Buoyant weight data collected during acute and chronic stress exposures in outdoor mesocosms in Hawaii from January 2022 to March 2024

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Project
» Collaborative Research: Reevaluating calcification response to changes in seawater chemistry by testing the Proton Flux Hypothesis and the Coral Metabolism Model (Proton Flux Hypothesis)

Contributors	Affiliation	Role
Bahr, Keisha D.	Texas A&M, Corpus Christi (TAMU-CC)	Principal Investigator
McNicholl, Conall	Hawaii Institute of Marine Biology (HIMB)	Scientist
Armstrong, David A.	Texas A&M, Corpus Christi (TAMU-CC)	Student
Bretzing-Tungate, Robert	Texas A&M, Corpus Christi (TAMU-CC)	Data Manager
York, Amber D.	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
Coral buoyant weight data collected from Montipora capitata (urn:lsid:marinespecies.org:taxname:287697) and Pocillopora acuta (urn:lsid:marinespecies.org:taxname:759099) during acute and chronic stress exposures in outdoor mesocosms in Hawaii from January 2022 to March 2024. This dataset includes demographic data (date measure was taken, experiment number, time point, season experiment was conducted in, treatment, mesocosm number, species code, coral individual) and corresponding buoyant weight value. Study description (discusses related data collected during the experiment): This project examined the fundamental connections between seawater chemistry and coral physiology by investigating the modulation of seawater chemistry in the microenvironment surrounding corals. Coral biological response variables were quantified during short-term incubations and long-term mesocosm manipulations to understand physiological implications across multiple scales (i.e., individual and community scales) and across boundary layers. This project generated several datasets to investigate coral response (biological and physiological), community response (ecosystem), environmental characteristics (water quality and seawater chemistry), and calibration data for quality control and standardization (see "Related Datasets" and "Supplemental Files" sections). All datasets are provided in .csv format for broad-scale accessibility and use. All biological data includes demographic variables to describe the experiment (experiment number, season experiment was conducted in, mesocosm number coral was held in, species code, coral individual number, and time point), while all water data includes a different set of demographic data (experiment number, season experiment was conducted in, and date measurements were taken). All datasets are organized in tidy format with each row equating to a single measurement and each column a separate variable.

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Coverage

Location: Hawaii Institute of Marine Biology, Kaneohe Bay, Hawaii

Spatial Extent: **Lat**:21.4327 **Lon**:-157.7877
Temporal Extent: 2022-06-07 - 2024-03-18

Methods & Sampling

Overview

Seasonal mesocosm incubations of Montipora capitata and Pocillopora acuta were conducted over a two-year period (June 2022–March 2024) at the Hawai'i Institute of Marine Biology (HIMB). Coral were collected from fringe reefs located around the institute. Experiments were carried out during winter (January 10–March 17) and summer (June 10–August 17) in outdoor flow-through mesocosms at the Hawaii Institute of Marine Biology in Kane`ohe, Hawai`i. Due to facility constraints, all treatments could not run simultaneously. Therefore, each seasonal experiment consisted of two consecutive 30-day exposures, each using newly collected coral colonies. All procedures were standardized across replicates, except for adjustments in total alkalinity (TA) and pH required to create the treatments.

Coral Collection and Pre-Experiment Holding

Coral colonies of Montipora capitata and Pocillopora acuta were collected by hand while snorkeling from shallow (1 m) fringing reef habitat surrounding HIMB in Kāne'ohe Bay, Hawai'i. Colonies were transported immediately to the mesocosm facility and held for one week in flow-through tanks that were identical in design to the experimental mesocosms.

One day prior to placement in experimental mesocosms, all colonies were stained for eight hours with alizarin red to mark the initial skeletal growth band (reference: Jokiel and Morrissey 1993).

At the start of each 30-day experiment, colonies were randomly assigned to mesocosms, buoyant weighed following Jokiel (1978), and placed into a standardized grid layout. Each mesocosm contained 20 colonies of each species arranged in alternating species order to avoid species-level clustering effects.

Mesocosm Facility Design

The flow-through mesocosm facility (Jokiel et al. 2014) consisted of twelve 450 L fiberglass tanks operating on continuous seawater flow. Tanks were arranged in four rows of three mesocosms, with each row supplied by a dedicated header tank. Water was pumped at ~3 m depth from the adjacent bay, supplying unfiltered seawater that preserved natural diel and seasonal variability.

Each header tank fed seawater into a 100 L mixing reservoir positioned above the system to facilitate chemical manipulation before seawater entered the mesocosms. Mesocosm residence time was greater than one hour. Each mesocosm contained two submersible circulation pumps and an airstone that ran continuously to maintain water movement and oxygenation.

Treatment Conditions and Carbonate Chemistry Manipulation

Four carbonate chemistry conditions were generated across consecutive 30-day exposures by altering total alkalinity (TA), dissolved inorganic carbon (DIC), and pH. Target conditions included:

1. Ambient control

2. Low pH (ΔpH ~ -0.3 from ambient)

3. High or low TA (±100 µmol kg-1 from ambient)

4. Combined low pH and TA alteration

TA adjustments were made using a peristaltic pump delivering either 1.0 M HCl (for low TA) at ~3 mL min-1 or 1.0 M Na2CO3 (for high TA) at ~2 mL min-1 directly into each row's mixing tank.
pH reductions were achieved by bubbling pure CO2 or a CO2-air mixture directly into designated mesocosms

using a Maxi-Jet 1600 pump-driven venturi injector.

Because each row was supplied by a single header tank, only one direction of TA manipulation (increase or decrease) could be applied per row in a given 30-day experiment. The subsequent 30-day experiment performed the opposite TA manipulation, ensuring all conditions were tested each season.

Seawater Sampling and Measurements

Daily Parameters

Temperature, salinity, dissolved oxygen, and pHNBS were measured daily at mid-day in all mesocosms and header tanks using a YSI multimeter (YSI ProDSS or YSI 556 MPS).

pH electrodes were calibrated daily using NIST-traceable pH 4, 7, and 10 buffers and corrected to the total scale using Tris buffer from A. Dickson (Scripps Institution of Oceanography). Dissolved oxygen calibrations followed manufacturer water-saturated air calibration protocols.

Total Alkalinity Sampling and Storage

Discrete seawater samples (100 mL) for total alkalinity (TA) were collected twice weekly.

Sampling procedures followed best practices described in Dickson et al. (2007):

Rinsed sample bottles three times with sample water.

Collected seawater in acid-cleaned 100 mL borosilicate bottles.

No headspace was left to prevent gas exchange.

Samples were analyzed within 12 hours of collection and stored at ambient temperature in the dark until analysis.

TA Determination

TA was determined using open-cell potentiometric titration on a Metrohm 877 Titrino Plus equipped with a Metrohm 9101 Herisau glass pH electrode.

All titrations were standardized using certified reference materials (CRM, batch number provided by Dickson Laboratory). Electrode slope, offset, and drift were checked daily.

Carbonate system variables (DIC, pCO2, HCO3-, CO32-, and Omega aragonite) were calculated using the R package seacarb.

Calcification Measurements

Calcification rates were quantified using buoyant weighing following Jokiel (1978). Colonies were weighed on day 0 and day 30. Calcification rate (G) was calculated as:

$G\ (g\ CaCO3\ d\text{-}1) = 1.54 \times (Wf - Wi) / d$

where 1.54 g cm-3 is the density of aragonite, Wf and Wi are final and initial buoyant weights, and d is the exposure duration in days.

Three colonies per species per mesocosm were randomly selected for further biometric analysis. Three branch tips per colony were sampled immediately before and after the exposure period. Collected fragments were frozen at -20 C until post-processing.

Mortality was low and did not differ across treatments; dead colonies were excluded from analysis. All corals were returned to the reef at the conclusion of experiments.

Post-Hoc Processing of Coral Samples

At the end of each experimental season, all frozen branch tips were transported to Texas A&M University–Corpus Christi for tissue, symbiont, pigment, and skeletal analysis.

Tissue Removal and Homogenization

Branch tips were thawed and sprayed with phosphate-buffered saline (PBS) using a Paasche airbrush.

Tissue slurry (25 mL) was sonicated for 20 seconds (Qsonica ultrasonic processor).

Samples were vortexed and centrifuged (VWR International centrifuge) to separate host tissue and symbionts.

Host Tissue Analyses

Aliquots of the host fraction were used to quantify protein concentration using spectrophotometry (SpectraMax M3). PBS was used as a blank.

Symbiont Analyses

Symbiont pellets were resuspended and counted using a Bright-Line hemocytometer (Hausser Scientific) under 10× magnification on a Leica ICC50W microscope.

Pigment Analyses

Chlorophyll a and c were extracted using 100 percent acetone. Absorbance was measured on a SpectraMax M3 spectrophotometer, with acetone blanks. Calculations followed Parsons et al. (1984).

Skeletal Preparation and Biometric Measurements

After tissue removal:

Skeletons were soaked in 10 percent bleach to remove remaining tissue.

Samples were dried for 4 hours at 60 C (DX302C drying oven).

Dry weights were recorded using a VWR-4002B2 analytical balance.

Skeletal volume was measured by water displacement in a 100 mL graduated cylinder.

Three-dimensional skeletal scans were generated using an Einscan-SE scanner and processed in MeshLab to estimate total surface area.

Data Processing Description

Overview

Data processing included organization of raw seawater chemistry measurements, conversion of pH values to the total scale, calculation of carbonate system parameters, buoyant-weight based calcification calculations, and post-hoc biochemical and skeletal data extraction. All steps described below represent the procedures used to generate the submitted dataset. No statistical analysis, modeling, or graphing is included here.

1. Seawater Chemistry Data Processing

pH Conversion

Daily pHNBS measurements from the YSI multiparameter meter were converted to total scale (pHT) using Tris buffer reference standards prepared and certified by the Dickson Laboratory. Temperature-dependent conversion equations followed Dickson et al. (2007).
All pH values were standardized across seasons using:

pHT = pHNBS + (Δbuffer offset)

where Δbuffer offset was determined daily from Tris-buffer calibration measurements.

Total Alkalinity (TA)

Raw potentiometric titration files from the Metrohm 877 Titrino Plus were exported using Metrohm tiamo software (version 2.5).
Processing included:

Drift correction and electrode slope verification.

CRM standardization using certified values for TA (Dickson Laboratory, batch number provided in metadata).

Extraction of equivalence point and TA value using open-cell titration algorithms embedded in tiamo.

TA values were reported in µmol kg-1.
If sample density was required, seawater density was calculated using temperature and salinity data following Fofonoff and Millard (1983).

Carbonate System Calculations

Carbonate chemistry parameters (DIC, pCO2, HCO3-, CO32-, and Omega aragonite) were calculated using the R package seacarb (version 3.3.1).
Inputs included:

pHT (corrected)

Total alkalinity

Temperature

Salinity

Default seacarb constants (k1k2 = "l", kf = "pf", ks = "d") were applied unless otherwise noted.

Outputs were merged into the final seawater chemistry dataset with unique sample IDs matching mesocosm, date, and header source.

2. Coral Calcification Data Processing

Buoyant Weight

Raw buoyant weight values (g) were recorded at day 0 and day 30 for each colony.
Calcification rate (G) was calculated following Jokiel (1978):

G (g $CaCO_3$ d-1) = 1.54 × (Wf – Wi) / d

where 1.54 g cm-3 is aragonite density, Wf is final buoyant weight, Wi is initial buoyant weight, and d is the exposure duration (30 days).
Results were merged into a calcification table by colony ID, species, treatment, mesocosm, and season.

3. Tissue and Symbiont Processing

Coral Tissue Extraction

Lab-generated tissue datasets included:

Host protein concentration

Symbiont cell density

Chlorophyll a and c concentrations

Processing steps:

Tissue slurry absorbance values were exported from the SpectraMax M3 software (SoftMax Pro, version 7.0).

Blanks (PBS for protein; acetone for chlorophyll) were subtracted from all readings.

Protein concentrations were calculated using standard curves prepared from bovine serum albumin (BSA).

Chlorophyll concentrations were calculated following Parsons et al. (1984) using:

Chl a = 11.85 A664 - 1.54 A647 - 0.08 A630
Chl c = 24.52 A630 - 1.67 A664

Symbiont cell counts obtained via hemocytometer were converted to cells cm-2 by dividing by coral fragment surface area (see 3D scan section below).

All tissue-based metrics were matched to corresponding colony IDs using unique fragment barcodes.

4. Skeletal Processing

Dry Weight and Volume

Dry weight (g) and volume (cm3) were combined to calculate skeletal density:

Density (g cm-3) = Dry mass / Volume

Volume was determined by water displacement using a 100 mL graduated cylinder.
Each cylinder was checked against manufacturer volume tolerances.

3D Scanning

Skeletal fragments were scanned using the Einscan-SE scanner.
Processing included:

Scan alignment and surface reconstruction in Einscan software (version 3.1).

Export of 3D mesh files (.stl format).

Mesh cleaning and area estimation in MeshLab (version 2022.02).

Surface area values (cm2) were merged into the biometric dataset using fragment ID numbers.

5. Data Organization and Quality Control

Data Entry and Validation

All raw files (TA titrator outputs, YSI logs, spectrophotometer outputs, and 3D scanner files) were manually inspected before integration.
Data validation steps included:

Removal of erroneous YSI readings (known sensor failure events, >3 SD outliers).

Titration rejection if CRM recovery was outside ±1 percent.

Elimination of samples with visible air bubbles during buoyant weighing.

Duplicate measurements averaged when replicates differed by <5 percent; otherwise re-measured.

Data Integration

All processed data were merged into the submission dataset using unique identifiers:

Season

Mesocosm number

Treatment

Species

Colony ID

Fragment ID (for biometrics)

Integrated datasets were exported as .csv files using R version 4.2.0.

6. Software Used

R version 4.2.0

seacarb version 3.3.1

Metrohm tiamo version 2.5

Molecular Devices SoftMax Pro version 7.0

Einscan-SE Scanner Software version 3.1

MeshLab version 2022.02

Microsoft Excel 365 (data organization and file formatting only)

BCO-DMO Processing Description

Primary Data File:
- Loaded "biomass.csv" as resource "995474_v1_buoyant-wgt" in CSV format with header row 1; treated empty strings, "nd", and "NA" as missing values
- Converted field "date" from format "%m/%d/%Y" to ISO 8601 date format "%Y-%m-%d" (output type: date)
- Set field types: "bwt" as number; "coral", "experiment", "mesocosm", "tp" as integer; "date" as date ("%Y-%m-%d"); "season", "species", "treatment" as string
- Applied BCO-DMO metadata to all fields: "bwt" (buoyant weight, grams), "coral" (coral ID number), "date" (sampling date, ISO 8601), "experiment" (experiment ID), "mesocosm" (tank ID), "season" (season of measurement; smr=summer, wntr=winter), "species" (coral species; mc=Montipora capitata, pa=Pocillopora acuta), "tp" (timepoint sampling day since experiment start), "treatment" (treatment ID; amb=control, co2=elevated pCO2, lta=lowered total alkalinity, hta=elevated total alkalinity)
- Updated package-level metadata with field-level statistics (count, min, max) for all 9 fields across 7,680 rows
- Output written to "995474_v1_buoyant-wgt.csv"

Tables added as supplemental files:
* sw_chem.csv
- In supplemental tables, converted field "date" from format "%m/%d/%Y" to ISO 8601 date format "%Y-%m-%d" (output type: date)
- Adjusted column names to meet BCO-DMO naming standards

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Parameters

Parameters for this dataset have not yet been identified

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Project Information

Collaborative Research: Reevaluating calcification response to changes in seawater chemistry by testing the Proton Flux Hypothesis and the Coral Metabolism Model (Proton Flux Hypothesis)

Coverage: Kaneohe, Hawaii

NSF Award Abstract:
Corals build calcium carbonate skeletons to maintain the three-dimensional structure of a coral reef, which provides habitat for many organisms and protects shorelines from bioerosion and storm damage. However, changes in ocean chemistry threaten the ability of corals to build and sustain these ecological important structures. To further the understanding of how climate change impacts coral reefs, this project investigates how changes in ocean carbonate chemistry directly influence coral calcification. The researchers are conducting a series of experiments on corals grown in seawater tanks to study corals responses to seawater chemistry in a changing ocean. Broader impacts of the project include student research opportunities, science-inquiry labs, and virtual learning. This project supports the training of several early career researchers, Ph.D. students, undergraduates, and high school students in the disciplines of chemistry, engineering, and marine ecology. Researchers partner with the Texas State Aquarium to communicate with the general public through a virtual research expedition series that will focus on coral reef health. This series includes interviews, behind the scene tours, and virtual dives on coral reefs in Hawaii.

This project examines the fundamental connections between seawater chemistry and coral physiology by investigating the modulation of seawater chemistry in the microenvironment surrounding corals. Specifically, this project 1) examines the response of corals to differing carbonate chemistry and 2) characterizes the proton gradient across the corals' boundary layers under differing ocean acidification conditions. Results of this work isolate whether carbonate ions or hydrogen ions have a stronger influence on calcification rates. This work utilizes a state-of-the-art experimental mesocosm facility that combines an automated systems to simultaneously and independently control both total alkalinity and carbon dioxide in the tanks to examine coral response under different carbon chemistry scenarios. Small-scale gradients in carbon chemistry surrounding the corals are being characterized using an innovative solid-state, reagentless sensor capable of making simultaneous measurements of two critical carbon system parameters. Coral biological response variables are quantified during short-term incubations and long-term mesocosm manipulations to understand physiological implications across multiple scales (i.e., individual and community scales) and across boundary layers.

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.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-2049406
NSF Division of Ocean Sciences (NSF OCE)	OCE-2049407

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