

Trichodesmium AHL amendment metatranscriptomic reads accessions and metadata

Website: <https://www.bco-dmo.org/dataset/746654>

Data Type: Cruise Results

Version: 1

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Project

» [Dissolved Phosphorus Processing by Trichodesmium Consortia: Quantitative Partitioning, Role of Microbial Coordination, and Impact on Nitrogen Fixation](#) (P Processing by Tricho)

Contributors	Affiliation	Role
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Abstract

Trichodesmium is a marine, diazotrophic cyanobacterium that plays a central role in the biogeochemical cycling of carbon and nitrogen. Colonies ubiquitously co-occur with a diverse microbiome of heterotrophic bacteria. Here we show that manipulation of the microbiome with quorum sensing acyl homoserine lactone (AHL) molecules significantly modulated rates of N₂ fixation by Trichodesmium collected from the western North Atlantic, with both positive and negative effects of varied magnitude. Changes in Trichodesmium N₂ fixation did not clearly correlate with changes in microbiome composition or geochemical patterns.

Metatranscriptome sequencing revealed significant changes in the relative abundance of microbiome transcripts encoding metabolic functions consistent with quorum sensing responses in model bacteria. There was little overlap in specific microbiome transcriptional responses to AHL addition between stations, and this variability in microbiome gene expression may underpin the heterogeneous changes in Trichodesmium N₂ fixation. These data suggest the microbiome could play a large and previously overlooked role in modulating Trichodesmium N₂ fixation. This metadata form describes the metatranscriptomic sequencing reads that were used in the study.

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Coverage

Spatial Extent: N:30.4177 E:-52.1795 S:7.4618 W:-64.9988

Temporal Extent: 2014-05-08 - 2014-05-26

Dataset Description

The samples are composed of raw metatranscriptomic reads from acyl homoserine lactone (AHL) (quorum sensing) addition incubation experiments performed on *Trichodesmium* colonies collected in May 2014 on the 'PABST' cruise (R/V Atlantic Explorer AE1409) in the Sargasso Sea. We extracted prokaryotic RNA from triplicate control and +AHL samples, pooling together triplicate samples and sequencing 60 million paired end reads.

Links to the NCBI GenBank BioProjects are provided.

Raw reads can also be found on the NCBI SRA under accession code [PRJNA450995](https://www.ncbi.nlm.nih.gov/sra/PRJNA450995).

Acquisition Description

Samples were collected on cruise AE1409. *Trichodesmium* colonies were obtained by net tow (130 micron mesh) and serially washed in sterile surface seawater. Clean colonies were then incubated with or without a cocktail of quorum sensing molecules. After four hours of incubation, colonies were placed onto filters and stored in liquid nitrogen until RNA was extracted and submitted for sequencing at the Columbia University Genome Center.

Methods: We extracted prokaryotic RNA from triplicate control and +AHL samples by first adding approximately 500 μ L of glass beads to each cryotube and bead beating with a vortex adaptor for 5 minutes. We extracted total RNA using the yeast protocol from the Qiagen RNeasy Mini Kit with the added on-column DNase digestion using the RNase-free DNase Kit (Qiagen, Hilden, Germany). We processed DNase-treated total RNA through a MICROBEnrich kit following the manufacturer's instructions (ThermoFisher Scientific, Waltham, MA, USA). We removed ribosomal RNA using a Ribo-Zero Magnetic Kit optimized for bacteria (Illumina, San Diego, CA, USA) following the manufacturer's instructions. Finally, we purified the prokaryotic RNA extract using the RNeasy MinElute Cleanup Kit by following manufacturer instructions and eluting in 14 μ L water (Qiagen). We pooled together triplicate samples, and pooled RNA extracts were quantified using the Take3 Nucleic Acid Quantification program and a Biotek plate reader. To further assess quality of pooled triplicate RNA samples, we used a BioAnalyzer and the RNA 6000 Nano Kit (Agilent Technologies, Santa Clara, CA, USA). The JP Sulzberger Genome Center at Columbia University carried out RNA sequencing with a depth of 60 million paired end reads using an Illumina HiSeq protocol.

Quality control: We trimmed sequence reads and normalized following the Eel Pond Protocol for mRNAseq assembly. To obtain read counts for each sample, we mapped cleaned forward and reverse reads to metagenome assemblies from the same sampling locations that were previously characterized and clustered into orthologous groups (OGs).

We carried out mapping using RSEM with the paired-end and Bowtie2 parameters. We summed counts for previously determined OGs for *Trichodesmium* and epibiont genome bins separately. We determined significant changes in OG relative abundance between control and +AHL samples by comparing control and sample treatments using a stringent empirical Bayes approach called Analysis of Sequence Counts (ASC). This approach evaluates the posterior probability associated with a given fold change across pooled triplicates, and performs similarly, but conservatively, on replicated and unreplicated sample datasets. OGs were considered significantly higher or lower if they had a 95% or higher posterior probability of a fold change greater than 2 between treatment and control. Taxonomic relative abundance estimates for metagenome samples were previously calculated by multiplying the length of each contig in a genome bin by read mapping coverage, and then summing those values for all contigs. Please refer to the manuscript related to this metadata for more details and references.

Processing Description

BCO-DMO Processing: (to be edited)

Added conventional header with dataset name, PI name, version date.

Modified parameter names to conform with BCO-DMO naming conventions.

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Related Publications

Frischkorn, K. R., Rouco, M., Van Mooy, B. A. S., & Dyhrman, S. T. (2018). The *Trichodesmium* microbiome can modulate host N₂ fixation. *Limnology and Oceanography Letters*, 3(6), 401–408. doi:[10.1002/lol2.10092](https://doi.org/10.1002/lol2.10092)

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Parameters

Parameter	Description	Units
bioproject_accession	collection of biological data related to a single initiative	unitless
biosample_accession	code for accessing short read sequence data from NCBI	unitless
library_ID	sample name code	unitless
title	description and type of the sample	unitless
library_strategy	type of sequencing performed to generate the sample	unitless
library_source	type of sequence data represented by the sample	unitless
library_selection	how reads were prescreened (unspecified indicates reads were not screened)	unitless
library_layout	whether sequenced reads were single or paired-end	unitless
platform	sequencing machine platform used to generate reads	unitless
instrument_model	make and model of the sequencing machine platform used	unitless
design_description	quick methods description detailing how genetic material was prepared prior to sequencing	unitless
filetype	type of file the reads are stored as	unitless
assembly	whether or not there is a linked assembly (blank indicates that no assembly is provided)	unitless
filename	name of the first read file	unitless
filename2	name of the second read file	unitless
filename3	name of the third read file	unitless
filename4	name of the fourth read file	unitless
filename5	additional read file (if present)	unitless
filename6	additional read file (if present)	unitless
filename7	additional read file (if present)	unitless
filename8	additional read file (if present)	unitless

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Instruments

Dataset-specific Instrument Name	net
Generic Instrument Name	Plankton Net
Dataset-specific Description	The net had 130 micron mesh and was used to collect Trichodesmium colonies.
Generic Instrument Description	A Plankton Net is a generic term for a sampling net that is used to collect plankton. It is used only when detailed instrument documentation is not available.

Dataset-specific Instrument Name	Illumina Miseq platform
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	PCR Thermal Cycler
Generic Instrument Description	General term for a laboratory apparatus commonly used for performing polymerase chain reaction (PCR). 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. (adapted from http://serc.carleton.edu/microbelife/research_methods/genomics/pcr.html)

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Deployments

AE1409

Website	https://www.bco-dmo.org/deployment/565190
Platform	R/V Atlantic Explorer
Start Date	2014-05-08
End Date	2014-05-26
Description	May 2014 cruise conducted as part of the "Dissolved Phosphorus Processing by Trichodesmium Consortia: Quantitative Partitioning, Role of Microbial Coordination, and Impact on Nitrogen Fixation" project.

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Project Information

Dissolved Phosphorus Processing by Trichodesmium Consortia: Quantitative Partitioning, Role of Microbial Coordination, and Impact on Nitrogen Fixation (P Processing by Tricho)

Coverage: Western Tropical North Atlantic

Description from NSF award abstract: Colonies of the cyanobacterium Trichodesmium are responsible for a large fraction of N₂ fixation in nutrient-poor, open-ocean ecosystems, ultimately fueling primary production in both Trichodesmium and in the broader planktonic community. However, in some parts of the ocean, the scarcity of dissolved phosphorus limits rates of Trichodesmium N₂ fixation. Trichodesmium colonies employ an arsenal of strategies to mitigate the effects of phosphorus limitation, and the consortia of epibiotic bacteria in the colonies may play a significant role in phosphorus acquisition. In this study, researchers from Woods Hole Oceanographic Institution and Columbia University will use metagenomic and metatranscriptomic sequencing to investigate how phosphorus metabolism is coordinated in Trichodesmium consortia, and to discern the role of quorum sensing in phosphorus acquisition and partitioning. Results from this study are expected to expand understanding of Trichodesmium from a monospecific colony whose primary function is fixing CO₂ and N₂ toward a unique planktonic consortium with a diverse, complex, and highly coordinated overall metabolism that exerts profound control over the cycling of inorganic and organic nutrients in the oligotrophic upper ocean.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1332898
NSF Division of Ocean Sciences (NSF OCE)	OCE-1332912

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