SPOT Microbial Observatory Bacterial ARISA relative peak abundance data from 2014 (Bacterial, Archaeal, and Protistan Biodiversity project, Marine Viral Dynamics project, Mar. Microbial Communities project)

Website: https://www.bco-dmo.org/dataset/553345
Version: 2015-01-29

Project
» Pattern and Process in Marine Bacterial, Archaeal, and Protistan Biodiversity, and Effects of Human Impacts (Bacterial, Archaeal, and Protistan Biodiversity)
» Marine viral dynamics and incorporation into microbial association networks (Marine Viral Dynamics)
» MO: Assembly of Marine Microbial Communities (Mar. Microbial Communities)

Program
» Dimensions of Biodiversity (Dimensions of Biodiversity)

Contributors	Affiliation	Role
Fuhrman, Jed A.	University of Southern California (USC-WIES)	Principal Investigator
Chow, Cheryl-Emiliane T.	University of Southern California (USC-WIES)	Contact
Copley, Nancy	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

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Dataset Description

Bacterial community composition was determined by Automated Ribosomal Intergenic Spacer Analysis (ARISA) (Fisher and Triplett, 1999; Brown et al., 2005; Chow et al., 2013; Needham et al., 2013). ARISA products were run in duplicate on non-adjacent lanes on an ABI377 by slab gel electrophoresis with internal size standards (Bioventures, Murfreesboro, TN, USA) every 25 bp (50–900 bp) or 50 bp (900–1400 bp). Peaks were identified in DAx (van Mierlo, Inc, Eindhoven, The Netherlands). Fragments (400–1210 bp) were rounded to the nearest 0.1 bp and dynamically binned (Ruan, et al., 2006b; Chow and Fuhrman, 2012). The resulting bins were manually curated to merge bins <0.1 bp wide with the nearest neighbor. ARISA OTUs were assigned an identity by matching ARISA lengths with known sequences and their ARISA products (Chow et al., 2013; Needham et al., 2013. Unique fragment lengths were considered as individual OTUs. Relative abundance of each OTU was calculated by dividing a peak's area by the total area within the monthly fingerprint. Bacterial OTUs <0.1% of the community were removed from further analysis, and the remaining peaks were normalized by sample to determine relative abundance per month; each community thus totaled to 100%.

Relevant References:

* Chow CET, Kim DY, Sachdeva R, Caron DA, and Fuhrman JA. (2014). Top-down controls on bacterial community structure: microbial network analysis of bacteria, T4-like viruses and protists. ISME Journal, 8: 816-829

and

Brown MV, Schwalbach MS, Hewson I, Fuhrman JA.(2005). Coupling 16S-ITS rDNA clone libraries and automated ribosomal intergenic spacer analysis to show marine microbial diversity: development and application to a time series. Environ Microbiol 7: 1466–1479.

Chow C-ET, Fuhrman JA. (2012). Seasonality and monthly dynamics of marine myovirus communities. Environ Microbiol 14: 2171–2183.

Chow C-ET, Sachdeva R, Cram JA, Steele JA, Needham DM, Patel A et al. (2013). Temporal variability and coherence of euphotic zone bacterial communities over a decade in the Southern California Bight. ISME J; epub ahead of print 18 July 2013; doi:10.1038/ismej.2013.122.

Countway PD, Gast RJ, Savai P, Caron DA. (2005). Protistan diversity estimates based on 18S rDNA from seawater incubations in the western north Atlantic1. J Eukaryot Microbiol 52: 95–106.

Countway PD, Vigil PD, Schnetzer A, Moorthi SD, Caron DA. (2010). Seasonal analysis of protistan community structure and diversity at the USC Microbial Observatory (San Pedro Channel, North Pacific Ocean). Limnol Oceanogr 55: 2381–2396.

Fisher MM, Triplett EW. (1999). Automated approach for ribosomal intergenic spacer analysis of microbial diversity and its application to freshwater bacterial communities. Appl Environ Microbiol 65: 4630–4636.

Methods & Sampling

Seawater (~20 l) was collected approximately monthly at 0 or 5m at the University of Southern California's Microbial Observatory at SPOT (33' 331 N, 118' 241 W) and filtered for free-living protistan (0.7–20 mm), bacterial (0.22–1 mm) and viral (30 kDa– 0.22 mm) community DNA from March 2008 to January 2011, as previously described (Countway et al., 2005; Fuhrman et al., 2006; Vigil et al., 2009; Countway et al., 2010; Steele et al., 2011; Chow and Fuhrman, 2012; Kim et al., 2012; Chow et al., 2013). Molecular data was unavailable for October 2008 (virus), January 2009 (all), March 2009 (bacteria), October–November 2009 (bacteria) and January 2011 (protist). Bulk seawater samples were also collected and analyzed for bacterial and viral abundance by SYBR green epifluorescence microscopy, bacterial production by thymidine and leucine incorporation, and nutrient concentrations using colorimetric methods (Chow et al., 2013).

Cruise logs are available at: http://dornsife.usc.edu/spot/document-access/

Data Processing Description

Peaks were identified in DAx (van Mierlo, Inc, Eindhoven, The Netherlands). Fragments (50–500 bp) were rounded to the nearest 0.1 bp and dynamically binned (Ruan, et al., 2006b; Chow and Fuhrman, 2012). The resulting bins were manually curated to merge bins <0.1 bp wide with the nearest neighbor. Terminal fragments from in silico analysis of publicly available T4-like viral genomes were used to assign identities to environmental g23-TRFLP OTUs. Unique fragment lengths were considered as individual OTUs. Relative abundance of each OTU was calculated by dividing a peak's area by the total area within the monthly fingerprint. Viral OTUs <0.1% of the community were removed from further analysis, and the remaining peaks were normalized by sample to determine relative abundance per month; each community thus totaled to 100%.

BCO-DMO Processing:

original file: Cheryl.data.inverted.for.BCO.DMO.xlsx
- added conventional header with dataset name, PI name, version date
- renamed parameters to BCO-DMO standard or more descriptive terms
- replaced na with nd
- reformated data from m/d/yyyy to yyyy-mm-dd
- reduced number of digits for most parameters

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Data Files

File
SPOT_virus_arisa.csv (Comma Separated Values (.csv), 50.20 KB) MD5:fc9fe292ee9a01eec4f64a12b015784b Primary data file for dataset ID 553345

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Parameters

Parameter	Description	Units
cruise_id	cruise identification	unitless
platform	vessel name	unitless
date	date of cruise	yyyy-mm-dd
time_start	cruise start time	HH:MM
time_end	cruise end time	HH:MM
lat	station latitude; north is positive	decimal degrees
lon	station longitude; east is positive	decimal degrees
ARISA__####_#	Bacterial ARISA relative peak abundance	proportion

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Instruments

Dataset-specific Instrument Name	Automated Sequencer
Generic Instrument Name	Automated DNA Sequencer
Generic Instrument Description	A DNA sequencer is an instrument that determines the order of deoxynucleotides in deoxyribonucleic acid sequences.

Dataset-specific Instrument Name	
Generic Instrument Name	Fluorescence Microscope
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of fluorescence and phosphorescence instead of, or in addition to, reflection and absorption of visible light. Includes conventional and inverted instruments.

Dataset-specific Instrument Name	Niskin bottle
Generic Instrument Name	Niskin bottle
Generic Instrument Description	A Niskin bottle (a next generation water sampler based on the Nansen bottle) is a cylindrical, non-metallic water collection device with stoppers at both ends. The bottles can be attached individually on a hydrowire or deployed in 12, 24, or 36 bottle Rosette systems mounted on a frame and combined with a CTD. Niskin bottles are used to collect discrete water samples for a range of measurements including pigments, nutrients, plankton, etc.

Dataset-specific Instrument Name	Thermal Cycler
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 http://serc.carleton.edu/microbelife/research_methods/genomics/pcr.html)

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Deployments

lab_Fuhrman_2014

Website	https://www.bco-dmo.org/deployment/535519
Platform	USC
Start Date	2014-10-17
End Date	2014-10-17
Description	Microbial diversity laboratory studies. Monthlly cruises to collect water samples in Los Angeles, California area.

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Project Information

Pattern and Process in Marine Bacterial, Archaeal, and Protistan Biodiversity, and Effects of Human Impacts (Bacterial, Archaeal, and Protistan Biodiversity)

Website: http://dornsife.usc.edu/labs/usc-microbial-observatory/

Coverage: San Pedro Ocean Time Series; approx. 33N, 118W

Description from NSF award abstract:
Bacteria, Archaea, and Protists dominate global elemental cycling and are immensely diverse genetically, taxonomically, and functionally. Yet the extent of marine microbial diversity, its patterns, and relationships among genetic, taxonomic, and functional diversity are very poorly characterized, even though the ocean covers 70% of the planet's surface. Among the least well known variables is the effect of human impacts on native marine microbial systems, although it is recognized that impacted systems are more prone to events like harmful algal blooms. Knowledge of these relationships and impacts are necessary to anticipate the responses of biota to global changes and feedback mechanisms that may alter the extents, rates, and even pathways of such changes. This project will expand upon an existing NSF-funded 10+-year monthly ocean time series (Microbial Observatory) that has focused on a single site midway between Los Angeles and Santa Catalina Island, to also include quarterly sampling adjacent to the impacted LA Harbor region to the barely-impacted Catalina coast. USC already runs facilities in LA Harbor and Catalina, with daily boats between (no cost). Measurements include (1) Genetic diversity: high throughput DNA sequences of "housekeeping" and functional genes. (2) Taxonomic diversity: high throughput tag sequences of small subunit ribosomal RNA genes, flow cytometry, automated image analysis (3) Functional Diversity: (a) Functional measurements (carbon fixation and respiration rates, microbial growth and grazing rates, cell size, morphology, and biomass variations), (b) distribution and expression of particular target functional genes involved with processes central to the cycles of carbon, nitrogen, and sulfur, (c) exploratory metatranscriptomics to explore functionalities that were not anticipated. (4) Integrating these: Multivariate statistical and network approaches including newly developed techniques (e.g. Bayesian networks to examine cause-effect relationships), and high speed computational approaches to assess the relationships among the genetic, taxonomic, and functional aspects of biodiversity observed. The PIs will also examine the collected data for signatures and specific effects (on organism identity and functions) associated with human impacted harbor site vs. the relatively pristine one.

The PIs will use network and time series analysis, along with other statistical tools to integrate "classical" microbial and oceanographic rate process measurements, flow cytometric and microscopic characterizations of communities, along with targeted as well as untargeted metagenomics and metatranscriptomics to relate genetic and taxonomic diversity with specific functions (at organismal, food web, and system levels). For example, they should be able to determine how different variants of particular taxa (e.g. at resolution levels ranging from what might be considered near the subspecies to genus levels) would differ in their association with particular measured functions, functional genes, or particular other taxa - or they might see how particular clusters of related organisms behave similarly or differently in their associations. This project offers an unprecedented and potentially transformative opportunity to combine and integrate measurements of genetic, taxonomic, and functional diversity along with direct measurements of system function in a well studied marine system that includes a gradient from one of the world's busiest harbors to a largely pristine ocean habitat. Far beyond just describing the distributions of organisms and functions (itself a necessary first step), they will specifically link spatial and temporal variations in a variety of functions with variations in genetic and taxonomic community composition.

Marine viral dynamics and incorporation into microbial association networks (Marine Viral Dynamics)

Website: http://dornsife.usc.edu/labs/fuhrmanlab/research/

Coverage: Southern California between Los Angeles and Santa Catalina Island; Approx. 33.5N, 118.5 W

Description from NSF award abstract:

Marine microbes are tremendously abundant and are major players and driving forces in global biogeochemical cycles of carbon, nitrogen, phosphorus, and iron. We learned over the past two decades that viruses are pervasive elements in marine systems, with significant ecological, biogeochemical, genetic, and evolutionary effects on cellular marine organisms, but we have remarkably little information about the dynamics of marine viral community structure and how it relates to the community structure of their hosts (largely bacteria and phytoplankton). Such information is critical for developing proper conceptual and practical models of the roles of viruses and how these change over time and space. The goals of this project are:
(1) primarily, to characterize a significant subset of the natural virus community and its dynamics, along with bacterial host communities, as they change over daily to monthly time scales at the USC well-studied marine Microbial Observatory site (midway between Los Angeles and Santa Catalina Island), testing hypotheses regarding repeating patterns, host range effects, and taxa-time relationships, and
(2) secondarily, to incorporate these viruses into microbial association networks by statistically connecting particular types of viruses to specific potential hosts.

Approaches for this study include:
(a) nested daily, weekly, and monthly collection of bacteria and viruses for nucleic acid samples,
(b) amplification of conserved genes, as proxy phylogenetic markers, from a few moderately-well-characterized broad viral groups previously readily found in seawater (i.e. the T4-like myoviruses, T7-like podoviruses), as well as bacterial rRNA genes,
(c) extensive sequencing, after screening by community fingerprinting, from the mixed amplified products,
(d) binning of the sequences or fingerprint fragments into operational taxonomic units (OTUs) at different levels of resolution,
(e) evaluation of the results with statistical approaches to examine temporal patterns, relationships (including time-lagged ones) with other viral OTUs, bacteria, protists (monthly only), and environmental parameters,
(f) incorporating the viral OTUs mathematically into microbial association networks.

Data on environmental parameters, bacteria, and protists are already being collected monthly for an existing Microbial Observatory, so the viral work is complementary to this project, providing a major value-added component. Similarly, this project will add selected daily and weekly microbial data to the Microbial Observatory. Data from the literature and from the PI's preliminary results show they have the technology and capability to meet the first goal, and to our knowledge this would be the first such data set of its scope and kind. The investigators have already published in 2006 that the bacterial communities at the 5m depth of this site show a predictable repeating annual cycle in bacterial community composition, so the expectation of a predictable repeating viral community is not unreasonable. They also have some preliminary data showing some repeated viral occurrences. The second goal requires that there are indeed significant statistical relationships between the viruses and other measured parameters, which the PI anticipates to be the case, but of course cannot predict; if they cannot be demonstrated, this result itself would be informative and would constrain the possible modes of microbial/viral interactions.

MO: Assembly of Marine Microbial Communities (Mar. Microbial Communities)

Coverage: San Pedro Channel and Basin off southern California coast

Abstract:

The USC Microbial Observatory was established in 2000. The research focus of this observatory is an investigation of the microbial diversity and microbial community composition at a study site in the San Pedro Channel and Basin off the coast of southern California. The Channel area encompasses a diversity of coastal ocean habitats. The near-coast region borders one of the most highly urbanized areas of the country (greater Los Angeles) while open ocean waters impinge on the Channel Island archipelago that extends to within 30 km of the mainland. The San Pedro Basin is a deep-water environment (approximately 890 m) that exhibits very low oxygen concentration. The overarching objective of this project is the derivation of fundamental understanding of how microbial communities in the ocean are organized spatially (with depth) and temporally (at scales of months-to-years), and how environmental and biological factors shape this organization. The basic premise of the research is that "guilds" or "consortia" of microbial species exist that constitute functional subunits within the huge diversity of taxa that comprise planktonic microbial communities. The microbial species forming these guilds are functionally interdependent, and act as ecological units that replace one another in time and space as environmental conditions change. The program consists of monthly sampling at four depths in order to document the abundance, biomass and species composition of all planktonic

microorganisms at the mid-channel sampling station. A variety of microscopical and molecular biological approaches are employed to examine archaeal, bacterial and microeukaryote (microalgal, protozoan, micrometazoan) diversity. The observatory is unique in that it entails an assessment of the complete spectrum of microorganisms (from viruses to the largest protists) in the water column. Genetic fingerprinting of the total microbial community is the primary tool for revealing the trophic roles and relationships among microbial taxa (predation, mutualism, commensalism, parasitism/infection), and to generate hypotheses on the interdependences among these species. Experimental studies involve manipulative food web experiments to test hypotheses concerning the relationships and interactions among the various microbial species. The data support extensive statistical analyses to identify relationships between microbial taxa, and with environmental parameters.

This research program strives to develop a fundamental understanding of the factors controlling the structure of microbial communities in aquatic ecosystems. Therefore, the results have far-reaching consequences for predicting biogeochemical processes mediated by microbial activities in nature. The project also incorporates a strong educational component aimed at reaching students ranging from elementary school children to graduate students. The information resulting from the research is incorporated into undergraduate and graduate courses taught by the principal investigators, and both types of students participate actively in the research. In addition, the principal investigators and graduate students supported by this project participate directly in an ongoing teacher education program (Centers for Ocean Science Education Excellence; COSEE-West) that reaches middle and high school students, many of whom are Hispanic, African-American or other ethnic minorities and most of whom are economically disadvantaged. This education/outreach goal is accomplished through an existing teacher enhancement and student enrichment program that incorporates the research from this microbial observatory into a learning experience that enhances student awareness of environmental science, microbiology and the natural world. The observatory principal investigators work is featured and publicly available on the internet as a part of the USC Microbial Observatory Website at http://www.usc.edu/microbialobservatory)

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Program Information

Dimensions of Biodiversity (Dimensions of Biodiversity)

Website: http://www.nsf.gov/funding/pgm_summ.jsp?pims_id=503446

Coverage: global

(adapted from the NSF Synopsis of Program)
Dimensions of Biodiversity is a program solicitation from the NSF Directorate for Biological Sciences. FY 2010 was year one of the program. [MORE from NSF]

The NSF Dimensions of Biodiversity program seeks to characterize biodiversity on Earth by using integrative, innovative approaches to fill rapidly the most substantial gaps in our understanding. The program will take a broad view of biodiversity, and in its initial phase will focus on the integration of genetic, taxonomic, and functional dimensions of biodiversity. Project investigators are encouraged to integrate these three dimensions to understand the interactions and feedbacks among them. While this focus complements several core NSF programs, it differs by requiring that multiple dimensions of biodiversity be addressed simultaneously, to understand the roles of biodiversity in critical ecological and evolutionary processes.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1136818
NSF Division of Molecular and Cellular Biosciences (NSF MCB)	MCB-0703159
NSF Division of Ocean Sciences (NSF OCE)	OCE-1031743

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