

# Incubation experiments were conducted in St. John, US Virgin Islands to investigate the macronutrient drawdown response of reef seawater microbial communities to exudates released from the coral species *Porites astreoides* and *Gorgonia ventalina*.

**Website:** <https://www.bco-dmo.org/dataset/865193>

**Data Type:** experimental

**Version:** 1

**Version Date:** 2022-11-10

## Project

» [Signature exometabolomes of Caribbean corals and influences on reef picoplankton](#) (Coral Exometabolomes)

Contributors	Affiliation	Role
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## Abstract

Incubation experiments were conducted in St. John, US Virgin Islands to investigate the composition of exudates released from different species of benthic organisms, and the response of reef seawater microbial communities to mixed exudates released from different species and to specific metabolites. Exudates were collected from the stony coral *Porites astreoides*, and the octocoral *Gorgonia ventalina* after an 8 hour incubation. Reef seawater microbial communities were incubated separately in the presence of exudates from *P. astreoides* and *G. ventalina* for 48 hours and samples were collected to monitor changes in macronutrient concentrations.

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## Coverage

**Spatial Extent:** Lat:18.315773476842 Lon:-64.727413483487

## Dataset Description

Note: The macronutrient concentrations in this dataset reflect the raw data and have not been normalized (as is presented in the manuscript Weber et al., 2022)

Other relevant files and publications:

The targeted and untargeted metabolomics data and metadata associated with this study are located on the MetaboLights database under accession numbers MTBLS2855 and MTBLS3286.

The 16S rRNA gene sequencing data and metadata associated with this study are located on the NCBI Sequence Read Archive (SRA) under BioProject PRJNA739882. BioSample accession numbers are not linked

with the data submitted to BCO-DMO because samples for flow cytometry and macronutrients were not always collected at the same time as samples collected for microbial community analyses, meaning that only some of the samples collected for microbial community analyses have affiliated microbial abundances and nutrient concentrations.

## Methods & Sampling

Experiments were conducted to monitor responses of reef seawater microbial communities to concentrated exudates from *P. astreoides* and *G. ventalina*. After the organism incubations were conducted, excess filtrate (2 l) from 3 of the 6 colony/fragment incubations (selected randomly) were pooled into an acid-washed, 10 l carboy and mixed. Excess filtrate (~2 l) from the three control incubations were also pooled into a second, acid-washed, 10 l carboy and mixed.

For the *P. astreoides* experiment, surface seawater was collected from the offshore site and coarsely filtered through a GF/A filter (1.6 µm nominal pore size) using peristalsis to remove larger cells and minimize heterotrophic grazing, but retain bacteria and archaea. Approximately 2.4 l of this inoculum was added separately to each of the 10 l carboys (pooled coral and control filtrate) to create a 5:2 ratio of filtrate: inoculum. After this addition, each carboy was mixed and a suite of samples were collected for different analyses including cell counts, inorganic and organic macronutrient quantification, and 16S rRNA gene sequencing. This collection marked the first timepoint (0 hours [hrs]) for the exudate uptake experiment. For the *G. ventalina* experiment, reef seawater inoculum was collected from Tektite reef (Table S1).

For each experiment, coral metabolite or control filtrate seawater were transferred into separate 1 l acid-washed polycarbonate bottles (6 bottles per treatment). Within each treatment (coral and control), 3 of the 6 bottles were blackened to block light. The bottles were placed into a flow-through seawater table. PAR readings in the seawater table for the *P. astreoides* and *G. ventalina* experiments ranged from 250 – 1000 and 164 – 530 µmol quanta m<sup>-2</sup> s<sup>-1</sup>, respectively, with variation caused by shading and cloud cover. Over the course of 48 hours, samples were collected for cell counts (1 ml) at all timepoints (0, 12, 24, 36, and 48 hrs), macronutrient analyses (30 – 40 ml) at 0 and 48 hrs, and microbial community analyses (60 – 300 ml) at 0, 24, and 48 hrs.

Samples (40 ml) collected for total organic carbon (TOC) and total nitrogen (TN) analyses were aliquoted into combusted, borosilicate EPA vials and acidified using 75 µl of phosphoric acid. Samples were stored at room temperature for two weeks and then refrigerated at 4 °C until analysis.

For inorganic nutrient analyses, seawater (30 ml) was allocated into acid-washed, polypropylene bottles (Nalgene) and frozen at -20 °C.

## Data Processing Description

Samples collected for total organic carbon (TOC) and total nitrogen (TN) analyses were investigated at the Woods Hole Oceanographic Institution using a Shimadzu TOC-VCSH total organic carbon analyzer with a TNM-1 module [4].

Samples collected for inorganic nutrient analyses were shipped to Oregon State University and the concentrations of nitrite, nitrite + nitrate, ammonium, silicate, and phosphate were obtained using a continuous segmented flow system (described in 18). Total organic nitrogen concentrations were obtained by subtracting the sum of the inorganic nitrogen species (nitrite and nitrate + ammonium) from the total nitrogen concentrations. Values measured below the detection limits of the instruments (ammonium = 0.02 M, phosphate = 0.01 M, nitrite + nitrate = 0.07 M, nitrite = 0.01 M) were reported as zero.

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## Data Files

File
<b>macronutrient.csv</b> (Comma Separated Values (.csv), 3.71 KB) MD5:6c8e4e700436d213340f8e68a21c9f82
Primary data file for dataset ID 865193

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## Related Publications

Apprill, A., & Rappé, M. (2011). Response of the microbial community to coral spawning in lagoon and reef flat environments of Hawaii, USA. *Aquatic Microbial Ecology*, 62(3), 251–266. doi:[10.3354/ame01471](https://doi.org/10.3354/ame01471)

*Methods*

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*

*Methods*

Callahan, B. J., McMurdie, P. J., Rosen, M. J., Han, A. W., Johnson, A. J. A., & Holmes, S. P. (2016). DADA2: High-resolution sample inference from Illumina amplicon data. *Nature Methods*, 13(7), 581–583. doi:[10.1038/nmeth.3869](https://doi.org/10.1038/nmeth.3869)

*Software*

*Software*

Campbell, L., & Vaulot, D. (1993). Photosynthetic picoplankton community structure in the subtropical North Pacific Ocean near Hawaii (station ALOHA). *Deep Sea Research Part I: Oceanographic Research Papers*, 40(10), 2043–2060. doi:[10.1016/0967-0637\(93\)90044-4](https://doi.org/10.1016/0967-0637(93)90044-4)

*Methods*

*Methods*

Campbell, L., Nolla, H. A., & Vaulot, D. (1994). The importance of *Prochlorococcus* to community structure in the central North Pacific Ocean. *Limnology and Oceanography*, 39(4), 954–961. doi:[10.4319/lo.1994.39.4.0954](https://doi.org/10.4319/lo.1994.39.4.0954)

*Methods*

Davis, N. M., Proctor, D. M., Holmes, S. P., Relman, D. A., & Callahan, B. J. (2018). Simple statistical identification and removal of contaminant sequences in marker-gene and metagenomics data. *Microbiome*, 6(1). doi:[10.1186/s40168-018-0605-2](https://doi.org/10.1186/s40168-018-0605-2)

*Methods*

*Methods*

Hansell, D. A., & Carlson, C. A. (2001). Biogeochemistry of total organic carbon and nitrogen in the Sargasso Sea: control by convective overturn. *Deep Sea Research Part II: Topical Studies in Oceanography*, 48(8-9), 1649–1667. doi:[10.1016/S0967-0645\(00\)00153-3](https://doi.org/10.1016/S0967-0645(00)00153-3)

*Methods*

*Methods*

Martin BD, Witten D, Willis AD (2019) Modeling microbial abundances and dysbiosis with beta-binomial regression. arXiv:[1902.02776](https://arxiv.org/abs/1902.02776) [stat].

*Methods*

*Methods*

Monger, B. C., & Landry, M. R. (1993). Flow Cytometric Analysis of Marine Bacteria with Hoechst 33342 †. *Applied and Environmental Microbiology*, 59(3), 905–911. doi:[10.1128/aem.59.3.905-911.1993](https://doi.org/10.1128/aem.59.3.905-911.1993)

*Methods*

*Methods*

Oksanen J. *Vegan: ecological diversity*. R Packag Version 2.4-4 . 2017. [https://cran.r-project.org/src/contrib/Archive/vegan/vegan\\_2.4-4.tar.gz](https://cran.r-project.org/src/contrib/Archive/vegan/vegan_2.4-4.tar.gz)

*Software*

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/10.1111/1462-2920.13023)

*Methods*

*Methods*

Weber, L., Gonzalez-Díaz, P., Armenteros, M., & Apprill, A. (2019). The coral ecosphere: A unique coral reef habitat that fosters coral-microbial interactions. *Limnology and Oceanography*, 64(6), 2373–2388. doi:[10.1002/lno.11190](https://doi.org/10.1002/lno.11190)

*Methods*

*Methods*

Weber, L., Soule, M. K., Longnecker, K., Becker, C. C., Huntley, N., Kujawinski, E. B., & Apprill, A. (2022).

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## Parameters

Parameter	Description	Units
Sample_ID	Sample ID	unitless
sample_type	Sample Type	unitless
LD	Samples were either incubated in the presence or absence of light.	unitless
Incubation	description	unitless
Time	This metadata category reflects the timepoints when each sample was collected. The timepoints include na (before the beginning of the experiment), T0 (at 0 hr after the experiment was started) and so on.	unitless
Total_Organic_Carbon	Total organic carbon	micrometers (um)
Total_Nitrogen	Total nitrogen	micrometers (um)
Total_Organic_Nitrogen	Total organic nitrogen	micrometers (um)
Phosphate	Phosphate	micrometers (um)
Nitrite_Nitrat	Nitrite and Nitrate	micrometers (um)
Silicate	Silicate	micrometers (um)
Nitrite	Nitrite and Nitrate	micrometers (um)
Ammonium	Ammonium	micrometers (um)
Nitrate	Nitrate	micrometers (um)

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## Project Information

## Signature exometabolomes of Caribbean corals and influences on reef picoplankton (Coral Exometabolomes)

**Coverage:** U.S. Virgin Islands

### *NSF Award Abstract:*

Coral reefs are some of the most diverse and productive ecosystems in the ocean. Globally, reefs have declined in stony (reef-building) coral abundance due to environmental variations, and in the Caribbean this decline has coincided with an increase in octocoral (soft coral) abundance. This phase shift occurring on Caribbean reefs may be impacting the interactions between the sea floor and water column and particularly between corals and picoplankton. Picoplankton are the microorganisms in the water column that utilize organic matter released from corals to support their growth. These coral-picoplankton interactions are relatively unstudied, but could have major implications for reef ecology and coral health. This project will take place in the U.S. territory of the Virgin Islands (USVI) and will produce the first detailed knowledge about the chemical diversity and composition of organic matter released from diverse stony coral and octocoral species. This project will advance our understanding of coral reef microbial ecology by allowing us to understand how different coral metabolites impact picoplankton growth and dynamics over time. The results from this project will be made publically accessible in a freely available online magazine, and USVI minority middle and high school students will be exposed to a lesson about chemical-biological interactions on coral reefs through established summer camps. This project will also contribute to the training of USVI minority undergraduates as well as a graduate student.

Coral exometabolomes, which are the sum of metabolic products of the coral together with its microbiome, are thought to structure picoplankton communities in a species-specific manner. However, a detailed understanding of coral exometabolomes, and their influences on reef picoplankton, has not yet been obtained. This project will utilize controlled aquaria-based experiments with stony corals and octocorals, foundational species of Caribbean reef ecosystems, to examine how the exometabolomes of diverse coral species differentially influence the reef picoplankton community. Specifically, this project will capitalize on recent developments in mass spectrometry-based metabolomics to define the signature exometabolomes of ecologically important and diverse stony corals and octocorals. Secondly, this project will determine how the exometabolomes of these corals vary with factors linked to coral taxonomy as well as the coral-associated microbiome (Symbiodinium algae, bacteria and archaea). With this new understanding of coral exometabolomes, the project will then apply a stable isotope probe labeling approach to the coral exometabolome and will examine if and how (through changes in growth and activity) the seawater picoplankton community incorporates coral exometabolomes from different coral species over time. This project will advance our ability to evaluate the role that coral exometabolomes play in contributing to benthic-picoplankton interactions on changing Caribbean reefs.

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1736288</a>

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