

# Metagenomic sequences from Lau Basin low temperature vents and Loihi Seamount; from R/V Roger Revelle and R/V Thomas G. Thompson cruises RR1211 and TN225 in the Northeast Lau Basin, Loihi Seamount from 2008-2012

**Website:** <https://www.bco-dmo.org/dataset/636430>

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## Project

» [Understanding microbial manganese-oxidizing communities and physiological mechanisms in metal oxide-rich hydrothermal sediments using a metagenomic and metatranscriptomic approach](#) (Vent Mn-Fe Microbes)

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## Dataset Description

Metagenomic sequences from Lau Basin low temperature vents and Loihi Seamount; collected on the 2012 "Submarine Ring of Fire" (SRoF) cruise and the 2008 FeMO cruise.

## Methods & Sampling

Samples were collected with a closable PVC scoop sampler that was designed to sample mat material along with bottom waters while minimizing cross-contamination from other samples or with ambient seawater. The sampler was constructed with 3" PVC pipe and was washed with 70% ethanol before being rinsed and filled with filter-sterilized deionized water and sealed with a ball valve before deployment. The valve was opened immediately prior to sampling and closed directly after the sample collected and remained sealed until the sample was recovered on the ship. All samples were allowed to settle at 4 degrees C for approximately 2 hours before the overlying seawater was decanted and the mat material frozen at -80 degrees C until DNA was extracted in the lab.

Note: The J2-373 dive video can be found on the virtual van (<http://4dgeo.whoi.edu/jason/>).

**DNA extraction:** Total genomic DNA (gDNA) was extracted from each sample in duplicate using the FastDNA Spin Kit for Soil following the manufacturer's protocol (Qbiogene, Irvine, CA). Extracted gDNA from each sample was pooled, cleaned, and concentrated using Montage PCR centrifugal filter devices (Millipore, Bedford, MA). The gDNAs were then quantified using a Nanodrop ND-1000 spectrophotometer and were diluted to 10 ng DNA/ml using filter sterilized 10 mM Tris 0.1 mM EDTA (pH 8.0).

**Metagenome sequencing:** Two micrograms of gDNA was sheared using a Covaris S2 sonicator and 400bp fragments were size selected for library construction. The library was constructed using the TruSeq DNA

sample prep kit (Illumina) following the manufactures protocol. Sequencing was performed using an Illumina HiSeq 2000 sequencer.

## Data Processing Description

**Metagenome quality control and assembly:** Sequences were quality controlled and trimmed using the program Sickle. The trimmed sequences were then digitally normalized using the program Khmer utilizing a two-pass strategy. The sequences were first normalized by median to reduce sequences with high-abundance kmers using a kmer size of 20bp and a maximum coverage overlap value of 20. The sequences were then filtered to remove low-abundance kmers and Illumina sequencing artifacts using the Khmer script filter-below-abund with a minimum overlap of 2 kmers. The remaining sequences after preprocessing were used for sequence assembly.

The normalized reads were assembled using the de Bruijn graph-based assembler Velvet. Four independent assemblies were constructed with kmer sizes of 41, 51, 61, and 71. The contigs were assembled into "supercontigs" using the program Geneious with a minimum overlap of 25bp. Contigs longer than 1000bp were exported and used in further analysis.

The sequence data has been submitted to NCBI under Bioproject numbers PRJNA298367 (Lau sequences) and PRJNA297446 (Loihi sequences).

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## Data Files

File
<b>metagenomes.csv</b> (Comma Separated Values (.csv), 822 bytes) MD5:95750607c921f3baeab53dfb3e7c2d74 Primary data file for dataset ID 636430

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## Parameters

Parameter	Description	Units
cruise	Cruise identifier. TN225 also known as FeMO-3. RR1211 also known as SRoF-12.	dimensionless
description	Description of sample.	dimensionless
BioProject	NCBI's BioProject ID number.	dimensionless
location	Description of location.	dimensionless
dive	Dive number.	dimensionless
sample	Sample number.	dimensionless
date	Date (year, month, and day) of sampling.	YYYYmmdd
lat	Latitude of sample.	decimal degrees
lon	Longitude of sample.	decimal degrees
depth	Depth of sample.	meters
BioProject_URL	Hyperlink to NCBI BioProject page.	dimensionless

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## Instruments

<b>Dataset-specific Instrument Name</b>	Illumina HiSeq 2000 sequencer
<b>Generic Instrument Name</b>	Automated DNA Sequencer
<b>Generic Instrument Description</b>	A DNA sequencer is an instrument that determines the order of deoxynucleotides in deoxyribonucleic acid sequences.

<b>Dataset-specific Instrument Name</b>	
<b>Generic Instrument Name</b>	ROV Jason
<b>Generic Instrument Description</b>	The Remotely Operated Vehicle (ROV) Jason is operated by the Deep Submergence Laboratory (DSL) at Woods Hole Oceanographic Institution (WHOI). WHOI engineers and scientists designed and built the ROV Jason to give scientists access to the seafloor that didn't require them leaving the deck of the ship. Jason is a two-body ROV system. A 10-kilometer (6-mile) fiber-optic cable delivers electrical power and commands from the ship through Medea and down to Jason, which then returns data and live video imagery. Medea serves as a shock absorber, buffering Jason from the movements of the ship, while providing lighting and a bird's eye view of the ROV during seafloor operations. During each dive (deployment of the ROV), Jason pilots and scientists work from a control room on the ship to monitor Jason's instruments and video while maneuvering the vehicle and optionally performing a variety of sampling activities. Jason is equipped with sonar imagers, water samplers, video and still cameras, and lighting gear. Jason's manipulator arms collect samples of rock, sediment, or marine life and place them in the vehicle's basket or on "elevator" platforms that float heavier loads to the surface. More information is available from the operator site at URL. <a href="https://ndsf.whoi.edu/jason/">https://ndsf.whoi.edu/jason/</a>

<b>Dataset-specific Instrument Name</b>	Nanodrop ND-1000 spectrophotometer
<b>Generic Instrument Name</b>	Spectrophotometer
<b>Generic Instrument Description</b>	An instrument used to measure the relative absorption of electromagnetic radiation of different wavelengths in the near infra-red, visible and ultraviolet wavebands by samples.

<b>Dataset-specific Instrument Name</b>	Covaris S2 sonicator
<b>Generic Instrument Name</b>	ultrasonic cell disrupter (sonicator)
<b>Generic Instrument Description</b>	Instrument that applies sound energy to agitate particles in a sample.

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## Deployments

### RR1211

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/527560">https://www.bco-dmo.org/deployment/527560</a>
<b>Platform</b>	R/V Roger Revelle
<b>Report</b>	<a href="http://dmoserv3.whoi.edu/data_docs/Vent_Mn-Fe_Microbes/SRoF12-cruisereport-final.pdf">http://dmoserv3.whoi.edu/data_docs/Vent_Mn-Fe_Microbes/SRoF12-cruisereport-final.pdf</a>
<b>Start Date</b>	2012-09-09
<b>End Date</b>	2012-09-26
<b>Description</b>	2012 "Submarine Ring of Fire" (SRoF) cruise in the Northeast Lau Basin. More information is available on the NOAA Ocean Explorer website: <a href="http://oceanexplorer.noaa.gov/explorations/12fire/welcome.html">http://oceanexplorer.noaa.gov/explorations/12fire/welcome.html</a> This cruise is also affiliated with the project, "Understanding microbial manganese-oxidizing communities and physiological mechanisms in metal oxide-rich hydrothermal sediments using a metagenomic and metatranscriptomic approach". Original cruise data are available from the NSF R2R data catalog

## TN225

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/636449">https://www.bco-dmo.org/deployment/636449</a>
<b>Platform</b>	R/V Thomas G. Thompson
<b>Start Date</b>	2008-09-22
<b>End Date</b>	2008-10-12
<b>Description</b>	See more information: <a href="https://earthref.org/FEMO/cruises/2008/main.htm">https://earthref.org/FEMO/cruises/2008/main.htm</a> Additional cruise data available from R2R.

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## Project Information

### **Understanding microbial manganese-oxidizing communities and physiological mechanisms in metal oxide-rich hydrothermal sediments using a metagenomic and metatranscriptomic approach (Vent Mn-Fe Microbes)**

**Coverage:** Northeast Lau Basin

*Project description from the NSF award abstract:*

Hydrothermal systems are important sources of dissolved Mn to the oceans. Upon oxidation of Mn(II), Mn(III,IV) oxides are deposited at the sea floor as crusts, nodules and sediments both near and far from the sources. Microbial activity has long been recognized as being important to the fate of Mn in these hydrothermal systems, yet we know very little about the organisms that catalyze Mn oxidation, the mechanisms by which Mn is oxidized or the physiological function that Mn oxidation serves. The overarching goals of this project are to reveal the organisms and mechanism(s) underlying Mn(II) oxidation, to evaluate whether hydrothermal Mn oxidizers may obtain energy from Mn oxidation, and test whether thermophilic Mn oxidizers exist. Specifically, the project will: 1) evaluate whether we can identify certain genomic sequences that correlate to the presence/concentration of Mn oxides (and hence Mn(II)- oxidizing bacteria) by comparing the metagenomes of ferromanganese (containing both Mn and Fe oxides) microbial mats with ferruginous (Fe oxide only) mats from Lau Basin and Loihi Seamount; 2) use peptide probes bound to magnetic particles for selectively binding and capturing Mn oxide particles and characterizing the particles using phylogenetic and functional gene (PCR and FISH) and transcriptomic analysis; 3) assess the main pathways of carbon fixation in ferromanganese microbial mats as compared to ferruginous mats as a possible indicator of Mn-based auto/mixotrophy using genomic approaches and substrate stimulated (e.g., addition of Mn(II)) CO<sub>2</sub> fixation measurements and stable isotope probing (SIP) genomic analysis; and 4) isolate and characterize Mn(II)-oxidizing bacteria and determine whether thermophilic Mn oxidizers exist.

The results of this research will increase our understanding of Mn(II) oxidation in hydrothermal sediments, identify microorganisms that are the environmentally relevant Mn oxidizers and begin to address the long standing question of whether Mn auto/mixotrophy exists using approaches not based on the biases associated with cultivation. Ultimately this information is critical to our understanding of biogeochemical cycles (Mn oxidation and Mn oxides impact many other elemental cycles, including carbon, sulfur, and heavy metals) and the natural attenuation of toxic metal and organic compounds; this may lead to improved technologies for environmental remediation. Because Mn oxides are believed to be an analog to the ancestral Mn centers in photosystem II, this research may also lend new insights into ancient biogeochemistry occurring before the Great Oxidation Event.

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1129553</a>

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