Data from common garden experiment containing three populations of T. rotula

Website: https://www.bco-dmo.org/dataset/860381
Data Type: Other Field Results
Version: 1
Version Date: 2021-09-13

Project
» Dimensions: Collaborative Research: Genetic, functional and phylogenetic diversity determines marine phytoplankton community responses to changing temperature and nutrients (Phytoplankton Community Responses)

Program
» Dimensions of Biodiversity (Dimensions of Biodiversity)

Contributors	Affiliation	Role
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Abstract
This dataset includes NCBI identifiers and sample descriptions from the common garden experiment containing three populations of T. rotula.

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Coverage

Spatial Extent: **Lat**:41.57 **Lon**:-71.38
Temporal Extent: 2018-09 - 2018-09

Methods & Sampling

Eight of the freshly-isolated *T. rotula* cells (3 cells each from PopA and PopB, 2 from PopC) were brought into culture and maintained at 18°C, ~100 µmol photons $m^{-2}s^{-1}$, and an 18:6 light to dark cycle. The isolates underwent antibiotic treatment 1-4 months before the start of the experiment. Antibiotic treatment consisted of 1.5 mL of culture and 150 mL of F/2 media with 100 µg L^{-1} of imipenem (Fisher Scientific) in 250 mL flasks on shaker tables at about 70 RPM for up to 96 hours before being transferred into F/2 media. Successful antibiotic treatment was identified by staining live cultures with 1% SYBR and visually verifying the absence of bacteria on a Nikon inverted Epifluorescent microscope both after the initial antibiotic treatments and before the start of the experiment. Isolates were maintained in exponential growth prior in F/2 media to the beginning of the experiment.

To begin the common garden experiment, 10L of whole seawater was collected from the pier from the URI Graduate School of Oceanography (41.49°, - 71.42°) on September 15, 2018. Whole seawater was filtered twice through 1µm polycarbonate track etch membrane filters (PCTE; Sterlitech) into autoclaved flasks to capture the free-living *in situ* bacterial community and remove grazers and large phytoplankton. Following filtration, 150 mL aliquots of the 1µm filtered seawater were added to sterile 250 mL Erlenmeyer flasks and amended with sterile F/10 vitamins and nutrients. All treatments were conducted in triplicate and included single strain flasks that were inoculated with a final concentration of 1,000 *T. rotula* cells mL^{-1}. We also had two control treatments: a bacterial control consisting of 150mL quadruplicates of the *in situ* bacterial community amended with F/10 vitamins and nutrients and a media control consisting of 150 mL triplicates of sterile seawater amended with F/10 vitamins and nutrients. All treatments were maintained at 18°C, 100 µmol photons $m^{-2}s^{-1}$, an 18:6 light:dark cycle, and 70 RPM on shaker tables.

On day 5, 100 mL of each culture flask was filtered onto a 0.2-µm Express Filter (Millipore), flash frozen in liquid nitrogen and kept at -80°C until DNA extraction using the Gentra Puregene Yeast/Bacteria kit (Qiagen) following the manufacturer's protocol.

The V4-V5 region of the bacterial 16S rRNA gene was amplified using the 515F-Y and 926R primers. PCR reactions were performed in triplicate 20 µl reactions containing 20 ng template DNA, 1X Taq Buffer, 0.5 µM of each primer, 200 µM of dNTPs, and 0.4 U of Taq DNA polymerase (Lucigen). The thermal cycling conditions were 2 min at 95°C, followed by 30 cycles of 1 min at 95°C, 1 min at 50°C, 30 s at 72°C, a final extension of 72°C for 10 min. Triplicate PCR reactions were pooled and gel purified. Libraries were sequenced at the Duke Center for Genomic and Computational Biology on a MiSeq 2x250bp run (Illumina).

Data Processing Description

Raw paired-end libraries were quality controlled using the ea-utils V.1.04.807 script *fastq-mcf* to a mate-pair quality mean of 25 with a 10 basepair (bp) sliding window and a minimum length of 150 bp. Forward and reverse reads were merged using the USEARCH V.8.1.1861 script *fastq_mergepairs* with a minimum overlap length of 10 bp and zero differences in the overlap region followed by clustering using Minimum Entropy Decomposition (MED) V.2.0 with standard settings. Chloroplasts were filtered using the QIIME V.1.9.1 script *filter_fasta.py*. Chimeras were identified and removed using the USEARCH V.11.0.667 script *uchime2_ref* in sensitive mode against the Silva v128 database. Silva database v128 was used to assign taxonomy to OTUs and libraries were then rarefied using the script *multiple_rarefactions_even_depth.py* ten times to an even depth of 5,827 sequences per sample and OTU tables were merged using *merge_otu_tables.py* in QIIME V.1.9.1 (10).

BCO-DMO Processing:
- renamed fields to conform with BCO-DMO naming conventions.

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Data Files

File
common_garden_expt.csv (Comma Separated Values (.csv), 3.39 KB) MD5:a2cbe7ca7ddf82acc5e96d4eada5678c Primary data file for dataset ID 860381

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Related Publications

Ahern, O. M., Whittaker, K. A., Williams, T. C., Hunt, D. E., & Rynearson, T. A. (2021). Host genotype structures the microbiome of a globally dispersed marine phytoplankton. Proceedings of the National Academy of Sciences, 118(48). https://doi.org/10.1073/pnas.2105207118
Results

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Parameters

Parameter	Description	Units
Experiment_Accession	Experiment accession number	unitless
Description	Description of 16S Library	unitless
SRA_Library_Name	Library identifier	unitless
Study_Accession	NBCI SRA accession for project	unitless
Sample_Accession	NCBI SRA library identifier	unitless
BioProject	NCBI BioProject identifier	unitless

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Instruments

Dataset-specific Instrument Name	Illumina MiSeq 2x250 bp PE reads
Generic Instrument Name	Automated DNA Sequencer
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	
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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Project Information

Dimensions: Collaborative Research: Genetic, functional and phylogenetic diversity determines

marine phytoplankton community responses to changing temperature and nutrients (Phytoplankton Community Responses)

Coverage: Narragansett Bay, RI and Bermuda, Bermuda Atlantic Time-series Study (BATS)

NSF Award Abstract:
Photosynthetic marine microbes, phytoplankton, contribute half of global primary production, form the base of most aquatic food webs and are major players in global biogeochemical cycles. Understanding their community composition is important because it affects higher trophic levels, the cycling of energy and elements and is sensitive to global environmental change. This project will investigate how phytoplankton communities respond to two major global change stressors in aquatic systems: warming and changes in nutrient availability. The researchers will work in two marine systems with a long history of environmental monitoring, the temperate Narragansett Bay estuary in Rhode Island and a subtropical North Atlantic site near Bermuda. They will use field sampling and laboratory experiments with multiple species and varieties of phytoplankton to assess the diversity in their responses to different temperatures under high and low nutrient concentrations. If the diversity of responses is high within species, then that species may have a better chance to adapt to rising temperatures and persist in the future. Some species may already be able to grow at high temperatures; consequently, they may become more abundant as the ocean warms. The researchers will incorporate this response information in mathematical models to predict how phytoplankton assemblages would reorganize under future climate scenarios. Graduate students and postdoctoral associates will be trained in diverse scientific approaches and techniques such as shipboard sampling, laboratory experiments, genomic analyses and mathematical modeling. The results of the project will be incorporated into K-12 teaching, including an advanced placement environmental science class for underrepresented minorities in Los Angeles, data exercises for rural schools in Michigan and disseminated to the public through an environmental journalism institute based in Rhode Island.

Predicting how ecological communities will respond to a changing environment requires knowledge of genetic, phylogenetic and functional diversity within and across species. This project will investigate how the interaction of phylogenetic, genetic and functional diversity in thermal traits within and across a broad range of species determines the responses of marine phytoplankton communities to rising temperature and changing nutrient regimes. High genetic and functional diversity within a species may allow evolutionary adaptation of that species to warming. If the phylogenetic and functional diversity is higher across species, species sorting and ecological community reorganization is likely. Different marine sites may have a different balance of genetic and functional diversity within and across species and, thus, different contribution of evolutionary and ecological responses to changing climate. The research will be conducted at two long-term time series sites in the Atlantic Ocean, the Narragansett Bay Long-Term Plankton Time Series and the Bermuda Atlantic Time Series (BATS) station. The goal is to assess intra- and inter-specific genetic and functional diversity in thermal responses at contrasting nutrient concentrations for a representative range of species in communities at the two sites in different seasons, and use this information to parameterize eco-evolutionary models embedded into biogeochemical ocean models to predict responses of phytoplankton communities to projected rising temperatures under realistic nutrient conditions. Model predictions will be informed by and tested with field data, including the long-term data series available for both sites and in community temperature manipulation experiments. This project will provide novel information on existing intraspecific genetic and functional thermal diversity for many ecologically and biogeochemically important phytoplankton species, estimate generation of new genetic and functional diversity in evolution experiments, and develop and parameterize novel eco-evolutionary models interfaced with ocean biogeochemical models to predict future phytoplankton community structure. The project will also characterize the interaction of two major global change stressors, warming and changing nutrient concentrations, as they affect phytoplankton diversity at functional, genetic, and phylogenetic levels. In addition, the project will develop novel modeling methodology that will be broadly applicable to understanding how other types of complex ecological communities may adapt to a rapidly warming world.

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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-1638834

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