Progress Report, September 2014

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Passive Acoustic Monitoring of Fish Spawning Aggregations in Puerto Rico

Background and Activity Summary

Large predators are important in healthy coral reef ecosystems. However, many populations of these species are depleted on the insular shelves of the U.S. Caribbean. Commercially important species, *e.g.*, large groupers, that gather annually to spawn, are highly susceptible to overfishing, and managers need better data on aggregation locations and dynamics. This project, in partnership with the University of Puerto Rico Mayagüez (UPRM) and the Caribbean Coral Reef Institute (CCRI), is supporting continued use of passive acoustic methods off western Puerto Rico, to locate, assess, and monitor reef fish spawning aggregations (FSAs). Over the next three years, we will also apply these research techniques in the NE Reserves of Puerto Rico, confirming spawning locations, species, aggregation timing, and species abundances.

Field Activities from October 2013-September 2014:

1. Completed deployment of digital spectrogram (DSG) long-term acoustic recorders (Loggerhead Instruments Inc.) for passive acoustic monitoring at each study site (presumed pre-spawning)
2. Completed recovery of DSGs from each study site (presumed post-spawning), except one (MI) to allow extended (year-long) recording
3. DSG data downloaded, processed, and backed up in UPRM (Appeldoorn) lab
4. DSG data analysis underway (project-supported UPRM grad student)
5. Underwater visual surveys for species, abundance, size structure, and spawning behavior completed
6. Began collection of information on NE spawning sites and outlined methods to proceed with more comprehensive information gathering

Project Basics

Puerto Rico's priority coral reef management goals include - *Protect coral reef ecosystems from large- and small-scale fisheries impacts* – and – *Enhance management programs to reduce fishing impacts to coral reef ecosystems*. These needs are especially keen in marine protected areas (MPAs) and depend on robust knowledge of reef fish populations. Many commercial fishery stocks on the insular shelves of the U.S. Caribbean, including large predators such as groupers that contribute ecological resistance and resilience in healthy coral reef ecosystems, are depleted or are near depletion. Annual aggregative spawning behaviors have made them easy to overfish. Managers know far less about these sites and their status than fishers, who have exploited these aggregations for decades, even to the point of fishing-out aggregations. To enhance management, we need better data on site locations and dynamics. We need answers to key questions:

- Where and when do aggregations occur?
- Which species use each location?
- How abundant are spawners during the aggregation?
- Can abundance at aggregations be used as an indicator of population abundance at resident reefs?

For successful management, effective conservation, and to recover these critical reef resources, spatial boundaries and seasonal fishing regulations must match the timing and locations of FSAs to reduce the threat of overexploitation. Prior attempts to locate and monitor aggregations were highly dependent on fishermen interviews and diver or other visual surveys, which have limited spatial coverage and depend on good weather and adequate light. The current research project continues and expands the successful use of passive acoustic monitoring, in combination with diver surveys for groundtruthing, in order to quantify baseline characteristics at known and reported FSA sites off western PR. Passive acoustic receivers (*i.e.*, DSGs) record, and researchers identify, species-specific "calls" of fish at aggregation sites allowing continuous high temporal resolution monitoring. Calls of five grouper species have been differentiated and the "vocalizations" generally indicate courtship and spawning behavior. We are also working on protocols to extrapolate sound levels to numbers of spawning individuals. In cooperation with partners, SERO, CCRI, University of Puerto Rico-Mayagüez (UPRM), and Loggerhead Instruments, we will transfer techniques proven on red hind (*Epinephelus guttatus*), Nassau (*E. striatus*), and yellowfin (*Mycteroperca venenosa*) grouper aggregations at Mona (MI), Bajo de Sico (BDS) and Abrir la Sierra

(ALS) to sites off northeastern (NE) Puerto Rico. Purported sites will be surveyed for active aggregations and acoustics will be used to locate and define temporal and spatial boundaries. Coordinated companion projects are ongoing with UVI (R. Nemeth) and PR DNER (SEAMAP-C) and additional funding sources are continuously pursued to expand and extend this research across the U.S. Caribbean.

Monitoring and Research Setting

Fish spawning aggregation sites in the waters off western Puerto Rico have been studied with diverse, constantly improving methodologies for almost a decade leading towards standardization of methods. We are supporting continuation and adaptation of that work. Sites (Fig. 1), including known FSAs at Abrir la Sierra (ALS), Mona Island (MI) and Monito, Bajo de Sico (BDS), and Tourmaline (B-8) are monitored with passive acoustics, underwater visual surveys, and autonomous audio/video recording methods. Two of these locations (ALS and MI) have been monitored with passive acoustic methods continuously since 2007. Four FSA sites (MI, ALS, B-8, plus El Seco (off Vieques on the PR east coast)) were determined to be priority sites for red hind and data are being collected in collaboration with/on behalf of SEAMAP-C. BDS is known as a multi-species spawning aggregation site (Schärer et al. 2014b) and is also considered a priority site for Nassau grouper, which has recently been listed as *threatened* under the Endangered Species Act. An overlapping research project has been funded by the CFMC (Schärer). Several of the FSA study sites have been documented to be used by multiple species of groupers, snappers, and parrotfishes. El Seco (ES), off of Vieques, PR, the Hind Bank, and the Grammanik Bank, both off St. Thomas, USVI, are being investigated, as well, using consistent technological approaches, with logistics handled collaboratively by Dr. Rick Nemeth (UVI) but data archiving and analysis by UPRM.

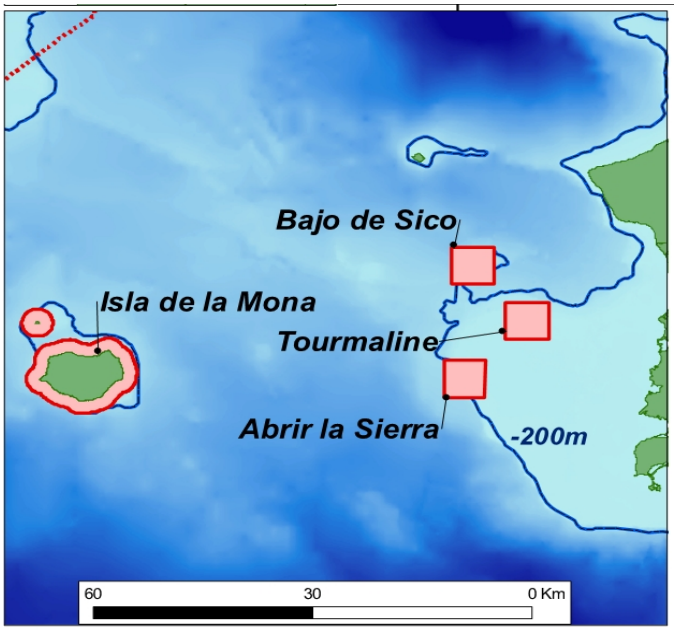

Figure 1. Map of western Puerto Rico fish spawning aggregation sites currently being researched and monitored. Bathymetry is color shaded and shelf edge is marked with 200 m depth contour.

Passive acoustic monitoring is based on recording, identifying, and quantifying sounds produced by groupers during reproductive displays, *i.e.*, courtship and

spawning. As far as we can discern, these sounds are almost exclusively associated with behaviors exhibited during aggregative spawning, *e.g.*, spawning migrations, territorial establishment and defense, mate attraction and defense, courtship, and spawning. During earlier phases of this research, sound signals (frequencies and patterns) were confirmed for red hind, Nassau, yellowfin, and black grouper (*M. bonaci*) (Mann et al. 2010, Schärer et al. 2012a, Schärer et al. 2012b, Schärer et al. 2014a). The species-specific sounds serve to document the presence at spawning aggregation sites and for the most abundant species a high-resolution (hourly) time series can be generated to determine peaks in reproductive activity. Other species such as *M. tigris* (tiger grouper), *M. interstitialis* (yellowmouth grouper), *Scarus guacamaia* (rainbow parrotfish), and *Canthidermis sufflamen* (ocean triggerfish) have also been documented at spawning aggregations with reproductive behaviors, but sound identification has not yet been confirmed. This year, long-term passive acoustic monitoring was conducted at multiple sites simultaneously and underwater visual surveys were conducted at two priority sites off western Puerto Rico.

Activities and Accomplishments for FY-14

Preparations for fieldwork were begun during November of 2013. This included meetings with partner institutions to identify priority locations and seasons for study, conditioning and staging equipment, and acquisition of materials. Study sites selected for the season (2013-14) were visited prior to the beginning of the expected spawning season of most groupers (December). At each location at least one digital spectrogram long-term acoustic recorder (DSG; Loggerhead Instruments Inc.) was deployed. At two important multi-species FSA sites, MI and BDS, more than one DSG were deployed in order to record at different locations where different species are known to congregate.

Each DSG was programmed to record continuously for at least six months with a sampling schedule of 20 seconds every 5 minutes. Data were recorded onto removable

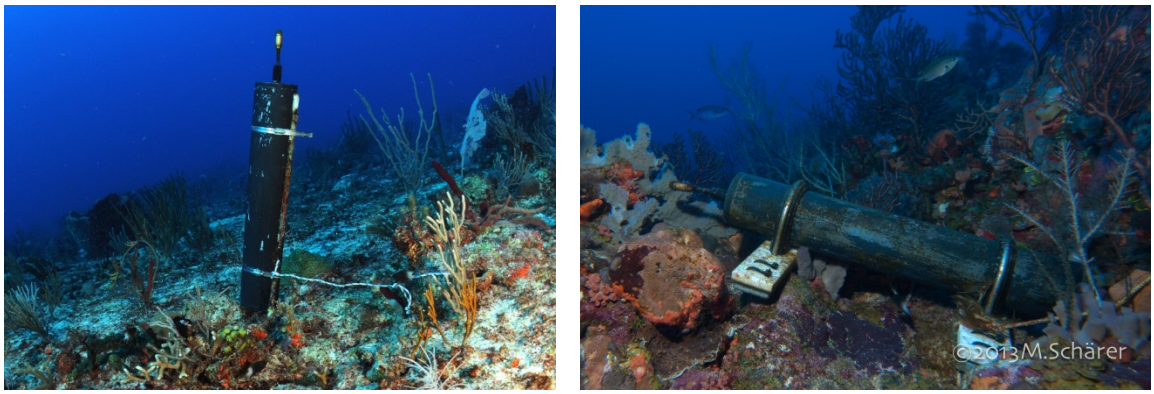

Figure 2. Deployment methods for DSGs on the seafloor, vertical rebar attachment at left (photo by J. Zegarra), horizontal weighted base attachment at right (photo by M. Schärer).

32-gigabyte secure digital high capacity (SDHC) flash memory cards. Files were digitized at a sample rate of 10 KHz. Units were powered by an array of 24 D-cell batteries for the duration of the deployment. DSG recorders were attached within the aggregation sites either with hose clamps to rebar inserted into the substrate or secured to weighted bases (Fig. 2).

The deployment schedule of the DSGs is summarized in Table 1. After deployment the DSGs were inspected for proper functioning whenever diver surveys were conducted at the site by observing the LED lights on the tip of the unit's hydrophone *in situ*. Only one DSG, at BDS-1.5, wasn't functioning properly, as indicated by a continuously lit, rather than a blinking, LED on the hydrophone. This DSG was recovered on 25-Feb-2014 and another unit was deployed there on 27-Feb-2014. The recorder had malfunctioned due to a faulty SD card (128 GB capacity) and did not record properly. Standard method now prescribes 32 GB cards for all units.

Table 1. Deployment and recording schedule for DSG units during the 2013-14 season.

Site	Unit #	Dates Recording	Date Deployed
Abrir la Sierra	1138	13-Dec-13 to 30-June-14	18-Dec-13
BDS-00	1018	3-Jan-14 to 26-July-14	3-Jan-14
BDS-1.5	1289	27-Feb-14 to 31-Aug-14	28-Feb-14
BDS-1.5	1275	03-Dec-13 to 31-May-14	18-Dec-13
BDS -01	1274	03-Dec-13 to 31-May-14	18-Dec-13
BDS -09	1014	13-Dec-13 to 31-July-14	18-Dec-13
BDS-2.5	57	13-Dec-13 to 31-July-14	18-Dec-13
El Seco	1288	2-Feb-2014 to 31-Aug-2014	15-Mar-14
Mona-Elbow	1273	03-Dec-13 to 31-May-14	27-Dec-13
Mona-H5	1017	25-Dec-13 to 6-May-14	27-Dec-13
Mona-H6	55	25-Dec-13 to 6-May-14	27-Dec-13
Monito	1276	03-Dec-13 to 31-May-14	27-Dec-13
Tourmaline	1137	13-Dec-13 to 30-June-14	18-Dec-13

Surveys to document the species present and relative abundances were conducted at two priority sites, BDS and ALS. The multi-species FSA site at BDS was visited repeatedly throughout the spawning season but ALS only on the day when peak spawning was expected for red hind. Surveys at BDS were conducted by UPR divers with closed circuit rebreather (CCR) technology due to the depths at which the groupers

aggregate (100' to 170'), while the surveys at ALS were conducted with open circuit SCUBA diving (60' to 90' depth). Three types of survey methodologies were employed at BDS, one with video and laser calipers (Fig. 3), a second counting the maximum abundance of groupers by species, and a third on peak spawning days to document the different color-phases of Nassau grouper (BDS only).

Underwater visual surveys to estimate abundance and size structure are conducted with complementary methods. One diver carries a slate and records all grouper species seen within a drifting belt transect, recording species, estimated size, and time, synchronized with a GPS unit towed on a dive flag at the surface. The second diver conducts video and laser caliper surveys (*GoPro® camera and underwater lasers attached) as the pair drifts unidirectionally through the aggregation site (Fig. 3).

Lasers are set to a calibrated width (20 cm). Each grouper encountered during the dive is recorded on video and the lights of the parallel red lasers are pointed at and, ideally, held on the side of the fish at a perpendicular angle. Frames of each grouper are extracted from the video, processed to get still images, and then analyzed with Adobe Acrobat Measure Tool to estimate the size of each individual. Length frequency distributions are generated for each species.

Figure 3. Video and laser caliper method at left (photo by M. Schärer) and lights from red laser on Nassau grouper at right (photo by H. J. Ruiz).

The density of each species is calculated for each transect survey performed through the FSA. The length of the drift transect over which groupers were surveyed is estimated by calculating the distance based on the geographic coordinates of the track recorded on the handheld GPS unit attached to the surface buoy towed by the diver. The length of each transect is multiplied by the estimated transect width, *e.g.*, 10m, to calculate the total area surveyed. Surveys also provide the relative spatial locations of

* Mention of any trade name or product should not be considered an endorsement by NOAA, NMFS or UPR.

groupers for mapping by extracting the timecode from the video and matching with the respective coordinates.

The remaining two types of count surveys were limited to a 300m by 100m area, defined as the courtship arena where DSGs are located. Due to the geomorphology of the site and strong currents that impede any type of belt transect or cylinder area estimate a roving search method is employed. Surveys were completed according to the schedule in Table 2.

Table 2. Number and type of underwater surveys conducted by divers

Date	Site	Color phases	Max count
22-Jan-14	BDS	2	
23-Jan-14	BDS	2	
24-Jan-14	BDS	1	
25-Jan-14	BDS	1	
27-Jan-14	BDS		1
28-Jan-14	BDS	1	1
23-Feb-14	BDS	1	
25-Feb-14	BDS	1	
28-Feb-14	BDS	1	
2-Mar-14	BDS		1
23-Mar-14	BDS	1	
25-Mar-14	BDS	1	
26-Mar-14	BDS	2	
26-Mar-14	ALS		1
30-Mar-14	BDS	1	
Total		**15**	**4**

Laboratory Analysis and Planning

All DSGs deployed during the 2013-2014 spawning season have been recovered and data from data cards were processed and have been backed up (Appendix-Table 3). These data, in excess of 256 gigabytes (GB), are currently being verified for recording accuracy and analyzed to quantify the number of grouper calls per species for the whole season. Figure 4 displays two examples of the digital sound signals derived from DSG recordings of black grouper and red hind. Data from videos and counts from underwater surveys will be processed and analyzed in order to get size and abundance estimates for each day surveyed. Finally the sound and abundance datasets will be

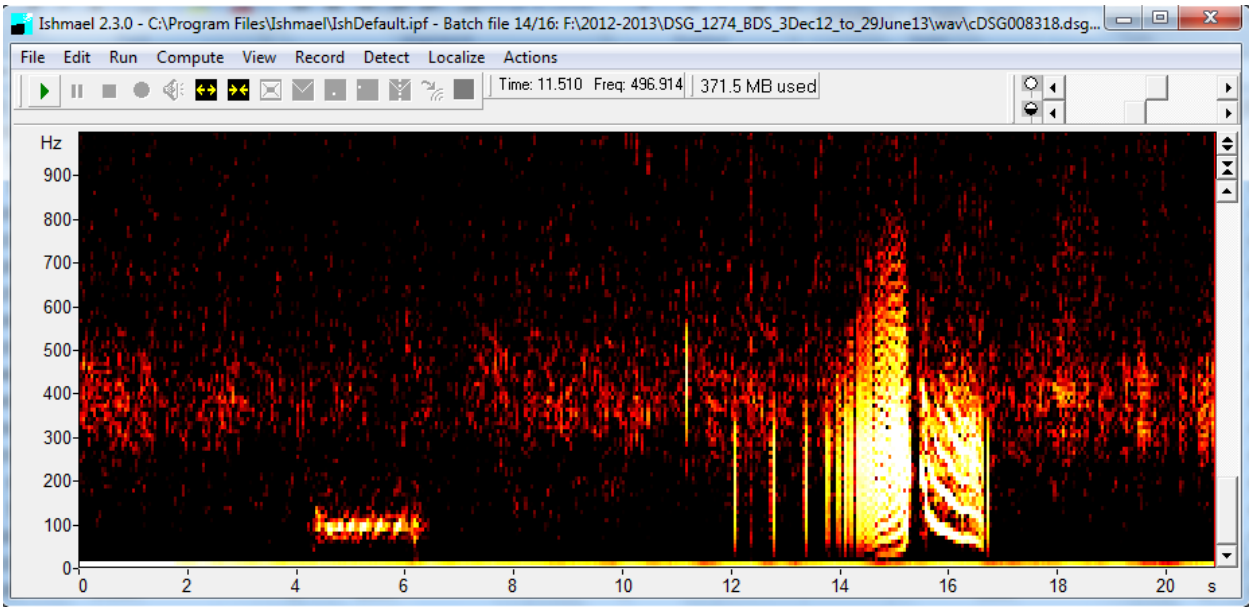

Figure 4. Digital spectrogram of grouper sounds from DSG deployment at BDS. Sound between 4-6 seconds is from a black grouper, sound between 12 and 17 seconds is from red hind (image courtesy of K. Clouse).

correlated in order to determine the patterns of temporal dynamics of the 2013-14 spawning season for groupers. In November 2014 preparations will begin to deploy DSGs and determine the survey schedule for the upcoming 2014-15 spawning season.

Fisher Input

A number of contacts have been made with fishers and divers familiar with purported spawning aggregation sites in the NE Reserves. Data from these participants are being compiled and will be examined to examine appropriate study sites going into next season's surveys. Prior data from Ojeda et al. 2007 (example, Fig. 5) are being collected into a comprehensive database of likely locations for surveys.

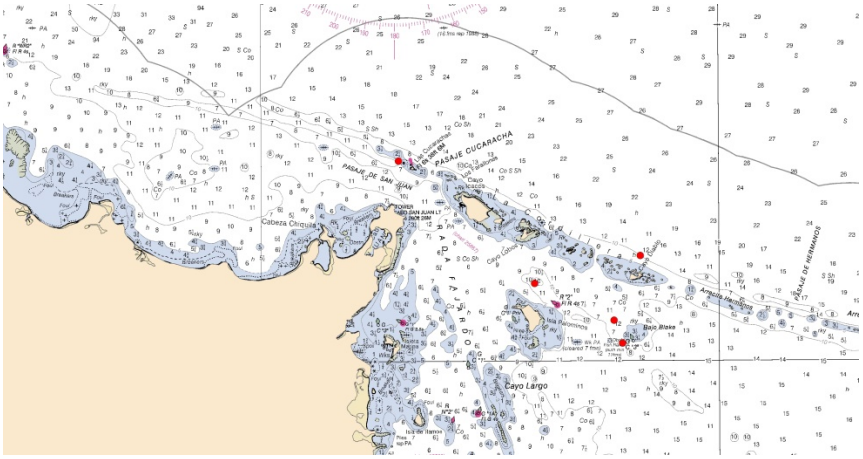

Figure 5. Purported red hind spawning aggregations sites to be examined for viability and confirmation, as identified in Ojeda et al. 2007.

Contributing Partner Institutions

SEFSC: Passive acoustics research, western and NE Reserves, PR (CRCP funding, FY-14-16; R. Hill and J. Doerr)
CCRI: Passive acoustic research of spawning aggregations (Year 9; PI - M. Schärer)
CFMC: Nassau grouper project (Year 2; PI - M. Schärer)
UVI-CMES: Fish spawning aggregation research (Year 2; PI - R. Nemeth)
SEAMAP-C: Red hind passive acoustic monitoring (Year 3; PI - R. Appeldoorn)

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Appendix-Table 3. August 2014 report on DSG data downloads for 2013-2014 spawning season. (During subsequent fieldwork, the remaining DSGs were successfully retrieved and data were downloaded, as planned.)

2013-2014 Spawning Season Passive Acoustic Recorders Data Downloads	
As of August 2014, the UPRM Dept. of Marine Sciences has recovered 9-DSG dataloggers from the field, including:	
	GB Data
Recovered by UPR-Mayaguez Team (9):	
DSG – 1138 (12-Dec-13 to 5-June-14) at Abrir la Sierra	19.7
DSG – 1289 (27-Feb-14 to 28-July-14) at Bajo de Sico	17.2
DSG – 1018 (3-Jan-14 to 26-July-14) at Bajo de Sico	23.2
DSG – 1274 (18-Dec-13 to 28-July-14) at Bajo de Sico	25.2
DSG – 1014 (18-Dec-13 to 23-July-14) at Bajo de Sico	22.1
DSG – 55 (25-Dec-13 to 5-June-14) at Mona-H6	18.4
DSG – 1273 (25-Dec-13 to 5-June-14) at Mona-Elbow	18.4
DSG – 1017 (25-Dec-13 to 5-June-14) at Mona-H5	18.4
DSG – 1276 (25-Dec-13 to 7-Aug-14) at Monito	25.6
DSG – 1275 (18-Dec-13 to 23-July-14) at Bajo de Sico	22.1
Recovered by USVI Team (2):	
DSG – 1104 (6-Dec-13 to 27-June-14) at Grammanik Bank	23.1
DSG – 1086 (6-Dec-13 to 27-June-14) at Red Hind Bank	23.1
TOTAL RECORDING DOWNLOADS thus far:	**256.5**
Left to Recover*:	
DSG – 57 (18-Dec-13 to _____ 2014) at Bajo de Sico	
DSG – 1137 (18-Dec-13 to _____ 2014) at Tourmaline	
DSG – 1016 (3-June-14 to _____ 2015) at Mona for year-round recording	
DSG – 1288 (1-Feb-13 to _____ 2014) at El Seco	
*As of September 2014, all data had been downloaded and secured.	
Recordings are 20-second duration, on a 5-min interval between the schedule dates provided.	
Multi-species analysis, using spectrograms and acoustic software, details behavioral characteristics and bio-distribution of Nassau, black and yellowfin grouper, as well as red hind, for a variety of study sites known to support aggregations: Bajo de Sico, Abrir la Sierra, Mona, Monito, Tourmaline, Grammanik Bank and the MCD.	