Investigating the Symbiotic Community of Coral Species in Puerto Rico

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Introduction

- Coral reefs in Puerto Rico have experienced bleaching due to rising ocean temperatures caused by climate change that destroy their symbiotic relationship with the algae called zooxanthellae (Smith 1978).
- Two coral species found in Puerto Rico are *Montastraea cavernosa* (great star coral) and *Orbicella faveolata* (mountainous star coral).
- Corals are believed to be able to modify their symbiont communities when facing temperature stress by either switching algae with the surrounding environment or shuffling their pre-existing background algae (Adaptive Bleaching Hypothesis; Buddemeier and Fautin, 1993
- Recent advances in research (e.g. quantitative PCR) now allow for more detailed observations into the dynamics of symbiont shuffling.

Cayo Enrique, and Cayo Mario, in La Parguera, Puerto Rico, where the samples were collected.(Google Earth Pro)

Objectives

- Do the two Caribbean coral species *Montastraea cavernosa* and *Orbicella faveolata* exhibit the same symbiont clades?
- Compare 2 species of corals' symbiotic algae communities (clades C and D), to gain a better understanding of the symbiotic community for coral conservation.
- Will both species exhibit clades C and D, as seen in previous research by Cunning and Baker 2013, who investigated the symbiotic algae community in Florida?

Image of *Orbicella faveolata* Photo Credit: Florida Keys National Marine Sanctuary

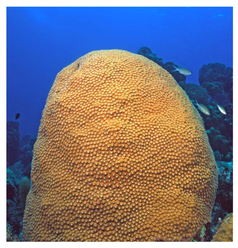
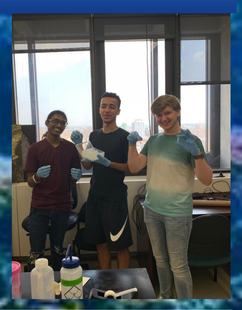


Image of *Montastraea cavernosa* Photo Credit: Marion Haarsma

Methods



- DNA was extracted from twelve coral tissue samples of both coral species (sites 4, 5, and 6), following Cunning and Baker's modified organic extraction protocol (2013)
- Procedure involves incubating the samples at 1% SDS at 65°C, Proteinase K digest at 55°C, incubation with 1% CTAB at 65°C, mixing chloroform, 100% ethanol precipitation at -20°C, adding 0.3M sodium acetate to dried pellet and then second ethanol precipitation at -20°C, ethanol wash in 70% ethanol, and then Tris-EDTA buffer (TE) was added. Samples were stored at -20°C.

Methods (Continued)

- After this, the DNA is ready for quantitative polymerase chain reaction (qPCR) to identify the symbionts.
- The qPCR protocol we followed was adapted from Cunning and Baker 2013: all reactions were carried out in 10 µL volumes (with 5 µL Taqman Genotyping MasterMix and 2 µL DNA template) on a Roche LightCycler 96 Instrument.
- Thermal cycling conditions were set at: an initial incubation at 50°C for 2 min and 95°C for 10 min, followed by 40 cycles of 95°C for 10s and 60°C for 1 min.

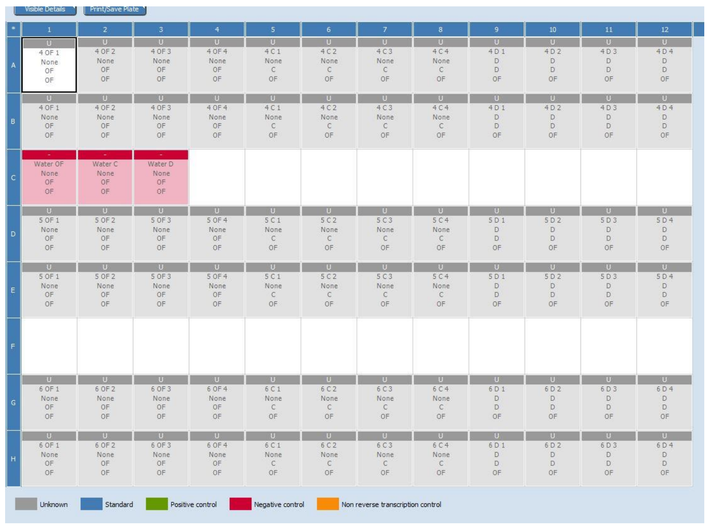


Fig. 1: Image is depicting a qPCR assay we set up for each coral species to determine if clade C or D was present.

Results

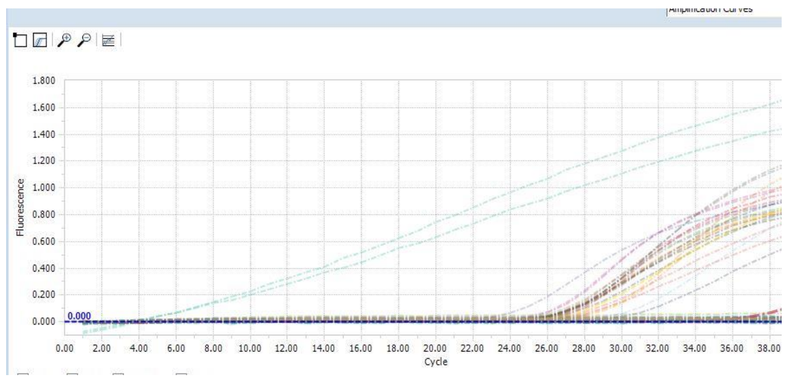


Fig. 2: Fluorescence graph representing the quantitative polymerase chain reaction process for the *M. cavernosa* samples. The curves indicate amplification.

Fig. 3: Table showing the results of the *M. cavernosa* qPCR assay. Green represents the sample that tested positive for clade C from columns 5-8. Red represents the samples that tested negative for clade D from columns 9-12.

Results (Continued)

- Both *Orbicella faveolata* and *Montastraea cavernosa* presented amplifications confirming the species being studied.
- *O. faveolata* did not exhibit any amplification for clades C or D, indicating that they did not contain symbionts from either clades C or D.
- Samples from site 4 for both species did not present any amplification for the species being studied nor the clades we screened for
- Some samples from sites (4-6) of the *M. cavernosa* exhibited symbionts from clade C, but not D.

Discussion

- Finding clade C in samples of *M. cavernosa* collected from sites 5 and 6 but not site 4 is unusual considering that the samples were collected from sites in close proximity.
- The corals that did not have symbionts from clades C or D could have symbionts from the other clades (A, B, or E-H), which we did not screen for, or they could possess a new clade, which is still unclassified.
- Some possible explanations for the samples from site 4 showing no amplification are that the samples from this site were contaminated, or there was human error during the DNA extraction/qPCR analysis, causing the results to be negative.
- This shows that corals in similar environments do not necessarily contain symbionts from the same clades, including corals of different species and different colonies of the same species.
- Future studies should look further into these clade classifications and determine what factors influence the clades of each coral population.

Literature Cited

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Acknowledgements

This study is partially supported and monitored by The National Oceanic and Atmospheric Administration – Cooperative Science Center for Earth System Sciences and Remote Sensing Technologies under the Cooperative Agreement Grant #: NA16SEC4810008. The author(s) would like to thank The City College of New York and NOAA Office of Education, Educational Partnership Program with Minority Serving Institutions (EPP/MSI) for support for Andrea Gomez and Ricardo Peralta. The statements contained within the poster are not the opinions of the funding agency or the U.S. government, but reflect the authors' opinions. The authors also thank the Pinkerton Foundation and the American Museum of Natural History for funding and supporting Sajiv Mahase and Connor Rabinowitz through the High School Initiative in Remote Sensing of the Earth Systems Science & Engineering (HIRES) program.