Physiology of Montipora capitata and Porites compressa from October 2019 to September 2022 in response to marine heatwaves (2015 and 2019) in Hawai‘i

Website: https://www.bco-dmo.org/dataset/904962
Data Type: Other Field Results
Version: 1
Version Date: 2023-07-21

Project
» RAPID: Collaborative Research: Disentangling the effects of heat stress versus bleaching phenotype on coral performance (Mcap pairs time series)

Contributors	Affiliation	Role
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Brown, Kristen	University of Pennsylvania (Penn)	Co-Principal Investigator
York, Amber D.	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
Increasingly frequent marine heatwaves are devastating coral reefs. Corals that survive these extreme events must rapidly recover if they are to withstand subsequent events, and long-term survival in the face of rising ocean temperatures may hinge on recovery capacity and acclimatory gains in heat tolerance over an individual's lifespan. To better understand coral recovery trajectories in the face of successive marine heatwaves, we monitored the responses of bleaching-susceptible and bleaching-resistant individuals of two dominant coral species in Hawai‘i, Montipora capitata and Porites compressa, over a decade that included three marine heatwaves. This dataset includes all of the physiological data for both species from October 2019 to September 2022.

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Coverage

Spatial Extent: **Lat**:21.4509 **Lon**:-157.7954
Temporal Extent: 2019-10-01 - 2022-09-30

Dataset Description

This dataset and other data from this study will be published in the results paper "Divergent bleaching and recovery trajectories in reef-building corals following a decade of successive marine heatwaves." (see pre-print Brown, et al. (2023), doi: 10.1101/2023.07.16.549193).

All BCO-DMO datasets related to this publication can be found on the page https://www.bco-dmo.org/related-resource/915300.

Methods & Sampling

Location:
patch reef 13 in Kāne'ohe Bay, O'ahu, Hawai'i (21.4509, -157.7954)

Physiological analyses:

A total of 276 fragments were collected from colonies of bleaching-resistant and bleaching-susceptible M. capitata and P. compressa (n= 7–10 pairs per phenotype per species) across eight timepoints from October 2019 to September 2022, incorporating the peak of the 2019 heatwave and three years (35 months) of recovery during non-heatwave years (Fig. S1, Brown et al. (2023)). Fragments (4–5 cm in length) were collected by hand using bone cutters, transported to the Hawai'i Institute of Marine Biology (HIMB) in ambient seawater and held in flow-through seawater aquaria for 24–72 hours until measurements of coral performance. For four timepoints (October 2019, October 2021, March 2022, and September 2022), metabolic rates were assessed via changes in oxygen evolution using oxygen optodes (PSt7, PreSens) connected to an optical analyser (OXY-10, PreSens) (Innis et al., 2021). Oxygen optodes were calibrated on each day with a 0% oxygen solution (0.01 g ml−1 NaSO3) and 100% air saturated seawater. Coral fragments were analyzed between 08:00 and 17:00 within 250 ml clear acrylic chambers on top of a magnetic stirrer to allow for continuous mixing. Seawater temperatures were replicated to those experienced on the reef by using ambient seawater and a water jacket to maintain temperatures within the incubation chambers. Temperature and dissolved oxygen concentrations were recorded every 3 seconds at increasing increments of light over a total of 45–50 minutes to determine maximum net photosynthesis (Pmax). Light levels ranged from 112–726 µmol m-2 sec-1 in October 2019 (Innis et al., 2021) (step size ~100 µmol m-2 sec-1; ~10 minutes step-1), and from 387–1800 µmol m-2 sec-1 in 2020–2021 (step size ~300 µmol m-2 sec-1; ~10 minutes step-1). After measurements were completed at the maximum light levels, the lights were turned off (0 µmol m-2 sec-1) to measure light-enhanced dark respiration (LEDR) as defined by (Edmunds and Davies, 1988). Photosynthesis-irradiance curves were fitted using the Platt model to extract Pmax and LEDR (Innis et al., 2021; Platt et al., 1981), 3). Upon completion of these in vivo analyses, coral fragments were flash frozen in liquid nitrogen and stored at −80°C until further processing.

Coral tissues containing symbionts were removed from skeletons with a waterpik using 50 mL of 0.1 M phosphate buffered saline (PBS) solution. The resulting holobiont homogenate was centrifuged at 4°C for 5 minutes at 2500 x g to separate the coral host fraction (supernatant) from the intracellular endosymbiont cells (family Symbiodiniaceae; pellet). Symbiodiniaceae were resuspended in PBS and cell abundances were quantified using a Millipore Guava flow-cytometer (Guava easyCyte 5HT) following the methodology of (Innis et al., 2021). Symbiodiniaceae cells were excited with a blue laser (488 nm) and identified by analyzing forward scatter and red autofluorescence in GuavaSoft 3.4 with the same gating for all samples. Symbiont densities were standardized to skeletal surface area (cm2), which was determined using the wax-dipping technique (Holmes, 2008; Stimson and Kinzie, 1991). Endosymbiont photopigments were extracted in 100% acetone for 24 hours and the concentration of chlorophyll-a was determined via absorbance at 630, 663, and 750 nm using the equation in (Jeffrey and Humphrey, 1975) and subtracting the absorbance at 750 nm from both A663 and A630:

Chlorophyll a =(11.43 (A663-A750)) -(0.64 (A630-A750))

Photopigment concentrations were standardized to both skeletal surface area and symbiont densities. Endosymbiont community composition (proportion Durusdinium vs. Cladocopium) of M. capitata colonies was determined by Dilworth et al. 2021 from samples collected in July 2019 using quantitative PCR.

Host tissue biomass was determined as ash-free dry weight (AFDW) from the coral fraction (i.e. symbionts removed as described above). First, 1 mL of the coral fraction was dried at 60°C for 24 hours until a constant weight was achieved. After the dry weight was recorded, the samples were then burned in a muffle furnace at 450°C for 6 hours. The samples were allowed to cool in the furnace before being weighed and the weight of the resulting ash was recorded. The difference between the ash weight and the dry weight was calculated to determine the AFDW of each sample. Host soluble protein content of each sample was determined via the Bradford method. Specifically, 10 µL of the coral fraction was pipetted into each of three wells of a 96-well plate (technical replicates), followed by the addition of 300 µL of Coomassie Plus Bradford reagent (Thermo Fisher Scientific) to each well. The plate was incubated for 10 minutes at room temperature followed by the collection of an absorbance reading at 595 nm on a spectrophotometer (BioTek PowerWave XS2). Each plate contained a set of internal standards with known concentrations of bovine serum albumin (0–2000 µg mL-1), which were used to generate a second-degree polynomial standard curve relating absorbance (x) to protein concentration

(y). The standard curve equation was used to calculate the protein concentrations of the samples. The concentration of lipids in each sample was determined via a modified method of (Dunn et al., 2012). First, ~45 mL of the coral fraction was lyophilized to produce dry tissue. A subsample of the lyophilized tissue was weighed (~90 mg), and 2 mL of chloroform-methanol (2:1) was added. The sample was then mixed using a homogenizer at 15000 rpm for 5 seconds, and an additional 2 mL of chloroform-methanol was used to rinse the homogenizer into the sample, and the tubes were then vigorously vortexed before being placed at 4°C for 2 hours in the dark. After incubation, tubes were again vortexed and the chilled homogenates were then passed through 0.22 µm syringe filters into new pre-weighed tubes. An additional 1 mL of chloroform-methanol was passed through the filters into each tube to ensure the full passage of sample material. Next, 1 mL of 0.1 M KCl was added to the samples, which were then vortexed and placed at 4°C for at least 1 hour in the dark until two phases formed. The aqueous (top) phase was discarded, and 5 mL of 50% methanol was added to the organic (bottom) phase. The samples were then placed at 4°C for 1 hour in the dark, followed by removal of the aqueous (top) phase. Washes with 50% methanol were repeated twice more, and the remaining organic phases were dried under a fume hood until the solvent had completely evaporated, at which point the lipid pellet was weighed. Coral host biomass, protein and lipid content were standardized to skeletal surface area.

The total antioxidant capacity of the coral homogenate was assessed using a kit from Cell Biolabs (STA-360) according to the manufacturer's instructions. First, 20 µL of the coral fraction was transferred to each of two wells of a 96-well plate (technical replicates), followed by the addition of 1x reaction buffer (20 µL) from the kit. An initial absorbance reading was collected with a spectrophotometer at 490 nm. Next, 50 µL of 1x copper ion reagent were added to each well, followed by incubation for 5 minutes with gentle shaking. Following shaking, 50 µL of 1x stop solution was added to each well, and a second absorbance reading was collected at 490 nm. Each plate contained a set of standards with known concentrations (0–0.1 mM) of uric acid. For analysis, the initial absorbance readings were subtracted from the final absorbance readings for each standard well, and the resulting values were used to generate a linear standard curve relating the absorbance (x) to the concentration of uric acid (y). The standard curve equation was used with the average change in absorbance across the two technical replicates for each sample to first determine the concentration of uric acid equivalents in the samples, which was then converted to a concentration of copper reducing equivalents (CRE) using the equivalence of 1 mM of uric acid to 2189 µM CREs. Finally, the CRE values were normalized to the amount of protein in each well, for a final value of TAC expressed as µM CRE mg protein-1.

To assess the melanin content of the coral homogenate, a protocol was adapted from (Wall et al., 2021). First, 1.5 mL tubes were filled with approximately 20 mg of the dry coral fraction and the weight recorded. Next, 300 µL of 10 M NaOH was added to each tube, followed by vortexing for 20 seconds. Samples were left overnight at room temperature in the dark, then vortexed again for 10 seconds before being centrifuged at 7000 x g for 5 minutes. Following centrifugation, 100 µL of supernatant was transferred to each of two wells of a 96-well plate (technical replicates), which was then read for absorbance on a spectrophotometer at 490 nm. Each plate contained a set of internal standards with known concentrations (0–0.01 mg mL-1) of synthetic melanin (M8631, Sigma Aldrich) in 10 M NaOH, which were used to generate a linear standard curve relating absorbance (x) to melanin concentration (y). The standard curve equation was then used to calculate the concentration of melanin in each sample, which was standardized to the original weight of the dry coral host tissue used in the assay.

A protocol was adapted from (Wall et al., 2021; Mydlarz and Palmer, 2011; Fuess et al., 2018) to assess the content of prophenoloxidase (PPO) in each coral sample. First, 150 µL of the coral fraction was transferred to each of two wells of a 96-well plate (technical replicates). Next, 23 µL of 0.2 mg mL-1 trypsin was added to each well, and the plate was incubated for 5 minutes at room temperature with shaking. Following incubation, 60 µL of 10 mM L-1,3- dihydroxyphenylalanine (L-DOPA) was added to all wells, and the plate was again incubated at room temperature for 5 minutes with shaking. Next, the plate was read for absorbance on a spectrophotometer at 490 nm every minute for 15 minutes. The absorbance of each well at the start of the 15 minutes was subtracted from the absorbance of the same well at the end, and values were divided by 15 to determine the change in absorbance per minute, which were then averaged over the two technical replicates. Finally, these values were standardized to the amount of protein in each well for a final quantification of PPO activity expressed as the change in absorbance minute-1 mg protein-1

Wax-dipping was used to determine calcium carbonate ($CaCO_3$) bulk density, where the skeleton was cleaned, dried to a constant mass and weighed, sealed with a coat of wax, dry weighed with the wax and then buoyant weighed in DI water at 20°C (Tambutté, et al. 2015; Brown and Mello-Athayde, 2022). The difference between dry weight (with wax) and buoyant weight (divided by the density of the DI water medium of 1 mg cm-3) was calculated to determine the total volume enclosed. The dry skeletal mass (wax free) was then divided by the total volume enclosed to yield bulk density (g cm-3).

Taxonomic Identifiers:
Montipora capitata, LSID (urn:lsid:marinespecies.org:taxname:287697)
Porites compressa, LSID (urn:lsid:marinespecies.org:taxname:207236)

Data Processing Description

See results publication Brown, et al. (2023) for more detailed information on analysis and results.

BCO-DMO Processing Description

* table within file "Phys_data_for_BCO-DMO.csv" was imported into the BCO-DMO data system.
** 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]

* Date converted to ISO 8601 format

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Data Files

File	
904962_v1_coral_physiology.csv	(Comma Separated Values (.csv), 60.03 KB) MD5:2aac8a20f355ef733687f9cecb3b635a
Primary data file for BCO-DMO dataset 904962 version 1.	

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Related Publications

Arbizu, Pedro Martinez. 2020. "pairwiseAdonis: Pairwise Multilevel Comparison Using Adonis." 2020. https://github.com/pmartinezarbizu/pairwiseAdonis.
Software

Brown, K. T., Lenz, E. A., Glass, B. H., Kruse, E., McClintock, R., Drury, C., Nelson, C. E., Putnam, H. M., & Barott, K. L. (2023). Divergent recovery trajectories in reef-building corals following a decade of successive marine heatwaves. bioRxiv preprint. https://doi.org/10.1101/2023.07.16.549193
Results

Brown, K. T., Mello-Athayde, M. A., Sampayo, E. M., Chai, A., Dove, S., & Barott, K. L. (2022). Environmental memory gained from exposure to extreme pCO2 variability promotes coral cellular acid–base homeostasis. Proceedings of the Royal Society B: Biological Sciences, 289(1982). https://doi.org/10.1098/rspb.2022.0941
Methods

Cunning, R., Parker, K. E., Johnson-Sapp, K., Karp, R. F., Wen, A. D., Williamson, O. M., Bartels, E., D'Alessandro, M., Gilliam, D. S., Hanson, G., Levy, J., Lirman, D., Maxwell, K., Million, W. C., Moulding, A. L., Moura, A., Muller, E. M., Nedimyer, K., Reckenbeil, B., … Baker, A. C. (2021). Census of heat tolerance among Florida's threatened staghorn corals finds resilient individuals throughout existing nursery populations. Proceedings of the Royal Society B: Biological Sciences, 288(1961). https://doi.org/10.1098/rspb.2021.1613
Methods

Dunn, S. R., Thomas, M. C., Nette, G. W., & Dove, S. G. (2012). A Lipidomic Approach to Understanding Free Fatty Acid Lipogenesis Derived from Dissolved Inorganic Carbon within Cnidarian-Dinoflagellate Symbiosis. PLoS ONE, 7(10), e46801. https://doi.org/10.1371/journal.pone.0046801

Methods

Evensen, N. R., Voolstra, C. R., Fine, M., Perna, G., Buitrago-López, C., Cárdenas, A., Banc-Prandi, G., Rowe, K., & Barshis, D. J. (2022). Empirically derived thermal thresholds of four coral species along the Red Sea using a portable and standardized experimental approach. Coral Reefs, 41(2), 239–252. https://doi.org/10.1007/s00338-022-02233-y
Methods

Fuess, L. E., Mann, W. T., Jinks, L. R., Brinkhuis, V., & Mydlarz, L. D. (2018). Transcriptional analyses provide new insight into the late-stage immune response of a diseased Caribbean coral. Royal Society Open Science, 5(5), 172062. https://doi.org/10.1098/rsos.172062
Methods

Holmes, G. (2008). Estimating three-dimensional surface areas on coral reefs. Journal of Experimental Marine Biology and Ecology, 365(1), 67–73. https://doi.org/10.1016/j.jembe.2008.07.045
Methods

Innis, T., Allen-Waller, L., Brown, K. T., Sparagon, W., Carlson, C., Kruse, E., Huffmyer, A. S., Nelson, C. E., Putnam, H. M., & Barott, K. L. (2021). Marine heatwaves depress metabolic activity and impair cellular acid–base homeostasis in reef-building corals regardless of bleaching susceptibility. Global Change Biology, 27(12), 2728–2743. Portico. https://doi.org/10.1111/gcb.15622
Methods

Jeffrey, S. W., & Humphrey, G. F. (1975). New spectrophotometric equations for determining chlorophylls a, b, c1 and c2 in higher plants, algae and natural phytoplankton. Biochemie Und Physiologie Der Pflanzen, 167(2), 191–194. doi:10.1016/s0015-3796(17)30778-3 https://doi.org/10.1016/S0015-3796(17)30778-3
Methods

Marzonie, M. R., Bay, L. K., Bourne, D. G., Hoey, A. S., Matthews, S., Nielsen, J. J. V., & Harrison, H. B. (2022). The effects of marine heatwaves on acute heat tolerance in corals. Global Change Biology, 29(2), 404–416. Portico. https://doi.org/10.1111/gcb.16473
Methods

Mydlarz, L. D., & Palmer, C. V. (2011). The presence of multiple phenoloxidases in Caribbean reef-building corals. Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology, 159(4), 372–378. https://doi.org/10.1016/j.cbpa.2011.03.029
Methods

Oksanen, Jari, F. Guillaume Blanchet, Roeland Kindt, Pierre Legendre, Peter R. Minchin, R. B. O'hara, Gavin L. Simpson, Peter Solymos, M. Henry H. Stevens, and Helene Wagner. 2013. "Package 'vegan.'" Community Ecology Package, Version 2 (9): 1-295. Available from https://cran.r-project.org/package=vegan
Software

Platt, T., C. L. Gallegos, and W. G. Harrison. 1980. “Photoinhibition of Photosynthesis in Natural Assemblages of Marine Phytoplankton.” Journal of Marine Research, 16.
https://elischolar.library.yale.edu/journal_of_marine_research/1525/
Methods

R Core Team (2021). "R: A Language and Environment for Statistical Computing." 2021. https://www.R-project.org/
Software

Ritz, C., Baty, F., Streibig, J. C., & Gerhard, D. (2015). Dose-Response Analysis Using R. PLOS ONE, 10(12), e0146021. https://doi.org/10.1371/journal.pone.0146021
Methods

Tambutté, E., Venn, A. A., Holcomb, M., Segonds, N., Techer, N., Zoccola, D., Allemand, D., & Tambutté, S. (2015). Morphological plasticity of the coral skeleton under CO2-driven seawater acidification. Nature Communications, 6(1). https://doi.org/10.1038/ncomms8368
Methods

Voolstra, C. R., Buitrago-López, C., Perna, G., Cárdenas, A., Hume, B. C. C., Rädecker, N., & Barshis, D. J. (2020). Standardized short-term acute heat stress assays resolve historical differences in coral thermotolerance across microhabitat reef sites. Global Change Biology, 26(8), 4328-4343. Portico. https://doi.org/10.1111/gcb.15148
Methods

Wall, C. B., Ricci, C. A., Wen, A. D., Ledbetter, B. E., Klinger, D. E., Mydlarz, L. D., Gates, R. D., & Putnam, H. M. (2021). Shifting baselines: Physiological legacies contribute to the response of reef corals to frequent

heatwaves. Functional Ecology, 35(6), 1366–1378. Portico. https://doi.org/10.1111/1365-2435.13795
Methods

Wickham, H. (2016). Data Analysis. Ggplot2, 189–201. https://doi.org/10.1007/978-3-319-24277-4_9
Software

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Parameters

Parameter	Description	Units
Coral_ID	Coral_ID (genotype). numeric identifier	unitless
Species	Genus and species	unitless
Bleach	Bleaching phenotype ("Bleach" or "Non-bleach")	unitless
Date	Date	unitless
Period	Period (heatwave or recovery)	unitless
Months	Months (post-heat stress). Numeric	unitless
Protein	Protein	micrograms per centimeter squared (mg cm-2)
Sym_density	Endosymbiont density	10^-6 cells per centimeter squared (10-6 cells cm-2)
Chl	Chlorophyll a	micrograms per centimeter squared (ug cm-2)
CaCO3	CaCO3 density	grams ler milliliter (g mL-1)
internal_volume_percent	Internal volume percent	percent (%)
TAC_CRE	Total antioxidant capacity; copper reducing equivalents (CRE)	uM copper reducing equivalents (CRE) mg protein-1
ug_Chl_a_per_zoox	Chlorophyll a per zoox (µg cell-1)	micrograms per cell (ug cell-1)
pg_Chl_a_per_zoox	Chlorophyll a per zoox (pg cell-1)	picograms per cell (ug cell-1)
Citrate_synthase	Citrate synthase mU mg protein-1	citrate synthase mg protein-1

AFDW	Ash-free dry weight	milligrams per centimeter squared (mg cm-2)
Endolithic	Endolithic(presence or absense of endolithic algae)	unitless
PPO	PPO(prophenoloxidase)	change in absorbance minute-1 mg protein-1
Melanin_per_tissue	Melanin per tissue (see methodology for details of the melanin concentration calculation in each sample standardized to the original weight of the dry coral host tissue)	unitless
Lipid	Lipids	grams per cenitmeter squared (g cm-2)
Pmax	Max photosynthesis (Pmax)	micromoles of oxygen per centimeter squared per hour (umol O2 cm2 hr-1)
R	light-enhanced dark respiration (LEDR)	micromoles of oxygen per centimeter squared per hour (umol O2 cm2 hr-1)
PR	PR (photosynthesis to respiration ratio)	unitless

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Instruments

Dataset-specific Instrument Name	Guava easyCyte 5HT
Generic Instrument Name	Flow Cytometer
Dataset-specific Description	endosymbiont densities: Millipore Guava flow-cytometer (Guava easyCyte 5HT)
Generic Instrument Description	Flow cytometers (FC or FCM) are automated instruments that quantitate properties of single cells, one cell at a time. They can measure cell size, cell granularity, the amounts of cell components such as total DNA, newly synthesized DNA, gene expression as the amount messenger RNA for a particular gene, amounts of specific surface receptors, amounts of intracellular proteins, or transient signalling events in living cells. (from: http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm)

Dataset-specific Instrument Name	Diving-PAM (Walz GmbH)
Generic Instrument Name	Fluorometer
Dataset-specific Description	photochemical yield (Fv/Fm): Diving-PAM (Walz GmbH)
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	OXY-10 (PreSens) optical analyser
Generic Instrument Name	PreSens OXY-10 Mini oxygen meter
Dataset-specific Description	net photosynthesis and light-enhanced dark respiration: oxygen optodes connected to an OXY-10 (PreSens) optical analyser
Generic Instrument Description	The OXY-10 mini is a precise multi-channel oxygen meter for up to 10 'in-house' sensors, simultaneously controlling and reading them. The meter is used with oxygen sensors based on a 2mm optical fibre. The meter is compatible with sensors that are type PSt3 which has a detection limit 15 ppb, 0 - 100% oxygen.

Dataset-specific Instrument Name	BioTek PowerWave XS2
Generic Instrument Name	Spectrophotometer
Dataset-specific Description	host-soluble protein concentration, chlorophyll a concentrations, total antioxidant capacity, melanin content, prophenoloxidase content, citrate synthase content: spectrophotometer (BioTek PowerWave XS2)
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.

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Project Information

RAPID: Collaborative Research: Disentangling the effects of heat stress versus bleaching phenotype on coral performance (Mcap pairs time series)

Coverage: Coral reefs of Kaneohe Bay, Oahu, Hawaii

NSF Award Abstract:

NSF Award Abstract:
Coral bleaching has become increasingly common on reefs worldwide as rising sea surface temperatures associated with climate change disrupt the coral-algal symbiosis. This dramatic heat stress response turns the normally colorful corals bright white, and yet during these heat stress events not all corals undergo bleaching. This project focuses on assessing the effects of bleaching by comparing pairs of corals side-byside on the reef during an ongoing heat wave, where one has bleached and the other has not, despite experiencing the same temperatures. These coral pairs have been monitored throughout three bleaching events in the past five years, providing a unique resource to address whether corals with consistently different bleaching susceptibilities have the capacity to acclimate in response to disturbances through epigenetic changes, or changes in gene expression not due to change in DNA bases. To address this, the project will characterize the impacts of bleaching or not bleaching on coral physiology, gene expression, and epigenetic patterns using coral pairs in their natural habitat during a marine heatwave. This project also provides research support for graduate student trainees, as well as data and materials for the research and training of undergraduate and high school students. This project will recruit underrepresented minority students from URI and UPenn area high schools and university undergraduates for work on computer analysis of images (benthic and colony photographs, brightfield and confocal micrographs) and sequencing data. It will also support the training of an undergraduate student at the University of Hawai'i in coral ecology and physiology, and the development of her senior thesis.

This project will investigate the effects of repeated heat stress events on the performance of Montipora capitata, a dominant reef builder throughout Hawai'i. It utilizes the timely context of paired colonies of M. capitata with bleached vs. unbleached histories that have been monitored through two past bleaching events in Hawai'i (2015 and 2019) and the currently ongoing 2020 event. This system allows for the unique opportunity to disentangle the consequences of heat stress versus bleaching on coral performance through time, an essential feature of reef resilience. The contrasting physiological and energetic processes these two phenotypes undergo during a heatwave are likely to result in alterations to the cellular environment within the animal that impacts epigenetic transcriptional regulation. These regulatory and energetic changes, if persistent over time, have the potential to alter coral fitness beyond the duration of the heatwave differentially between corals with contrasting bleaching phenotypes. Specifically, the project will: 1) quantify the effect of the 2020 heatwave on coral physiology during bleaching and recovery, 2) generate a corresponding archive of coral tissues and nucleic acids as a resource for future work characterizing how bleaching phenotype alters energetics and non-genetic inheritance, and 3) characterize how bleaching phenotype alters intra-generational inheritance of epigenetic marks (i.e., DNA methylation) and gene expression, and the duration of these marks and expression patterns following heat stress. This project represents an urgent assessment of an ideal system to test the legacy of coral bleaching phenotype on coral fitness. The results of this project will therefore lay the foundation for intra and cross-generational effects of bleaching vs. heat stress, which is essential for understanding coral resilience to climate change.

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-2102989
NSF Division of Ocean Sciences (NSF OCE)	OCE-2103067

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