Examining the efficacy of *Diadema antillarum* enhancement for restoration of coral reefs in the Florida Keys

"Protect Our Reef" Grant -Interim Report (August 16, 2006 to March 1, 2007)

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Project Justification

Prior to their region-wide die-off in 1983, the long-spined sea urchin *Diadema antillarum* was a keystone species on Caribbean coral reefs, credited with maintaining low algal cover and high coral cover, even in locations where overfishing had depleted herbivorous fish populations. Their mass mortality was one of the major causes of a phase shift from coral-dominated to algal-dominated communities on many Caribbean reefs (Hughes 1994). More than ten years after the mass mortality, *D. antillarum* densities were still extremely low in most locations, and in many locations densities remain low. However, *D. antillarum* densities have been increasing in recent years in some locations in the Caribbean (e.g., St. Croix, USVI (Miller et al. 2003); Jamaica (Edmunds and Carpenter 2001)), with concurrent increases in coral recruitment and decreases in algal cover. For unknown reasons, *D. antillarum* densities have not increased in the Florida Keys. Given the importance of *D. antillarum* as a keystone species on coral reefs, and the economic and ecological importance of coral reefs to Florida, it is imperative to understand the factors affecting *D. antillarum* in the Florida Keys. This understanding will contribute toward a longer term goal of facilitating the return of *D. antillarum* to Florida Keys reefs as an essential component of coral reef restoration. This research proposal requests funds for the second year of a multi-year project to address the ecology and restoration of *D. antillarum* in the Florida Keys.

Florida’s barrier coral reef, the third largest in the world, requires large scale ecological restoration. And since “reef restoration is a fertile field of study necessary to determine effective and efficient ways to restore degraded coral reef ecosystems” (NOAA 2005), aiding the recovery of *D. antillarum* has the potential to contribute to the ecological restoration of our coral reefs. This proposed research is a continuation of an ongoing and long-term experimental, step-wise approach to understanding the ecology of *D. antillarum* in the Florida Keys, determining the importance of *D. antillarum* to recovery of Florida Keys coral reefs, and examining the efficacy of enhancement as a tool to increase *D. antillarum* abundance and contribute to coral reef restoration. This proposal requests funds for field and culture research for 2007. This will support the second year of this ongoing and long-term project. Future years will see the addition of components to address conservation and education.

The broad goals of this research program will be achieved via a two-pronged approach. First, in situ monitoring and experimentation with wild *D. antillarum* will be conducted to examine ecological aspects of *D. antillarum*’s lack of recovery in the Florida Keys relative to
many Caribbean locations. Such an understanding is essential prior to any restorative efforts involving *D. antillarum*. Second, lab-based experimental and pilot-scale research trials will continue the development of aquaculture techniques to rear *D. antillarum* for stock enhancement trials on coral reef sites in the Florida Keys.

**Goals**

Based upon previous research in the Caribbean and Florida Keys, this study was undertaken to determine the extent that recovery of *Diadema antillarum* would positively affect Florida Keys coral reefs. And if so, how might a stock enhancement program be conducted to assist *D. antillarum* recovery. The specific objectives of the research are to: 1) test in the Florida Keys the findings of Caribbean-based research showing *D. antillarum* densities are negatively correlated with algal cover, and positively correlated with coral cover; (2) determine whether juvenile *D. antillarum* survival increases with increasing adult *D. antillarum* density; (3) determine whether *D. antillarum* density influences abundance of juvenile herbivorous fishes; and (4) develop and experimentally refine procedures for rearing *D. antillarum* for use in experimental enhancement research. The results of this research are prerequisite to determining causes of continued low *D. antillarum* densities in the Keys and testing the efficacy of a stock enhancement recovery program.

**Field Research Component**

**Task 1, experimental reef construction:** A joint permit was acquired from Florida Fish and Wildlife Conservation Commission and from Florida Keys National Marine Sanctuary for deploying the live-rock reef materials and for conducting visual surveys on the reef. After much consultation, it was determined that a permit from the Army Corps of Engineers was not needed. In July 2006, a contractor was hired to deploy 20 Supersacks of limestone rock to the study site. This rock was used as a base layer for each reef. Also in July 2006, SCUBA divers placed the rock in equal piles in the assigned 10 x 3 reef configuration in 8m depth. The divers then harvested seasoned live-rock from a nearby lease site, owned by Ken Nedimyer, transported the live-rock to the study site, and placed it on the experimental reefs. The reefs were allowed to season for two months.

**Objectives 1, 2, 3:** In September 2006, adult and juvenile *D. antillarum* were stocked on the reefs in pre-determined densities: 0, 1, and 4 adults per reef; 0 and 3 juveniles per reef (a figure showing the reef-specific urchin densities is in the appendix). Prior to stocking urchins on the reefs, the first fish censuses, and algal and coral cover assessments were completed. During this first census, numerous potential predators were recorded: four species each of Haemulidae, Lutjanidae, and Labridae, and one species each of Balistidae and Palinuridae. Few potential competitors (two species of Scaridae) were recorded. The benthic cover was dominated by turf algae.

The first monitoring event occurred on January 3, 2007. Fish censuses and estimates of percent benthic cover were completed for each of the 30 experimental live rock reefs. During this first monitoring event, it was noted that 100% of the wild *D. antillarum* stocked on the reefs were gone. It was not possible to determine whether they had died, been preyed upon, or emigrated from the site. No test or spine remains were observed. After this monitoring event, it was determined that the originally planned experimental strategy should be postponed, with all
efforts first being dedicated to determining the causes of 100% *D. antillarum* loss in three months.

A new experiment was designed to determine the cause of *D. antillarum* loss from the experimental reefs, planned to begin January 21, 2007. Five *D. antillarum* would be stocked on each reef on Project Day 1. The reefs would be checked, and *D. antillarum* counted, one, two, four, seven, 10, and 30 days after the stocking. This would allow a measure of short-term and longer-term loss of *D. antillarum* from the reefs, and estimates of the proportional contribution of predation, mortality, and emigration. Due to inclement weather, this experiment was postponed, and will take place beginning March 13, 2007.

**Experimental Aquaculture Research (Objective 4)**

*Methods*

The three phases proposed to be accomplished under this objective included developing the spawning and larval rearing techniques for small-scale production (SSP), developing the technology and facilities for pilot-scale production (PSP) and developing the technology and facilities for large-scale production (LSP). During the first year of this project, a culture facility was established for larval rearing at the SSP on Lower Matecumbe Key, the DACS culture permit was acquired, culture systems were tested, Diadema broodstock were collected, and three spawning trials were conducted. The location for the PSP culture system at Mote’s Tropical Research Lab (TRL) was identified and the area was prepared for raceways to be established. The location and tanks for the LSP macroalgae culture system were identified.

*Findings to Date*

**Small-Scale Production (SSP) Facility Construction:** The SSP *Diadema* culture facility was planned and constructed in a 25 by 16 foot concrete room. The culture laboratory includes 5 indoor systems and an outdoor system. The indoor facility and systems include two urchin broodstock systems (five tanks) (Figure 1), a larval culture and experimental system (17 tanks ranging from 8-55 gallons) (Figure 2), two 300 gallon growout systems (two 150 gallon tanks) and laboratory support for microalgae culture (Figure 3). Each of the 5 systems has an independent water system. All are gravity flow after pumping and three have surge tanks to provide ample water movement. All tanks have individual lighting to provide natural spectrum illumination, and individual tank aeration, which although not necessary for normal life support, functions to maintain oxygenated water during power outages. A new upwelling system has been constructed for larval culture (Figure 4). A water system was designed and installed to provide sterile, temperature and salinity adjusted salt water for the indoor culture lab and with exception of temperature control, for the outdoor culture facility as well. The outside culture facility includes four 150 gallon tanks, 4 Kalwell algae culture tubes, and a variable number of smaller tanks. For the *Diadema* projects, this facility is used to hold algae collected for adult and juvenile food for a week or two to eliminate more frequent collecting trips for *Hypnea* and turtle grass and to culture larger quantities of algae for feeding of *Diadema* larvae. These facilities were permitted for urchin aquaculture by the Florida Department of Agriculture in July 2006.

**SSP Culture Research:** The SSP lab has maintained and grown about 20 *Diadema* broodstock from about June through to early December of 2006. Additional broodstock were obtained in December of 2006 to compare gonadal development of urchins from the reef with
those that have been under culture for six months. Spawning was obtained from both groups. The techniques and material required for maintenance and growth of juvenile and adult *Diadema* in this facility have been well established. Tom Capo has visited and consulted on spawning techniques for *Diadema*. Three spawning attempts have been conducted. The first spawning attempt (08/10/06) did not result in the release of any gametes, probably because the urchins were not reproductively mature. The second spawning attempt (11/28/06) resulted in the release of sperm by 6 males; however, it appeared that none of the females were mature enough to produce eggs. *Diadema* cannot be sexually differentiated until spawning is observed. This may be because many of the urchins were juveniles at the time of capture and needed time to become reproductively active adults. The third spawning attempt (12/18/06) was successful and eggs were obtained from two females and sperm from 6 males (Figures 5 & 6). The eggs were fertilized successfully and development has proceeded normally, and the larvae lived for 7 days (through the four arm pluteus stage – Figure 7). The larvae of *Diadema* are similar to other sea urchin larvae and that they do not suspend in the water column without sufficient water motion. This rearing trial demonstrated that aeration in an aquarium is not adequate to suspend and sustain this species through larval development. Rearing sea urchins, however, is not an unknown science. It has been done in many instances for commercial, educational, conservation, and restoration purposes and *Diadema*, although smaller and of longer larval life than most species, can certainly be reared through the larval stage in large numbers. Two methods that have been employed successfully in sea urchin larval culture are upwelling systems and paddle systems, both supply the gentle agitation necessary to suspend the larvae. The upwelling system will be used during the next spawning trial.

**Pilot Scale Production (PSP) Facility Construction:** The PSP has been constructed at the Mote Tropical Research Laboratory in Summerland Key. It consists of five 50 gallon culture tubes, four of which are set up for phytoplankton culture with smaller inoculation flasks (Figure 8). One or two of these culture tubes will be used for larval production tests. In addition shallow and deep raceways have been established to handle nursery production trials. Additional outside tanks have been set up for macro-algae production and larger phytoplankton and larval trails (Figure 9). This pilot scale production facility is flexible enough to handle small to medium spawns, micro and macro algal feed production and nursery raceways. This facility is now already past phase II construction and development and will focus on production and operation in 2007. Dave Vaughan will focus on operation of the Pilot Scale Production Facility and will assist the Small Scale and Large Scale production facilities with information and assistance in micro and macro algal production technologies and design.
Figure 1. One of two SSP broodstock tanks.

Figure 2. SSP larval rearing tank systems.

Figure 3. SSP microalgae culture lab.

Figure 4. SSP larval rearing upwelling system.

Figure 5. Removal of gametes from broodstock.

Figure 6. *Diadema* eggs.

Figure 7. Pluteus larva.
Figure 8. PSP Indoor algal culture system.

Figure 9. PSP Outdoor macroalgae culture tanks.