Chronoamperometry data from cathodic poised potential experiments with subsurface crustal samples from CORK borehole observatories at North Pond on the Mid-Atlantic Ridge during R/V Atlantis cruise AT39-01

Website: https://www.bco-dmo.org/dataset/780127
Data Type: Cruise Results, experimental
Version: 1
Version Date: 2019-10-30

Project
» Collaborative Research: Completing North Pond Borehole Experiments to Elucidate the Hydrology of Young, Slow-Spread Crust (North Pond 2017)

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

Contributors	Affiliation	Role
Orcutt, Beth N.	Bigelow Laboratory for Ocean Sciences	Principal Investigator
York, Amber D.	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
Chronoamperometry data from cathodic poised potential experiments with subsurface crustal samples from CORK borehole observatories at North Pond on the Mid-Atlantic Ridge during R/V Atlantis cruise AT39-01.

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Coverage

Spatial Extent: **N**:22.8023 **E**:-46.053 **S**:22.7564 **W**:-46.0817
Temporal Extent: 2018-01-03 - 2019-01-10

Dataset Description

Chronoamperometry data from cathodic poised potential experiments with subsurface crustal samples from CORK borehole observatories at North Pond on the Mid-Atlantic Ridge during R/V Atlantis cruise AT39-01.

These results were published in Jones et al. (2020).

Related datasets from the same experiment:
AT39-01 CathodicEET CyclicVoltametry: https://www.bco-dmo.org/dataset/780248
AT39-01 CathodicEET Amplicons: https://www.bco-dmo.org/dataset/780255
AT39-01 CathodicEET SEM: https://www.bco-dmo.org/dataset/780261
AT39-01 CathodicEET Experimental Metadata:
https://www.bco-dmo.org/dataset/780225

Methods & Sampling

Samples for this study come from CORK observatories installed at IODP Holes U1382A and U1383C as described elsewhere (Edwards et al., 2012b). In brief, instrument strings containing OsmoSampler systems (Wheat et al., 2011) were deployed at different depths within the holes from 2011-2017. Additional OsmoSampler systems were deployed at the wellhead of the

CORKs in 2014 and were inoculated with bottom seawater, making them useful for identifying differences between crustal subsurface and bottom seawater inoculated microbial communities. OsmoSampler systems included Flow-through Osmo Colonization Systems (FLOCS) for mineral colonization experiments, as described elsewhere (Ramrez et al., 2019). Each FLOCS contained sterile (autoclaved and ethanol-rinsed) substrates including crushed basalts, pyrite, pyrrhotite, or inert glass beads housed within polycarbonate cassettes and sleeves. Fluids were introduced into the FLOCS via the OsmoSampler pumps, allowing fluid microbial communities to colonize the substrates.

All FLOCS were recovered in October 2017 during cruise AT39-01 of the RV Atlantis with the ROV Jason (Woods Hole Oceanographic Institution) following methods described elsewhere (Ramrez et al., 2019). In brief, the instrument strings with the downhole FLOCS were pulled up to the ship on a wire, then immediately disassembled to store the FLOCS in a cold room. Wellhead FLOCS incubated in milkcrates attached to crustal fluid umbilicals at the seafloor were also collected on this cruise with the ROV. FLOCS contents were distributed inside an ethanol-rinsed and UV-irradiated HEPA-filtered hood using sterile instruments. Substrates for the experiment were stored cold (2-4C) and in the dark in sterile centrifuge tubes with ultrafiltered (0.22 m-mesh Millipore Sterivex and 0.02 m-mesh Whatman Anotop 25 filters) crustal fluid until incubation at the shore-based laboratory. Parallel samples for initial characterization of the substrate biofilms were transferred to sterile cyrovials, flash frozen with liquid nitrogen, and stored at -80C until analysis. In addition to the FLOCS samples, 4-10L samples of raw and unfiltered crustal fluids were collected into ethanol-rinsed cubitainers after collection into gamma-irradiated bags on the seafloor using a Mobile Pumping System. Cubitainers were stored cold (2-4C) and in the dark for approximately one year before beginning the experiments.

A detailed description of the cathodic poised potential protocol used is available elsewhere (Jones and Orcutt, 2019). In brief, glass two-cell, three-electrode MFC systems (Adams and Chittenden, CA, USA) were used as incubations chambers, run in parallel with a multichannel potentiostat (model CHI1030C, CHI Instruments, TX, USA). Nafion 117 proton exchange membrane (Fuel Cell Store, TX) separated the half-cells. Cells were filled with distilled water and autoclaved prior to filling with media and sample inocula. Ag/AgCl reference electrodes (Gamry Instruments, PA, USA part 932-00018 and/or Analytical Instrument Systems, NY, USA) were calibrated before each run by immersion in 3 M NaCl and compared against a known electrode kept only for that purpose (max +/- 20 mV drift from the value of lab master electrode). Working electrodes (WEs) were 2 4 cm2 Indium Tin Oxide (ITO) coated glass slides (Delta Technologies Ltd, CO, USA part CB-50IN-1111), constructed for MFC as described elsewhere (Rowe et al., 2015). Counter electrodes (CEs) were carbon cloth with a 4 4 cm2 surface area. Electrodes were sterilized by rinsing with 80% ethanol, air-drying, then exposing to UV radiation for 15 minutes per side.

Each experiment consisted of five treatments: Three MFCs filled with buffered and double autoclaved crustal fluid (cool and oxic), inoculated with samples, and incubated with a WE set poised at -200 mV versus a standard hydrogen electrode (SHE) referred to as the Echem treatments; one MFC filled with the same fluid and WE but without any sample inoculum referred to as the Fluid treatment; and one glass bottle with a mixed sample inoculum and ITO electrode without any voltage applied referred to as the Offline treatment. Samples were transferred into the MFCs in HEPA-filtered, UV-irradiated biosafety cabinet or hood using sterile tools. An aliquot of each sample was also collected into a sterile plastic tube for microbial community analysis representing a time zero (T0) condition that may have changed from the Shipboard (SB) initial condition. For the Echem and Fluid treatments, the counter cell redox couple was H2O/O2. Testing (data not shown) determined that a strong kill control method was necessary to achieve sterility of the crustal fluid, consisting of 1 h autoclaving at 121C, incubation at room temperature in the dark for ~24 h to allow for spore germination, and then another 1 h autoclaving at 121C before cooling down to 4C for the addition of substrate. Sodium bicarbonate was added to the media (0.1 M) as a buffer. MFCs were incubated at 4C in the dark.

Once the MFCs were constructed, a cyclic voltammetry (CV) sweep was performed for each cell with the potentiostat before each poised potential experiment at a scan rate of 0.1 V s-1 and sample interval of 0.001 V, across -1 to 1 V range. Then the chronoamperometry (CA) experiments began by applying a constant voltage (-200 mV versus SHE) to the Echem and Fluid MFCs. Current generation was monitored and recorded until current changes began to decline after a period of approximately 15 days. At the end of each CA experiment, two further CV sweeps were performed: one with the incubated ITO electrode (Tend i) and one with a fresh ITO electrode dipped into the incubated media (Tend ii).

Data Processing Description

BCO-DMO Data Manager Processing Notes:
* added a conventional header with dataset name, PI name, version date
* modified parameter names to conform with BCO-DMO naming conventions
* blank values in this dataset are displayed as "nd" for "no data." nd is the default missing data identifier in the BCO-DMO system.
* Converted Date to ISO 8601 format yyyy-mm-dd
* Concatenated all the submitted csv files together and added column "ExperimentID" which was extracted from the csv file names.
* Added several columns from the CA experiment metadata table (https://www.bco-dmo.org/dataset/780225) to this dataset: FluidSource,FluidcollectionCruise,FluidcollectionDate,Latitude,Longitude,WaterDepth

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Data Files

File
ca.csv(Comma Separated Values (.csv), 16.92 MB) MD5:1aaae488b53d6287eba1ea2e610552df Primary data file for dataset ID 780127

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Related Publications

Edwards, K. J., Wheat, C. G., Orcutt, B. N., Hulme, S., Becker, K., Jannasch, H., ... Klaus, A. (2012). Design and deployment of borehole observatories and experiments during IODP Expedition 336, Mid-Atlantic Ridge flank at North Pond. Proceedings of the IODP. doi:10.2204/iodp.proc.336.109.2012
Methods

Jones, R. M., D'Angelo, T., & Orcutt, B. N. (2020). Using Cathodic Poised Potential Experiments to Investigate Extracellular Electron Transport in the Crustal Deep Biosphere of North Pond, Mid-Atlantic Ridge. Frontiers in Environmental Science, 8. doi:10.3389/fenvs.2020.00011
Results

Jones, R., & Orcutt, B. (2019). Bioelectrochemistry protocol For CHI Potentiostat v1 (protocols.io.xihfkb6). Protocols.io. doi:10.17504/protocols.io.xihfkb6
Methods

Ramírez, G. A., Garber, A. I., Lecoeuvre, A., D'Angelo, T., Wheat, C. G., & Orcutt, B. N. (2019). Ecology of Subseafloor Crustal Biofilms. Frontiers in Microbiology, 10. doi:10.3389/fmicb.2019.01983
Methods

Wheat, C. G., Jannasch, H. W., Kastner, M., Hulme, S., Cowen, J., Edwards, K. J., ... Glazer, B. (2011). Fluid sampling from oceanic borehole observatories: design and methods for CORK activities (19902010). Proceedings of the IODP. doi:10.2204/iodp.proc.327.109.2011
Methods

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Parameters

Parameter	Description	Units
Time_s	time in seconds (s) from start of experiment	seconds (s)
Time_min	time in minutes (min) from start of experiment	minutes (min)
Time_h	time in hours (h) from start of experiment	hours (h)
Time_d	time in days (d) from start of experiment	days (d)
CH1_A	current of the Channel 1 electrode at each time point	amps (A)
CH2_A	current of the Channel 2 electrode at each time point	amps (A)
CH3_A	current of the Channel 3 electrode at each time point	amps (A)
CH4_A	current of the Channel 4 electrode at each time point	amps (A)
CH1_uA_per_cm2	current density for the Channel 1 electrode at each time point	microamps per square centimeter (uA cm-2)

CH2_uA_per_cm2	current density for the Channel 2 electrode at each time point	microamps per square centimeter (uA cm-2)
CH3_uA_per_cm2	current density for the Channel 3 electrode at each time point	microamps per square centimeter (uA cm-2)
CH4_uA_per_cm2	current density for the Channel 4 electrode at each time point	microamps per square centimeter (uA cm-2)
ExperimentID	text description of experiment with microbial fuel cells; values of NP11, NP12, NP13, NP14 or NP15	unitless
FluidSource	name of the IODP borehole location where crustal fluid was collected for preparing microbial fuel cell media; values of U1382A (from IODP Hole U1382A shallow horizon), U1383Cdeep (from IODP Hole U1383C deep horizon), or U1383Cshallow (from IODP Hole U1383C shallow horizon)	unitless
FluidcollectionCruise	name of the cruise and ROV Jason dive number when the crustal fluid was collected; values of AT39-01 (R/V Atlantis cruise AT39-01) + J2-#### (ROV Jason dive number)	unitless
FluidcollectionDate	date of the collection of the crustal fluids, in ISO 8601 format yyyy-mm-dd	unitless
Latitude	Latitude of the FLOCS experiment	decimal degrees
Longitude	Longitude of the FLOCS experiment	decimal degrees
WaterDepth	water depth to seafloor of the IODP Hole	meters (m)

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Instruments

Dataset-specific Instrument Name	multichannel potentiostat (model CHI1030C, CHI Instruments, TX, USA)
Generic Instrument Name	Voltammetry Analyzers
Generic Instrument Description	Instruments that obtain information about an analyte by applying a potential and measuring the current produced in the analyte.

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Deployments

AT39-01

Website	https://www.bco-dmo.org/deployment/723337
Platform	R/V Atlantis
Report	http://datadocs.bco-dmo.org/docs/Subseafloor_Microbial_Carbon_Cycling/data_docs/North_Pond_2017_Expedition%20Report_FINAL.pdf
Start Date	2017-10-02
End Date	2017-11-02

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Project Information

Collaborative Research: Completing North Pond Borehole Experiments to Elucidate the Hydrology of Young, Slow-Spread Crust (North Pond 2017)

Website: http://www.darkenergybiosphere.org/research-activities/field-sites/

Coverage: North Pond, Mid-Atlantic Ridge flank CORKs

NSF Award Abstract:
Seawater circulates through the upper part of the oceanic crust much like groundwater flows through continental aquifers. However, in the ocean this seawater circulation, many times heated by buried magmatic bodies, transports and releases 25% of the Earth's heat. The rate of fluid flow through ocean crust is estimated to be equal to the amount of water delivered by rivers to the ocean. Much of what we know of this subseafloor fluid flow comes from studies in the eastern Pacific Ocean on ocean crust created by medium and fast spreading mid-ocean ridges. These studies indicate that seawater and its circulation through the seafloor significantly impact crustal evolution and biogeochemical cycles in the ocean and affect the biosphere in ways that are just now beginning to be quantified and understood. To expand this understanding, this research focuses on fluid flow of seafloor generated by slow spreading ridges, like those in the Atlantic, Indian and Arctic Oceans because it is significantly different in structure, mineralogy, and morphology than that formed at fast and intermediate spreading ridges. This research returns to North Pond, a long-term; seafloor; fluid flow monitoring site, drilled and instumented by the Ocean Drilling Program in the Atlantic Ocean. This research site was punctured by boreholes in which fluid flow and geochemical and biological samplers have been deployed for a number of years to collect data and samples. It also provides resources for shipboard and on-shore geochemical and biological analysis. Broader impacts of the work include sensor and technology development, which increases infrastructure for science and has commercial applications. It also provides training for students and the integration of education and research at three US academic institutions, one of which is an EPSCoR state (Mississippi), and supports a PI whose gender is under-represented in sciences and engineering. Public outreach will be carried out in conjunction with the Center for Dark Energy Biosphere Investigations.

This project completes a long-term biogeochemical and hydrologic study of ridge flank hydrothermal processes on slow-spreading, 8 million year old crust on the western flank of the Mid-Atlantic Ridge. The site, North Pond, is an isolated northeast-trending sediment pond, bounded by undersea mountains that have been studied since the 1970s. During Integrated Ocean Drilling Program Expedition 336 in 2011 and an expedition five months later (2012), sensors, samplers, and experiments were deployed in four borehole observatories drilled into the seafloor that penetrated into volcanic crust, with the purpose of monitoring changes in hydrologic properties, crustal fluid composition and mineral alteration, among other objectives. Wellhead sampling in 2012 and 2014 already revealed changes in crustal fluid compositions; and associated pressure data confirm that the boreholes are sealed and overpressured, reflecting a change in the formation as the boreholes recover from drilling disturbances. This research includes a 13-day oceanographic expedition and use of on-site robotically operated vehicles to recover downhole instrument packages at North Pond. It will allow the sampling of crustal fluids, recovering pressure data, and measuring fluid flow rates. Ship- and shore-based analyses will be used to address fundamental questions related to the hydrogeology of hydrothermal processes on slow-spread crust.

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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-1536539

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