FINAL REPORT

Award Number: NA13NOS4820023

Project title: *Do increased nutrients increase the vulnerability of reef corals to climate change? A field and experimental study in the Florida Keys using novel genetic tools.*

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Project Dates: 1 July 2013 – 30 June 2015 *(including no-cost extension)*

Project objectives:

This project tests the hypothesis that elevated nutrients cause corals to increase the densities of their algal symbionts, and that this change lowers their thermal tolerance. It uses a novel molecular assay to measure changes in the symbiont:host (S:H) cell ratio of corals, and undertakes a 12-month monitoring program in the Florida Keys to assess the variability of S:H ratios in the field, how they vary between species, and how they vary over seasonal timescales. It also includes laboratory experiments in which corals raised in elevated nutrients are then challenged with thermal stress to assess changes in thermal tolerance as a result of nutrient exposure. Finally, it includes an outreach component to help disseminate our findings and their relevance for management.

Project Findings:

Field work

We were delayed starting the project (see *No Cost Extension*, below), and so annual sampling for the project did not begin until 28 August 2014 and ended on 1 September 2015 (in order to cover a full 12 months). This sampling period includes a very unusual period of stress on these reefs, beginning during the onset of severe bleaching in the Florida Reef Tract in August 2014, continuing through the recovery (or, in some cases, mortality) of these bleached corals, and following on to a second bleaching episode which began in August 2015 and is still ongoing. It also covers a disease outbreak in the fall of 2014 that followed the initial bleaching. To date, we have collected 969 samples from ~220 tagged colonies of 6 coral species (*Orbicella faveolata, Montastraea cavernosa, Porites astreoides, Diploria strigosa,* and *Siderastrea siderea*) at our study site at Emerald Reef in the northern Florida Keys. These colonies have been sampled approximately monthly through January 2015, and semi-monthly from February 2015 onwards. The DNA from

these samples is being extracted and purified, and we have begun qPCR analysis on a newly-calibrated instrument. Although these samples amplify well using standard PCR (of ITS-2 rDNA), we are having problems obtaining reliable amplifications using qPCR. Due to the fact that we have only just finished collecting the first samples from the second bleaching event we are still in the process of resolving this problem. We will write up and publish our findings regarding seasonal changes in S:H ratio in different coral species as soon as the analyses are complete. We also anticipate that these samples will be very helpful in understanding how symbiont communities respond to a natural bleaching event, an unanticipated value-added component of the project. In addition to resulting in potential publication(s), these data will also form part of Ph.D. student Ana Palacio's doctoral dissertation.

Laboratory experiments

Our first round of laboratory experiments exposed replicate samples of *Orbicella faveolata*, *Montastraea cavernosa*, and *Porites astreoides* to elevated (10uM) N concentrations for a period of >3 months prior to exposing them to thermal stress. During this time we took measurements of photochemical efficiency using an Imaging Pulse Amplitude Modulated (I-PAM) fluorometer to assess heat tolerance, and removed small biopsies of tissue for qPCR analysis of the *Symbiodinium* communities. We also added an additional treatment to our experiment in which we raised replicate samples of *O. faveolata* under elevated Fe to see whether additional nutrient sources influence symbiont densities and bleaching tolerance. Preliminary analysis of our data, as well as visual observations made on the corals, indicate that corals exposed to higher N concentrations contained higher symbiont densities and bleached more severely than controls, as we hypothesized. The effect of Fe-enrichment on symbiont densities does not appear to be as strong as adding N. However, we still need to correct our molecular data for actin copy number and re-run samples to ensure consistency.

A second experiment on *Siderastrea siderea* and *Montastraea cavernosa* is currently underway. These experiments include the addition of N, P, and Fe to corals under controlled temperatures, together with feeding. However, we experienced high variability in the appearance of our experimental colonies in our new experimental facility (see below), perhaps due to new LED light sources we are using. As a result of this, we decided to extend our nutrient exposures in order for the corals to stabilize prior to exposing them to thermal stress. We will write up the results of these experiments for publication as soon as they are complete. They will also form part of Ph.D. student Ana Palacio's doctoral dissertation.

We also ran an experiment with a visiting Master's student (M-O. Guillermard) from France to compare the symbiont to host cell ratio assay against cell counts using a haemocytometer, in order to determine the comparability of the two methods in measuring symbiont density. These data were used for the intern's Master's thesis poster at the University of Paris, and we will incorporate them into a future publication, together with the results of the rest of the experiments described above as soon as they are complete.

Finally, we also ran a competition experiment to better understand why the zoanthid *Palythoa caribaeorum* is becoming increasingly common on our study reefs, particularly after the summer bleaching of 2014. We found that *Palythoa* hosts *Symbiodinium* D1a (*S. trenchi*, a thermally tolerant symbiont not previously recorded in *Palythoa*) but that *Palythoa* does not compete well against corals under non-stressful laboratory conditions. These findings suggest that *Palythoa* may be becoming more prevalent on Florida's reefs because it is relatively more resistant to heat-

induced mortality compared to its neighboring corals, and not because it is directly outcompeting or smothering healthy corals directly.

No Cost Extension

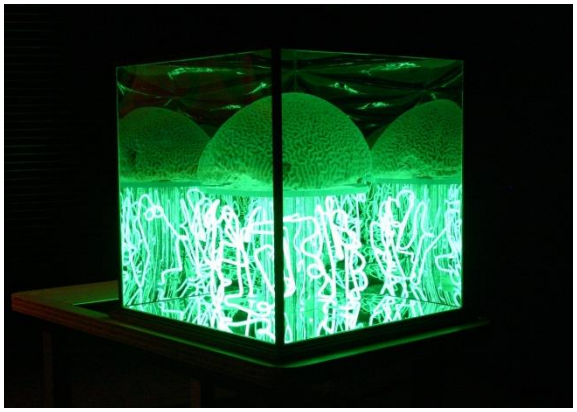
Due to a delay in award announcement and receipt of funds we were not able to officially start our project on its original start date of 1st July 2013. We originally anticipated beginning the project with trial experiments in our old running seawater building (prior to it being replaced with a new Marine Technology and Life Sciences research building at the University of Miami's Rosenstiel School). However, with the delay in being notified of an award, we decided to delay the start of the project until the new building was opened, which was scheduled for January 2014. However, the final construction of the building was delayed multiple times and the building was not officially opened until October 2014, more than a year after the original start of the award. We successfully applied for a one-year No Cost Extension in early May 2014 and planned to begin the field component of the award immediately. However, on 21 May 2014 we were notified of the coral rescue from the Port of Miami deep dredge, with a start date of 26 May 2014. The urgency of this rescue meant that it took precedence over our field monitoring project, which eventually began in August 2014.

These delays meant that the start of the project was eventually delayed for over one year. During the NCE period we were further delayed because many of the corals we had collected from the Port of Miami in June 2014 (which we were holding in our large outdoor tanks for these experiments) also began to bleach as a result of the same high summer temperatures that were causing bleaching on the reefs (see above). Because of this, we could not use these corals for our intended experiment (although they will be useful for other experiments not related to this grant). To compensate for this, we collected (under permit) additional colonies for the bleaching experiment, and these experiments are currently underway (see above).

Outreach

Science-Art Collaboration at Miami Museum of Science

In the summer of 2013 PI Baker designed and produced (with artist collaborator Sinisa Kukec) an art installation entitled *Do brain corals dream of their algal symbionts?*, as part of *The Curious Vault*: *Collaborations* at the Miami Museum of Science (now Frost Museum of Science). This



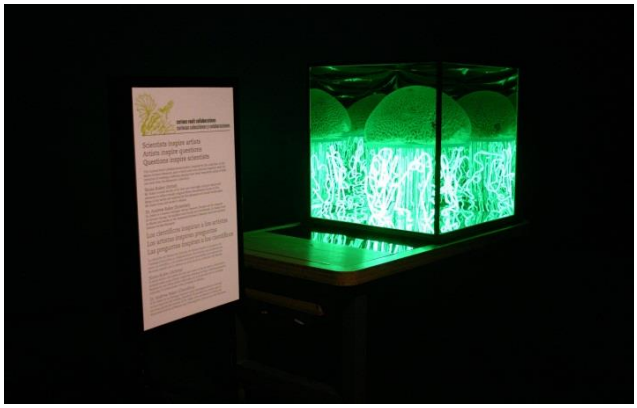


Figure 1: *Do brain corals dream of their algal symbionts? (*2013). Sinisa Kukec and Andrew Baker, Frost Science Museum, Miami, FL. Brain coral, neon, acrylic two way mirror, red oak, soundtrack, and electronics. 33 x 34 x 34 inches

was the inaugural collaboration between a local scientist and a local artist to produce a collaborative installation at the Museum involving objects from the Museum's collection. The installation (Fig. 1) features two-way mirrors and a set of voice recordings describing corals value to marine ecosystems, the threats they face, and the fascinating history of corals in art, literature, and mythology. It was on display at the Museum from September 2013 through August 2015, and is in the process of being relocated for exhibit at the University of Miami's Rosenstiel School.

Public Panels on Water Quality and Coral Reefs

In April 2014 PI Baker participated in two public panels on water quality in the Florida Keys organized by Last Stand and the Everglades Law Center. Both panels were in Monroe county (the Key Largo Library on April 16th, and the Eco-Discovery Center Key West on April 22nd). Baker presented a synthesis of nutrient effects on corals and the compounding effects of nutrient pollution and climate change. Also on the panels were Billy Causey, Ph.D. (Southeast Regional Director for NOAA's Office of National Marine Sanctuaries), Bob Johnson (Director of South Florida Natural Resources Center for Everglades National Park), Tom Van Lent, Ph.D. (Director of Science and Policy for the Everglades Foundation), and Jerry Lorenz, Ph.D. (Director of Research for Audubon Florida).

Coral Rescue from the Port of Miami "deep dredge"

In May-June 2014 the entire project team participated in a coral rescue project in the Port of Miami to save corals from the "deep dredge", which was being used to deepen and widen the shipping channel to accommodate post-Panamax cargo ships. We assembled a team of volunteer divers from UM/RSMAS and NOAA and were able to collect ~1,200 colonies (ranging in diameter from <1" to > 12") from the Port channel that would otherwise have been killed by the dredge. Saving these corals from the dredge became a *cause celebre* locally and went on to get national and international press attention because of the unanticipated environmental impacts of the dredge, including television (e.g., *Fox News, Al-Jazeera, CBS-4*), newspapers (e.g., *The New York Times, Miami Herald, Sun Sentinel, Christian Science Monitor, Seattle Post Intelligencer*) and radio (e.g., Miami's WLRN and several other NPR outlets). This helped raise the profile of corals and the various threats, including nutrients, that they face worldwide.

Google Hangout in the Lab

In January 2015 we hosted a Google Hangout in the lab, organized by the National Geographic Society. This was live streamed to K-8 schools nationwide and also archived online (https://www.youtube.com/watch?v=arS9mqPL7Fk&feature=youtu.be). During this Hangout we talked about our research, the various threats coral reefs face, and their value to human societies. The lab's research was also selected for a series of brief informative videos as part of the coral reef exhibit at the new Frost Museum of Science in Miami (opening in 2016), and PI Baker also appeared briefly in a short NSF-funded documentary on symbiosis aimed at schools (https://vimeo.com/128943934).

Miami Science Barge

In 2015, PI Baker also joined the Advisory Committee of the Miami Science Barge, a floating environmental education center on Biscayne Bay, which educates about STEM and sustainability through marine science. It won the inaugural Knight Cities Challenge in March 2015 and is scheduled to open in 2016.

Professional Development:

As a result of this award, the following 18 personnel have been provided with educational opportunities and/or professional development, or will be involved in collaborative research products (publications):

Postdocs:	Dr. P. Jones, Dr. X. Serrano
Ph.D. students:	A. Palacio, R. Winter, P. Kushlan, J. Bartz, E. Towle, C. Drury
Master's students:	D. Moanga, B. Jensen
Undergraduates:	Z. Gapayao, G. Snyder, S. Wedelich, J. Destache, S. May, M. Coogan, M. Kendi
High school students:	H. Waxman

Publications (underlined = graduate student in PI Baker's lab)

The principal research findings from this project still have to be fully written up and submitted for publication. However, the following publications from the award period are also related to the project as they investigate the use of S:H ratios to investigate symbiont community dynamics in our experimental corals. We anticipate that future publications will be added to this list.

Aswani S, Mumby PJ, **Baker AC**, Christie P, McCook LJ, Steneck RS, Richmond RH (2015) Scientific frontiers in the management of coral reefs. *Frontiers in Marine Science* 2:50 doi: 10.3389/fmars.2015.00050

Cunning R, Vaughan N, Gillette P, Capo T, Maté J, **Baker AC** (2015). Dynamic regulation of partner abundance mediates response of reef coral symbioses to environmental change. *Ecology* 96(5): 1411–1420

Silverstein RN, Cunning R, **Baker AC** (2015) Change in algal symbiont communities after bleaching, not prior heat exposure, increases heat tolerance of reef corals. *Global Change Biology* 21: 236–249, doi: 10.1111/gcb.12706doi: 10.1111/gcb.12706

Lirman D, Schopmeyer S, Galvan V, Drury C, **Baker AC**, Baums IB (2014) Growth dynamics of the threatened Caribbean staghorn coral *Acropora cervicornis*: Influence of host genotype, symbiont identity, colony size, and environmental setting. *PLoS One* doi: 10.1371/journal.pone.0107253

Cunning R, Gillette P, Capo T, Galvez K, **Baker AC** (2014). Growth tradeoffs associated with thermotolerant symbionts in the coral *Pocillopora damicornis* are lost in warmer oceans. *Coral Reefs* doi 10.1007/s00338-014-1216-4

Cunning R, **Baker AC** (2014) Not just who, but how many: The importance of partner abundance in reef coral symbioses. *Frontiers in Microbiology* 5:400.

Baker A (2014) What is natural? Shifting baselines in marine conservation. *Red Flag* 8: 36-39

Baker A (2014) Blue carbon and coral reefs. *In:* AGEDI (2014) *Building Blue Carbon Projects - An Introductory Guide*. AGEDI/EAD. Published by AGEDI. Produced by GRID-Arendal, A Centre Collaborating with UNEP, Norway. p. 42-45 *(contributed section)*