CO2 experiment physiology and carbonate chemistry from laboratory experiments with Pseudo-nitzschia australis conducted from 2021 to 2022

Website: https://www.bco-dmo.org/dataset/906938
Data Type: experimental
Version: 1
Version Date: 2023-08-28

Project
» MCA: Developing transcriptomics as a tool to investigate toxic diatom responses to ocean heatwave and upwelling events (Toxic diatoms and heatwaves)

Contributors	Affiliation	Role
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Abstract
These raw data contain physiological data collected from laboratory experiments with Pseudo-nitzschia australis. This dataset includes replicate data for CO2 experiment physiology and carbonate chemistry. See "Related Datasets" for other physiological measurements published as part of these experiments. See the results publication Kelley et al. (2003) for more detail. The following description provides details for all related physiological measurement datasets. These physiological measurements include: growth rates, domoic acid quotas, domoic acid production rates, net primary productivity, and nitrogen use efficiencies. Also included are pH and DIC measurement used to characterize the carbonate system. These data revealed novel insights into P. australis bloom dynamics and may be useful to harmful algal bloom modelers and were collected and analyzed by Kyla Kelly, Amjad Mansour, Chen Liang, Andrew Kim, Lily Mancini, Dr. Matthew Bertin, Dr. Bethany Jenkins, Dr. David Hutchins, and Dr. Fei-Xue Fu.

Table of Contents

Coverage

Spatial Extent: **Lat**:46.495103 **Lon**:-124.060591
Temporal Extent: 2020-11 - 2022-07

Methods & Sampling

These experiments were conducted with a strain of (strain NWFSC 731) isolated from Long Beach, Washington State, USA on November 3, 2020. The temperature and salinity were 14°C and 27 ppt, respectively at the time of collection. The data was collected in laboratory experiments at the University of Southern California. The experiments began in September 2021 and finished in July of 2022.

Treatments from the CO2 experiments:

Pre-industrial: treatment amended with low pCO2 (calculated as 201.8 µatm)
Ocean acidification: treatment amended with high pCO2 (calculated as 592.2 µatm)
Extreme ocean acidification: treatment amended with extremely high pCO2 (calculated as 1038.9 µatm)

The following section provides a methodology summary for this dataset and references related datasets collected as part of the same experiment (see "Related Datasets" section for data access). A full methodology was published in "Simulated upwelling and marine heatwave events promote similar growth rates but differential domoic acid toxicity in Pseudo-nitzschia australis" in Harmful Algae (Kelly et al., 2023).

Pseudo-nitzschia australis was grown under upwelling heatwave, and extreme heatwave conditions (e.g., combined temperature, nutrient, and carbon dioxide levels specific to each condition) and in single-factor response curves for carbon dioxide, temperature, and varying nitrogen:phosphorus (N:P) ratios/total nutrient concentrations.

Samples for chlorophyll a (used to calculate growth rates) were filtered on GF/F filters, extracted in 6 mL of 90 % acetone at -20°C for 24 h, then analyzed using a Turner 10AU field fluorometer (Welschmeyer 1994; Fu et al. 2007).

For elemental analysis (particulate organic carbon and nitrogen, POC and PON), cells were filtered onto pre-combusted GF/F filters, dried, and analyzed on a Costech 4010 Elemental Analyzer (Fu et al. 2007).

Samples for particulate domoic acid were filtered onto Supor 0.2 µm 47 mm PES filters. Samples were analyzed using LC-MS/MS on a Prominence UFLC system (Shimadzu, Kyoto, Japan) coupled to a SCIEX 4500 QTRAP mass spectrometer (AB Sciex, Framingham, MA, USA). Methods described in Wang et al. 2012.

Primary production was determined by measuring the uptake of radiolabeled bicarbonate (Fu et al. 2008). 14C-bicarbonate was added to 45 mL sub-cultures at T24 h and incubated for 24 h (approximating net carbon fixation) under the respective experimental conditions. After the incubation period, cells were collected on GF/F filters and placed in a scintillation vial containing scintillation cocktail. Samples were stored for 24 h before being read on a Wallac System 1400 liquid scintillation counter.

pH measurements were made on a Mettler Toledo SevenCompact pH meter using a three-point calibration curve and total pH scale (Cooley and Yager 2006). Samples for total DIC analysis were collected at Tfinal. Seawater from undisturbed culture bottles was removed with a sterile syringe, ejected into pre-evacuated borosilicate Exetainers, and poisoned with 5% MgCl2. Total DIC was then measured using a Picarro cavity ring-down spectrophotometer according to Subhas et al. (2015).

For cell count samples (for normalizing cellular domoic acid), 1 mL of the final experimental culture was preserved with 40 ul glutaraldehyde and stored at 4°C in the dark. Cells were counted on a Olympus BX51 microscope using a Sedgewick Rafter Chamber.

Organism:
Pseudo-nitzschia australis, LSID (urn:lsid:marinespecies.org:taxname:246604)

Data Processing Description

Data was processed in using excel, which was used to calculate rates, averages, and standard deviations.

Experimental seawater pCO2 and total alkalinity were calculated from measured DIC and pH using CO2SYS version 2.1 software (Lewis and Wallace, 1998).

Problems/Issues:

The following samples were removed from analysis, as there were issues during data collection or processing:

One DA sample was removed from analysis in the pre-industrial treatment in the CO2 single factor experiment, and from the NP=50, high nutrient, 19°C treatment in the N:P ratio experiment, due to a sampling and/or analytical error. In the cluster experiment, one sample from the extreme heatwave treatment was removed from primary production analyses due to an error made during the assay.

These data points were not included in this dataset.

BCO-DMO Processing Description

* File CO2_experiment.csv was loaded into the BCO-DMO data system with missing identifier indicated by "NA".
** Missing data values are displayed differently based on the file format you download. They are blank in csv files, "NaN" in MatLab files, etc.
* Column names adjusted to conform to BCO-DMO naming conventions designed to support broad re-use by a variety of research tools and scripting languages. [Only numbers, letters, and underscores. Can not start with a number]

[table of contents | back to top]

Data Files

File
CO2 experiments filename: 906938_v1_p-australis-co2.csv (Comma Separated Values (.csv), 864 bytes) MD5:beec845d0a9d8e66667bdbc4fe59d352 Primary data table for dataset 906938 version 1. Replicate data for CO2 experiment physiology and carbonate chemistry.

[table of contents | back to top]

Related Publications

Cooley, S. R., & Yager, P. L. (2006). Physical and biological contributions to the western tropical North Atlantic Ocean carbon sink formed by the Amazon River plume. Journal of Geophysical Research, 111(C8). doi:10.1029/2005jc002954 https://doi.org/10.1029/2005JC002954
Methods

Fu, F.-X., Mulholland, M. R., Garcia, N. S., Beck, A., Bernhardt, P. W., Warner, M. E., Sañudo-Wilhelmy, S. A., & Hutchins, D. A. (2008). Interactions between changing pCO2, N2 fixation, and Fe limitation in the marine unicellular cyanobacterium Crocosphaera. Limnology and Oceanography, 53(6), 2472–2484. Portico. https://doi.org/10.4319/lo.2008.53.6.2472
Methods

Fu, F.-X., Zhang, Y., Feng, Y., & Hutchins, D. A. (2006). Phosphate and ATP uptake and growth kinetics in axenic cultures of the cyanobacteriumSynechococcusCCMP 1334. European Journal of Phycology, 41(1), 15–28. https://doi.org/10.1080/09670260500505037
Methods

Kelly, K. J., Mansour, A., Liang, C., Kim, A. M., Mancini, L. A., Bertin, M. J., Jenkins, B. D., Hutchins, D. A., & Fu, F.-X. (2023). Simulated upwelling and marine heatwave events promote similar growth rates but differential domoic acid toxicity in Pseudo-nitzschia australis. Harmful Algae, 127, 102467. https://doi.org/10.1016/j.hal.2023.102467
Results

Lewis, E., Wallace, D., & Allison, L. J. (1998). Program developed for CO2 system calculations (No. ORNL/CDIAC-105). Brookhaven National Lab., Dept. of Applied Science, Upton, NY (United States); Oak Ridge National Lab., Carbon Dioxide Information Analysis Center, TN (United States). doi: 10.2172/639712
Methods

Subhas, A. V., Rollins, N. E., Berelson, W. M., Dong, S., Erez, J., & Adkins, J. F. (2015). A novel determination of calcite dissolution kinetics in seawater. Geochimica et Cosmochimica Acta, 170, 51–68. https://doi.org/10.1016/j.gca.2015.08.011
Methods

Wang, Z., Maucher-Fuquay, J., Fire, S. E., Mikulski, C. M., Haynes, B., Doucette, G. J., & Ramsdell, J. S. (2012). Optimization of solid-phase extraction and liquid chromatography–tandem mass spectrometry for the determination of domoic acid in seawater, phytoplankton, and mammalian fluids and tissues. Analytica Chimica Acta, 715, 71–79. doi:10.1016/j.aca.2011.12.013
Methods

Welschmeyer, N. A. (1994). Fluorometric analysis of chlorophyll a in the presence of chlorophyll b and pheopigments. Limnology and Oceanography, 39(8), 1985–1992. doi:10.4319/lo.1994.39.8.1985
Methods

[table of contents | back to top]

Related Datasets

IsRelatedTo

Kelly, K. J., Fu, F., Hutchins, D. A., Bertin, M., Mansour, A., Mancini, L. A., Jenkins, B. D., Chen, L., Kim, A. (2023) **Cluster (combined temperature, nutrient concentration, and CO2) results from laboratory experiments with Pseudo-nitzschia australis conducted from 2021 to 2022.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-08-28 doi:10.26008/1912/bco-dmo.906949.1 [view at BCO-DMO]
Relationship Description: Related dataset published in the same results publication (Kelly et al., 2023).

Kelly, K. J., Fu, F., Hutchins, D. A., Bertin, M., Mansour, A., Mancini, L. A., Jenkins, B. D., Chen, L., Kim, A. (2023) **N:P ratio experiment physiology and carbonate chemistry laboratory experiments with Pseudo-nitzschia australis conducted from 2021 to 2022.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-08-28 doi:10.26008/1912/bco-dmo.906858.1 [view at BCO-DMO]
Relationship Description: Related dataset published in the same results publication (Kelly et al., 2023).

Kelly, K. J., Fu, F., Hutchins, D. A., Bertin, M., Mansour, A., Mancini, L. A., Jenkins, B. D., Chen, L., Kim, A. (2023) **Single-factor temperature experiment physiology and carbonate chemistry from laboratory experiments with Pseudo-nitzschia australis conducted from 2021 to 2022.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2023-08-28 doi:10.26008/1912/bco-dmo.906927.1 [view at BCO-DMO]
Relationship Description: Related dataset published in the same results publication (Kelly et al., 2023).

[table of contents | back to top]

Parameters

Parameter	Description	Units
Treatment	Treatment type (Upwelling= 13°C, high CO2, high nutrients; Heatwave= 19°C, low CO2, low nutrients; Extreme heatwave= 21°C, low CO2, low nutrients; LTCN (low temperature, CO2, and nitrogen) = 13°C, low CO2, low nutrients).	unitless
Replicate	replicate number (1..3)	unitless
Growth_rate	Growth rate per day. Chlorophyll a samples were collected at T-initial and T-final and used for determination of growth rates. The following equation was used to calculate specific growth rates: growth rate = ln(Tfinal-Tinitial)/2 (where Tfinal and Tinitial are the chlorophyll a samples collected at their respective times, and 2 is the number of days between sampling). Null values indicate data point is missing as there was an issue during data collection or processing.	per day (d-1)
Particulate_DA	Particulate domoic acid. The amount of intracellular domoic acid normalized to particulate organic carbon. Null values indicate data point is missing as there was an issue during data collection or processing.	nanograms of domoic acid per micromole of carbon (ng DA/umol C)
DA_production_rate	Domoic acid production rate. Domoic acid production rates were calculated by multiplying specific growth rates by DA quotas. This value provides an estimate of how toxic a bloom might be, based on the ability of Pseudo-nitzschia to increase cell abundances and produce high DA quotas (per mol POC). Null values indicate data point is missing as there was an issue during data collection or processing.	nanograms of domoic acid per micromole of carbon per day (ng DA/umol C/day)
Measured_DIC	Measured dissolved inorganic carbon (DIC). The amount of aqueous carbon dissolved in seawater. Null values indicate data point is missing as there was an issue during data collection or processing.	miromoles per kilogram (umol/kg)
Measured_pH	Measured pH. Null values indicate data point is missing as there was an issue during data collection or processing.	total pH scale
Calculated_bulk_alkalinity	Calculated bulk alkalinity. The buffering capacity of seawater comprised on weak acids and their conjugate bases. This was calculated using CO2sys. Null values indicate data point is missing as there was an issue during data collection or processing.	miromoles per kilogram (umol/kg)
Calculated_pCO2	Calculated pCO2. The partial pressure of carbon dioxide in seawater. This was calculated using CO2sys. Null values indicate data point is missing as there was an issue during data collection or processing.	microatmospheres (uatm)

[table of contents | back to top]

Instruments

Dataset-specific Instrument Name	SCIEX 4500 QTRAP mass spectrometer (AB Sciex, Framingham, MA, USA)
Generic Instrument Name	Accelerator Mass Spectrometer
Generic Instrument Description	An AMS measures "long-lived radionuclides that occur naturally in our environment. AMS uses a particle accelerator in conjunction with ion sources, large magnets, and detectors to separate out interferences and count single atoms in the presence of 1x1015 (a thousand million million) stable atoms, measuring the mass-to-charge ratio of the products of sample molecule disassociation, atom ionization and ion acceleration." AMS permits ultra low-level measurement of compound concentrations and isotope ratios that traditional alpha-spectrometry cannot provide. More from Purdue University: http://www.physics.purdue.edu/primelab/introduction/ams.html

Dataset-specific Instrument Name	Costech 4010 Elemental Analyzer
Generic Instrument Name	Elemental Analyzer
Generic Instrument Description	Instruments that quantify carbon, nitrogen and sometimes other elements by combusting the sample at very high temperature and assaying the resulting gaseous oxides. Usually used for samples including organic material.

Dataset-specific Instrument Name	Turner 10AU field fluorometer
Generic Instrument Name	Fluorometer
Generic Instrument Description	A fluorometer or fluorimeter is a device used to measure parameters of fluorescence: its intensity and wavelength distribution of emission spectrum after excitation by a certain spectrum of light. The instrument is designed to measure the amount of stimulated electromagnetic radiation produced by pulses of electromagnetic radiation emitted into a water sample or in situ.

Dataset-specific Instrument Name	Prominence UFLC system (Shimadzu, Kyoto, Japan)
Generic Instrument Name	High-Performance Liquid Chromatograph
Dataset-specific Description	Prominence UFLC system (Shimadzu, Kyoto, Japan) coupled to a SCIEX 4500 QTRAP mass spectrometer (AB Sciex, Framingham, MA, USA)
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	Wallac System 1400 liquid scintillation counter
Generic Instrument Name	Liquid Scintillation Counter
Generic Instrument Description	Liquid scintillation counting is an analytical technique which is defined by the incorporation of the radiolabeled analyte into uniform distribution with a liquid chemical medium capable of converting the kinetic energy of nuclear emissions into light energy. Although the liquid scintillation counter is a sophisticated laboratory counting system used the quantify the activity of particulate emitting (ß and a) radioactive samples, it can also detect the auger electrons emitted from 51Cr and 125I samples. Liquid scintillation counters are instruments assaying alpha and beta radiation by quantitative detection of visible light produced by the passage of rays or particles through a suitable scintillant incorporated into the sample.

Dataset-specific Instrument Name	Olympus BX51 microscope
Generic Instrument Name	Microscope - Optical
Generic Instrument Description	Instruments that generate enlarged images of samples using the phenomena of reflection and absorption of visible light. Includes conventional and inverted instruments. Also called a "light microscope".

Dataset-specific Instrument Name	Mettler Toledo SevenCompact pH meter
Generic Instrument Name	pH Sensor
Dataset-specific Description	Mettler Toledo SevenCompact pH meter (calibrated using a three-point calibration curve and total pH scale)
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	Picarro cavity ring-down spectrophotometer
Generic Instrument Name	Spectrophotometer
Generic Instrument Description	An instrument used to measure the relative absorption of electromagnetic radiation of different wavelengths in the near infra-red, visible and ultraviolet wavebands by samples.

[table of contents | back to top]

Project Information

MCA: Developing transcriptomics as a tool to investigate toxic diatom responses to ocean heatwave and upwelling events (Toxic diatoms and heatwaves)

Coverage: University of Southern California

NSF Award Abstract:
The diatom Pseudo-nitzschia forms large, toxic harmful algal blooms along the U.S. West Coast, killing wildlife and harming valuable ocean fisheries. Understanding the causes of these blooms and predicting their occurrence, both now and under future changing climate conditions, is critical to coastal environmental and economic health. Puzzlingly, these blooms seem to happen during periods when coastal seawater upwelling results in cold, nutrient-rich, low pH sea surface conditions, and also during times when heat wave events cause warm, nutrient-poor, high pH conditions. These two extremes are forecast to get even more intense with climate change. This project is experimentally testing how Pseudo-nitzschia responds to upwelling and heat wave events using measurements of cell growth, toxin production, and gene expression. Broader impacts of this project include training the principal investigator in new gene expression methods, graduate and undergraduate research training, high school research mentoring experiences, and outreach and communications activities aimed at the commercial fishing industry. Societal benefits include obtaining a better understanding of the causes of damaging toxic algal blooms, and how they may change in the future coastal ocean.

The toxic diatom Pseudo-nitzschia causes annual harmful blooms along the US West Coast, a region where wind-driven upwelling brings rich nutrient supplies into the euphotic zone. However, this region is also experiencing unprecedented episodic ocean heatwave events linked to global warming. Thus, future climate trends in this region suggest an exaggeration of current physio-chemical extremes between colder, more nutrient-rich, low pH upwelling, and warmer, more nutrient-depleted, higher pH heatwaves. Surprisingly, toxic Pseudo-nitzschia spp. can bloom under both upwelling and heatwave conditions, despite opposite trends in key environmental controls like nutrients, temperature, and carbonate chemistry. This project is testing how this happens by first obtaining full response curves for each of the individual factors, temperature, pCO2,

phosphorus, nitrogen, and silicon for two Pseudo-nitzschia isolates. Then, these variables are combined in holistic upwelling and heatwave scenario incubation experiments, to compare how growth and toxicity is affected in both cultures and natural blooms of Pseudo-nitzschia. The PI is assessing toxic diatom responses in these experiments using her existing expertise in algal physiology, as well as by expanding her professional horizons to develop new skills in transcriptome bioinformatics in partnership with Dr. Bethany Jenkins from the University of Rhode Island. Experiments are conducted to test the physiological responses of Pseudo-nitzschia to changes in nutrient concentrations, temperature and pCO2 during simulated upwelling or heatwave occurrences, and measure expression of key metabolic pathway genes such as toxin synthesis pathways. This project is helping to understand and interpret the surprising niche flexibility of toxic Pseudo-nitzschia in a changing ocean, and at the same time offers the PI a new avenue forward for her future career development.

This award reflects NSF's statutory mission and has been deemed worthy of support through evaluation using the Foundation's intellectual merit and broader impacts review criteria.

[table of contents | back to top]

Funding

Funding Source	Award
National Oceanic and Atmospheric Administration (NOAA)	NA18OAR4170094
NSF Division of Ocean Sciences (NSF OCE)	OCE-2120619
National Institutes of Health (NIH)	NIH-P20GM103430

[table of contents | back to top]