

Experimental coral physiological and $\delta^{15}\text{N}$ isotopic measurements in October 2012 at Reef Systems Coral Farm, Ohio.

Website: <https://www.bco-dmo.org/dataset/839920>

Data Type: experimental

Version: 1

Version Date: 2022-03-21

Project

» [Interactive Effects of Temperature, Nutrients, and Ocean Acidification on Coral Physiology and Calcification](#) (OA_coral_physiology)

Program

» [Science, Engineering and Education for Sustainability NSF-Wide Investment \(SEES\): Ocean Acidification \(formerly CRI-OA\)](#) (SEES-OA)

Contributors	Affiliation	Role
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Abstract

Experimental coral physiological and $\delta^{15}\text{N}$ isotopic measurements in October 2012 at Reef Systems Coral Farm, Ohio.

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Coverage

Spatial Extent: Lat:40.123 Lon:-82.782

Temporal Extent: 2012-10-06 - 2012-10-13

Dataset Description

Details of instruments used during the experiment can be found in Hoadley et al. (2016).

Other comments on the data:

- No control data (treatment 1) for *A. millepora*.
- No DOC or POC samples were collected for *A. millepora*.

- No $\delta^{15}\text{N}$ coral host data was collected for *T. reniformis*, and no $\delta^{15}\text{N}$ whole data was collected for *A. millepora*.

Methods & Sampling

Prior to the start of this experiment in September 2012, six colonies of *A. millepora* and *T. reniformis* originally collected from 3-10m depth in Fiji (17°29'19"S, 177°23'39"E) were maintained for 18 months at 26.4°C \pm 0.04 SE in 3785 L recirculating indoor aquaria with artificial seawater (Instant Ocean Reef Crystals) in a greenhouse (700–1000 $\mu\text{mol quanta}^{-1} \text{m}^{-2} \text{s}^{-1}$) at the Reef Systems Coral Farm mariculture facility in New Albany, Ohio (40°07'24"N, 82°46'55"W). In January 2012, *A. millepora* and *T. reniformis* colonies were divided into coral ramets (n=8 per colony), mounted on 5cm PVC tiles using EcoTech coral glue, and allowed to recover. On 6 August 2012, coral ramets were placed into indoor experimental tanks (57 L) with artificial light (Tek Light T5 actinic lights, 275 $\mu\text{mol quanta}^{-1} \text{m}^{-2} \text{s}^{-1}$, 10:14 hours light:dark diurnal cycle) under ambient conditions (26.4°C, 402 μatm) and allowed to acclimate for four weeks.

The experimental systems in this study were previously outlined in Hoadley *et al.* (2016). Briefly, from 7 – 16 September 2012, treatment conditions were initiated: temperature, $p\text{CO}_2$, and nutrients were gradually increased over the course of a week until target conditions in each treatment were reached to minimize shocking any of the corals. The treatments consisted of a control (26.4°C, 402 μatm), elevated $p\text{CO}_2$ (26.4°C, 760 μatm), elevated temperature (29.8°C, 402 μatm), and combined elevated temperature and $p\text{CO}_2$ (29.8°C, 760 μatm). Half of the tanks in each treatment were under ambient nutrient concentrations (0.41 $\mu\text{mol L}^{-1}$ NO_3^- and 0.25 $\mu\text{mol L}^{-1}$ PO_4^{3-}) and the other half under moderate nutrient concentrations (3.56 $\mu\text{mol L}^{-1}$ NO_3^- and 0.31 $\mu\text{mol L}^{-1}$ PO_4^{3-}). Experimental conditions (ramping plus target conditions) lasted for 30 days, and coral were fed fresh, two-day old *Artemia nauplii* (Carolina Biological Supply) twice each week.

Data Processing Description

Calcification. Net calcification was determined using the buoyant weight technique (Jokiel *et al.* 1978). Each coral fragment was weighed at the beginning and end of the experiment. Buoyant weight was normalized to dry weight.

Gross photosynthesis. Gross photosynthesis was calculated from net photosynthesis and respiration; these measurements were made on the live coral fragments incubated in sealed chambers (Hoadley *et al.* 2016).

CZAR, CHARToc, CTAR. Gross photosynthesis and respiration were used to calculate CZAR (Muscatine *et al.* 1981). Dissolved and particulate organic carbon samples (DOC and POC) were collected following incubations. DOC samples were analyzed using a Shimadzu TOC-L total organic carbon analyzer (using the 680°C combustion catalytic oxidation method) (Levas *et al.* 2015). POC filters (GF/F) were acid fumigated (Levas *et al.* 2015) and combusted in an Elementar Vario EL Cube/Micro Cube elemental analyzer interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer at the stable isotope facility at the University of California - Davis. CHARToc was calculated using methods modified from Levas *et al.* (2015). CZAR and CHARZOO were summed to yield CTAR (Grottoli *et al.* 2014).

For tissue analyses, corals were frozen at -80 degrees C and samples were either ground or airbrushed for further analyses.

Surface area. Surface area was measured on *Acropora millepora* using the wax-dipping technique (Veal *et al.* 2010) and on *Turbinaria reniformis* using the aluminum foil method (Marsh 1970).

Tissue biomass. Ash free dry weight was calculated using methods in McLachlan *et al.* (2020) on ground growing tips of *A. millepora* and water-picked tissue slurry of *T. reniformis*.

Chlorophyll a. *A. millepora* chlorophyll a was measured from tissue slurry using 100% acetone (Jeffrey & Humphrey 1975). *T. reniformis* chlorophyll data was from Hoadley *et al.* (2016).

Protein. Protein was measured using the bicinchoninic method (Smith *et al.* 1985) with bovine serum albumin as a standard (Pierce BCA Protein Assay Kit). This was carried out on air-brushed tissue slurry of *A. millepora* and water-picked and freeze-dried tissue slurry for *T. reniformis*. Protein was reported in Joules (Gnaiger & Bitterlich, 1984) and standardized to total biomass of the sub-sample.

Carbohydrates. Carbohydrates were measured using the phenol-sulfuric acid spectrophotometric method (Dubois et al. 1956) with glucose standards. This was carried out on air-brushed tissue slurry of *A. millepora* and ground *T. reniformis*. Carbohydrates were reported in Joules (Gnaiger & Bitterlich, 1984) and standardized to total biomass of the sub-sample.

Total lipids. Total lipids were measured using chloroform:methanol (2:1, v:v) with two KCl rinses (Baumann et al. 2014). This was carried out on air-brushed tissue slurry of *A. millepora* and water-piked and freeze-dried tissue slurry for *T. reniformis*. Total lipids were reported in Joules (Gnaiger & Bitterlich, 1984) and standardized to total biomass of the sub-sample.

$\delta^{15}\text{N}$ isotopes (coral host, algal endosymbiont, whole tissue). Samples were prepped for isotopic analysis using a method modified from Hughes and Grottoli (2010). All *A. millepora* host and algal endosymbiont fractions, partitioned from airbrushed tissue slurry, were combusted in a Costech elemental analyzer and the resulting N_2 gas analyzed with a Thermo Finnigan Delta IV stable isotope ratio mass spectrometer (SIRMS) via a ConFlow open split interface in the Grottoli Stable Isotope Biogeochemistry Lab at The Ohio State University. All *T. reniformis* whole (freeze-dried tissue slurry) and algal endosymbiont fractions were combusted in an Elementar Vario EL Cube/Micro Cube elemental analyzer interfaced to a PDZ Europa 20-20 SIRMS at the stable isotope facility at University of California - Davis. Repeated measurements of an internal standard had an average SD $\pm 0.08\text{‰}$ $\delta^{15}\text{N}$. The $\delta^{15}\text{N}$ values of both species are reported as the per mil deviation of the ratio of stable nitrogen isotopes $^{15}\text{N}:^{14}\text{N}$ relative to air. Approximately 10% of *A. millepora* samples were run in duplicate with an average SD $\pm 0.21\text{‰}$.

A Euclidean distance-based resemblance matrix was constructed using normalized data of primary physiological variables: P, calcification, biomass, protein, and total lipids. Non-metric multi-dimensional scaling (NMDS) plots were generated to visualize relationships between each coral ramet across all treatments, for each species. Analysis of similarities (ANOSIM) was used to evaluate the degree of dissimilarity among treatments. All multivariate analyses were conducted using the software package Primer v6 (Clarke & Gorley 2006). Prior to further statistical analysis, all data were tested for normality by a Shapiro-Wilk's test and homogeneity of variance was assessed with plots of expected vs. residual values. Univariate three-way analysis of variance (ANOVA) was used to test the effects of temperature, $p\text{CO}_2$, nutrients, and genotype on each measured variable for each species. Temperature was fixed with 2 levels (26.4°C, 29.8°C), $p\text{CO}_2$ fixed with 2 levels (401 μatm , 760 μatm), nutrients fixed with 2 levels (ambient, moderate). All univariate parametric statistics were generated using SAS software, Version 9.3 of the SAS System for Windows. Values of $p \leq 0.05$ were considered significant.

Additional details of the statistical analysis are outlined in Dobson et al. 2020.

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Data Files

File
nutrients.csv (Comma Separated Values (.csv), 12.69 KB) MD5:8ff32ee405fa84a1a8e351081682560b
Primary data file for dataset ID 839920

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Related Publications

Cai, W.-J., Ma, Y., Hopkinson, B. M., Grottoli, A. G., Warner, M. E., Ding, Q., ... Wang, Y. (2016). Microelectrode characterization of coral daytime interior pH and carbonate chemistry. *Nature Communications*, 7(1).

doi:[10.1038/ncomms11144](https://doi.org/10.1038/ncomms11144)

Methods

Dobson, K. L., Levas, S., Schoepf, V., Warner, M. E., Cai, W.-J., Hoadley, K. D., Yuan, X., Matsui, Y., Melman, T. F., & Grottoli, A. G. (2021). Moderate nutrient concentrations are not detrimental to corals under future ocean conditions. *Marine Biology*, 168(7). <https://doi.org/10.1007/s00227-021-03901-3>

Results

Grottoli, A. G., Dalcin Martins, P., Wilkins, M. J., Johnston, M. D., Warner, M. E., Cai, W.-J., ... Schoepf, V. (2018). Coral physiology and microbiome dynamics under combined warming and ocean acidification. PLOS ONE, 13(1), e0191156. doi:[10.1371/journal.pone.0191156](https://doi.org/10.1371/journal.pone.0191156)

Methods

Hoadley, K. D., Pettay, D. T., Grottoli, A. G., Cai, W.-J., Melman, T. F., Levas, S., ... Warner, M. E. (2016). High-temperature acclimation strategies within the thermally tolerant endosymbiont *Symbiodinium trenchii* and its coral host, *Turbinaria reniformis*, differ with changing pCO₂ and nutrients. Marine Biology, 163(6). doi:[10.1007/s00227-016-2909-8](https://doi.org/10.1007/s00227-016-2909-8)

Methods

Levas, S., Grottoli, A., Warner, M., Cai, W., Bauer, J., Schoepf, V., ... Wang, Y. (2015). Organic carbon fluxes mediated by corals at elevated pCO₂ and temperature. Marine Ecology Progress Series, 519, 153–164.

doi:[10.3354/meps11072](https://doi.org/10.3354/meps11072)

Methods

Schoepf, V., Grottoli, A. G., Warner, M. E., Cai, W.-J., Melman, T. F., Hoadley, K. D., ... Baumann, J. H. (2013). Coral Energy Reserves and Calcification in a High-CO₂ World at Two Temperatures. PLoS ONE, 8(10), e75049.

doi:[10.1371/journal.pone.0075049](https://doi.org/10.1371/journal.pone.0075049)

Methods

Yuan, X., Cai, W.-J., Meile, C., Hopkinson, B. M., Ding, Q., Schoepf, V., ... Grottoli, A. G. (2018). Quantitative interpretation of vertical profiles of calcium and pH in the coral coelenteron. Marine Chemistry, 204, 62–69.

doi:[10.1016/j.marchem.2018.06.001](https://doi.org/10.1016/j.marchem.2018.06.001)

Methods

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Parameters

Parameter	Description	Units
Date_Collected	Collection date in ISO format (yyyy-mm-dd)	unitless
Latitude	Latitude of sampling location, south is negative	decimal degrees
Longitude	Longitude of sampling location, west is negative	decimal degrees
Species	Taxonomic name of the coral species	unitless
Sample_ID	ID number of the coral sample	unitless
Treatment	No description provided	units
Temperature_Water	Water temperature	units
pCO2	Partial pressure of CO2	micro-atmospheres (μatm)
Nutrient_Concentration	Nutrient concentration of the water	mol/L
Genotype	Genotype (parent colony) identifier	unitless
Total_Calcification	Calcification rate across the entire experiment using buoyant weight, normalized to dry weight, and standardized to the surface area of the coral fragment	milligrams per day per square centimeters (mg/day/cm ²)
Gross_Photosynthesis	Gross photosynthesis of the coral fragment in amount of oxygen produced, standardized to time and surface area of the fragment	micromoles per minute per square centimeters ($\mu\text{mol}/\text{min}/\text{cm}^2$)
CZAR	Contribution of Zooxanthellae to Animal Respiration	percentage (%)
CHARTOC	Contribution of Heterotrophy to Animal Respiration from total organic carbon, measured in percentage	percentage (%)
CTAR	Contribution of Total Acquired fixed carbon relative to animal Respiration, measured in percentage	percentage (%)
Tissue_Biomass	The ash-free dry weight of the whole tissue (animal host and algal endosymbiont) standardized to surface area	milligrams per square centimeters (mg/cm ²)
Chlorophyll_a	The chlorophyll a content of the endosymbiont fraction, standardized to surface area of the fragment	micrograms per square centimeters ($\mu\text{g}/\text{cm}^2$)
Protein	The soluble protein content of the animal host fraction, converted into Joules and standardized to ash-free dry weight	Joules per g ash-free dry weight (J/gdw)
Carbohydrates	The soluble carbohydrate content of the animal host fraction, converted into Joules and standardized to ash-free dry weight	Joules per g ash-free dry weight (J/gdw)
Total_lipids	The soluble lipid content of the combined animal host and algal endosymbiont fraction, converted to Joules and standardized to ash-free dry weight	Joules per g ash-free dry weight (J/gdw)
Delta15N_Whole	Stable nitrogen isotopes of the combined animal host and algal endosymbiont, relative to air	parts per thousand (‰)
Delta15N_Algal_Endosymbionts	Stable nitrogen isotopes of the isolated algal endosymbiont, relative to air	parts per thousand (‰)
Delta15N_Coral_Host	Stable nitrogen isotopes of the isolated animal host, relative to air	parts per thousand (‰)

Instruments

Dataset-specific Instrument Name	Costech elemental analyzer
Generic Instrument Name	Elemental Analyzer
Dataset-specific Description	A. millepora $\delta^{15}\text{N}$ samples measured on Costech elemental analyzer coupled to a Thermo Finnigan Delta IV stable isotope ratio mass spectrometer (EA-SIRMS).
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	Elementar Vario EL Cube/Micro Cube elemental analyzer
Generic Instrument Name	Elemental Analyzer
Dataset-specific Description	T. reniformis $\delta^{15}\text{N}$ samples measured on Elementar Vario EL Cube/Micro Cube elemental analyzer interfaced to a PDZ Europa 20-20 stable isotope ratio mass spectrometer (EA-SIRMS).
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	Thermo Finnigan Delta IV stable isotope ratio mass spectrometer
Generic Instrument Name	Isotope-ratio Mass Spectrometer
Dataset-specific Description	A. millepora $\delta^{15}\text{N}$ samples measured on Costech elemental analyzer coupled to a Thermo Finnigan Delta IV stable isotope ratio mass spectrometer (EA-SIRMS).
Generic Instrument Description	The Isotope-ratio Mass Spectrometer is a particular type of mass spectrometer used to measure the relative abundance of isotopes in a given sample (e.g. VG Prism II Isotope Ratio Mass-Spectrometer).

Dataset-specific Instrument Name	PDZ Europa 20-20 stable isotope ratio mass spectrometer
Generic Instrument Name	Isotope-ratio Mass Spectrometer
Dataset-specific Description	T. reniformis $\delta^{15}\text{N}$ samples measured on Elementar Vario EL Cube/Micro Cube elemental analyzer interfaced to a PDZ Europa 20-20 stable isotope ratio mass spectrometer (EA-SIRMS).
Generic Instrument Description	The Isotope-ratio Mass Spectrometer is a particular type of mass spectrometer used to measure the relative abundance of isotopes in a given sample (e.g. VG Prism II Isotope Ratio Mass-Spectrometer).

Dataset-specific Instrument Name	Thermo Scientific Genesys
Generic Instrument Name	Spectrophotometer
Dataset-specific Description	A Thermo Scientific Genesys spectrophotometer was used to measure the intensity of light transmission at specific wavelengths for chlorophyll a, protein and carbohydrate samples of both species.
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.

Dataset-specific Instrument Name	Shimadzu TOC-L total organic carbon analyzer
Generic Instrument Name	Total Organic Carbon Analyzer
Dataset-specific Description	DOC samples measured on Shimadzu TOC-L total organic carbon analyzer.
Generic Instrument Description	A unit that accurately determines the carbon concentrations of organic compounds typically by detecting and measuring its combustion product (CO_2). See description document at: http://bcodata.whoi.edu/LaurentianGreatLakes_Chemistry/bs116.pdf

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Project Information

Interactive Effects of Temperature, Nutrients, and Ocean Acidification on Coral Physiology and Calcification (OA_coral_physiology)

Coverage: Reef Systems Coral Farm, New Albany, Ohio, USA

Extracted from the NSF award abstract:

Atmospheric and sea surface CO_2 concentrations are expected to continue to increase substantially over the

coming decades resulting in warmer and more acidic oceans, which will greatly stress the health of coral reefs. In addition, ocean margins where most corals live will also see continued increases in human-produced nutrient inputs. While there has recently been a considerable focus on how ocean acidification (due to higher CO₂ alone) could negatively impact the growth of reef-building corals due to the projected loss in calcification, the combined impacts of CO₂, temperature, and nutrients on coral physiology and calcification are poorly understood. This project will investigate the possible synergistic and antagonistic effects of elevated temperature, CO₂, and nutrients on the physiology and internal calcifying chemistry of several species of corals in a laboratory setting. Research tools will include the assessment of coral energy reserves and metabolic demand, symbiotic algal physiology and molecular diversity, coral calcification, and direct measurement of the internal coral pH and carbonate concentration via microprobes. The results from this project have the potential to supply broad scientific impacts regarding how (or if) reef-building corals will survive future climate change scenarios, and will help establish several parameter ranges that could be used to strengthen ocean acidification and coral reef growth models.

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Program Information

Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES): Ocean Acidification (formerly CRI-OA) (SEES-OA)

Website: https://www.nsf.gov/funding/pgm_summ.jsp?pims_id=503477

Coverage: global

NSF Climate Research Investment (CRI) activities that were initiated in 2010 are now included under Science, Engineering and Education for Sustainability NSF-Wide Investment (SEES). SEES is a portfolio of activities that highlights NSF's unique role in helping society address the challenge(s) of achieving sustainability. Detailed information about the SEES program is available from NSF (https://www.nsf.gov/funding/pgm_summ.jsp?pims_id=504707).

In recognition of the need for basic research concerning the nature, extent and impact of ocean acidification on oceanic environments in the past, present and future, the goal of the SEES: OA program is to understand (a) the chemistry and physical chemistry of ocean acidification; (b) how ocean acidification interacts with processes at the organismal level; and (c) how the earth system history informs our understanding of the effects of ocean acidification on the present day and future ocean.

Solicitations issued under this program:

[NSF 10-530](#), FY 2010-FY2011

[NSF 12-500](#), FY 2012

[NSF 12-600](#), FY 2013

[NSF 13-586](#), FY 2014

NSF 13-586 was the final solicitation that will be released for this program.

PI Meetings:

[1st U.S. Ocean Acidification PI Meeting](#) (March 22-24, 2011, Woods Hole, MA)

[2nd U.S. Ocean Acidification PI Meeting](#) (Sept. 18-20, 2013, Washington, DC)

3rd U.S. Ocean Acidification PI Meeting (June 9-11, 2015, Woods Hole, MA – Tentative)

NSF media releases for the Ocean Acidification Program:

[Press Release 10-186 NSF Awards Grants to Study Effects of Ocean Acidification](#)

[Discovery Blue Mussels "Hang On" Along Rocky Shores: For How Long?](#)

[Discovery nsf.gov - National Science Foundation \(NSF\) Discoveries - Trouble in Paradise: Ocean Acidification This Way Comes - US National Science Foundation \(NSF\)](#)

[Press Release 12-179 nsf.gov - National Science Foundation \(NSF\) News - Ocean Acidification: Finding New](#)

[Answers Through National Science Foundation Research Grants - US National Science Foundation \(NSF\)](#)

[Press Release 13-102 World Oceans Month Brings Mixed News for Oysters](#)

[Press Release 13-108 nsf.gov - National Science Foundation \(NSF\) News - Natural Underwater Springs Show How Coral Reefs Respond to Ocean Acidification - US National Science Foundation \(NSF\)](#)

[Press Release 13-148 Ocean acidification: Making new discoveries through National Science Foundation research grants](#)

[Press Release 13-148 - Video nsf.gov - News - Video - NSF Ocean Sciences Division Director David Conover answers questions about ocean acidification. - US National Science Foundation \(NSF\)](#)

[Press Release 14-010 nsf.gov - National Science Foundation \(NSF\) News - Palau's coral reefs surprisingly resistant to ocean acidification - US National Science Foundation \(NSF\)](#)

[Press Release 14-116 nsf.gov - National Science Foundation \(NSF\) News - Ocean Acidification: NSF awards \\$11.4 million in new grants to study effects on marine ecosystems - US National Science Foundation \(NSF\)](#)

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Funding

Funding Source	Award
NSF Emerging Frontiers Division (NSF EF)	EF-1041124

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