

# Microbiome composition of coral species collected from reefs in Mo'orea, French Polynesia and exposed to various experimental treatments in August 2018

**Website:** <https://www.bco-dmo.org/dataset/843188>

**Data Type:** Other Field Results

**Version:** 1

**Version Date:** 2021-03-11

## Project

» [Collaborative Research: Viral Reefscapes: The Role of Viruses in Coral Reef Health, Disease, and Biogeochemical Cycling](#) (Moorea Virus Project)

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## Abstract

Coral samples from reefs in Mo'orea, French Polynesia were exposed to various experimental treatments to quantify how different environmental stressors impact the coral health and microbial community structure of the corals. Environmental stressors included increased temperature and nutrients as well as exudate released from corals when they bleach.

## Table of Contents

- [Coverage](#)
- [Dataset Description](#)
  - [Methods & Sampling](#)
  - [Data Processing Description](#)
- [Data Files](#)
- [Related Publications](#)
- [Related Datasets](#)
- [Parameters](#)
- [Instruments](#)
- [Project Information](#)
- [Funding](#)

## Coverage

**Spatial Extent:** Lat:-17.4754 Lon:-149.84

**Temporal Extent:** 2018-08 - 2021-08

## Methods & Sampling

### Sampling and analytical procedures:

Corals of two species (*Pocillopora cf. meandrina* and *Acropora hyacinthus*) were collected from the reef crest on the north side of the island of Mo'orea, French Polynesia (GPS location: 17.47541 S, 149.8402 W). The fragmented corals were transported to Gump Research Station where they equilibrated for >24 hours in shaded flowing seawater tables.

Coral fragments were exposed to slightly elevated bicarbonate (+1 mM) and Ammonium (0.5 mM NH<sub>4</sub>) for 24 hours prior to being sampled (T0 controls) and then exposed to various experimental treatments to better

understand the response of reefs to factors that trigger viral outbreaks.

Treatments were:

- Control: no modification
- Nutrient: addition of two millimolar nitrate (2mM NO<sub>3</sub>)
- Temperature: increase of 3 degrees Celsius (+3°C)
- Coral exudate: addition of a mean of 2 millimolar dissolved organic carbon (DOC) + particulate organic carbon (POC) from a bleaching coral (co-occurring con-specific coral head)
- Exudate: exudate blank is the coral exudate only, with no coral sample added.
- Blank: water with no coral added

*Note: Water was filtered (0.2 micron filtration) before being used in the experiment.*

Corals were kept in individual recirculating water baths. Fragments from a single coral head were exposed to all treatments, with replicate samples of the same coral head sharing the same letter designation. At specified time points (6, 12, 24, and 48 hours since exposure to treatment began) corals were sacrificed and water from the water baths (microcosm water) was sampled for microbial community composition by filtering ca. 0.5L water through a 0.2 micron filter (Sterivex-GP 0.22 µm, SVGP01050, Millipore) after which they were frozen.

Coral exudate blank clogged the Sterivex filter, resulting in a marked decrease in the amount of water that was filtered. Coral pieces were placed in Zymo DNA/RNA shield. Initial processing of the corals included bead beating and then freezing at -80°C, followed by shipping to either Rice University or Oregon State University (OSU) for DNA extraction.

### **DNA sequencing**

DNA was extracted using either the Zymo quick-DNA extraction kit (corals) or the Qiagen Powerwater kit (water samples) following manufacturer protocols. DNA was amplified (at OSU) following the Earth Microbiome Project protocols using the updated primers of 515f (Parada et al. 2016) and 806r (Apprill et al. 2015).

Due to co-amplification of eukaryotic 12S rRNA genes, DNA was size selected using Blue Pippin (Sage Scientific) prior to sequencing to minimize 12S sequencing. While we used dual indexed primers during sequencing, reverse read quality scores were not acceptable and only forward reads were used and uploaded to the Sequence Read Archive (SRA).

Sequencing was performed on the Illumina MiSeq platform using the V.2 chemistry at the Center for Genome Research and Biocomputing at Oregon State University.

All data associated with this submission has been made available through the NCBI Short Read Archive under BioProject PRJNA684406 (<https://www.ncbi.nlm.nih.gov/bioproject/PRJNA684406>). The accompanying data files list the accessions numbers and sampling information.

### **Sample ID/ Library ID details:**

B = blank  
C = control  
V = exudate  
VB = exudate blank  
N = nutrient  
T = temperature  
ACR = Acropora  
POC = Pocillopora

"Vd-12\_water\_Experiment\_d" means Exudate, replicate d, 12 hours into treatment, water experiment, replicate d

"ACR-Cb-0-20\_b2" means Acropora, Control, replicate b, zero time (0 hours into treatment), sequential sample number 20, replicate b2

"POC-Cc-0-44\_c3" means Pocillopora, Control, replicate c, 0 hours into treatment, sample number 44, replicate c3

## Data Processing Description

The only data processing was done by the sequencing facility which included stripping of sequencing primers and bar codes. No other data manipulation was done.

BCO-DMO processing description:

- Split single lat\_lon column into separate latitude and longitude columns and converted to decimal degrees
- Removed extraneous row with single value of '1' and blanks elsewhere
- Added column for BioProject
- Added column for BioSample number and populated it with data found at NCBI
- Adjusted field/parameter names to comply with database requirements
- Added a conventional header with dataset name, PI names, version date

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

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## Data Files

File
<b>experimental_treatments.csv</b> (Comma Separated Values (.csv), 28.48 KB) MD5:345ce86a2543f7ab43847800b090a904
Primary data file for dataset ID 843188

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

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## Related Publications

Apprill, A., McNally, S., Parsons, R., & Weber, L. (2015). Minor revision to V4 region SSU rRNA 806R gene primer greatly increases detection of SAR11 bacterioplankton. *Aquatic Microbial Ecology*, 75(2), 129–137. doi:[10.3354/ame01753](https://doi.org/10.3354/ame01753)

*Methods*

Marotz, C., Amir, A., Humphrey, G., Gaffney, J., Gogul, G., & Knight, R. (2017). DNA extraction for streamlined metagenomics of diverse environmental samples. *BioTechniques*, 62(6), 290–293.

<https://doi.org/10.2144/000114559>

*Methods*

Parada, A. E., Needham, D. M., & Fuhrman, J. A. (2015). Every base matters: assessing small subunit rRNA primers for marine microbiomes with mock communities, time series and global field samples. *Environmental Microbiology*, 18(5), 1403–1414. doi:10.1111/1462-2920.13023 <https://doi.org/http://doi.org/10.1111/1462-2920.13023>

*Methods*

Schwartz, T., Thompson, L., Humphrey, G., Gogul, G., Gaffney, J., Amir, A., & Knight, R. (2018). Earth Microbiome Project (EMP) high throughput (HTP) DNA extraction protocol v1. *Protocols.io*.

<https://doi.org/10.17504/protocols.io.pdmd46>

*Methods*

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

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## Related Datasets

### IsRelatedTo

Oregon State University. Moorea Virus Project - Longitudinal Coral Microbiome Study at the Mo'orea LTER. 2020/12. In: BioProject [Internet]. Bethesda, MD: National Library of Medicine (US), National Center for Biotechnology Information; Available from: <https://www.ncbi.nlm.nih.gov/bioproject/PRJNA684406>. NCBI: BioProject PRJNA684406.

## Parameters

Parameter	Description	Units
Collection_Date	Collection date month and year	unitless
Latitude	Latitude	decimal degrees
Longitude	Longitude	decimal degrees
BioProject	NCBI BioProject number	unitless
BioSample	NCBI BioSample number	unitless
SRA_accession	NCBI Short Read Archive Accession reference	unitless
Library_ID	Sample identification (see metadata descriptions for details)	unitless
Sample_Type	Type of sample (water or coral)	unitless
Species	Species of Coral from which DNA was extracted, or Water	unitless
Treatment	Treatment (control, blank, nutrient, temperature, coral exudate, exudate blank)	unitless
Timepoint	Hours since initial exposure to treatment	hours
Replicate	Replicate samples of a single coral head represented by alphanumeric designation	unitless

## Instruments

<b>Dataset-specific Instrument Name</b>	Illumina MiSeq platform
<b>Generic Instrument Name</b>	Automated DNA Sequencer
<b>Dataset-specific Description</b>	Sequencing was performed on the Illumina MiSeq platform using the V.2 chemistry at the Center for Genome Research and Biocomputing at Oregon State University.
<b>Generic Instrument Description</b>	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.

<b>Dataset-specific Instrument Name</b>	recirculating water bath
<b>Generic Instrument Name</b>	circulating water bath
<b>Dataset-specific Description</b>	Corals were kept in individual recirculating water baths
<b>Generic Instrument Description</b>	A device designed to regulate the temperature of a vessel by bathing it in water held at the desired temperature. [Definition Source: NCI]

<b>Dataset-specific Instrument Name</b>	
<b>Generic Instrument Name</b>	Sage Science BluePippin DNA size selection device
<b>Generic Instrument Description</b>	An automated DNA size selection instrument, with pulsed-field electrophoresis for resolving and collecting high molecular weight DNA. The instrument is used to automatically extract DNA fragments of a user selected size for downstream technologies such as miRNA isolation, DNA sequencing, RNA-seq, genotyping, DNA sequencing, ChIP-seq, and Long-read sequencing. The instrument uses electrophoresis along with laser detection or other imaging technology to determine when to start collecting DNA based on size ranges entered by the user. Once the DNA is no longer in the desired size range, collection ceases. The instrument has electrophoresis voltage options: 25V, 100V or 150V constant, or 100V pulsed field. The optical detection wavelength is 470 nm excitation, and 525 nm emission. The instrument can run up to 5 samples/gel cassettes at a time, with no possibility of cross contamination.

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

## Project Information

### **Collaborative Research: Viral Reefscapes: The Role of Viruses in Coral Reef Health, Disease, and Biogeochemical Cycling (Moorea Virus Project)**

**Coverage:** Moorea, French Polynesia, Pacific 17 S 150 W

Ecologically and economically, coral reefs are among the most valuable ecosystems on Earth. These habitats are estimated to harbor up to nine million species, contribute ~30 billion US dollars annually to the global economy, and are tropical epicenters of biogeochemical cycling. Global (climate change) and local (nutrient pollution and overfishing) stressors are drivers of coral reef decline that can disrupt the symbiotic associations among corals and resident microbial communities, including dinoflagellate algae, bacteria, and viruses. Viruses interact with all living cellular organisms, are abundant in oceans, and integral to marine ecosystem functioning. This project will be the first to quantify the variability of viral infection in corals across different reef habitats and across time. This will increase our understanding of the total diversity of coral viruses and illuminate the full suite of factors that trigger viral outbreaks on reefs. At the same time the project will evaluate how carbon and nitrogen cycling are altered on coral reefs as a result of global and local stressors that trigger viral infection. This project will ultimately broaden our understanding of the impacts of viruses on reefs beyond their role as putative disease agents. Results of the project will be communicated broadly in scientific arenas, in K-12, undergraduate, and graduate education and training programs, and to the general public through video and multimedia productions, as well as outreach events. 2-D Reef Replicas from our field sites across Moorea will be constructed, allowing children and adults in the US and French Polynesia to 'become' marine scientists and use quadrats, transect tapes, and identification guides to quantify metrics of reef change. Three graduate students will be involved in all aspects of the research and an effort will be made to recruit and support minority students. All datasets will be made freely available to the public and newly developed methods from this project will serve as an important set of springboard tools and baselines for future lines of inquiry into the processes that influence reef health.

Coral reefs, found in nutrient-poor shallow waters, are biodiversity and productivity hotspots that provide substantial ecological and societal benefits. Corals energetically subsidize these oligotrophic ecosystems by releasing significant amounts of mucus (an organic carbon and nitrogen-rich matrix) into the surrounding seawater. Viral production in reef waters can be a significant portion of total reef carbon cycling, accounting for ~10% of gross benthic carbon fixation in reef ecosystems. Viruses are also ~10 times more abundant on coral surfaces than in the water column meaning that viral infection experienced by corals during stress likely results in an increase in carbon and perhaps nitrogen flux to the water column. Thus phages and eukaryotic viruses may be responsible for shifting reef health and function directly via coral and symbiont infection and by altering biogeochemical cycling in host colonies and the adjacent reef system. The main goal of this project is to experimentally interrogate and then model the links among viral infections, declines in coral and reef health, and associated shifts in biogeochemical cycling in reef ecosystems. Lab and field experiments will be conducted at the Moorea Coral Reef LTER to characterize the spatiotemporal dynamics of viruses within two dominant reef-building coral species that differ in their susceptibility to abiotic stress. A novel viral infection and induction approach will be coupled with stable isotopic pulse-chase experiments to quantify and track carbon and nitrogen flux out of coral holobionts (host and microbial symbionts) and into dissolved and particulate pools. In these experiments, virus, bacteria, and symbiont abundance, diversity, and function will be measured simultaneously with the health and activity of the host. Pulse-chase techniques, as well as flux- and niche-based modeling, will result in a holistic understanding of how corals and associated viruses impact reef energy budgets and the ramifications of carbon and nitrogen flux for reef communities. Ultimately, this project will quantify and describe an integrated mechanism by which environmental stressors alter viral, microbial, and coral diversity and, consequently, ecosystem function.

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

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1635798</a>
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1635913</a>

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