV). Blank tubes were similarly prepared but not inoculated; these were analyzed to establish background levels of metabolites.

MPN assays were successful for all incubations using oxygen, Mn(IV) or nitrate as electron acceptors. However, the Mn(II) assay suffered from unidentified interferences in many cases. Growth as identified by CO2 accumulation was used where results of the Mn(II) assay were ambiguous. MPN results were converted to estimates of viable cell concentrations as follows. The highest dilution exhibiting evidence of growth was multiplied by the initial 1.6667-fold dilution used to make the sediment slurry. This count was considered the minimum MPN; the next higher dilution was considered the maximum MPN. For example, an assay exhibiting growth at 10^3 dilution but not at 10^4 dilution was considered to have a viable cell concentration of between 1,667-16,667 cells per cm^3 of sediment. For any subsequent calculations, such as cell turnover, the geometric mean of these values was used; e.g., 52,705 cells per cm^3 in this example case. In cases where no growth occurred at the lowest dilution or positive growth occurred at the highest dilution, MPN can only be constrained to be lower or higher than these estimates, respectively, and the MPN column is left blank.

For each electron acceptor/donor combination from each core section, the highest dilution MPN tube exhibiting growth was targeted for further culture transfers and eventual microbial identification/isolation. We have noted where PCR products were successfully obtained from these tubes, providing additional validation of the corresponding MPN result.

Data Processing Description

BCO-DMO Processing:
- modified parameter names to conform with BCO-DMO naming conventions;
- re-formatted date to yyyy-mm-dd;
- replaced missing data with "nd";
- replaced "R/V Knorr" with "RV_Knorr" in data.

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Data Files

File
MPN.csv (Comma Separated Values (.csv), 11.61 KB) MD5:b7c13a22ef2c19a0056e0b4cc0960f84 Primary data file for dataset ID 686389

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Related Datasets

IsRelatedTo

Boyd, E. S., Amenabar, M. J. (2021) **3H-leucine and thymidine incorporation of North Atlantic subseafloor sediments from cruise KN223 on R/V Knorr in the North and West Atlantic Ocean in November 2014.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2017-03-27 doi:10.26008/1912/bco-dmo.685944.1 [view at BCO-DMO]

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Parameters

Parameter	Description	Units
site	Site name	unitless
lat	Site latitude	decimal degrees
lon	Site longitude	decimal degrees
depth_w	Water depth	meters (m)
sed_thickness	Sediment thickness	meters (m)
basement_age	Basement age	millions of years ago (Ma)
date_coring	Date of coring in yyyy-mm-dd format	unitless
cruise_id	Cruise identifier	unitless
platform	Platform (e.g. vessel) name	unitless
core	Core identifier. LC = long core; GC = gravity core.	unitless
core_section	Core section	unitless
depth_mbsf	Sample depth	meters below sea floor (mbsf)
electron_donor	Organic compound added as an electron donor (acetate or peptone)	unitless
electron_acceptor	Inorganic addition serving as an electron acceptor (nitrate, oxygen, or manganese(IV) oxide)	unitless
MPN_min	Minimum MPN; true viable (under these culture conditions) cell concentration is likely to be greater than or equal to this figure.	viable cells per milliliter (cells/mL)
MPN_max	Maxiumum MPN; True viable (under these culture conditions) cell concentration is likely to be less than or equal to this figure.	viable cells per milliliter (cells/mL)
notes	Indicates samples for which no growth was noted at the lowest dilution tested as well as samples for which positive growth was detected at the highest dilution tested.	unitless
PCR_products	PCR products obtained? Yes = highest dilution exhibiting growth yielded PCR products; No = highest dilution exhibiting growth did not yield PCR products; N/A = not applicable because no growth was noted at any dilution and therefore no attempt was made to extract DNA for PCR amplification.	unitless

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Instruments

Dataset-specific Instrument Name	infrared gas detector
Generic Instrument Name	Gas Analyzer
Dataset-specific Description	Activity in MPN assays was evaluated by determining the headspace concentration of CO2 using an infrared gas detector.
Generic Instrument Description	Gas Analyzers - Instruments for determining the qualitative and quantitative composition of gas mixtures.

Dataset-specific Instrument Name	gravity core
Generic Instrument Name	Gravity Corer
Dataset-specific Description	The sediment subsamples were collected from long piston cores or shorter gravity cores.
Generic Instrument Description	The gravity corer allows researchers to sample sediment layers at the bottom of lakes or oceans. The coring device is deployed from the ship and gravity carries it to the seafloor. (http://www.whoi.edu/instruments/viewInstrument.do?id=1079).

Dataset-specific Instrument Name	long piston core
Generic Instrument Name	Piston Corer
Dataset-specific Description	The sediment subsamples were collected from long piston cores or shorter gravity cores.
Generic Instrument Description	The piston corer is a type of bottom sediment sampling device. A long, heavy tube is plunged into the seafloor to extract samples of mud sediment. A piston corer uses a "free fall" of the coring rig to achieve a greater initial force on impact than gravity coring. A sliding piston inside the core barrel reduces inside wall friction with the sediment and helps to evacuate displaced water from the top of the corer. A piston corer is capable of extracting core samples up to 90 feet in length.

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Deployments

KN223

Website	https://www.bco-dmo.org/deployment/567408
Platform	R/V Knorr
Start Date	2014-10-25
End Date	2014-12-02

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Project Information

Defining the interplay between oxygen, organic carbon, and metabolism in subseafloor sediment communities (Subseafloor metabolisms)

Website: http://www.darkenergybiosphere.org/award/defining-the-interplay-between-oxygen-organic-carbon-and-metabolism-in-subseafloor-sediment-communities/

Coverage: SW North Atlantic sediments (13-42° N, 48-71° W)

Abstract from C-DEBI:

Deep marine sediments harbor an abundance of microbial cells that, if active, are likely to exert a strong influence on element biogeochemical cycling. Despite decades of study, our understanding of the fraction of cells that are active in situ and the metabolic processes that sustain them remain under-explored. We propose an integrated set of analyses aimed at unraveling the links between geochemical heterogeneity, cellular viability and synthesis, and metabolism along a vertical depth profile in four sediment cores collected during the North Atlantic long coring expedition. These sediment columns exhibit varying levels of organic carbon and differences in the degree of oxygen penetration along the depth profile which we hypothesize exert strong influence on the extent and nature of microbial life. Most probable number assays containing nine different selective enrichment conditions were initiated using subsamples from these cores in Nov. 2014. Separate subsamples were preserved for use in measuring rates of secondary production. Multivariate modeling tools will be applied to integrate these measurements with co-registered geochemical measurements, cell counts, and molecular data provided by collaborators. This work will provide new insight into the dynamic interplay between O2 and organic carbon and microbial activity, viability, and productivity in deep marine sediments.

NOTE: This project follows the C-DEBI program Data Management Plan (PDF).

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Program Information

Center for Dark Energy Biosphere Investigations (C-DEBI)

Website: http://www.darkenergybiosphere.org

Coverage: Global

The mission of the Center for Dark Energy Biosphere Investigations (C-DEBI) is to explore life beneath the seafloor and make transformative discoveries that advance science, benefit society, and inspire people of all ages and origins.

C-DEBI provides a framework for a large, multi-disciplinary group of scientists to pursue fundamental questions about life deep in the sub-surface environment of Earth. The fundamental science questions of C-DEBI involve exploration and discovery, uncovering the processes that constrain the sub-surface biosphere below the oceans, and implications to the Earth system. What type of life exists in this deep biosphere, how much, and how is it distributed and dispersed? What are the physical-chemical conditions that promote or limit life? What are the important oxidation-reduction processes and are they unique or important to humankind? How does this biosphere influence global energy and material cycles, particularly the carbon cycle? Finally, can we discern how such life evolved in geological settings beneath the ocean floor, and how this might relate to ideas about the origin of life on our planet?

C-DEBI's scientific goals are pursued with a combination of approaches:
(1) coordinate, integrate, support, and extend the research associated with four major programs—Juan de Fuca Ridge flank (JdF), South Pacific Gyre (SPG), North Pond (NP), and Dorado Outcrop (DO)—and other field sites;

(2) make substantial investments of resources to support field, laboratory, analytical, and modeling studies of the deep subseafloor ecosystems;
(3) facilitate and encourage synthesis and thematic understanding of submarine microbiological processes, through funding of scientific and technical activities, coordination and hosting of meetings and workshops, and support of (mostly junior) researchers and graduate students; and
(4) entrain, educate, inspire, and mentor an interdisciplinary community of researchers and educators, with an emphasis on undergraduate and graduate students and early-career scientists.

Note: Katrina Edwards was a former PI of C-DEBI; James Cowen is a former co-PI.

Data Management:
C-DEBI is committed to ensuring all the data generated are publically available and deposited in a data repository for long-term storage as stated in their Data Management Plan (PDF) and in compliance with the NSF Ocean Sciences Sample and Data Policy. The data types and products resulting from C-DEBI-supported research include a wide variety of geophysical, geological, geochemical, and biological information, in addition to education and outreach materials, technical documents, and samples. All data and information generated by C-DEBI-supported research projects are required to be made publically available either following publication of research results or within two (2) years of data generation.

To ensure preservation and dissemination of the diverse data-types generated, C-DEBI researchers are working with BCO-DMO Data Managers make data publicly available online. The partnership with BCO-DMO helps ensure that the C-DEBI data are discoverable and available for reuse. Some C-DEBI data is better served by specialized repositories (NCBI's GenBank for sequence data, for example) and, in those cases, BCO-DMO provides dataset documentation (metadata) that includes links to those external repositories.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-0939564

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