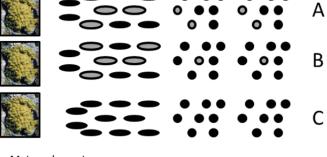
Final Report: Ocean Life Institute (6/1/09-5/31/11)

The genetic basis of physiological variation among coral larvae

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Scientists have become increasingly concerned about the degree to which reef-building corals will be able to persist and continue to build reefs in the face of increasing thermal stress and acidification associated with climate change. A few recent studies indicate stress responses can vary greatly among *individual* corals of the same species. It is still unclear how much of this variability is due to environmental effects and how much is due to genetic diversity. For example, corals growing in an environment with more food may be healthier and better able to tolerate a given stressor. In contrast, some coral genotypes may be better adapted to responding to the stressor. Both processes are important to understanding how coral individuals and populations and ultimately reef ecosystems will be impacted by environmental change. I proposed to evaluate performance (survival, growth) of coral larvae with different genetic backgrounds ("families" containing larvae produced by self-and/or crossfertilization) under conditions of ambient and elevated CO₂.

We conducted an experiment at the Bermuda Institute of Ocean Science in summer 2012 to investigate the role of maternal effects (genetic and environmental) on the response of juvenile corals (*Porites astreoides*) to ocean acidification (Figs 1-2).



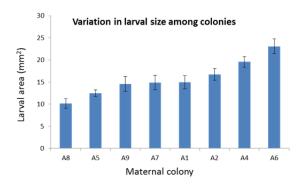
Maternal Larvae Ambient CO₂ Elevated CO₂ colonies Juveniles

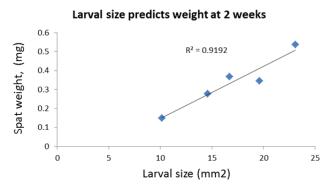
Fig 1. Simplified sampling design. Coral larvae were collected from several maternal colonies (3 shown). Larvae may be a mixture of cross- (black) and self-fertilized (gray) individuals (A,B), or may be homogeneous (e.g., "C" all cross-fertilized). Growth (not shown) and survival were monitored under ambient and elevated CO_2 (acidification). Possible survival outcomes would be no change in genetic composition (A), a change in proportions (increase in cross-fertilized, B), or no change in homogenous populations (C). Larvae and maternal tissue were sampled for genetic analysis.



Fig. 2. (Left) Aquarium containing four small coral colonies, each isolated within a mesh chamber. When the corals spawn, the larval float to the top and are collected with a pipette. (Right) Undergraduate students adding coral larvae to aquaria with terra cotta tiles. The aquaria contain either normal or acidified seawater. The larvae settle on the tiles and grow under these conditions for about two weeks.

From this experiment, we have found a large variation in the size of coral larvae (2.3-fold difference in projected area) released from different maternal colonies (Fig. 3). Under normal conditions (ambient seawater that isn't acidified), large larvae generally grow into large juvenile corals (Fig. 4).





Figs. 3 and 4. At the time of spawning, larvae released from different mother colonies have a big range in size (Fig. 3, left). Under normal conditions (ambient seawater), the larval size has a large influence on the size of the juvenile corals ("spat") after two weeks (Fig. 4, right). Each diamond in Fig. 4 represents the average for the larvae and juveniles released by one maternal colony.



Fig. 5. Example of three juvenile corals settled on a tile. These corals are each about 2 weeks old and consist of a single polyp. The white edge visible around some of the colonies is the newly formed skeleton. The circle in the center of the colonies is the mouth and gut, which are surrounded by a ring of tentacle used for feeding. The brownish-gold color is produced by symbiotic algae inside the coral tissue.

We have found that larvae from these different maternal colonies vary in their responses to ocean acidification. We might have predicted that larger larvae would be more resistant...they would use their stored energy to compensate for the effects of acidification and continue growing, but this is the opposite of what we observed!

We found that the largest larvae were **most** strongly affected by acidification, exhibiting smaller size and weight after two weeks in acidified conditions relative to control larvae from the same maternal colony (Fig. 6). One possible explanation is that the largest larvae have a greater scope for growth, which is negatively impacted by acidification. This means the largest larvae would normally grow a lot and with acidification, they would perhaps slow down their metabolism and grow less. In contrast, the smallest larvae aren't able to grow much, even under normal

conditions. Their metabolism is perhaps already slow and they are not able to form heavy skeletons. It is possible that mortality is higher among these smaller larvae, but mortality was not specifically measured in this study. In one case we even observed a positive impact of acidification: the juveniles belonging to the "family" with the smallest larval size were larger in the acidified tanks. One possible reason is that the acidification provided more carbon for the symbionts in the coral tissue to produce food for the coral animal.

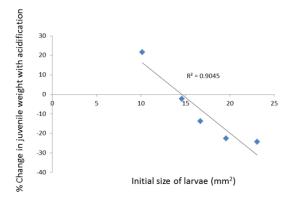
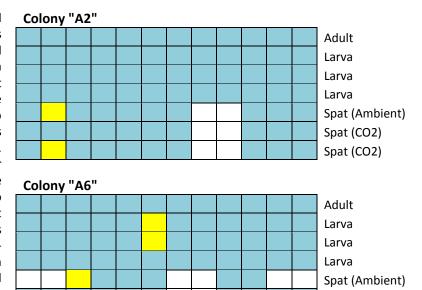


Fig. 6. Percent change in juvenile weight with acidification in relation to initial size of larvae. A positive number on the y-axis means that acidified tanks contained larger juvenile corals, and a negative number (generally observed) means the acidified tanks contained smaller corals. Corals with a larger initial size were more negatively impacted by acidification.

Genetic analyses were conducted in collaboration with Dr. Iliana Baums (Penn State University). A few studies indicate that some brooding can corals exhibit a high degree of self-fertilization. We have begun microsatellite-based genetic analysis of larval and juvenile corals from the acidification experiment described above (Fig. 7). Preliminary results indicate a high degree of clonality, but we have been able to find evidence of cross-fertilization as well as genetic patterns consistent with self-fertilization. In the future, we will need to identify more variable genetic markers to increase the sensitivity of this parentage analysis and analyze a larger number of samples. We hope to continue this work with the goal of determining whether and how the mode of fertilization affects the fitness of the offspring.

Fig. 7. Sample results of initial genetic analysis. The two clusters represent two different maternal colonies. Each row represents a different sample: the adult (maternal) colony or one of the offspring (larvae or juveniles, also called spat). Each column represents a different microsatellite marker. Blue indicates that the larval or juvenile type is the same as the parent, not necessarily that the two parental colonies are the same. Spat and juveniles with only blue squares may have resulted from selffertilization. Spat or juveniles with yellow squares most likely resulted from cross-fertilization. Because the overall amount of diversity in our markers is low (all the colonies we



Spat (Ambient)

Spat (Ambient)

have studied are relatively similar to one another), we are still limited in our ability to reliably detect cross-fertilization (some larvae may appear to have resulted from self-fertilization because their parents similar genotypes. With future development of additional markers, we will improve our power of detection.

Other Impacts of this Project

Training was provided for two undergraduate students:

Ms. Caitlin Church, a recent graduate of the University of San Diego (WHOI Guest Student). She assisted with field/experimental studies at BIOS and with laboratory analysis at WHOI. In April 2014, Caitlin presented research conducted within this project to fulfill her requirement for a senior seminar presentation at USD.

Mr. Javar Henry, now graduated from Savannah State University (WHOI Guest Student). He assisted with laboratory analysis (measurement of coral spat, lipid extractions). In February 2013, Javar presented work related to this project as a poster at the ASLO Aquatic Sciences Meeting.

Presentations at Scientific Meetings:

Tarrant AM, McCorkle DC, dePutron SJ, Church C, Henry J, Cohen AL. 2013. Variation in size of juvenile corals and sensitivity to ocean acidification. Society of Integrative and Comparative Biology (SICB) Annual Meeting, San Francisco, CA, January 3-7, 2013. Abstract 695-109128. (Platform presentation)

Henry JE, **Tarrant AM**, dePutron SJ, McCorkle DC, Church C, Cohen AL. 2013. Maternal effects on skeletal size and sensitivity to ocean acidification in juvenile corals. ASLO Aquatic Sciences Meeting, New Orleans, LA, February 17-22, 2013. Abstract 416. (Poster presentation)

Other presentations related to this project:

Ocean Life Institute Committee. October 11, 2013

Children's School of Science, Woods Hole MA July 9, 2013.

WHOI Journalism Fellows. September 12, 2012.

Seminar at the Bermuda Institute of Ocean Sciences. July 30, 2012.

Sigma Xi Induction Ceremony, Keynote address. North Shore Chapter, Endicott College. April 26, 2012.

Future Plans

We are currently preparing a manuscript describing our results. We plan to submit this paper as part of a themed article set within the ICES Journal of Marine Science titled: *Towards a broader perspective on ocean acidification research - jumping off the bandwagon*. A specific focus of this article set is "Studies that emphasize the variability of responses to OA, including individuals that are not affected by the OA treatment, and how selection might act on that variability." I anticipate that this OLI-supported project will be an important contribution to this issue, and that the themed collection will provide good visibility for the work. Submissions are due in January 2015.

Along with collaborators, I plan to submit a pre-proposal to the Division of Environmental Biology (DEB, part of the Biological Sciences directorate at NSF, the National Science Foundation) in January 2015. We would propose continued development of sensitive genetic markers for parentage analysis and use these markers to explore the effects of inbreeding and reproductive mode (self- and cross-fertilization) on the fitness of juvenile corals and their ability to respond to stressors.