# Supplementary Table 1: Results of CHNS analyses in rock samples at different depths and sample images.

Website: https://www.bco-dmo.org/dataset/810902

Data Type: Other Field Results

Version: 1

Version Date: 2020-05-11

#### Project

» <u>Collaborative Research: Delineating The Microbial Diversity and Cross-domain Interactions in The Uncharted Subseafloor Lower Crust Using Meta-omics and Culturing Approaches</u> (Subseafloor Lower Crust Microbiology)

#### **Program**

» International Ocean Discovery Program (IODP)

Contributors	Affiliation	Role
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#### Abstract

Results of CHNS analyses in rock samples at different depths and sample images taken on board of the R/V JOIDES Resolution between November 30, 2015 and January 30, 2016

### **Table of Contents**

- Coverage
- Dataset Description
  - Methods & Sampling
  - Data Processing Description
- Data Files
- Related Publications
- <u>Parameters</u>
- <u>Instruments</u>
- Deployments
- Project Information
- Program Information
- Funding

# Coverage

**Spatial Extent**: Lat:-32.70567 Lon:57.278183 **Temporal Extent**: 2015-11-30 - 2016-01-30

# **Dataset Description**

Results of CHNS analyses in rock samples at different depths and sample images taken on board of the R/V JOIDES Resolution between November 30, 2015 and January 30, 2016.

## Methods & Sampling

Powderized rock material 462 from each sample (produced in the laminar flow hood on the *JOIDES* Resolution using a sterile mortar and pestle) was immediately transferred to sterile, muffled glass containers and stored in a desiccator until analysis of carbon and nitrogen according to established methods. Briefly, samples were

weighed into methanol rinsed silver boats (4 x 6 mm, Costech). 96-well glass plates (combusted 4 hrs at  $450^{\circ}$ C) holding these samples were placed in a vacuum desiccator that also contained an open dish with about 50 mL fresh, concentrated (12N) HCl. An inverted crystallization dish was placed over the samplesto protect them from water condensation. The desiccator was closed and pumped out with an airdriven aspirator, to a reading of about  $\sim 0.5$  atm and the desiccator was placed in an oven kept between 60 and 65 °C. Acidification was allowed to run for 60 to 72 hours. When acidification was complete, the samples were removed and set in the oven to dry (60-65 °C), and then placed in a vacuum desiccator charged with indicating silica gel (Fisher S162-500, activated by heating above 220 °C for several hours) and pumped down again and dried for about 24 hours.

## **Data Processing Description**

Samples were then analyzed on a Costech 4010 Elemental Analyzer connected via a Finningan-MAT Conflo-II interface to a DeltaPlus isotope ratio mass spectrometer.

BCO-DMO processing notes:

- Adjusted column headers to comply with database requirements
- Changed , to ; in observations to comply with database requirements

# [ table of contents | back to top ]

#### **Data Files**

F:1_	

#### chns analysis.csv

(Comma Separated Values (.csv), 1.44 KB) MD5:bd773855e0207e87bb5371968505a61d

Primary data file for dataset ID 810902

## Rock sample images associated with dataset 810902

filename: 810902 images.zip

(ZIP Archive (ZIP), 651.20 KB) MD5:97362e0e44148ec2636f67dab82b02d6

Rock sample images associated with dataset 810902; refer to column "Reference\_Picture" in dataset for the image corresponding to each Sample\_ID.

[ table of contents | back to top ]

# **Related Publications**

Pella, E. (1990). Elemental organic analysis. Part 1, Historical developments. American Laboratory 22,116–25. Methods

Pella, E. (1990). Elemental organic analysis. Part 2: State of the art. American Laboratory 22, 28-32 Methods

Whiteside, J. H., Olsen, P. E., Eglinton, T. I., Cornet, B., McDonald, N. G., & Huber, P. (2011). Pangean great lake paleoecology on the cusp of the end-Triassic extinction. Palaeogeography, Palaeoclimatology, Palaeoecology, 301(1-4), 1–17. doi:10.1016/j.palaeo.2010.11.025

Methods

[ table of contents | back to top ]

## **Parameters**

Parameter	Description	Units
Sample_ID	Sample ID	unitless
Depth	Depth below sea floor	meters (m)
TOC	Total Organic Carbon	percentage (%)
TON	Total Organic Nitrogen - bd = below detection limit	percentage (%)
Total_C_Total_N	Total Carbon/Total Nitrogen	percentage (%)
Ratio_13C_12C	Ratio of stable isotopes 13C/12C	percentage (%)
Clay_OH	wt% OH from clays (150-300°C)	percentage (%)
Observations	Lithology and alteration observations	unitless
Reference_Picture	Sample image. See "Data Files" section of metadata for .zip file of images.	unitless
Latitude	Latitude, south is negative	decimal degrees
Longitude	Longitude, west is negative	decimal degrees

# [ table of contents | back to top ]

# Instruments

Dataset- specific Instrument Name	Costech 4010 Elemental Analyzer	
Generic Instrument Name	Costech International Elemental Combustion System (ECS) 4010	
Generic Instrument Description	The ECS 4010 Nitrogen / Protein Analyzer is an elemental combustion analyser for CHNSO elemental analysis and Nitrogen / Protein determination. The GC oven and separation column have a temperature range of 30-110 degC, with control of +/- 0.1 degC.	

Dataset- specific Instrument Name	DeltaPlus isotope ratio mass spectrometer
Generic Instrument Name	Isotope-ratio Mass Spectrometer
Generic Instrument Description	The Isotope-ratio Mass Spectrometer is a particular type of mass spectrometer used to measure the relative abundance of isotopes in a given sample (e.g. VG Prism II Isotope Ratio Mass-Spectrometer).

# [ table of contents | back to top ]

# **Deployments**

IODP-360

Website	https://www.bco-dmo.org/deployment/810905	
Platform	R/V JOIDES Resolution	
Report	http://publications.iodp.org/scientific_prospectus/360/index.html	
Start Date	2015-11-30	
End Date	2016-01-30	

[ table of contents | back to top ]

# **Project Information**

Collaborative Research: Delineating The Microbial Diversity and Cross-domain Interactions in The Uncharted Subseafloor Lower Crust Using Meta-omics and Culturing Approaches (Subseafloor Lower Crust Microbiology)

**Coverage**: SW Indian Ridge, Indian Ocean

#### **NSF** abstract:

The lower ocean crust has remained largely unexplored and represents one of the last frontiers for biological exploration on Earth. Preliminary data indicate an active subsurface biosphere in samples of the lower oceanic crust collected from Atlantis Bank in the SW Indian Ocean as deep as 790 m below the seafloor. Even if life exists in only a fraction of the habitable volume where temperatures permit and fluid flow can deliver carbon and energy sources, an active lower oceanic crust biosphere would have implications for deep carbon budgets and yield insights into microbiota that may have existed on early Earth. This is all of great interest to other research disciplines, educators, and students alike. A K-12 education program will capitalize on groundwork laid by outreach collaborator, A. Martinez, a 7th grade teacher in Eagle Pass, TX, who sailed as outreach expert on Drilling Expedition 360. Martinez works at a Title 1 school with ~98% Hispanic and ~2% Native American students and a high number of English Language Learners and migrants. Annual school visits occur during which the project investigators present hands on-activities introducing students to microbiology, and talks on marine microbiology, the project, and how to pursue science related careers. In addition, monthly Skype meetings with students and PIs update them on project progress. Students travel to the University of Texas Marine Science Institute annually, where they get a campus tour and a 3-hour cruise on the R/V Katy, during which they learn about and help with different oceanographic sampling approaches. The project partially supports two graduate students, a Woods Hole undergraduate summer student, the participation of multiple Texas A+M undergraduate students, and 3 principal investigators at two institutions, including one early career researcher who has not previously received NSF support of his own.

Given the dearth of knowledge of the lower oceanic crust, this project is poised to transform our understanding of life in this vast environment. The project assesses metabolic functions within all three domains of life in this crustal biosphere, with a focus on nutrient cycling and evaluation of connections to other deep marine microbial habitats. The lower ocean crust represents a potentially vast biosphere whose microbial constituents and the biogeochemical cycles they mediate are likely linked to deep ocean processes through faulting and subsurface fluid flow. Atlantis Bank represents a tectonic window that exposes lower oceanic crust directly at the seafloor. This enables seafloor drilling and research on an environment that can transform our understanding of connections between the deep subseafloor biosphere and the rest of the ocean. Preliminary analysis of recovered rocks from Expedition 360 suggests the interaction of seawater with the lower oceanic crust creates varied geochemical conditions capable of supporting diverse microbial life by providing nutrients and chemical energy. This project is the first interdisciplinary investigation of the microbiology of all 3 domains of life in basement samples that combines diversity and "meta-omics" analyses, analysis of nutrient addition experiments, high-throughput culturing and physiological analyses of isolates, including evaluation of their ability to utilize specific carbon sources, Raman spectroscopy, and lipid biomarker analyses. Comparative genomics are used to compare genes and pathways relevant to carbon cycling in these samples to data from published studies of other deep-sea environments. The collected samples present a rare and time-sensitive opportunity to gain detailed insights into microbial life, available carbon and energy sources for this life, and of dispersal of microbiota and connections in biogeochemical processes between the lower oceanic crust and the overlying aphotic water column.

#### About the study area:

The International Ocean Discovery Program (IODP) Expedition 360 explored the lower crust at Atlantis Bank, a 12 Ma oceanic core complex on the ultraslow-spreading SW Indian Ridge. This oceanic core complex represents a tectonic window that exposes lower oceanic crust and mantle directly at the seafloor, and the expedition provided an unprecedented opportunity to access this habitat in the Indian Ocean.

[ table of contents | back to top ]

# **Program Information**

International Ocean Discovery Program (IODP)

Website: <a href="http://www.iodp.org/index.php">http://www.iodp.org/index.php</a>

Coverage: Global

The International Ocean Discovery Program (IODP) is an international marine research collaboration that explores Earth's history and dynamics using ocean-going research platforms to recover data recorded in seafloor sediments and rocks and to monitor subseafloor environments. IODP depends on facilities funded by three platform providers with financial contributions from five additional partner agencies. Together, these entities represent 26 nations whose scientists are selected to staff IODP research expeditions conducted throughout the world's oceans.

IODP expeditions are developed from hypothesis-driven science proposals aligned with the program's <u>science</u> <u>plan</u> *Illuminating Earth's Past, Present, and Future*. The science plan identifies 14 challenge questions in the four areas of climate change, deep life, planetary dynamics, and geohazards.

IODP's three platform providers include:

- The U.S. National Science Foundation (NSF)
- Japan's Ministry of Education, Culture, Sports, Science and Technology (MEXT)
- The European Consortium for Ocean Research Drilling (ECORD)

More information on IODP, including the Science Plan and Policies/Procedures, can be found on their website at <a href="http://www.iodp.org/program-documents">http://www.iodp.org/program-documents</a>.

[ table of contents | back to top ]

# **Funding**

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1658031

[ table of contents | back to top ]