## Use of Open Accelerometer Tag to Detect Grouper Courtship Associated Sounds: A Pathway to Spawning-stock Size Determination

## Uso de Aaccelerometros para Detectar Sonidos Asociados a la Reproducción en Meros: Una Vía para Determinar el Tamaño de la Población Reproductora

# Utilisation de L'étiquette D'accéléromètre Ouvert pour Détecter les sons Liés à la Parade de Grouper: Un Chemin vers la Détermination de la Taille du Stock de Frai

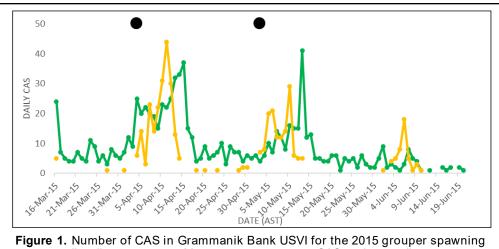
\*CARLOS M. ZAYAS-SANTIAGO<sup>1</sup>\*, RICHARD S. APPELDOORN<sup>1</sup>, MICHELLE T. SCHÄRER-UMPIERRE<sup>2</sup>, and JUAN J. CRUZ MOTTA<sup>1</sup> <sup>1</sup>Department of Marine Sciences, University of Puerto Rico Mayagüez, Puerto Rico 00681-9013 USA. <sup>2</sup>HJR Reefscaping, Cabo Rojo, Puerto Rico 00623 USA. \*<u>carlos.zayas3@upr.edu</u>

### **EXTENDED ABSTRACT**

Many species of marine fishes produce species-specific sounds associated with reproductive behaviors. Passive acoustic monitoring has been used to study groupers that produce courtship associated sounds (CAS) when they aggregate to spawn. This technique has revealed patterns of sound production during spawning aggregations with extremely high temporal resolution. A primary goal to continue to expand the application of passive acoustics is to measure trends in abundance over time and apply this to spawning stock or population estimates during critical life stages. Underwater visual census has been most commonly used to estimate fish abundances during aggregations, but with several limitations (i.e. weather constraints, depth, fish behavior, water quality) that restrict its long-term effectiveness. A factor limiting the extrapolation of CAS counts to fish abundance is that call rates of an individual fish are not known. Groupers have a sound production mechanism made up of cranial muscles vibrating onto the swimbladder and generating sound waves. During the 2017 spawning season, red hind (*Epinephelus guttatus*) where held in a 1,500 - gallon tank with an accelerometer tag and a low frequency hydrophone to record CAS. By placing the accelerometer tags near the fish, the vibrations generated during sound production were detected, and correlated with CAS. In additional experiments, stronger signals of induced alarm calls produced by red hind were simultaneously recorded by both methods, confirming the potential of this approach to make passive acoustic monitoring useful for assessing spawning aggregations of groupers.

### Introduction

Many species of marine fish produce species-specific sounds associated with reproductive behaviors. Passive acoustic monitoring has been used to study groupers that produce courtship associated sounds (CAS) when they aggregate to spawn. As slow growing, sex changing aggregate spawners, groupers are vulnerable to overfishing. A primary objective in studying fish spawning aggregations (FSA) is to estimate fish abundance and hence spawning stock. Historically, underwater visual census (UVC) has been used to determine abundance, but grouper spawning occurs at periods of low light, abundance peaks over a narrow time period, spawning can occur over one or multiple aggregations each year, access is often limited by wind/sea conditions, assessments are labor intensive, and divers are logistically limited to assessing only a few sites – all of which severely limit the use of UVC. Given the limitations of the UVC approach, there is a need to develop efficient, low cost remote monitoring techniques that can describe temporal spawning behavior and estimate sitespecific stock populations. Other methods, such as active acoustics (split-beam sonar), are either limited by the difficulty in hitting a moving aggregation with a narrow beam, such as with Nassau grouper Epinephelus striatus, or as in the case of red hind, Epinephelus guttatus, being able to detect fish near bottom or even hiding in caves and crevices within the bottom. Passive acoustic monitoring holds great promise in overcoming these difficulties. Hydrophones can be deployed relatively inexpensively to record these sounds over the full course of the spawning season, so sampling has high temporal resolution and is less affected by sea/wind state; additionally, automatic processing of sounds is becoming operational, thus greatly reducing processing time and costs. Furthermore, passive acoustic monitoring can be used to distinguish grouper species that are using the same spawning season and location (Figure 1). Studies in croakers suggest a positive relationship between sound pressure levels and fish density (Rowell et al. 2017, Sprague and Luczkovich 2012). However, in red hind that relationship may vary among and within spawning aggregations from year to year (Appeldoorn et al. 2013). The limiting factor in converting CAS counts to number of fish is an understanding of the calling behavior of the fish, specifically the call rate of an individual male, which may be subject to several factors in addition to the number of other males and females. Some known factors, such as the time of day (light-levels), lunar period or day of the aggregation can be controlled by limiting the period of analysis. Other suspected factors, such as current flow, water temperature, or swells need to be directly accounted for. A combination of passive acoustic monitoring and micro-accelerometer tags at a spawning aggregation site may help answer some of these questions. Micro-accelerometers have been used to study feeding strikes and escape responses in the great sculpin Myoxocephalus polyacanthocephalus (Broell et. al 2013). Furthermore, microaccelerometer tags have been used to study seal whisker vibrations at different frequencies (Murphy et al. 2016). Groupers such as *Epinephelus striatus* are known to use cranial muscles that vibrate onto the swimbladder to produce sound (Hazlett and Winn 1962). Consequently, a micro-accelerometer tag could be used to detect vibrations of these fast-contracting



**Figure 1.** Number of CAS in Grammanik Bank USVI for the 2015 grouper spawning season yellow line represents *Mycteroperca venenosa* CAS and green line represents *Epinephelus striatus* CAS. Black circles on top represent days of Full Moon.

muscles. By coupling passive acoustic monitoring with micro-accelerometer transmitters, we could improve our understanding of fish spawning aggregations, reproductive behaviors, sound production, and the factors affecting them. This information could provide estimates of spawning stock abundance and assess grouper spawning aggregations.

#### Methods

During the 2016 - 2017 spawning season five red hind, two males and three females were captured (DRNA permit 2016-IC-176) and placed in a 1,500 - gallon tank. The tank was set up with caves and flat areas to resemble the benthic habitat where they were captured, and equipped with a recording hydrophone (DSG-Ocean Loggerhead sensitivity of 180 dBVµPa<sub>-1</sub> frequency range 2 to 37 kHz) and two micro-accelerometers (Open Accelerometer Tag Loggerhead/Analog Devices accelerometer model adx1345 max output  $\pm 16$  g). The accelerometers recorded continuously on the Z-axis every five minutes at 1400 Hz (16-bit). The hydrophone recorded continuous 1-minute files at a sample rate of 10,000 Hz and sample size of 16-bits. A circular cage was placed inside the tank to separate a resident male with two females from another male periodically the nonresident male was introduced to induce calling activity on the resident male.

On October 25, 2017 a 383mm female red hind was captured off La Parguera shelf edge and placed in a 200 - gallon cement tank with the micro-accelerometer and the recording hydrophone synchronized by being tapped underwater at the same time. To induce alarm calls the fish was hand held next to both instruments. Holding the fish was enough to produce an alarm call. To describe the alarm call in both instruments Raven Pro 1.4 was used to measure four parameters; call duration (s) and amplitude (root mean square (RMS)) taken from oscillogram and peak frequency (Hz) and peak power (dB) taken from spectrogram. Hydrophone spectrograms were made with a 512-point Hann window 3 dB bandwidth = 28.1 Hz with 50% overlap and a 512-point DFT window size. Accelerometer spectrograms were made with a 163-point Hann

window 3 dB bandwidth = 12.4 Hz with 49% overlap and a 256-point DFT window size.

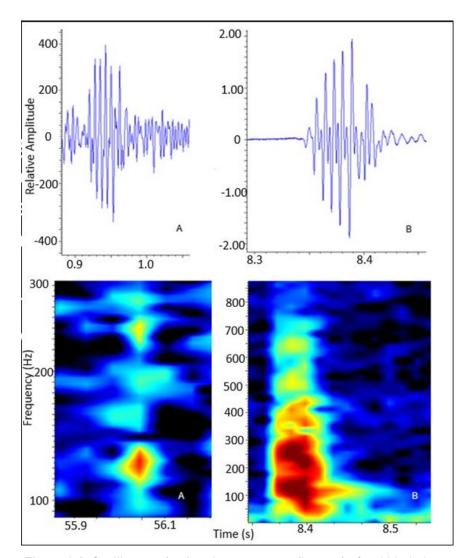
#### **Results and Discussion**

Captive male red hind produced CAS in the morning and dusk and during territory experiments. These sounds were recorded in the hydrophone but were too faint to be recorded in the micro-accelerometer tag for identification. No spectrogram measurements were taken from the initial experiments due to water pump acoustic interference, but CAS were clearly produced and recorded with the hydrophone. In the second experiment the micro-accelerometer and hydrophone recorded the alarm call (Figure 2.A). Both were confirmed visually and audibly. The alarm calls recorded in the accelerometer (n = 6) had a mean duration  $0.11 \pm 0.03$  s, mean peak power  $30.56 \pm 1.56$  dB, mean amplitude RMS  $23.53 \pm 2.28$  and mean peak frequency  $130.32 \pm 3.7$  Hz. The alarm call in the hydrophones (n = 6) (Figure 1.B) had a mean duration  $0.11 \pm 0.01$  s, mean peak power 92.2  $\pm$  1.51 dB, mean amplitude RMS 1919.35  $\pm$ 195.72 and a mean peak frequency 126.95  $\pm$  14.89 Hz. This study has proven that micro-accelerometer tags can record individual grouper sonic activity. The ongoing development of implantable accelerometer acoustic tags has the potential to enhance present passive acoustic monitoring programs by allowing them to quantitatively describe site-specific stock size. These data will have higher temporal resolution and greater standardization than other existing data available for the study of grouper spawning aggregations, making them easier to understand across stakeholders. The ability to remotely monitor individual locations will allow a greater number of aggregation sites to be concurrently monitored and/or aggregations to be studied in much greater detail. Such data will enhance assessments of site-specific spawning stocks, assess the effectiveness of (population response to) MPAs and other management strategies, and enhance our understanding of fish spawning aggregation sonic behavior.

KEYWORDS: Red hind, spawning aggregations, micro accelerometer, passive acoustics

### LITERATURE CITED

- Appeldoorn, R.S, M.T. Schärer-Umpierre, T.J. Rowell, and M. Nemeth. 2013. Measuring relative density of spawning Red hind *Epinephelus* guttatus from sound production: consistency within and among sites. *Proceedings of the Gulf and Caribbean Fisheries Institute* 65:284-286.
- Broell, F., T. Noda, S. Wright, P. Domenici, J.F. Fleng, J.P. Auclair, and C.T. Taggart. 2013. Accelerometer tags: detecting and identifying activities in fish and the effect of sampling frequency. *The Journal* of *Experimental Biology* **216**:1255-1264
- Hazlett, W and H. Winn. 1962. Sound producing mechanism of the Nassau grouper, *Epinephalus striatus*. *Copeia* **2**:447-449.
- Murphy, C., C. Reichmut, W. Eberhardt, B. Calhoun, and D. Mann. 2016. Seal whiskers vibrate over broad frequencies during hydrodynamic tracking. *Scientific Reports* 7(8350):1-6.
- Rowell, J.T., D.A. Demer, O. Auburto-Oropeza, J.J. Cota-Nieto, J.R. Hyde, and B.E. Erisman. 2017. Estimating fish abundance at spawning aggregations from courtship sound levels. *Scientific Reports* 7:3340.
- Sprague, M and J. Luczkovich. 2012. Modeling fish aggregation sounds in very shallow water to estimate numbers of calling fish in aggregations. Proceedings of the 161th Acoustical Society Meeting 12:1-8



**Figure 2.A** Oscillogram (top) and spectrogram (bottom) of red hind alarm call recorded by the micro accelerometer. **2.B** Oscillogram (top) and spectrogram (bottom) of red hind alarm call recorded by the hydrophone. Hydrophone spectrograms were made with a 512-point Hann window 3 dB bandwidth = 28.1 Hz with 50% overlap and a 512-point DFT window size. Accelerometer spectrograms were made with a 163-point Hann window 3 dB bandwidth = 12.4 Hz with 49% overlap and a 256-point DFT window size.