

Carbonate chemistry data collected as part of a study of the "Community context and pCO₂ impact the transcriptome of the "helper" bacterium *Alteromonas* in co-culture with picocyanobacteria"

Website: <https://www.bco-dmo.org/dataset/883120>

Data Type: experimental

Version: 1

Version Date: 2022-12-27

Project

» [Collaborative Research: Ecology and Evolution of Microbial Interactions in a Changing Ocean](#) (LTPE)

Contributors	Affiliation	Role
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Abstract

Carbonate chemistry data collected as part of a study of the "Community context and pCO₂ impact the transcriptome of the "helper" bacterium *Alteromonas* in co-culture with picocyanobacteria" (Barreto Filho et al., 2022). The following results abstract describes these data along with related datasets which can be accessed from the "Related Datasets" section of this page. Many microbial photoautotrophs depend on heterotrophic bacteria for accomplishing essential functions. Environmental changes, however, could alter or eliminate such interactions. We investigated the effects of changing pCO₂ on gene expression in co-cultures of 3 strains of picocyanobacteria (*Synechococcus* strains CC9311 and WH8102 and *Prochlorococcus* strain MIT9312) paired with the 'helper' bacterium *Alteromonas macleodii* EZ55. Co-culture with cyanobacteria resulted in a much higher number of up- and down-regulated genes in EZ55 than pCO₂ by itself. Pathway analysis revealed significantly different expression of genes involved in carbohydrate metabolism, stress response, and chemotaxis, with different patterns of up- or down-regulation in co-culture with different cyanobacterial strains. Gene expression patterns of organic and inorganic nutrient transporter and catabolism genes in EZ55 suggested resources available in the culture media were altered under elevated (800 ppm) pCO₂ conditions. Altogether, changing expression patterns were consistent with the possibility that the composition of cyanobacterial excretions changed under the two pCO₂ regimes, causing extensive ecophysiological changes in both members of the co-cultures. Additionally, significant downregulation of oxidative stress genes in MIT9312/EZ55 cocultures at 800 ppm pCO₂ were consistent with a link between the predicted reduced availability of photorespiratory byproducts (i.e., glycolate/2PG) under this condition and observed reductions in internal oxidative stress loads for EZ55, providing a possible explanation for the previously observed lack of "help" provided by EZ55 to MIT9312 under elevated pCO₂. The data stored in this archive permit the recalculation of the pH measurements shown in Table 1 of the results publication Barreto Filho et al. (2022).

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Coverage

Temporal Extent: 2016-10-01 - 2022-06-01

Methods & Sampling

Synechococcus cultures were grown under similar conditions to those described in our previous experiment with *Prochlorococcus* (Hennon et al., 2017). Briefly, all cultures were prepared in acid-washed conical-bottom glass centrifuge tubes containing 13 mL of artificial seawater (ASW) amended with nutrient stocks (Hennon et al., 2017) and with acid and/or base to control pCO₂. ASW (per L: 28.41 g NaCl, 0.79 g KCl, 1.58 g CaCl₂*2H₂O, 7.21 g MgSO₄*7H₂O, 5.18 g MgCl₂*6H₂O) was sterilized in acid-washed glass bottles, amended with 2.325 mM (final concentration) of filter-sterilized sodium bicarbonate, then bubbled with sterile air overnight. *Synechococcus* cultures were grown in SEv (per L: 32 μM NaNO₃, 2 μM NaH₂PO₄, 20 μL SN trace metal stock, and 20 μL F/2 vitamin stock). The primary differences between this medium and the PEv medium used in our earlier *Prochlorococcus* study are the nitrogen source (NO₃⁻ vs. NH₄⁺, with molar concentration of N and N:P ratios identical to PEv) and the addition of F/2 vitamins (Hennon et al., 2017). Carbonate chemistry of each media batch was determined prior to pCO₂ manipulations by measuring alkalinity and pH by titration and colorimetry, respectively (Dickson et al., 2007) and then using the *oa* function in *seacarb* package in R to determine how much hydrochloric acid and bicarbonate (for 800 ppm pCO₂) or sodium hydroxide (for 400 ppm pCO₂) was needed to achieve desired experimental conditions (Gattuso and Lavigne, 2009). Acid and base amendments were introduced immediately prior to inoculation. Cultures were grown in a Percival growth chamber at 21^o C under 150 μmol photons m⁻² s⁻¹ on a 14:10 light:dark cycle. *Synechococcus* cultures were grown on a rotating tissue culture wheel at approximately 60 rpm. After addition of amendments, absorbance values at 434, 578, and 730 nm were collected both before and after injection of m-cresol purple dye, and again after a second dye injection to correct for the impact of dye addition on solution pH. Equations from (Dickson et al., 2007) were used to compute the pH. The data are provided in the form of an Excel spreadsheet containing the necessary formulas for computing the pH from the absorbance data. pH assays were performed in microtiter plates using a BioTek H1 plate reader with an automated dye injection mechanism and temperature control.

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Data Files

File
carb_chem.csv (Comma Separated Values (.csv), 5.75 KB) MD5:fb8861fc038cf4322d40644bab2c7684
Primary data file for dataset ID 883120

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Related Publications

Barreto Filho, M. M., Lu, Z., Walker, M., & Morris, J. J. (2022). Community context and pCO₂ impact the transcriptome of the “helper” bacterium *Alteromonas* in co-culture with picocyanobacteria. *ISME Communications*, 2(1). <https://doi.org/10.1038/s43705-022-00197-2>
Results

Hennon, G. M., Morris, J. J., Haley, S. T., Zinser, E. R., Durrant, A. R., Entwistle, E., ... Dyhrman, S. T. (2017). The impact of elevated CO₂ on *Prochlorococcus* and microbial interactions with “helper” bacterium *Alteromonas*. *The ISME Journal*, 12(2), 520–531. doi:[10.1038/ismej.2017.189](https://doi.org/10.1038/ismej.2017.189).
Results

Related Datasets

IsRelatedTo

Morris, J. (2022) **Synechococcus (WH8102 and CC9311) growth and genetic sequence accessions from experiments with variable pCO₂ treatments from 2016 to 2018**. Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2022-10-13 doi:10.26008/1912/bco-dmo.882390.1 [[view at BCO-DMO](#)]
Relationship Description: Data from the same experiment.

Morris, J. J., Barreto Filho, M. M., Zhiying, L., Walker, M. (2022) **Pipelines for transcriptome analyses conducted as part of "Community context and pCO₂ impact the transcriptome of the "helper" bacterium Alteromonas in co-culture with picocyanobacteria"**. Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2022-10-04 doi:10.26008/1912/bco-dmo.881942.1 [[view at BCO-DMO](#)]
Relationship Description: Data from the same experiment.

Morris, J. J., Zhiying, L. (2022) **Pipeline for phylogenetic analysis of the GlcDEF, GOX/LOX, and tsar genes conducted as part of "Community context and pCO₂ impact the transcriptome of the "helper" bacterium Alteromonas in co-culture with picocyanobacteria"**. Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2022-10-25 doi:10.26008/1912/bco-dmo.882970.1 [[view at BCO-DMO](#)]
Relationship Description: Data from the same experiment.

Parameters

Parameter	Description	Units
Sample	Sample Identifier	unitless
Sample_Temp	Temperature of the sample at the time of pH measurement	degrees Celsius
Salinity	Salinity of the medium	PSU
volume_dye	Amount of dye injected at each injection point	milliliters (mL)
pH_sample_with_dye_corrections	pH of the sample taking into account both dye injections	unitless
pH_sample_without_dye_corrections	pH of the sample only considering the first dye injection	unitless
d730_1	Difference between initial and injection 1 A730 readings	unitless
d730_2	Difference between initial and injection 2 A730 readings	unitless
Sample_A_434	Absorbance at 434nm before dye injection	unitless
Sample_A_578	Absorbance at 578nm before dye injection	unitless
Sample_A_730	Absorbance at 730nm before dye injection	unitless
Sample_and_Dye_A_434	Absorbance at 434nm after 1 dye injection	unitless
Sample_and_Dye_A_578	Absorbance at 578nm after 1 dye injection	unitless
Sample_and_Dye_A_730	Absorbance at 730nm after 1 dye injection	unitless
Sample_and_Dye_x2_A_434	Absorbance at 434nm after 2 dye injections	unitless
Sample_and_Dye_x2_A_578	Absorbance at 578nm after 2 dye injections	unitless
Sample_and_Dye_x2_A_730	Absorbance at 730nm after 2 dye injections	unitless
A1_to_A2sub1	Ratio of A578:A434 after 1 injection	unitless
A1_to_A2sub2	Ratio of A578:A434 after 2 injections	unitless
pK2	pKa of the dye, corrected for salinity and temperature	unitless
delta_A1_to_A2	Difference in absorbance ratios between first and second dye injections	unitless
Constant_A	Constant A = $E1(HI-)/E2(HI-)$; extinction coefficient from Dickson et al. 2007 SOP6b eq. 7	unitless
Constant_B	Constant B = $E1(I-2)/E2(HI-)$; extinction coefficient from Dickson et al. 2007 SOP6b eq. 7	unitless
Constant_C	Constant C = $E2(I-2)/E2(HI-)$; extinction coefficient from Dickson et al. 2007 SOP6b eq. 7	unitless
Dye_Correction_Incerpt_a	Intercept of regression line for the two dye injections, for correcting for effect of dye addition	unitless
Dye_Correction_slope_b	Slope of regression line for the two dye injections, for correcting for effect of dye addition	unitless
A1_to_A2_corrected	Ratio of A578:A434 corrected for effect of dye addition	unitless

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Instruments

Dataset-specific Instrument Name	BioTek H1 plate reader
Generic Instrument Name	plate reader
Generic Instrument Description	<p>Plate readers (also known as microplate readers) are laboratory instruments designed to detect biological, chemical or physical events of samples in microtiter plates. They are widely used in research, drug discovery, bioassay validation, quality control and manufacturing processes in the pharmaceutical and biotechnological industry and academic organizations. Sample reactions can be assayed in 6-1536 well format microtiter plates. The most common microplate format used in academic research laboratories or clinical diagnostic laboratories is 96-well (8 by 12 matrix) with a typical reaction volume between 100 and 200 uL per well. Higher density microplates (384- or 1536-well microplates) are typically used for screening applications, when throughput (number of samples per day processed) and assay cost per sample become critical parameters, with a typical assay volume between 5 and 50 µL per well. Common detection modes for microplate assays are absorbance, fluorescence intensity, luminescence, time-resolved fluorescence, and fluorescence polarization. From: http://en.wikipedia.org/wiki/Plate_reader, 2014-09-0-23.</p>

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Project Information

Collaborative Research: Ecology and Evolution of Microbial Interactions in a Changing Ocean (LTPE)

Coverage: Lab work: Birmingham, Alabama and New York, New York. Field Work: Bermuda Atlantic Time Series.

NSF Award Abstract:

Carbon dioxide released from fossil fuels is causing the ocean to become more acidic. Much attention has been given to how this will affect shelled animals like corals, but acidification also affects the algae that form the base of the ocean food chain. It is possible that future algal communities will look very different than they do today, with potentially negative consequences for fisheries, recreation, and climate. Alternatively, it is possible that these algae will be able to adapt rapidly enough to avoid the worst of it. This study looks at algae adapting to acidification in real time in the lab, focusing on "marketplace" interactions between the algae and the bacteria they live alongside. The researchers also go to sea to learn whether adaptations from the lab experiments are beneficial under real-world conditions. Ultimately, this project is helping scientists better understand how the ocean's most important and most overlooked organisms will respond to the changes humans are causing in their habitat. The researchers also use their scientific work to create fun educational opportunities from grade school to college, including agar art classes where students learn about microbial ecology by "painting" with freshly-isolated ocean bacteria.

The effect of ocean acidification on calcifying organisms has been well-studied, but less is known about how changing pH will affect phytoplankton. Previous work showed that the mutualistic interaction between the globally abundant cyanobacterium *Prochlorococcus* and its "helper" bacterium *Alteromonas* broke down under projected future CO₂ conditions, leading to a strong decrease in the fitness of *Prochlorococcus*. It is possible that such interspecies interactions between microbes are important for many ecological processes, but a lack of understanding of how these interactions evolve makes it difficult to predict how important they are. This project is using laboratory evolution experiments to discover how evolution shapes the interactions between bacteria and algae like *Prochlorococcus*, and how these co-evolutionary dynamics might influence the biogeochemical processes that shape Earth's climate. Four research cruises to the Bermuda Atlantic Time Series are also planned to study how natural algal/bacterial communities respond to acidification, and whether evolved microbes from laboratory experiments have a competitive advantage in complex, natural communities exposed to elevated CO₂. The ultimate goal of this project is to gain a mechanistic understanding of microbial interactions that can be used to inform models of Earth's oceans and biological feedbacks on global climate.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1851085

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