

16S rRNA gene amplicon sequences 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/780255>

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
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Abstract

16S rRNA gene amplicon sequences 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. Illumina amplicon sequence data (V4-V5 region of the 16S rRNA gene) is publicly available through NCBI Sequence Read Archive under BioProject PRJNA564565 samples SAMN12723345-SAMN12723415. BioProject PRJNA564565: <https://www.ncbi.nlm.nih.gov/bioproject/PRJNA564565>

Table of Contents

- [Coverage](#)
- [Dataset Description](#)
 - [Methods & Sampling](#)
- [Supplemental Files](#)
- [Related Publications](#)
- [Parameters](#)
- [Instruments](#)
- [Deployments](#)
- [Project Information](#)
- [Program Information](#)
- [Funding](#)

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

16S rRNA gene amplicon sequences 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.

Illumina amplicon sequence data (V4-V5 region of the 16S rRNA gene) is publicly available through NCBI Sequence Read Archive under BioProject PRJNA564565 samples SAMN12723345-SAMN12723415.

<https://www.ncbi.nlm.nih.gov/bioproject/PRJNA564565>

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

Related datasets from the same experiment:

AT39-01 CathodicEET Chronoamperometry: <https://www.bco-dmo.org/dataset/780127>

AT39-01 CathodicEET CyclicVoltametry: <https://www.bco-dmo.org/dataset/780248>

AT39-01 CathodicEET SEM: <https://www.bco-dmo.org/dataset/780261>

AT39-01 CathodicEET Experimental Metadata:

Methods & Sampling

The FastDNA kit from MP Biomedicals (Irvine, CA, USA) was used for all DNA extractions following manufacturers instructions with the exception of using a Retsch MM 400 shaker (frequency 20 r/s for 5 min) instead of vortexing as described in detail elsewhere (D'Angelo, 2019). From the Echem and Fluid samples, the ITO swab end was cut into a sterile plastic tube and subjected to a freeze-thaw cycle (8 min at -80C then thawing at room temperature for 10 min or until ice barely melted into water) before transferring into a FastDNA kit tube for DNA extraction. For the Shipboard and NP12 samples, 0.232 g/l polyadenine (polyA, final concentration) was added to the sample at the first step to increase DNA yield from low biomass samples. For each batch of samples extracted, a no template control (NTC) was included as a check for contamination. DNA concentrations were measured by Q-bit Fluorometer 3.0 (Thermo Fisher Scientific, Waltham, MA USA) using the Qubit HS dsDNA kit (Thermo Fisher Scientific, Waltham, MA USA) according to manufacturer instructions. Aliquots of DNA extracts were sent to the Integrated Microbiome Resource facility at Dalhousie University (Halifax, Canada) for sequencing of the V4-V5 hypervariable region of the 16S rRNA gene via Illumina Mi-Seq technology using the 515F/926R primer pair. Sequencing was performed on an Illumina MiSeq using 300 300 bp paired-end V3 chemistry. Raw sequenced reads were processed at the same time as sequenced reads from mineral incubation experiments taken from the same environments, as well as additional no-template-control samples (data not shown, but full data available at the NCBI Sequence Read Archive BioProject PRJNA564565 samples SAMN12723399-489), using modifications of the standard DADA2 pipeline Version 1.12 to construct unique amplicon sequence variants (ASVs). This was done to directly link ASVs between the two datasets and to give the DADA2 algorithm the most possible data to use for sequence inference. Thirty base-pairs were trimmed off of the start of each read, forward reads were truncated at 250 base-pairs, and reverse reads were truncated at 200 base-pairs. This allowed for 50 BP of overlap for the forward and reverse reads to be merged after sequence inference. Approximately 100 million bases from ~500K reads were used by DADA2 to infer the base-call error rates in the data. Inference of ASVs, merging of forward and reverse reads, and chimera removal resulted in 2,011 ASVs ranging from 345 360 BP in length. These sequences were assigned taxonomy by the naive Bayesian classifier built into the DADA2 package using the Silva v132 database. Scripts used to process the data are available via github.

[[table of contents](#) | [back to top](#)]

Supplemental Files

File
North Pond FLOCs and Electrochemistry (.RMD file) GitHub release v1.0 filename: northpond-1.0.zip (ZIP Archive (ZIP), 2.39 KB) MD5:4c3211286870be67b0436cec4c25e9e7 North Pond FLOCs and Electrochemistry The .RMD file can be downloaded and ran with the fastq files in NCBI Sequence Read Archive BioProject PRJNA564565 to recreate the ASVs used for the North Pond Electrochemistry and FLOCs experiments. The .zip file contains github release v1.0 from https://github.com/BCODMO/northpond/tree/v1.0 which was forked from https://github.com/orcuttlab/northpond/commit/73aeab07d94d48c1b61ee50310fbb5a34bb60375 to BCO-DMO for curation purposes.

[[table of contents](#) | [back to top](#)]

Related Publications

D'Angelo, T. (2018). DNA Extraction using FastDNA Spinkit for Soil with PolyA Modifictaion v1 ([protocols.io.ur8ev9w](https://doi.org/10.17504/protocols.io.ur8ev9w)).
Protocols.io. doi:[10.17504/protocols.io.ur8ev9w](https://doi.org/10.17504/protocols.io.ur8ev9w)
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](https://doi.org/10.3389/fenvs.2020.00011)
Results

[[table of contents](#) | [back to top](#)]

Parameters

Parameters for this dataset have not yet been identified

Instruments

Dataset-specific Instrument Name	Illumina MiSeq
Generic Instrument Name	Automated DNA Sequencer
Dataset-specific Description	Aliquots of DNA extracts were sent to the Integrated Microbiome Resource facility at Dalhousie University (Halifax, Canada) for sequencing of the V4-V5 hypervariable region of the 16S rRNA gene via Illumina Mi-Seq technology using the 515F/926R primer pair. Sequencing was performed on an Illumina MiSeq using 300 300 bp paired-end V3 chemistry.
Generic Instrument Description	General term for a laboratory instrument used for deciphering the order of bases in a strand of DNA. Sanger sequencers detect fluorescence from different dyes that are used to identify the A, C, G, and T extension reactions. Contemporary or Pyrosequencer methods are based on detecting the activity of DNA polymerase (a DNA synthesizing enzyme) with another chemoluminescent enzyme. Essentially, the method allows sequencing of a single strand of DNA by synthesizing the complementary strand along it, one base pair at a time, and detecting which base was actually added at each step.

Dataset-specific Instrument Name	Q-bit Fluorometer 3.0
Generic Instrument Name	Fluorometer
Dataset-specific Description	Q-bit Fluorometer 3.0 (Thermo Fisher Scientific, Waltham, MA USA)
Generic Instrument Description	A fluorometer or fluorimeter is a device used to measure parameters of fluorescence: its intensity and wavelength distribution of emission spectrum after excitation by a certain spectrum of light. The instrument is designed to measure the amount of stimulated electromagnetic radiation produced by pulses of electromagnetic radiation emitted into a water sample or in situ.

Dataset-specific Instrument Name	Retsch MM 400 shaker
Generic Instrument Name	Shaker
Generic Instrument Description	A Shaker is a piece of lab equipment used to mix, blend, or to agitate substances in tube(s) or flask(s) by shaking them, which is mainly used in the fields of chemistry and biology. A shaker contains an oscillating board which is used to place the flasks, beakers, test tubes, etc.

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

[[table of contents](#) | [back to top](#)]

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 instrumented 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.

[[table of contents](#) | [back to top](#)]

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.

[[table of contents](#) | [back to top](#)]

Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1536539

[[table of contents](#) | [back to top](#)]