Trichodesmium growth rate and cell size related to pCO2 and nutrients from experiments conducted at the University of Southern California from 2011-2013 (HiCO2_AdaptCyano project)

Website: https://www.bco-dmo.org/dataset/649904
Data Type: experimental
Version: Final
Version Date: 2016-06-22

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

Contributors	Affiliation	Role
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Dataset Description

Trichodesmium erythraeum growth rates per day for all replicates in each treatment calculated from cell counts. *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).

These data were utilized in the following publications:

Walworth, N. G., Fu, F. X., Webb, E. A., Saito, M. A., Moran, D., McIlvin, M. R. & Hutchins, D. A. (2016a). 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

Walworth, N. G., Lee, M. D., Suffridge, C., Qu, P., Fu, F., 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*, 189. doi: 10.3389/fmicb.2018.00189

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

Walworth, N. G., Lee, M. D., Fu, F. X., Hutchins, D. A., ... & Webb, E. A.(2016b). Molecular and physiological evidence of genetic assimilation to high CO2 in the marine nitrogen fixer Trichodesmium. *Proceedings of the National Academy of Sciences*, *113*(47), E7367-E7374. doi: 10.1073/pnas.1605202113

Related Dataset: Trichodesmium proteomes and Hi CO2 adaptation

Methods & Sampling

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

"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. "

"Growth rates were calculated according to microscopic cell counts for reported values, or using *in vivo* chlorophyll fluorescence measurements with a Turner 10 AU fluorometer for semi-daily dilution calculations in real time during the experiments. Comparisons between cell counts and *in vivo*-based growth rates revealed no significant differences between the two methods of assessing biomass changes."

A picogram carbon/µm filament was used for all replicates in each treatment (cell size proxy).

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Data Files

File
cell_physio_size.csv (Comma Separated Values (.csv), 1.37 KB) MD5:8bce9923bfcc19905335a407352f7e91 Primary data file for dataset ID 649904

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Parameters

Parameter	Description	Units
Treatments	CO2 and Nutrient regimes replicated three times; +P and -P speak of non-limiting or limiting phosphorus (20 µM and 0.5 µM phosphate respectively); +Fe or -Fe speak of limiting or non-limiting iron (500 nM iron); 380 or 750 indicate the partial pressure of CO2	micromoles;nanomoles;uatm
Growth_rates	rates calculated from cell counts	reciprocal time - per day (d-1) - because the cell numbers cancel out
pg_C_per_um_filament	Picogram carbon per micron filament for all replicates in each treatment (cell size proxy)	picogram

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Instruments

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-CO_2 adapted ecotypes from each genus in well-replicated cell lines at 380 ppm and 750 ppm CO_2 for up to 1000 generations. Periodically, they will perform "switch" experiments to measure N_2 and CO_2 fixation rates and growth rates of high CO_2-selected cell lines grown briefly (one week) at low CO_2, 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_2-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_2 and CO_2 fixation. Finally, the research team will determine how long term selection by high CO_2 affects the iron and phosphorus requirements of *Trichodesmium* and *Crocosphaera*, since constitutive up-regulation of N_2 fixation would also have major implications for limitation of diazotrophs by these two critical nutrients in the future high CO_2 ocean.

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Funding

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

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