Quicklook Report Coral Spawning 2021: Activities and Observations

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Summary:

Spawning observations and gamete collections were made for the coral species *Diploria labyrinthiformis* (DLAB) in May, *Acropora palmata* (APAL) and *A. cervicornis* (ACER) in July, and *Orbicella faveolata* (OFAV) and *Montastraea cavernosa* (MCAV) in August of 2021. Observations of *Pseudodiploria strigosa* (PSTR), *Siderastrea siderea* (SSID), and *Eusmilia fastigiata* (EFAS) were made on one night in September of 2021 with no spawning activity observed. Batch cultures were made of all species except for *M. cavernosa*. Reciprocal egg-sperm crosses of *A. palmata* and *A. cervicornis* were done to yield both *A. prolifera* (APRO) morphotypes (bushy and palmate). Larvae were reared at the Coral Research and Assessment Lab (CoRAL) Nursery in Miami, Florida, and settled onto artificial substrates where they will be raised for future research and restoration projects. Additionally, in partnership with the University of Miami Rosenstiel School of Marine and Atmospheric Science (UM RSMAS) Coral Futures Lab, sperm was cryopreserved from DLAB, APAL and OFAV. Larvae from batch cultures were provided to other research and restoration partners including the Coral Futures Lab at the University of Miami Rosenstiel School of Marine and Atmospheric Science (UM RSMAS), SECORE, the University of Southern California (USC), Mote Marine Lab, and Florida International University (FIU).

Introduction:

The Southeast Fisheries Science Center (SEFSC) CoRAL team has observed spawning patterns and collected gametes from APAL and OFAV in the upper Florida Keys for the past two decades. Historically, our team has not been able to house coral recruits at the SEFSC Miami Lab. Instead, we have typically reared the larvae and maintained newly settled recruits in a temporary seawater system at a rental space in Key Largo, Florida, where they were either immediately transported and released to the reef or were provided to various research agencies and universities. In 2021, the CoRAL team began expanding capabilities to maintain



Figure 1. New seawater system with kreisels holding coral embryos.

recruits for grow-out in a land-based nursery at UM's Experimental Hatchery where the recruits can either be transferred to a seawater system capable of independent experimental replication or maintained in grow-out tanks until they are suitable for outplanting to the reef. This year, we were able to rear larvae and settle recruits in one 80 gallon and two 120-gallon seawater systems (Fig. 1) where they will be maintained for grow-out.

Species Observed:

Diploria labyrinthiformis (DLAB)

In collaboration with the Coral Restoration Foundation (CRF), we have monitored DLAB colonies for spawning activity on an unnamed patch reef in the upper Florida Keys since 2020. In 2020, spawning was not observed during the five months in which observations were made (May - September). On the first night that observations were made in 2021 (May 6th), a large spawning event was observed among DLAB colonies at that site. Spawning nets were placed on ~21 colonies, and spawn was collected from 18 of those colonies. Parent colonies ranged in size from 18cm to 70cm (maximum diameter). Spawning occurred at approximately 18:15, which is the early end of the predicted window. The colonies were watched again on May 7th, but spawning was not observed. Due to the large volume of gametes collected and the large proportion of colonies observed to spawn on May 6th, spawning was considered to be complete and no further observations for DLAB were made in 2021.

A batch culture of gametes from the 18 parents that spawned on May 6th was mixed on the boat. The bundles broke up allowing fertilization to begin while we were still on site. Aliquots of eggs and sperm were placed in flasks of seawater for transport to Miami, FL. A portion of sperm was reserved for cryopreservation, and the remainder of the gametes were poured back into the water immediately before leaving the site. Because fertilization had likely occurred on the boat, releasing the embryos likely resulted in a greater number of larvae being produced than would have occurred naturally at that site. The embryos transported to Miami appeared to have ~90% fertilization. The embryos were placed into kreisels in our seawater system for the larval phase, where they were also settled on ceramic plugs, and in our seawater system for ongoing maintenance.

Acropora palmata (APAL) & A. cervicornis (ACER)

Due to the timing of the full moon, it was difficult to anticipate the spawning windows for 2021. Typically, the spawning window for APAL and ACER occurs in August; however, observations were made following the July full moon, during which spawning occurred on multiple nights (<u>Table 1</u>). On night 2 following the July full moon, we collected spawn from both wild and outplanted APAL colonies at North Dry Rocks Reef and outplanted ACER colonies. Batch cultures of both species were made. Additionally, eggs and sperm from each



Figure 2. *Acropora palmata* colonies (outplanted in 2016) with spawn collectors.

species were separated on the boat so that APAL eggs and ACER sperm could be mixed to produce a batch of the 'palmate' morphotype of APRO (APRO-P). Similarly, ACER eggs and APAL sperm were mixed to produce a batch of the bushy morphotype of APRO (APRO-C). Fertilization of APRO-P was high (90%) while fertilization of APRO-C was lower (sub-batches ranged from 15-50%). Resulting larvae were placed into kreisels for rearing and settlement at the CoRAL team facility in Miami, Florida.

On night 3, we moved to Elbow Reef to monitor nine wild APAL genets and eight restored APAL genets (Fig. 2) that were outplanted in 2016. This was the first year these outplants had been monitored for spawning as most were not large enough to be expected to spawn prior to this year. Spawning was not observed on night 3 at Elbow Reef. On night 4, two wild genets and one outplanted genet of APAL were observed 'dribbling' a small number of gametes. Gametes from two APAL genets (CN2 & orange) were used to make a small batch of larvae. Night 5 after the full moon (AFM) at Elbow, 9 out of 11 wild APAL genets and 5 out of 8 outplanted APAL genets were observed to spawn. Some wild genets only partially spawned, yielding smaller volumes of gametes. One wild genet that has never been observed to spawn produced bundles this year. Observations were not possible during the window following the August full moon due to high winds and rough seas.

Batches of gametes were mixed on the boat and sperm was reserved for cryopreservation and a large volume of gametes/embryos was released back into the water before leaving the site. The night 5 spawning event was one of the larger events we have observed among the wild genotypes at that site, and the first ever observed among the outplants at that site. It was, by far, the greatest number of genets we have used to create a batch of larvae. Approximately 60 vials/batches of sperm were cryopreserved by the UM Coral Futures Lab (Liv Williamson). Embryo cultures were transported to Miami where they completed the fertilization process (90% fertilization was achieved). A large volume (68ml @ 90% fertilization = ~306,000 larvae) was given to SECORE and the remainder were placed into kreisels in our seawater system.

Orbicella faveolata (OFAV) & Montastraea cavernosa (MCAV)

OFAV and MCAV were observed at Horseshoe Reef (north mooring) on night 6 and 7 after the August full moon. The colonies at this site have not been genotyped, however, based on past observations and collections, there are numerous unique genotypes of OFAV. Gametes were collected from eight OFAV colonies on night 6 and dribbles from one colony on night 7. Gametes from all parents were mixed to create a batch culture of embryos on night 6 (Aug 28; Fig. 3), and a portion of the embryos along with uncrossed gametes were provided to researchers at USC. On night 7 (Aug 29), only dribbles of gametes from a single colony were collected. Researchers from USC were able to collect a similar amount from one OFAV parent at Cheeca Rocks and were able to cross the two parents to make a batch for research purposes.



Figure 3. Estimated 100,000 *Orbicella faveolata* embryos.

The CoRAL team's batch culture from night 6 had high fertilization rates (95%). The larvae were maintained and settled onto ceramic plugs (Fig. 4) within kreisels in our seawater system. Following settlement, the recruits were transferred from the kreisels and placed in the open raceways in our seawater system where they continue to be maintained.

Other Species

On night 9 after the August full moon, we monitored several (~ 8) *Pseudodiploria strigosa* (PSTR) colonies, numerous (20+) *Siderastrea siderea* (SSID) colonies, two *Colpophyllia natans* (CNAT) colonies and a single colony of *Eusmilia fastigiata* (EFAS) at the unnamed patch reef where DLAB spawn was collected in May 2021. Monitoring occurred from sunset until midnight with breaks to swap tanks, and no spawning was observed. EFAS eggs were observed in the tentacles of one colony, however we did not witness their release.



Figure 4. Settlement substrate with numerous 2 month old *Orbicella faveolata* recruits.

Collaborations

Our coral spawning collections were shared with multiple research and restorations groups listed below.

University of Miami (Coral Futures Lab):

1500 DLAB settled recruits, 48 aliquots of DLAB sperm 200 APAL settled recruits, 108 aliquots of APAL sperm 200 ACER settled recruits 200 APRO settled recruits 20,000 OFAV larvae, 48 aliquots of OFAV sperm

SECORE (M. Miller) 300,000 APAL larvae 1,000,000 OFAV larvae

University of Southern California (C. Kenkel): 10-15ml of OFAV gametes, 30,000 OFAV larvae

Mote Marine Lab (H. Koch): 47,000 OFAV larvae

Nova SE (J. Figueirdo): 20,000 OFAV larvae

Acknowledgements

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Table 1. Spawning observations for 2021

	dAFM	Reef Site	Wild/ Outplant	Species Observed	Spawning		# genets			
Date					Observed	Collected	Monitored	Spawned	Larvae produced	Notes
7/25	2	North Dry Rocks	wild	A. palmata	Yes	Yes	3	1	- 50,000	2 of 5 colonies
			outplant	A. palmata	Yes	Yes	4	2		~25% spawned
			outplant	A. cervicornis	Yes	Yes	unkn	unkn	14,250	~10% spawned
				A. prolifera					61,500	
7/26	3	Elbow	wild	A. palmata	No	No	9	0		
			outplant		No	No	8	0		
7/27	4	Elbow	wild	A. palmata	Yes	Yes	9	1	- 5,000	small dribbles
			outplant		Yes	Yes	8	2		from 3 genets
7/28	5	Elbow	wild	A. palmata	Yes	Yes	9	8	319,500	
			outplant		Yes	Yes	8	5		
8/28	6	Horseshoe	wild	O. faveolata	Yes	Yes	15	8	1,539,000	
8/29	7 Horsoshaa	wild	O. faveolata	Yes	Yes	15	1			
		norseshoe	wild	M. cavernosa	Yes	Yes	5	1		
8/31	9	9 "DLAB Site"	wild	P. strigosa	No	No	8	0		
			wild	S. siderea	No	No	20+	0		
			wild	E. fastigiata	No*	No	1	0		
			wild	C. natans	No	No	2	0		

*dAFM is the number of days after the full moon.