# Physiological parameters of eight corals species surveyed from six locations around O'ahu, HI from August to November of 2015

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# Project

» Phenotype and genotype of coral adaptation and acclimatization to global change (Coral Adaptation)

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### Abstract

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# Coverage

Spatial Extent: N:21.5925 E:-157.675 S:21.2875 W:-158.131 Temporal Extent: 2015-08-17 - 2015-11-13

#### Methods & Sampling

#### Coral collection

Corals were collected in 2015 from each of the six study sites (See Supplemental File "coral\_collection\_location\_list.csv). Unsafe weather conditions lead to several delays during sampling which caused coral collection to span an 11-week period (between 17 August and 13 November). Eight species of coral, from three genera were sampled: Montipora capitata (branching and encrusting), Montipora flabellata (encrusting), Montipora patula (encrusting), Porites compressa (branching), Porites lobata (massive), Porites evermanni (massive), Pocillopora meandrina (branching), and Pocillopora acuta (branching) (McLachlan et al. submitted, Fig. 1). The Y and B morphs of P. damicornis that were historically common in Kāne'ohe Bay (Richmond and Jokiel 1984), appear to correspond to the cryptic species P. damicornis and P. acuta (Johnston et al. 2018). Recent studies have found that P. acuta has become the dominant species and that P. damicornis is now rare in Kāne'ohe Bay (Gorospe et al. 2015; Johnston et al. 2018), so we focus on P. acuta in this study. Due to the natural zonation in coral species distributions, not all eight species were found in all six locations. To minimize the impact of collection, only the most commonly available species were collected from 0.5-5 m depth yielding a total of 422 samples (McLachlan et al. submitted, Table 1). Genets were confirmed by genotyping about half of the colonies (232 of the 422 colonies sampled) using available species-specific microsatellite markers (Concepcion et al. 2010; Gorospe and Karl 2013), and no identical multilocus genotypes were found within a site, suggesting very low probability that any were clonally derived. Each coral ramet was 5-10 cm in size and was removed from larger parent genets using a hammer and chisel. None of the coral ramets included in this study were visibly pale or severely bleached. Samples were immediately frozen at -20 °C, stored at HIMB, and later shipped to The Ohio State University, Ohio, USA, where they were stored at -80 °C.

# Physiological data

Encrusting algae and boring organisms were removed from the surface of each coral ramet using a Dremel tool fitted with a diamond tipped bit (Dremel Inc., Racine, WI). Each coral ramet was split into two pieces: one for biochemical and one for isotopic analysis.

The coral pieces designated for biochemical physiological analyses were photographed from all sides and the photographs processed in the software ImageJ (Rasband 1997) to estimate surface area using the geometric method (Naumann et al. 2009). Each coral piece was then individually ground into a fine homogenous paste using a chilled mortar and pestle, partitioned into subsamples designated for each biochemical analysis, and stored at -80 °C. Analyses of total chlorophyll *a* and *c2* concentration, total soluble protein, total tissue biomass, and total soluble lipid concentration (henceforth referred to as chlorophyll, protein, biomass, and lipid, respectively) were conducted based on methods modified from Jeffrey and Humphrey (1975), Bradford (1976), Grottoli et al. (2004), and Rodrigues and Grottoli (2007). Briefly, chlorophyll was extracted from ground coral samples using a double extraction in 100 % acetone, and the absorbance at 630, 663, and 750 nm wavelengths was measured using a spectrophotometer.

Detailed protocols for each of the other analyses are deposited in *Protocols.io. See:* McLachlan et al. 2020; doi:10.17504/protocols.io.bdyai7se McLachlan et al. 2020; doi:10.17504/protocols.io.bc4qiyvw McLachlan et al. 2020; doi:10.17504/protocols.io.bdc8i2zw

The coral pieces designated for stable isotope analyses were prepared using methods modified from Hughes et al. (2010) and a detailed protocol is deposited in *Protocols.io* (Price et al. 2020). All isotope samples were combusted using a PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK) at the University of California Davis Stable Isotope Facility. The carbon

isotopic signature of the animal host ( $\delta$ 13Ch), and algal endosymbiont ( $\delta$ 13Ce), were reported as the per mil deviation of the stable isotopes 13C:12C relative to Vienna Peedee Belemnite Limestone Standard. The nitrogen isotopic signature of the animal host ( $\delta$ 15Nh), and algal endosymbiont ( $\delta$ 15Ne), were reported as the per mil deviation of the stable isotopes 14N:15N relative to air. At least 10 % of all measurements were made in duplicate, and the standard deviation of duplicate sample analyses were ±0.09 ‰ for  $\delta$ 13Ch, ±0.21 ‰ for  $\delta$ 13Ce, ±0.06 ‰ for  $\delta$ 15Nh, and ±0.12 ‰ for  $\delta$ 15Ne. Differences between  $\delta$ 13Ch and  $\delta$ 13Ce of each ramet were used to assess the relative contribution of photosynthetically and heterotrophically derived C in coral tissues [*sensu* Muscatine et al. (1989) and Rodrigues and Grottoli (2006)]. The lower the  $\delta$ 13Ch -  $\delta$ 13Ce value, the greater the relative contribution of heterotrophically versus photoautotrophically derived carbon to coral tissues, and vice versa for larger  $\delta$ 13Ch -  $\delta$ 13Ce values (Muscatine et al. 1989; Rodrigues and Grottoli 2006; Levas et al. 2013; Schoepf et al. 2015; Grottoli et al. 2017). Conversely, the lower the  $\delta$ 15Nh-e –  $\delta$ 15Nh-e value, the lower the relative contribution of heterotrophically derived nitrogen to coral tissues (Conti-Jerpe et al. 2020).

#### Species list:

ScientificName, AphialD Montipora capitata, 287697 Montipora flabellata, 207174 Montipora patula, 207149 Pocillopora acuta, 759099 Pocillopora meandrina, 206964 Porites compressa, 207236 Porites evermanni, 288900 Porites lobata, 207225

#### **Data Processing Description**

BCO-DMO data manager processing notes:

- \* Data submitted in file data.csv
- \* data values that are a period "." imported as missing data values as indicated in the dataset metadata.
- \* Coral collection site information supplied by the submitter was turned into a csv file and added as a supplemental document to this dataset.
- \* Latitude, longitude, and collection depth min and max added to the dataset from the collection site information.
- \* Date converted to ISO 8601 format yyyy-mm-dd

\* Species names checked using the World Register of Marine Species taxa match tool. All names were exact matches to accepted names (as of 2021-02-18). List of species names and matched AphialD added to the metadata.

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# **Data Files**

File	
coral_physiology.csv(Comma Separated Values (.csv), 53.11 KB) MD5-6ae7d55f74488885495b8a8fdc47ae7f	
Primary data file for dataset ID 841242	

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#### Supplemental Files

File
Coral collection locations
$filename: coral\_collection\_location\_list.csv(Comma Separated Values (.csv), 531 bytes) \\ MD5:8cb7d848966bb4bd881840beec1682ea$
Coral collection site information including columns:
Site, Collection site short name, unitless
Site_full_name,Colleciton site full name including special characters
Island,Island name
State, State name
Country, Country name
lat,latitude,decimal degrees
lon,longitude,decimal degrees
collection_min_depth,Collection range minimum depth,meters
collection_max_depth,Collection range maximum depth,meters

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#### **Related Publications**

Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry, 72(1-2), 248–254. doi:<u>10.1016/0003-2697(76)90527-3</u> *Methods* 

Concepcion, G. T., Polato, N. R., Baums, I. B., & Toonen, R. J. (2010). Development of microsatellite markers from four Hawaiian corals: Acropora cytherea, Fungia scutaria, Montipora capitata and Porites lobata. Conservation Genetics Resources, 2(1), 11–15. doi:<u>10.1007/s12686-009-9118-4</u> Methods Conti-Jerpe, I. E., Thompson, P. D., Wong, C. W. M., Oliveira, N. L., Duprey, N. N., Moynihan, M. A., & Baker, D. M. (2020). Trophic strategy and bleaching resistance in reef-building corals. Science Advances, 6(15), eaaz5443. doi:<u>10.1126/sciadv.aaz5443</u> Methods

Gorospe, K. D., & Karl, S. A. (2013). Genetic relatedness does not retain spatial pattern across multiple spatial scales: dispersal and colonization in the coral, Pocillopora damicornis. Molecular Ecology, 22(14), 3721–3736. doi:<u>10.1111/mec.12335</u> Methods

Gorospe, K. D., Donahue, M. J., & Karl, S. A. (2015). The importance of sampling design: spatial patterns and clonality in estimating the genetic diversity of coral reefs. Marine Biology, 162(5), 917–928. doi:10.1007/s00227-015-2634-8 Methods

Grottoli, A. G., Rodrigues, L. J., & Juarez, C. (2004). Lipids and stable carbon isotopes in two species of Hawaiian corals, Porites compressa and Montipora verrucosa, following a bleaching event. Marine Biology, 145(3). doi:<u>10.1007/s00227-004-1337-3</u> Methods

Grottoli, A. G., Tchernov, D., & Winters, G. (2017). Physiological and Biogeochemical Responses of Super-Corals to Thermal Stress from the Northern Gulf of Aqaba, Red Sea. Frontiers in Marine Science, 4. doi:<u>10.3389/fmars.2017.00215</u> Methods

Hughes, A., Grottoli, A., Pease, T., & Matsui, Y. (2010). Acquisition and assimilation of carbon in non-bleached and bleached corals. Marine Ecology Progress Series, 420, 91–101. doi:<u>10.3354/meps08866</u> *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 <a href="https://doi.org/10.1016/S0015-3796">https://doi.org/10.1016/S0015-3796</a> (17)30778-3 <a href="https://doi.org/10.1016/S0015-3796">https://doi.org/10.1016/S0015-3796</a> (17)30778-3

Methods

Johnston, E. C., Forsman, Z. H., & Toonen, R. J. (2018). A simple molecular technique for distinguishing species reveals frequent misidentification of Hawaiian corals in the genus Pocillopora. PeerJ, 6, e4355. doi:<u>10.7717/peerj.4355</u> *Methods* 

Levas, S. J., Grottoli, A. G., Hughes, A., Osburn, C. L., & Matsui, Y. (2013). Physiological and Biogeochemical Traits of Bleaching and Recovery in the Mounding Species of Coral Porites lobata: Implications for Resilience in Mounding Corals. PLoS ONE, 8(5), e63267. doi:<u>10.1371/journal.pone.0063267</u> *Methods* 

McLachlan, R. H., Price, J. T., Muñoz-Garcia, A., Weisleder, N. L., Jury, C. P., Toonen, R. J., & Grottoli, A. G. (2021). Environmental gradients drive physiological variation in Hawaiian corals. Coral Reefs, 40(5), 1505–1523. https://doi.org/<u>10.1007/s00338-021-02140-8</u> *Results* 

Mclachlan, R., Dobson, K., & Grottoli, A. (2020). Quantification of Total Biomass in Ground Coral Samples v1 (protocols.io.bdyai7se). Protocols.io. doi:<u>10.17504/protocols.io.bdyai7se</u> Methods

Mclachlan, R., Munoz, A., & Grottoli, A. (2020). Extraction of Total Soluble Lipid from Ground Coral Samples v1 (protocols.io.bc4qiyvw). Protocols.io. doi:10.17504/protocols.io.bc4qiyvw Methods

Mclachlan, R., Price, J., Dobson, K., Weisleder, N., & Grottoli, A. (2020). Microplate Assay for Quantification of Soluble Protein in Ground Coral Samples v1 (protocols.io.bdc8i2zw). Protocols.io. doi:<u>10.17504/protocols.io.bdc8i2zw</u> Methods

Muscatine, L., Porter, J. W., & Kaplan, I. R. (1989). Resource partitioning by reef corals as determined from stable isotope composition. Marine Biology, 100(2), 185–193. doi:10.1007/bf00391957 <a href="https://doi.org/10.1007/BF00391957">https://doi.org/10.1007/BF00391957</a> Methods

Naumann, M. S., Niggl, W., Laforsch, C., Glaser, C., & Wild, C. (2009). Coral surface area quantification–evaluation of established techniques by comparison with computer tomography. Coral Reefs, 28(1), 109–117. doi:<u>10.1007/s00338-008-0459-3</u> Methods

Price, J. T., McLachlan, R. H., Jury, C. P., Toonen, R. J., & Grottoli, A. G. (2021). Isotopic approaches to estimating the contribution of heterotrophic sources to Hawaiian corals. Limnology and Oceanography, 66(6), 2393–2407. Portico. https://doi.org/<u>10.1002/ino.11760</u> Results

Price, J., Smith, A., Dobson, K., & Grottoli, A. (2020). Airbrushed Coral Sample Preparation for Organic Stable Carbon and Nitrogen Isotope Analyses v1 (protocols.io.bgi7juhn). Protocols.io. doi:<u>10.17504/protocols.io.bgi7juhn</u> *Methods* 

Richmond RH, Jokiel PL (1984) Lunar periodicity in larva release in the reef coral Pocillopora damicornis at Enewetak and Hawaii. Bull Mar Sci 34:280-287. <u>https://www.researchgate.net/publication/233692712\_Lunar\_Periodicity\_in\_Larva\_Release\_in\_the\_Reef\_Coral\_Pocillopora\_Damicornis\_at\_Enewetak\_and\_Hawaii</u> *Methods* 

Rodrigues, L. J., & Grottoli, A. G. (2006). Calcification rate and the stable carbon, oxygen, and nitrogen isotopes in the skeleton, host tissue, and zooxanthellae of bleached and recovering Hawaiian corals. Geochimica et Cosmochimica Acta, 70(11), 2781–2789. doi: 10.1016/j.gca.2006.02.014 Methods

Rodrigues, L. J., & Grottoli, A. G. (2007). Energy reserves and metabolism as indicators of coral recovery from bleaching. Limnology and Oceanography, 52(5), 1874–1882. doi:10.4319/lo.2007.52.5.1874 Methods

Schneider, C. A., Rasband, W. S., ... (n.d.). ImageJ. US National Institutes of Health, Bethesda, MD, USA. Available from <u>https://imagej.nih.gov/ij/</u> Software

Schoepf, V., Grottoli, A. G., Levas, S. J., Aschaffenburg, M. D., Baumann, J. H., Matsui, Y., & Warner, M. E. (2015). Annual coral bleaching and the longterm recovery capacity of coral. Proceedings of the Royal Society B: Biological Sciences, 282(1819), 20151887. doi:<u>10.1098/rspb.2015.1887</u> *Methods* 

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Parameters

Parameter	Description	Units
id	Coral fragment identifier code	unitless
full_species_name	Full species name. Genus and species name of coral	unitless
genus	Genus name of coral	unitless
collection_site_name	Collection site name location where coral was sampled from	unitless
lat	Collection site latitude; South is negative.	decimal degrees
lon	Collection site longitude; West is negative.	decimal degrees
collection_max_depth	Collection site depth range minimum	meters
collection_min_depth	Collection site depth range maximum	meters
coral_collection_date	Coral collection date date when coral was sampled in ISO 8601 format yyyy-mm- dd	unitless
biomass	Tissue biomass of each fragment	mg cm-2
lipid	Total lipid concentration of each fragment	kilojoules per gram of dry weight (kJ gdw-1)
protein	Total soluble protein concentration of each fragment	kilojoules per gram of dry weight (kJ gdw-1)
chl	Chlorophyll a & c2 concentration	micrograms per centimeter squared (æg cm- 2)
d13Ch_e	The d13C ( $\delta$ 13C) of the coral host minus the $\delta$ 13C of the algal endosymbiont	permil (0/00)
d15Nh_e	The d15N ( $\delta$ 15N) of the coral host minus the $\delta$ 15N of the algal endosymbiont	permil (0/00)

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# Instruments

Dataset-specific Instrument Name	PDZ Europa 20-20 isotope ratio mass spectrometer	
Generic Instrument Name	Name Elemental Analyzer	
	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.	

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# **Project Information**

# Phenotype and genotype of coral adaptation and acclimatization to global change (Coral Adaptation)

Coverage: Oahu, HI and Hawaii Institute of Marine Biology

#### **Project Summary:**

*Overview:* This study proposes to evaluate the adaptation and acclimatization capacity of eight species of Hawaiian corals to long-term exposure of elevated temperature and ocean acidification (OA) conditions using a two-part approach: 1) a survey of natural corals found across natural temperature and pCO2 gradients and 2) a two-year long mesocosm study which will expose corals collected in part 1 to a range of temperature and pCO2 conditions expected this century. In both approaches, the phenotypic (i.e., physiological and biogeochemical) responses of corals to future climate change will be measured in conjunction with the already funded genotypic (i.e., genomic and transcriptomic) responses of the same corals by Dr. Rob Tonen. This study will address variation at both the population and species level. It will also be the first study to examine the effects of elevated temperature and pCO2 on corals in replicated mesocosms over a multiannual timeframe with a comprehensive suite of physiological, biogeochemical, and genomic tools.

*Intellectual Merit:* Coral reefs are among the most diverse ecosystems on the planet, housing an estimated 25% of marine species. Yet, they appear to be especially susceptible to the effects of climate change and ocean acidification. To date, the assumption has been that corals will not be able to adapt because the rates of anthropogenically driven ocean acidification and climate change are too high. But there is little experimental evidence to evaluate that assumption. Recent models highlight the critical importance of that assumption in determining coral extinction risk, and several recent studies (including a couple of recent ones from Grottoli's group) indicate that previous studies may have underestimated the potential for corals to acclimatize or adapt to global change. Here, quantitative, empirical estimates of the potential for long-term coral acclimatization and adaptation under global change scenarios will be made. The proposed study includes ~97% of the corals in the Hawaiian archipelago, yielding extensive spatial and biological relevance for the study. Lastly, this research brings together the expertise of Grottoli at OSU (coral physiologist and biogeochemist), Toonen at UH (marine molecular biologist), and McCulloch at UWA (geochemist) in a unique collaboration that blends a large suite of genetic, physiological, and biogeochemical tools to build an unprecedentedly comprehensive picture of coral adaptation and acclimation to global change.

*Broader Impacts:* Half of the species in the Hawaiian archipelago are endemic, making Hawaiian coral reefs a high priority for biodiversity conservation. Results from the proposed work will be used for adaptive management plans that collaborator Dr. Toonen is involved in with the goal of preserving Hawaiian coral biodiversity in a UNESCO World Heritage Site -- the Papahanaumokuakea Marine National Monument (PMNM). PMNM encompasses the northwestern Hawaiian Islands, is renowned as one of the most pristine and highly protected coral reefs remaining on the planet, is the single largest conservation area under the U.S. flag, and one of the largest marine conservation areas in the world. This project will provide a bridge between shortterm, single-species studies and longer-term, multi-species responses to global change in reef community settings. Findings from this work will be communicated at scientific meetings, through peer-reviewed journal publications, and via press releases. Grottoli will also bring her research and enthusiasm for marine science into her classrooms and onto the podium when giving general audience and professional talks. She has an established track record of recruiting and promoting under-represented students and will continue to do so. This project will recruit 3 undergrads and 2 high school students for supported senior thesis/independent research and provide an environment that will foster their passion and skills necessary to pursue career options in STEM disciplines.

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# Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	<u>OCE-1459536</u>

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