# The eukaryotic (poly-A) metatranscriptome for Canterbury Basin from R/V JOIDES Resolution cruise JRES-317 in the Canterbury Basin from 2009-2010

Website: https://www.bco-dmo.org/dataset/654429

**Data Type**: Cruise Results **Version**: 19 Aug 2016 **Version Date**: 2016-08-19

#### **Project**

» Exploring of the Ecological Role(s) of Marine Fungi in the Deep Subseafloor (Fungal and prokaryotic activity in subseafloor)

## **Program**

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

Contributors	Affiliation	Role
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## **Dataset Description**

The eukaryotic (poly-A) metatranscriptome for Canterbury Basin; sampled from the eastern margin of the South Island of New Zealand on JOIDES Resolution IODP leg 317.

## Methods & Sampling

During IODP leg 317 expedition (JOIDES Resolution), a sediment core was drilled at Site U1352 in 344 m water depth. A total depth of 1927.5 mbsf was cored, spanning the Holocene to late Eocene periods. For this work, core sections were obtained from this site from 3.76 mbsf, 11.60 mbsf, 24.60 mbsf, 345.50 mbsf, and 403.10 mbsf.

RNA was extracted from 16 g of sediment from the Canterbury core samples using the RNA PowerSoil Total RNA Isolation Kit (MoBIO Laboratories, Carlsbad, CA) following a modified manufacturer's protocol. Modifications included: ten cycles of homogenization for 1-minute intervals with 1 minute rest between intervals using a FastPrep benchtop homogenizer (MP Biomedicals, Santa Ana, CA) set to 4.0 m/s and increasing the incubation time to 45 min following the addition of SR4 buffer. Trace DNA was removed by treatment with Turbo DNA-free (Life Technologies, Grand Island, NY) for 60 minutes at 37 degrees C. A final RNA purification step was performed using the MEGAclear kit (Life Technologies, USA). In order to avoid

contamination, all manipulations were carried out in a dedicated PCR hood (AirClean Systems, USA) for RNA work. An extraction blank was also carried through the entire procedure to control for kit contamination and served as "negative control". Removal of carry-over DNA in RNA extracts was confirmed by the absence of visible amplification of the V4 hypervariable region of SSU rDNA after 35 cycles of PCR using the RNA extracts as template with key-tagged bacterial primers (Flores et al., 2011). PCR conditions were: 95 degrees C for 2 minutes followed by 35 cycles of 95 degrees C for 15 seconds, 53 degrees C for 45 seconds and 68 degrees C for 45 seconds with a final incubation of 68 degrees C for 3 minutes. Total RNA was used as template for cDNA amplification using the Ovation 3'-DGE System (NuGEN, San Carlos, CA) to obtain an enriched eukaryotic metatranscriptome through selection of polyA transcripts. Double-stranded cDNA was purified using the DNA Clean & Concentrator kit (Zymo Research) as described in the modified user guide (<a href="https://www.biolynx.ca/pdf/admin/nu\_threedgeuserguide.pdf">https://www.biolynx.ca/pdf/admin/nu\_threedgeuserguide.pdf</a>) provided by NuGEN. The quantity of amplified cDNA was evaluated using a fluorometer (Qubit 2.0, Life Technologies).

## **Data Processing Description**

Raw read files were deposited in GenBankSRA under accession number SRP072032.

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## **Data Files**

File

polyA\_RNA.csv(Comma Separated Values (.csv), 282 bytes)
MD5:d9cb7157d5053f4614e6e443858a8c32

Primary data file for dataset ID 654429

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## **Parameters**

Parameter	Description	Units
cruise_id	Cruise identifier	unitless
location	Sampling location	unitless
lat	Latitude of sampling location	decimal degrees
lon	Longitude of sampling location	decimal degrees
core_depth	Depth of the sediment core	meters below seafloor (mbsf)
sample_depth	Depth from which the samples were taken	meters below seafloor (mbsf)
accession_num	GenBank SRA accession number	unitless
accession_link	Hyperlink to GenBank SRA for the accession number	unitless

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#### Instruments

Dataset- specific Instrument Name	
Generic Instrument Name	Advanced Piston Corer
	The JOIDES Resolution's Advanced Piston Corer (APC) is used in soft ooze and sediments. The APC is a hydraulically actuated piston corer designed to recover relatively undisturbed samples from very soft to firm sediments. More information is available from IODP (PDF).

Dataset- specific Instrument Name	Qubit 2.0, Life Technologies
Generic Instrument Name	Fluorometer
	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	
Generic Instrument Name	Thermal Cycler
Generic Instrument Description	A thermal cycler or "thermocycler" is a general term for a type of laboratory apparatus, commonly used for performing polymerase chain reaction (PCR), that is capable of repeatedly altering and maintaining specific temperatures for defined periods of time. The device has a thermal block with holes where tubes with the PCR reaction mixtures can be inserted. The cycler then raises and lowers the temperature of the block in discrete, pre-programmed steps. They can also be used to facilitate other temperature-sensitive reactions, including restriction enzyme digestion or rapid diagnostics. (adapted from <a href="http://serc.carleton.edu/microbelife/research_methods/genomics/pcr.html">http://serc.carleton.edu/microbelife/research_methods/genomics/pcr.html</a> )

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## **Deployments**

## JRES-317

Website	https://www.bco-dmo.org/deployment/654203
Platform	R/V JOIDES Resolution
Report	http://dmoserv3.whoi.edu/data_docs/C-DEBI/cruise_reports/317PR.PDF
Start Date	2009-11-04
End Date	2010-01-04
Description	More information is available from IODP: http://iodp.tamu.edu/scienceops/expeditions/canterbury_basin.html

## **Project Information**

## Exploring of the Ecological Role(s) of Marine Fungi in the Deep Subseafloor (Fungal and prokaryotic activity in subseafloor)

Coverage: Peru Margin and Canterbury Basin New Zealand

The deep sedimentary biosphere, extending hundreds of meters below the seafloor harbors unexpected diversity of Bacteria, Archaea and microbial eukaryotes. Far less is known about microbial eukaryotes in subsurface habitats, albeit several studies have indicated that fungi dominate microbial eukaryotic communities and fungal molecular signatures (of both yeasts and filamentous forms) have been detected in samples as deep as 1740mbsf. Here we compare and contrast fungal ribosomal RNA gene signatures and whole community metatranscriptomes present in sediment core samples from 6 and 95mbsf from Peru Margin site 1229A and from samples from 12 and 345 mbsf from Canterbury Basin site U1352. The metatranscriptome analyses reveal higher relative expression of amino acid and peptide transporters in the less nutrient rich Canterbury Basin sediments compared to the nutrient rich Peru Margin, and higher expression of motility genes in the Peru Margin samples. Higher expression of genes associated with metals transporters and antibiotic resistance and production was detected in Canterbury Basin sediments. A poly-A focused metatranscriptome produced for the Canterbury Basin sample from 345 mbsf provides further evidence for active fungal communities in the subsurface in the form of fungal-associated transcripts for metabolic and cellular processes, cell and membrane functions, and catalytic activities. Fungal communities at comparable depths at the two geographically separated locations appear dominated by distinct taxa. Differences in taxonomic composition and expression of genes associated with particular metabolic activities may be a function of sediment organic content as well as oceanic province. Microscopic analysis of Canterbury Basin sediment samples from 4 and 403 mbsf produced visualizations of septate fungal filaments, branching fungi, conidiogenesis and spores. These images provide another important line of evidence supporting the occurrence and activity of fungi in the deep subseafloor biosphere.

This project was funded by C-DEBI sub-award #49538097.

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## **Program Information**

Center for Dark Energy Biosphere Investigations (C-DEBI)

Website: <a href="http://www.darkenergybiosphere.org">http://www.darkenergybiosphere.org</a>

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 <u>Data Management Plan (PDF)</u> and in compliance with the <u>NSF Ocean Sciences Sample and Data Policy</u>. 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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