Phytoplankton MicroTOOLs microarray-based metatranscriptomes collected from a nutrient amendment experiment conducted during R/V New Horizon Cruise NH1417 in the California Current System from 2014-08-23 to 2014-08-26

Website: https://www.bco-dmo.org/dataset/773486
Version: 1
Version Date: 2019-07-18

Project
» Oligotrophic phytoplankton community response to changes in N substrates and the resulting impact on genetic, taxonomic and functional diversity (PhytoNsubResponse)

Program
» Dimensions of Biodiversity (Dimensions of Biodiversity)

Contributors	Affiliation	Role
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Coverage

Spatial Extent: **N**:33.502 **E**:-129.37 **S**:33.1237 **W**:-129.954
Temporal Extent: 2014-08-23 - 2014-08-26

Dataset Description

Transcription changes by surface microbial communities from the California Current System in response to added nitrogen and/or iron substrates were assessed using MicroTOOLs microarrays. Samples were collected during the R/V New Horizon Cruise NH1417 in 2014. These data are available at The National Center for Biotechnology Information (NCBI) under NCBI Gene Expression Omnibus (GEO) accession number GSE130464 https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE130464. BioProject https://www.ncbi.nlm.nih.gov/bioproject/PRJNA540297. The manuscript "Phytoplankton transcriptomic and physiological responses to fixed nitrogen in the California Current System" by Shilova and Magasin et al. (2020) describes the experiment, metatranscriptomic data, and results. The previously published manuscript details other measurements obtained from the same experiment and includes phytoplankton cell counts, Fast Repetition Rate Fluorometry measurements, nutrients and Chlorophyll a concentrations, and 16S rRNA gene sequences (Shilova et al., 2017).

Methods & Sampling

Methodology:

Nutrient amendments incubation experiments were conducted during the Nitrogen Effects on Marine microOrganisms (NEMO) Cruise NH1417 in 2014 and described in Shilova et al. (2017). Briefly, seawater collected from 25 m depth at Stn. 38 on 24 August was used in on-deck perturbation experiments with different nitrogen substrates (nitrate, ammonium, urea, iron, nitrate with iron, and also with filtered deep water collected from 600 m depth). Control bottles with no added nutrients were included. The experiment setup was done under trace metal clean conditions. Experimental bottles were incubated on deck for 48 h, with RNA collected at 24 h for all treatments and also at the start of the incubation for controls (no substrate added). Experimental setup and physiological responses to added nutrients observed at 48 h are described in detail in Shilova et al. 2017.

Sampling and analytical procedures:

After 24 h incubation with N and Fe substrates, 2 L of seawater from each incubation were collected before sunrise and filtered onto 0.2 µm Supor membrane filters (Pall Corp., Ann Arbor, Michigan, U.S.A.) using peristaltic pumps. Filters were flash-frozen in liquid nitrogen immediately after filtering and stored at -80C until processing in the lab. Total RNA was extracted from environmental samples using DirectZol (Zymo Research). DNA was removed in a solution using RNase-Free DNase Kit (Qiagen), and RNA was purified again with RNA Clean Concentrator-25 (Zymo Research) according to the manufacturers' protocols. The RNA quality and quantity were evaluated using the Agilent BioAnalyzer RNA Nano Kit and Qiagen Qubit. All samples with an RNA Integrity Number greater than 9 were processed for microarray analyses as described in Shilova et al., 2014. Cy3-labeled cDNA was hybridized at the Roy J. Carver Center for Genomics (The University of Iowa, USA) using a Gene Expression Hybridization Kit (Cat# 5188-5242) and following a protocol based on One-Color Microarray-Based Gene Expression Analysis: Low Input Quick Amp Labeling [Version 6.7, September 2014]. Microarray platform GPL24371 Agilent-073391 MicroTOOLs_171K_oligo_v2.0 was used in this study. Microarrays were scanned at the Roy J. Carver Center for Genomics at the University of Iowa using an Agilent SureScan Microarray Scanner G2600D (Serial #: SG13134301) and using the Agilent scanning protocol GE1_1200_Jun14 (Feature Extractor software version 11.5.1.1).

Data Processing Description

Microarray data was processed using the MicroTOOLs software pipeline (www.jzehrlab.com/microtools), and transcription changes were examined using the pipeline as well as an Ensemble of Gene Set Enrichment Analyses (Alhamdoosh et al. 2017) and Whole Genome Correlation Network Analysis (Langfelder and Horvath, 2008).

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Related Publications

Alhamdoosh, M., Ng, M., Wilson, N. J., Sheridan, J. M., Huynh, H., Wilson, M. J., & Ritchie, M. E. (2016). Combining multiple tools outperforms individual methods in gene set enrichment analyses. Bioinformatics, btw623. doi:10.1093/bioinformatics/btw623
Methods

Langfelder, P., & Horvath, S. (2008). WGCNA: an R package for weighted correlation network analysis. BMC Bioinformatics, 9(1). doi:10.1186/1471-2105-9-559
Methods

MicroTOOLs. (2015). Zehr Laboratory. Retrieved from http://www.jzehrlab.com/microtools
Software

Shilova IN, Magasin JD, Mills MM, Robidart JC, Turk-Kubo KA, et al. (2020) Phytoplankton transcriptomic and physiological responses to fixed nitrogen in the California current system. PLOS ONE 15(4): e0231771. doi: 10.1371/journal.pone.0231771
Results

Shilova, I. N., Mills, M. M., Robidart, J. C., Turk-Kubo, K. A., Björkman, K. M., Kolber, Z., … Zehr, J. P. (2017). Differential effects of nitrate, ammonium, and urea as N sources for microbial communities in the North Pacific Ocean. Limnology and Oceanography, 62(6), 2550–2574. doi:10.1002/lno.10590
Results

Shilova, I. N., Robidart, J. C., James Tripp, H., Turk-Kubo, K., Wawrik, B., Post, A. F., … Zehr, J. P. (2014). A microarray for assessing transcription from pelagic marine microbial taxa. The ISME Journal, 8(7), 1476–1491. doi:10.1038/ismej.2014.1
Methods

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Parameters

Parameters for this dataset have not yet been identified

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Instruments

Dataset-specific Instrument Name	Agilent BioAnalyzer RNA
Generic Instrument Name	Bioanalyzer
Dataset-specific Description	The RNA quality and quantity were evaluated using the Agilent BioAnalyzer RNA Nano Kit and Qiagen Qubit.
Generic Instrument Description	A Bioanalyzer is a laboratory instrument that provides the sizing and quantification of DNA, RNA, and proteins. One example is the Agilent Bioanalyzer 2100.

Dataset-specific Instrument Name	Agilent SureScan Microarray Scanner G2600D
Generic Instrument Name	microarray scanner
Dataset-specific Description	Agilent SureScan Microarray Scanner G2600D (Serial #: SG13134301)
Generic Instrument Description	Microarray scanners are instruments used to detect and quantify the intensity of fluorescence of spots on a microarray slide, by selectively exciting fluorophores with a laser and measuring the fluorescence. A microarray scanner typically consists of lasers, a special microscope, and a camera. The DNA material in the microarray is labeled with fluorescents which become excited by the lasers in the scanner. The microscope and camera work together to create a digital image of the array.

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Deployments

NH1417

Website	https://www.bco-dmo.org/deployment/544429
Platform	R/V New Horizon
Start Date	2014-08-18
End Date	2014-09-16
Description	NEMO cruise. Bounding box -- 35 degrees N to 21.5 degrees N, 117 degrees W to 157 degrees W NSF R2R data catalog

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Project Information

Oligotrophic phytoplankton community response to changes in N substrates and the resulting impact on genetic, taxonomic and functional diversity (PhytoNsubResponse)

Coverage: North Pacific Subtropical Gyre at Station ALOHA, and a transect from San Diego, CA to Hawaii

(Extracted from NSF award abstract)

Marine phytoplankton are a diverse group of Prokaryotic and Eukaryotic unicellular organisms that account for approximately 50% of global carbon fixation. Nitrogen (N) is an essential element for microbial growth, but concentrations of bioavailable nitrogen in vast regions of subtropical ocean gyres are extremely low (submicromolar to nanomolar concentrations), and generally limit phytoplankton growth. Phytoplankton taxa differ in their genetic capabilities to take up and assimilate nutrients, and thus competition for different chemical forms of N (NH4+, NO3- and urea) and supply of these N-containing compounds are important controls on phytoplankton growth, productivity, and ultimately ecosystem function. The form and supply of N to phytoplankton have already been altered by anthropogenic activities, and with increasing environmental perturbations the effects will accelerate. To date however, there is limited information on how the N forms and fluxes impact the marine phytoplankton community composition and primary production. Similarly, determining the mechanisms of the response are crucial to assessing how ocean ecosystem function will respond to global climate change.

This project seeks to determine how taxonomic, genetic and functional dimensions of phytoplankton diversity are linked with community-level responses to the availability of different N substrates (NH4+, NO3-, and urea) in one of Earth's largest aquatic habitats, the North Pacific Subtropical Gyre. The project will characterize phytoplankton community composition change and gene expression, photosynthetic performance, carbon fixation, and single-cell level N and C uptake in different taxa within the phytoplankton assemblage in response to different N compounds. The research project is unique in investigating community-to-single-cell level function and species (strain)-specific gene expression patterns using state-of-the-art methods including fast repetition rate fluorometry, nanoscale secondary ion mass spectrometry and a comprehensive marine microbial community microarray. The results will provide predictive understanding of how changes in the availability of key nitrogen pools (N) may impact phytoplankton dynamics and function in the ocean.

References:

Karl, D. M., Bjorkman, K. M., Dore, J. E., Fujieki, L., Hebel, D. V., Houlihan, T., Letelier, R. M., Tupas, L. M. 2001. Ecological nitrogen-to-phosphorus stoichiometry at station ALOHA. Deep-Sea Research II. 48:1529 - 1566.

Karl, D. M., Letelier, R., Tupas, L., Dore, J., Christian, J. & Hebel, D. 1997. The role of nitrogen fixation in biogeochemical cycling in the subtropical North Pacific Ocean. Nature. 388:533-538.

McCarthy, J., Taylor, W. R., Taft, J. 1997. Nitrogenous nutrition of the plankton in the Chesapeake Bay. Limnology and Oceanography. 35:822 - 829.

Letelier, R., Karl, D. M. 1996. Role of Trichodesmium spp. in the productivity of the subtropical North Pacific Ocean. Marine Ecology Progress Series. 133:263 - 273.

Lipschultz, F. 1995. Nitrogen-specific uptake rates of marine phytoplankton isolated from natura populations

of particles by flow cytometry. Marine Ecology Progress Series. 123:245-258.

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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-1241221

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