

# NCBI accessions for raw RNA-seq fastq in 380 and 750 uatm CO<sub>2</sub> from experiments conducted at the University of Southern California: deep metatranscriptomic sequencing of Trichodesmium enrichment cultures (HiCO<sub>2</sub>\_AdaptCyano project)

**Website:** <https://www.bco-dmo.org/dataset/726901>

**Data Type:** experimental

**Version:**

**Version Date:** 2018-02-14

## Project

» [Adaptation of key N<sub>2</sub>-fixing cyanobacteria to changing CO<sub>2</sub>](#) (HiCO<sub>2</sub>\_AdaptCyano)

» [Collaborative Research: Iron and phosphorus balanced limitation of nitrogen fixation in the oligotrophic ocean](#) (TriCoLim)

Contributors	Affiliation	Role
<a href="#">Hutchins, David A.</a>	University of Southern California (USC)	Principal Investigator
<a href="#">Webb, Eric A.</a>	University of Southern California (USC)	Co-Principal Investigator
<a href="#">Copley, Nancy</a>	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

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## Dataset Description

These data were derived from laboratory-maintained *Trichodesmium erythraeum* cultures and contain accessions and links to raw RNA-seq fastq files in NCBI's Gene Expression Omnibus, accessible through GEO Series accession number [GSE94951](#). The sample accession numbers corresponding to the low and high CO<sub>2</sub> samples from this work are [GSM2492342](#), [GSM2492343](#), [GSM2492344](#), and [GSM2492345](#).

These data were reported in the paper (Lee et al., 2017)

## Methods & Sampling

**Culturing:** Cultures of *Trichodesmium erythraeum* strain IMS101 were maintained at the University of Southern California, Los Angeles, California, USA, in modified Aquil medium with no fixed nitrogen provided. Cultures were grown at 26°C in a light:dark cycle of 12:12, and maintained under 120 μmol photons per m<sup>2</sup> s<sup>-1</sup>.

Samples GSM2492342 and GSM2492343 were grown as noted above, but under 380 μatm CO<sub>2</sub>.

Samples GSM2492344 and GSM2492345 were grown as noted above, but under 800 μatm CO<sub>2</sub>.

Sampling: Cultures were filtered onto 5 µm polycarbonate filters during the middle of the photoperiod, flash frozen, and stored in liquid nitrogen until RNA extraction.

## Data Processing Description

### BCO-DMO Processing Notes:

- created a table with a conventional header with dataset name, PI name, version date
- records were compiled from NCBI website; accessions obtained from (Lee et al., 2017).
- hyperlink were created to the NCBI accessions pages.

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## Data Files

File
<b>Tricho_FASTq_accessions.csv</b> (Comma Separated Values (.csv), 719 bytes) MD5:81cde9e6505eb560d97a578d4adf3b23
Primary data file for dataset ID 726901

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## Related Publications

Lee, M. D., Webb, E. A., Walworth, N. G., Fu, F.-X., Held, N. A., Saito, M. A., & Hutchins, D. A. (2017). Transcriptional Activities of the Microbial Consortium Living with the Marine Nitrogen-Fixing Cyanobacterium *Trichodesmium* Reveal Potential Roles in Community-Level Nitrogen Cycling. *Applied and Environmental Microbiology*, 84(1), e02026–17. doi:10.1128/aem.02026-17 <https://doi.org/10.1128/AEM.02026-17>

*Results*  
,  
*Methods*

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## Parameters

Parameter	Description	Units
organism_community	host organism from which the microbial community genomic material was extracted	unitless
pCO2	CO2 level of culture	microatmospheres
BioSample	NCBI BioSample accession number: descriptive information about the physical biological materials	unitless
SRA	NCBI Sequence Read Archive accession number	unitless
GSM	NCBI sample Gene Expression Omnibus accession number	unitless
GSM_link	link to NCBI BioSample (GSM) accession webpage for the GSM	unitless

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## Instruments

<b>Dataset-specific Instrument Name</b>	Illumina Hi-Seq
<b>Generic Instrument Name</b>	Automated DNA Sequencer
<b>Dataset-specific Description</b>	Used to sequence the extracted RNA, performed at UC San Diego Institute for Genomic Medicine
<b>Generic Instrument Description</b>	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.

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## Deployments

### lab\_Webb\_Hutchins\_Fu

<b>Website</b>	<a href="https://www.bco-dmo.org/deployment/59058">https://www.bco-dmo.org/deployment/59058</a>
<b>Platform</b>	Webb-Hutchins-Fu USC
<b>Start Date</b>	2011-08-15
<b>End Date</b>	2013-03-31
<b>Description</b>	Lab experiments of transcriptome samples (labeled 750) obtained from cultures grown in either projected year 2100 CO2 levels (~750ppm) or current 380ppm levels (labeled 380) for four years.

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## Project Information

### Adaptation of key N2-fixing cyanobacteria to changing CO2 (HiCO2\_AdaptCyano)

**Coverage:** Culture study at the University of Southern California, Los Angeles

*Description from NSF award abstract:*

This study will employ a novel combination of experimental evolution techniques and state-of-the-art molecular methods to yield unique insights into adaptive changes in the keystone marine cyanobacteria *Trichodesmium* and *Crocospaera* in response to selection by high CO<sub>2</sub>. Several studies have suggested that N<sub>2</sub>-fixation rates of the biogeochemically-critical cyanobacteria *Trichodesmium* and *Crocospaera* may increase dramatically in the future high CO<sub>2</sub> ocean, but these have all used the same limited set of cultured isolates and considered cells only briefly acclimated to elevated CO<sub>2</sub>. The investigator's new results, however, demonstrate that a broad diversity of high- and low-CO<sub>2</sub> adapted ecotypes exists within each diazotroph genus. Furthermore, in a preliminary four year experimental evolution study with *Trichodesmium*, the PIs observed large adaptive responses following 500-700 generations of selection by high CO<sub>2</sub>- but in a completely unexpected way. All of the six replicate high CO<sub>2</sub>-adapted cell lines exhibited strong constitutive up-regulation of N<sub>2</sub> fixation rates. These very elevated N<sub>2</sub> fixation rates continued, even though the cultures have been switched back to low-CO<sub>2</sub> conditions for many months. Expression of the nif operon and N assimilatory genes was also up-

regulated in these cell lines, as is expression of many intergenic regions of the genome.

**The investigators hypothesize that constitutive up-regulation of cellular N<sub>2</sub> fixation systems may be a common adaptive response of both *Trichodesmium* and *Crocospaera* under extended selection by elevated CO<sub>2</sub>. This project will test this hypothesis in a four-year experimental evolution study to determine the adaptive responses of both high- and low-CO<sub>2</sub> specialized ecotypes of these two diazotrophs to increased CO<sub>2</sub>.**

The investigators will grow representative high- and low-CO<sub>2</sub> adapted ecotypes from each genus in well-replicated cell lines at 380 ppm and 750 ppm CO<sub>2</sub> for up to 1000 generations. Periodically, they will perform "switch" experiments to measure N<sub>2</sub> and CO<sub>2</sub> fixation rates and growth rates of high CO<sub>2</sub>-selected cell lines grown briefly (one week) at low CO<sub>2</sub>, and vice versa. These switch experiments will allow screening for cell lines which exhibit adaptive changes in phenotypically-expressed rate parameters, such as those observed in the preliminary *Trichodesmium* study. Evolutionary mechanisms in the CO<sub>2</sub>-selected cell lines will be examined by comparison of changes in their genomes, transcriptomes, and proteomes over time relative to reference genomes, using frozen samples archived monthly during the preceding selection period. Examination of these molecular and biochemical changes will be coordinated with an in-depth array of physiological and biogeochemical analyses. This combined approach will allow an evaluation of potential adaptive mechanisms in diazotrophic cyanobacteria ranging from indel, duplication, single nucleotide polymorphism, and transposition mutations to altered putative non-coding RNA expression, protein expression, and post-translational protein modifications, and then allow the investigators to link these mechanisms directly with their potential impacts on ecosystem-level biogeochemical processes like N<sub>2</sub> and CO<sub>2</sub> fixation. Finally, the research team will determine how long term selection by high CO<sub>2</sub> affects the iron and phosphorus requirements of *Trichodesmium* and *Crocospaera*, since constitutive up-regulation of N<sub>2</sub> fixation would also have major implications for limitation of diazotrophs by these two critical nutrients in the future high CO<sub>2</sub> ocean.

### **Collaborative Research: Iron and phosphorus balanced limitation of nitrogen fixation in the oligotrophic ocean (TriCoLim)**

**Coverage:** Tropical Atlantic

NSF abstract:

Marine cyanobacteria are able to use or "fix" atmospheric nitrogen gas, and so supply much of the essential nutrient nitrogen that supports open ocean food chains. Oceanographers have usually thought that the growth of these nitrogen-fixing cyanobacteria is limited at any particular time and place by the supply of either iron, or of phosphorus. Preliminary experiments have shown, though, that these nitrogen fixers instead grow best when both iron and phosphorus are scarce at the same time. In this project, the researchers will use cellular indicators that are specific for iron and phosphorus limitation to determine how important this type of "balanced limitation" of nitrogen-fixing cyanobacteria is in controlling the productivity of ocean food chains in the tropical Atlantic Ocean. Two graduate students will be trained at the University of Southern California (USC) and Woods Hole Oceanographic Institution, as well as a postdoctoral researcher at USC. Educational outreach efforts will take place at a Los Angeles inner city high school with a student body that is over 98% Hispanic and African-American, and with underrepresented undergraduates in the USC Global Environmental Microbiology course. In addition, two Research Experiences for Undergraduates students will be supervised for summer research projects to help them learn about science career options.

The researchers will investigate the biological and biogeochemical consequences of this unique balanced iron/phosphorus-limited phenotype, using both laboratory and fieldwork approaches. During the first year of this project, the nitrogen-fixing cyanobacteria will be cultured under iron and/or phosphorus limitation, followed by application of proteomics and transcriptomics to identify genes that are potential diagnostic biomarkers for iron/phosphorus balanced limitation. Preliminary work has already identified one promising candidate biomarker in one cyanobacterium, an EzrA protein domain that appears to be associated with the cell size decreases seen specifically under balanced limitation, and the researchers have identified numerous other potential candidates for similar biomarkers. During the second year, these new co-limitation biomarkers and others previously validated for iron limitation (IsiB) and phosphorus limitation (SphX) will be used to investigate balanced limitation during a research cruise transecting from relatively high-iron, low-phosphorus North Atlantic waters, to the relatively high-phosphorus, low-iron South Atlantic. This fieldwork component will survey nitrogen fixing cyanobacteria populations across this natural iron/phosphorus gradient for genetic, proteomic, and

physiological indicators of balanced limitation, as well as testing their responses to iron and phosphorus manipulations in shipboard incubation experiments. The third year will be devoted to sample analysis, and publications exploring the responses of oceanic nitrogen fixers to simultaneous limitation by both iron and phosphorus.

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## Funding

Funding Source	Award
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1260233</a>
<a href="#">NSF Division of Ocean Sciences (NSF OCE)</a>	<a href="#">OCE-1657757</a>

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