Trichodesmium nutrient-limited proteomes in 380 and 750 uatm CO2 from experiments conducted at the University of Southern California from 2011-2013 (HiCO2_AdaptCyano project)

Website: https://www.bco-dmo.org/dataset/649873
Data Type: experimental
Version: 2
Version Date: 2018-03-15

Project
» Adaptation of key N2-fixing cyanobacteria to changing CO2 (HiCO2_AdaptCyano)

Contributors	Affiliation	Role
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Dataset Description

Trichodesmium erythraeum was grown in biological triplicate under two CO2 regimes (380 and 750 uatm pCO2) and four different nutrient regimes (replete, phosphorus-limited,iron-limited, iron/phosphorus colimited).

Raw sequence reads from the *Trichodesmium erythraeum* used in this experiment can be accessed at The National Center for Biotechnology Information (NCBI) under the BioProject PRJNA312342.

Protein sequences can be accessed through NCBI BioProject PRJNA318.

The fasta file IMG_Trichodesmium_IMS101_genes.fasta (5.4 MB) contains all of the Integrated Microbial Genomes (IMG)-annotated genes in the Trichodesmium erythraeum IMS101 genome. More information can be found at the IMG Trichodesmium genome portal, "genes page" and IMG Genome ID 637000329 page.

Raw spectra are available as dataset PXD010515 in the PRIDE proteome-exchange repository (doi: 10.6019/PXD010515). These results were published in: * Walworth et al., 2016a * Walworth et al., 2016b * Walworth et al., 2018

Related dataset: Trichodesmium physiology and cell growth

Methods & Sampling

The following are excerpts from Walworth et al. 2016a. Please refer to this reference for more details of the methodology used to generate these data.

"Protein spectral counts are generated by extracting proteins from cells, digesting into smaller peptides, separating them on HPLC (high-performance liquid chromatography), and measuring peptide masses on a mass spectrometer to ascertain mass-to-charge ratio. Peptide fragment ratios are then compared to a reference genome and spectral counts per protein are enumerated."

"Nitrogen fixation by cyanobacteria supplies critical bioavailable nitrogen to marine ecosystems worldwide; however, field and lab data have demonstrated it to be limited by iron, phosphorus and/or CO2. To address unknown future interactions among these factors, we grew the nitrogen-fixing cyanobacterium Trichodesmium for 1 year under Fe/P co-limitation following 7 years of both low and high CO2 selection. Fe/P co-limited cell lines demonstrated a complex cellular response including increased growth rates, broad proteome restructuring and cell size reductions relative to steady-state growth limited by either Fe or P alone. Fe/P co-limitation increased abundance of a protein containing a conserved domain previously implicated in cell size regulation, suggesting a similar role in Trichodesmium. Increased CO2 further induced nutrient-limited proteome shifts in widespread core metabolisms. Our results thus suggest that N2-fixing microbes may be significantly impacted by interactions between elevated CO2 and nutrient limitation, with broad implications for global biogeochemical cycles in the future ocean."

"Spectral counts compare a specific protein's abundance between treatments, rather than against other proteins, because the sensitivity of spectral counts can vary between proteins depending on the number of tryptic peptides within the sequence and their chemical characteristics."

Data Processing Description

BCO-DMO data manager notes:
* Data version 2: 2018-03-15 replaces version 1: 2016-06-22. Data version 2 removed column "accession_numbers" containing protein sequence accession numbers at NCBI which were no loger valid due to an annotation remapping effort. The appropriate protein accessions can instead be found through the NCBI BioProject page: PRJNA318. The data parameter "Gene ID" was changed to name "Locus Tag" to be consistent with terminology at IMG and NCBI.

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Data Files

File
trichodesmium380_750_v2.csv (Comma Separated Values (.csv), 469.89 KB) MD5:0eeb92a312a28a37f18ca810b26f11b5 Primary data file for dataset ID 649873

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Related Publications

Walworth, N. G., Fu, F.-X., Webb, E. A., Saito, M. A., Moran, D., McIlvin, M. R., … Hutchins, D. A. (2016b). Mechanisms of increased Trichodesmium fitness under iron and phosphorus co-limitation in the present and future ocean. Nature Communications, 7, 12081. doi:10.1038/ncomms12081
Results

,
Methods

Walworth, N. G., Lee, M. D., Fu, F.-X., Hutchins, D. A., & Webb, E. A. (2016a). Molecular and physiological evidence of genetic assimilation to high CO2in the marine nitrogen fixerTrichodesmium. Proceedings of the National Academy of Sciences, 113(47), E7367–E7374. doi:10.1073/pnas.1605202113

Results
,
Methods

Walworth, N. G., Lee, M. D., Suffridge, C., Qu, P., Fu, F.-X., Saito, M. A., ... Hutchins, D. A. (2018). Functional Genomics and Phylogenetic Evidence Suggest Genus-Wide Cobalamin Production by the Globally Distributed Marine Nitrogen Fixer Trichodesmium. Frontiers in Microbiology, 9. doi:10.3389/fmicb.2018.00189
Results
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Related Datasets

Related Research

ProteomeXchange dataset PXD010515. Trichodesmium LC-MSMS iron and phosphorous co-limitation in the ocean. doi:10.6019/pxd010515 https://doi.org/10.6019/PXD010515

Different Version

ProteomeXchange dataset PXD010515. Trichodesmium LC-MSMS iron and phosphorous co-limitation in the ocean. doi:10.6019/pxd010515 https://doi.org/10.6019/PXD010515

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Parameters

Parameter	Description	Units
Locus_Tag	Locus identfier	text
COG_category	Clusters of Orthologous Groups; generated by comparing the protein sequences of complete genomes; classified into functional categories	text
gene_product	protein identified by the gene in the first column	text
pCO2_380_1	these columns are spectral counts defined as the total number of spectra identified for a protein; a semiquantitative measure of protein abundance; 380 uatm CO2-replete nutrients (unlimited Fe and P); first replicate	counts
pCO2_380_2	380 uatm CO2-replete nutrients (unlimited Fe and P); second replicate	counts
pCO2_380_3	380 uatm CO2-replete nutrients (unlimited Fe and P); third replicate	counts
pCO2_750_1	750 uatm CO2-replete nutrients (unlimited Fe and P); first replicate	counts
pCO2_750_2	750 uatm CO2-replete nutrients (unlimited Fe and P); second replicate	counts
pCO2_750_3	750 uatm CO2-replete nutrients (unlimited Fe and P); third replicate	counts
pCO2_380plusPlim_1	380 uatm CO2 P limited; first replicate	counts
pCO2_380plusPlim_2	380 uatm CO2 P limited; second replicate	counts
pCO2_380plusPlim_3	380 uatm CO2 P limited; third replicate	counts
pCO2_750plusPlim_1	750 uatm CO2 P limited; first replicate	counts
pCO2_750plusPlim_2	750 uatm CO2 P limited; second replicate	counts
pCO2_750plusPlim_3	750 uatm CO2 P limited; third replicate	counts
pCO2_380plusFelim_1	380 uatm CO2 Fe limited; first replicate	counts
pCO2_380plusFelim_2	380 uatm CO2 Fe limited; second replicate	counts
pCO2_380plusFelim_3	380 uatm CO2 Fe limited; third replicate	counts
pCO2_750plusFelim_1	750 uatm CO2 Fe limited; first replicate	counts
pCO2_750plusFelim_2	750 uatm CO2 Fe limited; second replicate	counts
pCO2_750plusFelim_3	750 uatm CO2 Fe limited; third replicate	counts
pCO2_380plusFePlim_1	380 uatm CO2 iron/phosphorus co-limited; first replicate	counts
pCO2_380plusFePlim_2	380 uatm CO2 iron/phosphorus co-limited; second replicate	counts
pCO2_380plusFePlim_3	380 uatm CO2 iron/phosphorus co-limited; third replicate	counts
pCO2_750plusFePlim_1	750 uatm CO2 iron/phosphorus co-limited; first replicate	counts
pCO2_750plusFePlim_2	750 uatm CO2 iron/phosphorus co-limited; second replicate	counts
pCO2_750plusFePlim_3	750 uatm CO2 iron/phosphorus co-limited; third replicate	counts

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Instruments

Dataset-specific Instrument Name	CM140 Total Inorganic Carbon Analyzer
Generic Instrument Name	CO2 Coulometer
Dataset-specific Description	Dissolved inorganic carbon was measured with CO2 coulometry (model CM 140, UIC).
Generic Instrument Description	A CO2 coulometer semi-automatically controls the sample handling and extraction of CO2 from seawater samples. Samples are acidified and the CO2 gas is bubbled into a titration cell where CO2 is converted to hydroxyethylcarbonic acid which is then automatically titrated with a coulometrically-generated base to a colorimetric endpoint.

Dataset-specific Instrument Name	Shimadzu gas chromatograph GC-8a
Generic Instrument Name	Gas Chromatograph
Dataset-specific Description	Shimadzu Scientific Instruments, Columbia, Maryland. Equipped with a flame ionization detector.
Generic Instrument Description	Instrument separating gases, volatile substances, or substances dissolved in a volatile solvent by transporting an inert gas through a column packed with a sorbent to a detector for assay. (from SeaDataNet, BODC)

Dataset-specific Instrument Name	Michrom Advance nanoflow LC and autosampler
Generic Instrument Name	High-Performance Liquid Chromatograph
Dataset-specific Description	Michrom Bioresources. And a 100 µm inner diameter 15 cm long capillary column with a pulled tip (packed in-lab with MAGIC C18AQ 200 Å pore size 3 µm particle size from Michrom Bioresources).
Generic Instrument Description	A High-performance liquid chromatograph (HPLC) is a type of liquid chromatography used to separate compounds that are dissolved in solution. HPLC instruments consist of a reservoir of the mobile phase, a pump, an injector, a separation column, and a detector. Compounds are separated by high pressure pumping of the sample mixture onto a column packed with microspheres coated with the stationary phase. The different components in the mixture pass through the column at different rates due to differences in their partitioning behavior between the mobile liquid phase and the stationary phase.

Dataset-specific Instrument Name	Thermo Fusion mass spectrometer
Generic Instrument Name	Mass Spectrometer
Dataset-specific Description	With MS1 scans at an Orbitrap resolution of 60 K, 350–1,800 m/z scan range, 2.0e5 automatic gain control target, and a maximum injection time of 35 ms. MS2 scans were analysed on the linear ion trap in topN data dependent mode at a cycle time of 3 s using normal scan rate and range with a maximum injection time of 75 ms and a 0.7 m/z isolation window. Charge states of 2–7 were analysed with a dynamic exclusion of 15 s with a mass tolerance of 10 p.p.m. Monoisotopic precursor selection was used, and a user-defined lock mass of 445.12003 m/z.
Generic Instrument Description	General term for instruments used to measure the mass-to-charge ratio of ions; generally used to find the composition of a sample by generating a mass spectrum representing the masses of sample components.

Dataset-specific Instrument Name	Orion 5 STAR pH meter
Generic Instrument Name	pH Sensor
Dataset-specific Description	Thermo Fisher Scientific. With a combined glass electrode. The metre was calibrated with National Bureau of Standards buffer solutions of pH 4, 7 and 10.
Generic Instrument Description	An instrument that measures the hydrogen ion activity in solutions. The overall concentration of hydrogen ions is inversely related to its pH. The pH scale ranges from 0 to 14 and indicates whether acidic (more H+) or basic (less H+).

Dataset-specific Instrument Name	Turner 10 AU fluorometer
Generic Instrument Name	Turner Designs Fluorometer 10-AU
Generic Instrument Description	The Turner Designs 10-AU Field Fluorometer is used to measure Chlorophyll fluorescence. The 10AU Fluorometer can be set up for continuous-flow monitoring or discrete sample analyses. A variety of compounds can be measured using application-specific optical filters available from the manufacturer. (read more from Turner Designs, turnerdesigns.com, Sunnyvale, CA, USA)

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Deployments

lab_Webb_Hutchins_Fu

Website	https://www.bco-dmo.org/deployment/59058
Platform	Webb-Hutchins-Fu USC
Start Date	2011-08-15
End Date	2013-03-31
Description	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 *Crocosphaera* in response to selection by high CO2. Several studies have suggested that N2-fixation rates of the biogeochemically-critical cyanobacteria *Trichodesmium* and *Crocosphaera* may increase dramatically in the future high CO2 ocean, but these have all used the same limited set of cultured isolates and considered cells only briefly acclimated to elevated CO2. The investigator's new results, however, demonstrate that a broad diversity of high- and low-CO2 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 CO2- but in a completely unexpected way. All of the six replicate high CO2-adapted cell lines exhibited strong constitutive up-regulation of N2 fixation rates. These very elevated N2 fixation rates continued, even though the cultures have were switched back to low-CO2 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 N2 fixation systems may be a common adaptive response of both *Trichodesmium* and *Crocosphaera* under extended selection by elevated CO2. This project will test this hypothesis in a four-year experimental evolution study to determine the adaptive responses of both high- and low-CO2 specialized ecotypes of these two diazotrophs to increased CO2.

The investigators will grow representative high- and low-CO2 adapted ecotypes from each genus in well-replicated cell lines at 380 ppm and 750 ppm CO2 for up to 1000 generations. Periodically, they will perform "switch" experiments to measure N2 and CO2 fixation rates and growth rates of high CO2-selected cell lines grown briefly (one week) at low CO2, 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 CO2-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 N2 and CO2 fixation. Finally, the research team will determine how long term selection by high CO2 affects the iron and phosphorus requirements of *Trichodesmium* and *Crocosphaera*, since constitutive up-regulation of N2 fixation would also have major implications for limitation of diazotrophs by these two critical nutrients in the future high CO2 ocean.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1260490
NSF Division of Ocean Sciences (NSF OCE)	OCE-1260233

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