

Symbiont psbA and ORF host sequences generated as part of a study of pCO₂ variability on the reef-building coral *Pocillopora damicornis* conducted at Heron Island Research Station, Heron Island, southern Great Barrier Reef in 2021

Website: <https://www.bco-dmo.org/dataset/885679>

Data Type: Other Field Results

Version: 1

Version Date: 2022-12-20

Project

» [Influence of environmental pH variability and thermal sensitivity on the resilience of reef-building corals to acidification stress](#) (Coral Resilience)

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

Abstract

This dataset contains symbiont psbA and ORF host sequences and accession information at the National Center for Biotechnology Information (NCBI)'s Genbank database. These data were collected as part of a study of pCO₂ variability on the reef-building coral *Pocillopora damicornis* conducted at Heron Island Research Station, Heron Island, southern Great Barrier Reef in 2021 (Brown et al., 2022). Abstract for all data from the study (Brown et al., 2022) including this dataset: Ocean acidification is a growing threat to coral growth and the accretion of coral reef ecosystems. Corals inhabiting environments that already endure extreme diel pCO₂ fluctuations, however, may represent acidification resilient populations capable of persisting on future reefs. Here, we examined the impact of pCO₂ variability on the reef-building coral *Pocillopora damicornis* originating from reefs with contrasting environmental histories (variable reef flat vs. stable reef slope) following reciprocal exposure to stable (218 ± 9) or variable (911 ± 31) diel pCO₂ amplitude (μatm) in aquaria over eight weeks. This study measured: growth (net calcification, extension, CaCO₃ density) and physiology (dark respiration, light-enhanced dark respiration, host soluble protein, mycosporine-like amino acids, net photosynthesis, photosynthetic efficiency, endosymbiont density, chlorophyll a concentration, intracellular pH) of *P. damicornis* across treatment and origin. See all datasets related to this publication (<https://www.bco-dmo.org/related-resource/885684>).

Table of Contents

- [Coverage](#)
- [Dataset Description](#)
 - [Methods & Sampling](#)
 - [Data Processing Description](#)
- [Data Files](#)
- [Related Publications](#)
- [Related Datasets](#)
- [Parameters](#)
- [Project Information](#)
- [Funding](#)

Coverage

Spatial Extent: Lat:-23.27 Lon:151.55

Temporal Extent: 2021-01-06 - 2021-04-06

Dataset Description

The truncated sequences were submitted to GenBank under the following accession numbers (accessible from the below "popsets"):

"*Cladocopium latusorum* photosystem II D1/D2 subunit (psbA) gene, partial cds; chloroplast."

* popset: <https://www.ncbi.nlm.nih.gov/popset/2310683771>

* accessions OP279755 - OP279774

"*Pocillopora damicornis* ATP synthase subunit 6 (ATP6) gene, complete cds; and control region, complete sequence; mitochondrial."

* popset: <https://www.ncbi.nlm.nih.gov/popset/2310683883>

* accessions OP296521 - OP296503

This dataset contains two fasta files aggregating sequences from the above popsets at NCBI.

Methods & Sampling

Genetic analyses

A small chip (2–3mm) of each fragment (preserved in 100% ethanol and stored at -80°C) was used to extract gDNA, cf. methods described in (Lajeunesse et al., 2003) and the DNA eluted in ddH₂O. The coral species field-based identification using morphological characteristics (*sensu* *Pocillopora damicornis*) was confirmed with a genetic assay (Schmidt-Roach et al., 2013) using the mitochondrial ORF region. The DNA was diluted 1:20 and PCR was done using the FATP/RORF primers cf. (Flot and Tillier, 2007) and NEB TAQ Polymerase (M0320) following the manufacturer's routine PCR protocol under cycling conditions: 95°C 2 min initial denaturation, 35 cycles of 94°C 30s, 53°C 30s, 68°C 1 min, and a final extension of 68°C for 10 min. All but two specimens (one from each habitat) were successfully amplified; these were cleaned using ExoSap-IT and Sanger sequenced in a single direction using the forward FATP primer at the Australian Genome Research Facility (Brisbane, Australia). In addition, the species of resident coral endosymbionts (Symbiodiniaceae) were identified using PCR amplification of the ITS2 rDNA region followed by denaturing gradient electrophoresis alongside local 'marker' specimens from *Pocillopora* species with previously confirmed symbiont identifications (cf. Sampayo et al., 2009). To cross-confirm, dominant bands of characteristic profiles were excised, re-amplified and sequenced using the forward ITS2intfor and reverse ITS2reverse primers (cf. Lajeunesse et al., 2003; Sampayo et al., 2009). In addition, the chloroplast minicircle psbA non-coding and partial coding region were amplified using two different primer sets and conditions. Initially, the 7.4-Forw/7.8-Rev primers were used cf. (Moore et al. 2003) but some samples failed to amplify. Some of these yielded successful PCR amplicons with the psbAFor-1/psbARev-1 primers (Lajeunesse and Thornhill, 2011). Successful reactions were cleaned using ExoSap-IT and bi-directionally Sanger sequenced with their respective primers at the Australian Genome Research Facility. Sequence chromatograms for all regions (host mtORF; symbiont ITS2 rDNA and chloroplast psbA) were visually inspected and compared to holotype sequences from described *Pocillopora* species (Schmidt-Roach et al., 2013) and recently described symbiont species derived from these coral species (Turnham et al., 2021). The mtORF sequences from the study specimens all had 100% match with the *P. damicornis* (GenBank Accession numbers OP296503 - OP296521; 100% match to *Pocillopora* type alpha cf. (Schmidt-Roach et al., 2013), GenBank accession numbers JX985598 and JX985606). The ITS2 rDNA profiles as well as sequences matched previously described *Cladocopium* type 'C1b-c, 42a' from Heron Reef (Sampayo et al. 2007). For the psbA, 4 out of 24 samples returned poor sequence reads (2 from each environment). The remaining 20 psbA sequences were aligned with inclusion of reference sequences for recently described *Cladocopium latusorum* and *C. pacificum* (GenBank Accession numbers MW819755 - MW819779 and MW861711 - MW861727, respectively) as well as the sequence for *C. goreaii* (Turnham et al., 2021). Sequences were aligned using Geneious (MAFFT), checked and adjusted by eye, which was particularly important given the large number of indels present in the psbA sequence dataset. All sequences were truncated to the 7.4-Forw and 7.8-Rev primer binding sites, given that no differences were seen in the sequence regions extending outward to the psbA coding region psbAFor1/psbARev1 binding sites. The truncated sequences were submitted to GenBank under accession numbers OP279755 - OP279774 (<https://www.ncbi.nlm.nih.gov/popset/2310683771>). All indels were adjusted to represent a single base change, using gaps as a 5th character state, in subsequent phylogenetic analyses. A psbA (partial coding, and entire non-coding region) maximum parsimony majority-rule phylogeny was constructed using a heuristic search with bootstrap values calculated (TBR branch swapping, random addition, 1000 replicates) in PAUP vs. 4.0a (build 169; Turnham et al., 2021).

For more detailed information, please see: Brown et al. (2022).

Data Processing Description

See results publication Brown et al., 2022 for details of statistical analyses performed using these data.

The analysis code package for "Environmental memory gained from exposure to extreme pCO₂ variability promotes coral cellular acid-base homeostasis" published as Brown (2022, doi: 10.5281/zenodo.7373705) which is a publication of github repository [https://github.com/imkristenbrown/pCO₂-variability-promotes-coral-cellular-acid-base-homeostasis](https://github.com/imkristenbrown/pCO2-variability-promotes-coral-cellular-acid-base-homeostasis).

[[table of contents](#) | [back to top](#)]

Data Files

File
Symbiont ORF host sequences filename: KB_ORFseqs_GenBank.fasta (FASTA, 21.21 KB) MD5:72974616fc311f369b527e739a92b256 Sequences (fasta format) with the Genbank accession numbers for the deposited symbiont ORF host sequences. This fasta file aggregates data under NCBI popset, Accessions (OP296503.1 - OP296521.1): Brown, K. T., Mello-Athayde, M. A., Sampayo, E. M., Chai, A., Dove, S., & Barott, K. L. (2022). Pocillopora damicornis ATP synthase subunit 6 (ATP6) gene, complete cds; and control region, complete sequence; mitochondrial. The National Center for Biotechnology Information PopSet: 2310683883. Available from https://www.ncbi.nlm.nih.gov/popset/2310683883
Symbiont psbA host sequences filename: KB_psbbaseqs_Genbank.fasta (FASTA, 21.56 KB) MD5:e601d1b3f85037e63198e6deb853f0aa Sequences (fasta format) with the Genbank accession numbers for the deposited symbiont psbA host sequences. This fasta file aggregates data under NCBI popset 2310683771 Accessions (OP279755.1 - OP279774.1) Brown, K. T., Mello-Athayde, M. A., Sampayo, E. M., Chai, A., Dove, S., & Barott, K. L. (2022). Cladocopium latusorum photosystem II D1/D2 subunit (psbA) gene, partial cds; chloroplast. The National Center for Biotechnology Information PopSet: 2310683771. Available from https://www.ncbi.nlm.nih.gov/popset/2310683771

[[table of contents](#) | [back to top](#)]

Related Publications

Brown, K. T. (2022). Barott Lab/Heron pHi [Data set]. Zenodo. <https://doi.org/10.5281/ZENODO.7373705>
<https://doi.org/10.5281/zenodo.7373705>
Software

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>
Results

Flot, J.-F., & Tillier, S. (2007). The mitochondrial genome of Pocillopora (Cnidaria: Scleractinia) contains two variable regions: The putative D-loop and a novel ORF of unknown function. *Gene*, 401(1-2), 80–87.
doi:[10.1016/j.gene.2007.07.006](https://doi.org/10.1016/j.gene.2007.07.006)
Methods

Lajeunesse, T. C., & Thornhill, D. J. (2011). Improved Resolution of Reef-Coral Endosymbiont (Symbiodinium) Species Diversity, Ecology, and Evolution through psbA Non-Coding Region Genotyping. PLoS ONE, 6(12), e29013. <https://doi.org/10.1371/journal.pone.0029013>

Methods

Lajeunesse, T. C., Loh, W. K. W., van Woesik, R., Hoegh-Guldberg, O., Schmidt, G. W., & Fitt, W. K. (2003). Low symbiont diversity in southern Great Barrier Reef corals, relative to those of the Caribbean. Limnology and Oceanography, 48(5), 2046–2054. Portico. <https://doi.org/10.4319/lo.2003.48.5.2046>

Methods

Moore, R. B. (2003). Highly organized structure in the non-coding region of the psbA minicircle from clade C Symbiodinium. INTERNATIONAL JOURNAL OF SYSTEMATIC AND EVOLUTIONARY MICROBIOLOGY, 53(6), 1725–1734. <https://doi.org/10.1099/ijs.0.02594-0>

Methods

R Core Team (2021). R: A language and environment for statistical computing. R v4.0.0. R Foundation for Statistical Computing, Vienna, Austria. URL <https://www.R-project.org/>

Software

SAMPAYO, E. M., DOVE, S., & LAJEUNESSE, T. C. (2009). Cohesive molecular genetic data delineate species diversity in the dinoflagellate genus Symbiodinium. Molecular Ecology, 18(3), 500–519. <https://doi.org/10.1111/j.1365-294x.2008.04037.x> <https://doi.org/10.1111/j.1365-294X.2008.04037.x>

Methods

SAMPAYO, E. M., FRANCESCHINIS, L., HOEGH-GULDBERG, O., & DOVE, S. (2007). Niche partitioning of closely related symbiotic dinoflagellates. Molecular Ecology, 16(17), 3721–3733. <https://doi.org/10.1111/j.1365-294x.2007.03403.x> <https://doi.org/10.1111/j.1365-294X.2007.03403.x>

Methods

Schmidt-Roach, S., Lundgren, P., Miller, K. J., Gerlach, G., Noreen, A. M. E., & Andreakis, N. (2012). Assessing hidden species diversity in the coral Pocillopora damicornis from Eastern Australia. Coral Reefs, 32(1), 161–172. <https://doi.org/10.1007/s00338-012-0959-z>

Methods

Turnham, K. E., Wham, D. C., Sampayo, E., & Lajeunesse, T. C. (2021). Mutualistic microalgae co-diversify with reef corals that acquire symbionts during egg development. The ISME Journal, 15(11), 3271–3285. <https://doi.org/10.1038/s41396-021-01007-8>

Methods

Wickham, H. (2016). ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York. ISBN 978-3-319-24277-4, <https://ggplot2.tidyverse.org>. <https://doi.org/10.1007/978-3-319-24277-4>

Methods

[[table of contents](#) | [back to top](#)]

Related Datasets

IsRelatedTo

Brown, K. T., Mello-Athayde, M. A., Sampayo, E. M., Chai, A., Dove, S., & Barott, K. L. (2022). Cladocodium latusorum photosystem II D1/D2 subunit (psbA) gene, partial cds; chloroplast. The National Center for Biotechnology Information PopSet: 2310683771. Available from <https://www.ncbi.nlm.nih.gov/popset/2310683771>

Brown, K. T., Mello-Athayde, M. A., Sampayo, E. M., Chai, A., Dove, S., & Barott, K. L. (2022). Pocillopora damicornis ATP synthase subunit 6 (ATP6) gene, complete cds; and control region, complete sequence; mitochondrial. The National Center for Biotechnology Information PopSet: 2310683883. Available from <https://www.ncbi.nlm.nih.gov/popset/2310683883>

[[table of contents](#) | [back to top](#)]

Parameters

Parameters for this dataset have not yet been identified

Project Information

Influence of environmental pH variability and thermal sensitivity on the resilience of reef-building corals to acidification stress (Coral Resilience)

Coverage: Kaneohe Bay, Oahu, HI; Heron Island, Queensland, Australia

NSF Award Abstract:

Coral reefs are incredibly diverse ecosystems that provide food, tourism revenue, and shoreline protection for coastal communities. The ability of coral reefs to continue providing these services to society is currently threatened by climate change, which has led to increasing ocean temperatures and acidity that can lead to the death of corals, the animals that build the reef framework upon which so many species depend. This project examines how temperature and acidification stress work together to influence the future health and survival of corals. The scientists are carrying out the project in Hawaii where they have found individual corals with different sensitivities to temperature stress that are living on reefs with different environmental pH conditions. This project improves understanding of how an individual coral's history influences its response to multiple stressors and helps identify the conditions that are most likely to support resilient coral communities. The project will generate extensive biological and physicochemical data that will be made freely available. Furthermore, this project supports the education and training of undergraduate and high school students and one postdoctoral researcher in marine science and coral reef ecology. Hands-on activities for high school students are being developed into a free online educational resource.

This project compares coral responses to acidification stress in populations experiencing distinct pH dynamics (high diel variability vs. low diel variability) and with distinct thermal tolerances (historically bleaching sensitive vs. tolerant) to learn about how coral responses to these two factors differ between coral species and within populations. Experiments focus on the two dominant reef builders found at these stable and variable pH reefs: *Montipora capitata* and *Porites compressa*. Individuals of each species exhibiting different thermal sensitivities (i.e., bleached vs. pigmented) were tagged during the 2015 global coral bleaching event. This system tests the hypotheses that 1) corals living on reefs with larger diel pH fluctuations have greater resilience to acidification stress, 2) coral resilience to acidification is a plastic trait that can be promoted via acclimatization, and 3) thermally sensitive corals have reduced capacity to cope with pH stress, which is exacerbated at elevated temperatures. Coral cells isolated from colonies from each environmental and bleaching history are exposed to acute pH stress and examined for their ability to recover intracellular pH in vivo using confocal microscopy, and the expression level of proteins predicted to be involved in this recovery (e.g., proton transporters) is examined via Western blot and immunolocalization. Corals from each pH history are exposed to stable and variable seawater pH in a controlled aquarium setting to determine the level of plasticity of acidification resilience and to test for pH acclimatization in this system. Finally, corals with different levels of thermal sensitivity are exposed to thermal stress and recovery, and their ability to regulate pH is examined over time. The results of these experiments help identify reef conditions that promote coral resilience to ocean acidification against the background of increasingly common thermal stress events, while advancing mechanistic understanding of coral physiology and symbiosis.

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.

Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1923743