A characterization of microbes at the San Pedro Ocean Time-series (SPOT) from 2005 to 2018, using SSU rRNA gene sequencing from two size fractions, with a universal primer set that amplifies from prokaryotes and eukaryotes

Website: https://www.bco-dmo.org/dataset/885982
Data Type: Cruise Results, experimental
Version: 1
Version Date: 2022-12-29

Project
» Protistan, prokaryotic, and viral processes at the San Pedro Ocean Time-series (SPOT)

Contributors	Affiliation	Role
Fuhrman, Jed A.	University of Southern California (USC)	Principal Investigator
Yeh, Yi-Chun	University of Southern California (USC)	Scientist
Furtado, Laura	University of Southern California (USC)	Technician
Rauch, Shannon	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
This study aims to characterize microbes at the San Pedro Ocean Time-series (SPOT) from 2005 to 2018, using small subunit (SSU) rRNA gene sequencing from two size fractions (0.2-1 and 1-80 µm), with a universal primer set that amplifies both prokaryotic 16S and eukaryotic 18S rRNA genes. This allows for direct comparisons of diversity patterns in a single set of analyses. This dataset includes National Center for Biotechnology Information (NCBI) accession numbers and related sample information.

Table of Contents

Coverage

Spatial Extent: **Lat**:33.33 **Lon**:-118.24
Temporal Extent: 2005-01-19 - 2018-07-18

Acquisition Description

Monthly San Pedro Ocean Time-series (SPOT) cruises on R/V Yellowfin were conducted in the San Pedro Channel, off the coast of Los Angeles, California, USA (33 N, 118 W). Samples were collected monthly from five depths, including 5 meters (m), deep chlorophyll maximum (DCM), 150m, 500m, and 890 m, between the years 2005 and 2018. Ten to fifteen liters of seawater was sequentially filtered through an 80-micrometer (µm) mesh, a 1-µm A/E filter (Pall, Port Washington, NY), and a 0.2-µm Durapore filter (ED Millipore, Billerica, MA). Filters were stored at -80° Celsius (C) until DNA extraction. Durapore filters (collecting material 0.2 to 1 µm) were used for free-living prokaryotic community analysis, and A/E filters (collecting material between 1 to 80

µm) were used to analyze phytoplankton, microzooplankton, and particle-associated or larger prokaryotic communities. DNA was extracted from the Durapore filters using a hot SDS, phenol/chloroform/isoamyl alcohol, ethanol precipitation extraction protocol as described by Fuhrman et al. (1988). DNA on the A/E filters was extracted using a NaCl/CTAB bead-beating extraction protocol as described by Lie et al. (2013) with slight modification by adding an ethanol precipitation step after lysis to reduce the volume of crude extract, which helps minimize DNA loss during the subsequent purification.

The V4-V5 hyper-variable region of the 16S and 18S rRNA genes were amplified simultaneously using a universal primer set 515Y (GTGYCAGCMGCCGCGGTAA) and 926R (CCGYCAATTYMTTTRAGTTT). All DNA samples were amplified and purified using the same conditions described in Yeh et al. 2021). Purified PCR products were pooled in equal amount and then sequenced on Illumina HiSeq 2500 in PE250 mode or MiSeq PE300.

Processing Description

The code used for processing the raw sequence data and getting the reads into ASVs that can be used for downstream community composition analyses has been saved in Zenodo with DOI 10.5281/zenodo.7340378.

BCO-DMO Processing:
- concatenated two separate .csv files (one per BioProject) into one dataset.

[table of contents | back to top]

Related Publications

Fuhrman, J. A., Comeau, D. E., Hagström, Å., & Chan, A. M. (1988). Extraction from Natural Planktonic Microorganisms of DNA Suitable for Molecular Biological Studies. Applied and Environmental Microbiology, 54(6), 1426–1429. https://doi.org/10.1128/aem.54.6.1426-1429.1988
Methods

Lie, A., Kim, D., Schnetzer, A., & Caron, D. (2013). Small-scale temporal and spatial variations in protistan community composition at the San Pedro Ocean Time-series station off the coast of southern California. Aquatic Microbial Ecology, 70(2), 93–110. https://doi.org/10.3354/ame01652
Methods

McNichol, J., Aleman, M., & fletchec99. (2022). jcmcnch/eASV-pipeline-for-515Y-926R: qiime2-2019.4 archive release (Version v1.0.0) [Computer software]. Zenodo. https://doi.org/10.5281/ZENODO.7340378
Software

Yeh, Y., McNichol, J., Needham, D. M., Fichot, E. B., Berdjeb, L., & Fuhrman, J. A. (2021). Comprehensive single-PCR 16S and 18S rRNA community analysis validated with mock communities, and estimation of sequencing bias against 18S. Environmental Microbiology, 23(6), 3240–3250. Portico. https://doi.org/10.1111/1462-2920.15553
Methods

Yeh, Y.-C., & Fuhrman, J. A. (2022). Contrasting diversity patterns of prokaryotes and protists over time and depth at the San-Pedro Ocean Time series. ISME Communications, 2(1). https://doi.org/10.1038/s43705-022-00121-8
General

Yeh, Y.-C., & Fuhrman, J. A. (2022). Effects of phytoplankton, viral communities, and warming on free-living and particle-associated marine prokaryotic community structure. Nature Communications, 13(1). https://doi.org/10.1038/s41467-022-35551-4
General

[table of contents | back to top]

Related Datasets

IsRelatedTo

UNIVERSITY OF SOUTHERN CALIFORNIA. Comprehensive single-PCR 16S and 18S rRNA community analysis validated with mixed 16S and 18S rRNA mock communities and denoising algorithms. 2019/12. In: BioProject [Internet]. Bethesda, MD: National Library of Medicine (US), National Center for Biotechnology Information; 2011-. Available from: http://www.ncbi.nlm.nih.gov/bioproject/PRJEB35673. NCBI:BioProject: PRJEB35673. https://www.ncbi.nlm.nih.gov/bioproject/PRJEB35673

UNIVERSITY OF SOUTHERN CALIFORNIA. Monthly time-series analysis of the marine microbial community from two size fractions at the San Pedro Ocean Time-series station. 2021/11. In: BioProject [Internet]. Bethesda, MD: National Library of Medicine (US), National Center for Biotechnology Information; 2011-. Available from: http://www.ncbi.nlm.nih.gov/bioproject/PRJEB48162. NCBI:BioProject: PRJEB48162. https://www.ncbi.nlm.nih.gov/bioproject/PRJEB48162

[table of contents | back to top]

Parameters

Parameter	Description	Units
study_accession	National Center for Biotechnology Information (NCBI) project identifier	unitless
sample_accession	National Center for Biotechnology Information (NCBI) sample accession number	unitless
secondary_sample_accession	National Center for Biotechnology Information (NCBI) secondary sample accession number	unitless
run_accession	National Center for Biotechnology Information (NCBI) run accession number	unitless
tax_id	National Center for Biotechnology Information (NCBI) taxon ID	unitless
sequence_category	Sample category type	unitless
instrument_platform	Sequencing platform ("ILLUMINA")	unitless
instrument_model	Sequencing instrument model	unitless
library_layout	Library layout ("PAIRED")	unitless
library_strategy	Library strategy ("AMPLICON")	unitless
library_source	Library source ("METAGENOMIC")	unitless
library_selection	Library selection ("PCR")	unitless
study_title	Study title	unitless
fastq_ftp	Generated FASTQ files: FTP	unitless
submitted_ftp	Submitted files: FTP	unitless
submitted_format	File format ("FASTQ")	unitless
sample_title	Sample name	unitless

[table of contents | back to top]

Instruments

Dataset-specific Instrument Name	Illumina HiSeq 2500 sequencer
Generic Instrument Name	Automated DNA Sequencer
Dataset-specific Description	Purified PCR products were sequenced on an Illumina HiSeq 2500 in PE250 mode or MiSeq PE300.
Generic Instrument Description	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.

Dataset-specific Instrument Name	Illumina MiSeq sequencer
Generic Instrument Name	Automated DNA Sequencer
Dataset-specific Description	Purified PCR products were sequenced on an Illumina HiSeq 2500 in PE250 mode or MiSeq PE300.
Generic Instrument Description	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.

[table of contents | back to top]

Deployments

SPOT_Yellowfin_Cruises

Website	https://www.bco-dmo.org/deployment/754348
Platform	R/V Yellowfin
Start Date	2005-01-19
End Date	2018-07-18
Description	San Pedro Ocean Time Series (SPOT) station (33°33′N, 118°24′W) R/V Yellowfin, monthly SPOT cruises in the San Pedro Channel Deployment: SPOT Platform: RV Yellowfin Platform Type: vessel

[table of contents | back to top]

Project Information

Protistan, prokaryotic, and viral processes at the San Pedro Ocean Time-series (SPOT)

Coverage: San Pedro Channel off the coast of Los Angeles

Planktonic marine microbial communities consist of a diverse collection of bacteria, archaea, viruses, protists (phytoplankton and protozoa) and small animals (metazoan). Collectively, these species are responsible for virtually all marine pelagic primary production where they form the basis of food webs and carry out a large fraction of respiratory processes. Microbial interactions include the traditional role of predation, but recent research recognizes the importance of parasitism, symbiosis and viral infection. Characterizing the response of pelagic microbial communities and processes to environmental influences is fundamental to understanding and modeling carbon flow and energy utilization in the ocean, but very few studies have attempted to study all of these assemblages in the same study. This project is comprised of long-term (monthly) and short-term (daily) sampling at the San Pedro Ocean Time-series (SPOT) site. Analysis of the resulting datasets investigates co-occurrence patterns of microbial taxa (e.g. protist-virus and protist-prokaryote interactions, both positive and negative) indicating which species consistently co-occur and potentially interact, followed by examination gene expression to help define the underlying mechanisms. This study augments 20 years of baseline studies of microbial abundance, diversity, rates at the site, and will enable detection of low-frequency changes in composition and potential ecological interactions among microbes, and their responses to changing environmental forcing factors. These responses have important consequences for higher trophic levels and ocean-atmosphere feedbacks. The broader impacts of this project include training graduate and undergraduate students, providing local high school student with summer lab experiences, and PI presentations at local K-12 schools, museums, aquaria and informal learning centers in the region. Additionally, the PIs advise at the local, county and state level regarding coastal marine water quality.

This research project is unique in that it is a holistic study (including all microbes from viruses to small metazoa) of microbial species diversity and ecological activities, carried out at the SPOT site off the coast of southern California. In studying all microbes simultaneously, this work aims to identify important ecological interactions among microbial species, and identify the basis(es) for those interactions. This research involves (1) extensive analyses of prokaryote (archaean and bacterial) and eukaryote (protistan and micro-metazoan) diversity via the sequencing of marker genes, (2) studies of whole-community gene expression by eukaryotes and prokaryotes in order to identify key functional characteristics of microorganismal groups and the detection of active viral infections, and (3) metagenomic analysis of viruses and bacteria to aid interpretation of transcriptomic analyses using genome-encoded information. The project includes exploratory metatranscriptomic analysis of poorly-understood aphotic and hypoxic-zone protists, to examine their stratification, functions and hypothesized prokaryotic symbioses.

[table of contents | back to top]

Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1737409
Gordon and Betty Moore Foundation: Marine Microbiology Initiative (MMI)	GBMF3779
Simons Foundation (Simons)	CBIOMES 549943

[table of contents | back to top]