Measuring Relative Density of Spawning Red Hind (*Epinephelus guttatus*) from Sound Production: Consistency Within and Among Sites

Usando la Producción del Sonido como Medida de las Densidades Relativas del Mero Cabrilla (*Epinephelus guttatus*) durante la Agregación de Desove: Consistencia Dentro y entre Los Lugares

Mesure de la Densité Relative de le Vieille Rouge, (*Epinephelus guttatus*) d'Après les Sons: Cohérence dans et entre Emplacements

RICHARD S. APPELDOORN*, MICHELLE T. SCHÄRER-UMPIERRE, TIMOTHY J. ROWELL, and MICHAEL NEMETH

Department of Marine Sciences, University of Puerto Rico, Mayagüez, Puerto Rico 00681-9000 *richard.appeldoorn@upr.edu

ABSTRACT

Groupers are among the most important yet vulnerable fishes in the Caribbean and in tropical waters worldwide. Vulnerability comes from their habit of forming spawning aggregations predictable in space and time, yet these same factors make aggregations focal points for both management and assessment. However, *in situ* assessments are often difficult due to wind and wave conditions and limited resources, especially where multiple spawning sites exist. Passive acoustic recordings using benthic hydrophones offer a potential solution. Groupers are soniferous fishes, and males make specific sounds related to courtship activities. These calls can be distinguished among species. For the red hind at the Abrir la Sierra (ALS) aggregation site off western Puerto Rico, the number and sound level of calls were sufficiently correlated to overall fish density to be able to use sound as a predictor of density, or relative abundance. We address the reliability of this predictive capacity by comparing the relationship between red hind sound production to density at ALS over two years and between ALS and Mona Island, which is separated from ALS by 47 km and depths of 400 to 600 m. Red hind calls were recorded using a bottom-mounted hydrophone while density was measured using diver visual census surveys. Due to low sample size and greater inherent variability, the relationship between sound production and density at Mona Island in 2010 was not statistically significant, but the slope was similar to that observed at ALS in 2012. The relationships between sound production and density at ALS in 2011 and 2012 were both significant, but their slopes differed. In 2012 there was greater sound production at a lower density. Seasonal profiles of sound production during a spawning season are seen as useful predictors of density, but the dynamics of this across years may be variable.

KEY WORDS: Red hind, spawning aggregation, passive acoustics

INTRODUCTION

Groupers are among the most important yet vulnerable fishes in the Caribbean and in tropical waters worldwide. Their vulnerability comes from a combination of life history (long life, slow growth) and their reproductive behavior. Groupers form large seasonal aggregations for the purposes of spawning. These spawning aggregations are predictable in both space and time, which allows fishers to target their fishing effort for maximum efficiency, but without controls on overall fishing effort the result has been a decline in grouper populations throughout the Caribbean. Yet, maintenance of functional grouper populations is imperative if we are to maintain both viable fisheries and, perhaps more importantly, the biodiversity and trophic structures necessary to sustain healthy coral ecosystems that support overall fisheries production.

Assessments of grouper populations are often difficult because the adults are often typically solitary and in low abundance, particularly for overfished species such as Nassau, yellowfin and goliath groupers. As an extreme example, in the US Caribbean, catches of Nassau and goliath grouper are prohibited, so there are no effective fishery dependent data sets available. However, the habit of forming discrete spawning aggregations in space and time also make aggregations potential focal points for both management and assessment.

Key obstacles to this approach are that the location and timing of these aggregations must be known. Furthermore, given the limited time of each aggregation, and the fact that aggregations build over time until the actual day(s) of spawning, there is a narrow window wherein an assessment should be done. These are barriers that most fisheries agencies are poorly equipped to overcome. *In situ* surveys are labor intensive and dependent upon wind and wave conditions for access to sites at key times. This problem is multiplied when more than one aggregation site exists for a species, and multiple species are to be assessed. Traditional ecological knowledge from local fishermen will be extremely important in reducing the effort in locating spawning aggregations in space and time (Claro and Lindeman 2003, Ojeda et al. 2007), though even this may require significant follow-up (e.g., Schärer et al. 2010)

Passive acoustic recordings offer a potential solution. Groupers are soniferous fishes, and males make specific sounds related to courtship activities. These calls can be distinguished among species (Mann et al. 2010, Schärer et al. 2012a, b). Rowell et al. (2011) reported on the use of passive acoustic techniques for locating and mapping red hind (*Epinephelus guttatus*) spawning aggregations. Additional work (Rowell et al. 2012) indicated that the number and sound level of calls are sufficiently correlated to overall red hind density to be able to use sound as a predictor of density, or relative abundance.

In this study, we address the question as to whether this relationship is consistent between locations and year to year.

METHODS

We address the reliability of the predictive relationship between density and sound levels/call frequency by comparing the relationship between red hind calls to density at two sites. The main study site is Abrir la Sierra (ALS), located along the edge of the insular shelf off western Puerto Rico. Details of the methodology are given in Rowell et al. (2012). Briefly, a long-term recording hydrophone (DS-Ocean, Loggerhead Instruments) was mounted on the bottom within a known red hind spawning aggregation. During the spawning season periodic visual surveys over a fixed course, centered over the hydrophone, were conducted and the number, location and size of each red hind were recorded. Transects were targeted for the period 16:00 - 17:00. Recordings were made for 20 seconds every five minutes throughout the spawning season. For analysis, we monitored the hydrophone recordings over the period between 18:00 - 19:00, previously established as the peak of the diel cycle of sound production. The recordings were also processed with low and high band filters to isolate the frequency range containing red hind calls. Red hind calls above 105 dB re 1 mPa were counted for each day. Sound levels within that band (band levels) were averaged over the one-hour period.

The second site was located at Mona Island, which is separated from ALS by 47 km and depths of 400 to 600 m. Hydrophone recordings were processed as above. Transects at Mona were not standardized to time of day, nor over a fixed course, but data collection was the same as at ALS. Additionally, the days available for diving were more limited and more affected by sea conditions.

The relationship between red hind density and call counts/band levels were analyzed using linear regression.

RESULTS AND DISCUSSION

Figure 1 shows the variation in both band level and red hind density over the red hind spawning season at ALS in 2012. Use of call counts produced similar results. It is clear from the figure that noise levels produced by red hind reasonably tracked red hind densities. This is supported by regression analysis (Table 1), which showed a significant relationship (p < .05) between the two factors and which had an $r^2 = 0.482$. This relationship is shown in Figure 2, which also shows the regression line obtained for 2011 (Rowell et al. 2012). While the relationship is significant

for 2012, the slope and r^2 are lower than for 2011. The differences in slope are significant, but the residual mean square error is actually smaller for 2012. This indicates that differences in slope and r^2 are not due to greater variability in the 2012 data, but red hind in that year produced higher sound levels (and had higher call counts) per given density than in 2011.

Only four density estimates were available for the Mona Island site, and these were from 2010. Because these density estimates were not standardized by time of day or area, there is greater inherent variability represented within the data. There was a positive relationship between band

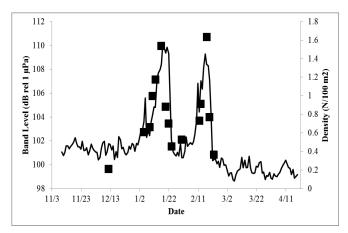


Figure 1. Band level (line) or sound production and density (squares) of red hind at Abrir la Sierra from November 2011 to April 2012.

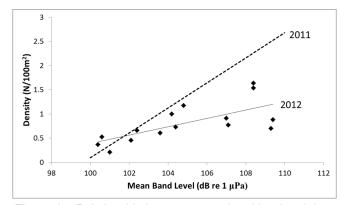


Figure 2. Relationship between mean band level and density of red hind density at Abrir la Sierra in 2012 (points and solid regression line). Dashed line is the regression line obtained for 2011 from Rowell et al. (2012). See Table 1 for the equations of the regression lines.

Table 1. Results of regression analysis on density (y) on band level (x) forred hind at Abrir la Sierra. Residual MS is the Residual Mean Square Error of the points from the regression line.

Year	N	Slope	Intercept	p-value	r²	Residual MS
2011	10	0.260	-25.908	0.00005	0.829	0.178
2012	15	0.087	-8.261	0.00745	0.482	0.089

level (and call count) and red hind density, with a slope similar to that seen at ALS in 2012. Yet, the relationship was not statistically significant. Maximum densities observed at Mona Island were only half those observed at ALS in 2012 and only one-fourth observed in 2011.

The results from ALS clearly indicate that band levels and call counts are good predictors of the relative abundance of red hind during spawning aggregations. In both 2011 and 2012 there was a strong, statistically valid relationship. Thus, these passive acoustic techniques provide high-resolution temporal data on the occurrence, build-up and abundance of red hind within a given year. This information is valuable for calibrating visual census surveys to timing of the aggregation cycle and for verification that management regulations related to closed seasons actually encompass the entire spawning season.

The data are less clear on the consistency of the relationship over time or between locations. For the latter, the relationship at Mona Island in 2010 was not statistically significant, although the slope was similar to that observed at ALS in 2012. The lack of significance was not surprising given the greater variability in the data and the few points available, but the results were promising. The results comparing ALS in 2011 and 2012 are more enigmatic. Maximum density at ALS in 2012 was just half that observed in 2011. Red hind are known to vary the proportion of fish occurring at the aggregation site depending on the timing of the full moon. According to Nemeth et al. (2007), if the full moon falls between January 10 and 30, there will likely be one dominant aggregation. If the full moon falls outside that period, there will be two aggregations, with one occurring in December if the full moon occurs prior to this period or one occurring in February if the full moon occurs after this period. Depending on the exact timing, the resulting two aggregations can be equal or unequal in fish density. At ALS, in 2011 there was one large aggregation, but in 2012 there were two aggregations, though not quite equal in size. Thus the lower densities observed in 2012 relative to 2011 could be explained by the population being roughly equally split between the first and second aggregations, with a resulting halving of the density at each one relative to 2011. At Mona in 2010 there were again two aggregations, with the second aggregation being larger than the first.

The comparison between 2011 and 2012, however, indicates that the problem is not related to density but to the calling behavior. In 2012, red hind sound production was almost the same as in 2011, but with half the density. Why this is so is unclear. Only males are believed to produce sounds during the aggregations, both to defend territories from other males and to attract females to their territory. Males are known to set up territories at the aggregation site before the arrival of females, and if more than two aggregations occur they continue to occupy the aggregation site. This contrasts with females, which come and go from aggregation sites, with their numbers gradually increasing toward the actual night(s) of spawning. That

calling activity increases in consort with the increase in density suggests that males become more active as more females arrive. What regulates this activity is unknown. Other factors can also affect the density-sound relationship, such as a shift in the location of peak abundance relative to the location of the hydrophone. Because the areas covered by the visual survey and the listening distance of the hydrophone are different, both in area and shape, there can be a mismatch between the two. However, examination of densities in the immediate vicinity of the hydrophone showed results similar to overall density values, suggesting this was not a problem.

Despite these difficulties, there are additional indications that there is a direct relationship between sound production and fish density, such that sound production may be used as an indicator of abundance across different aggregations. Examination of band level and call-count profiles across four red hind aggregations in Puerto Rico and the US Virgin Islands in 2012 (unpublished) suggests a consistent relationship between density and sound production. Among these sites, density varied by an order of magnitude, yet differences in sound production tracked both small and large differences in density. Continued monitoring coupled with targeted studies on the behavior of red hind relative to sound production will be needed to clarify these relationships.

LITERATURE CITED

Claro, R. and K.C. Lindeman. 2003. Spawning aggregation sites for snappers and groupers species (Lutjanidae and Serranidae) on the insular shelf of Cuba. Gulf and Caribbean Research 14:91-106.

Mann, D., J. Locascio, M. Schärer, M. Nemeth, and R. Appeldoorn. 2010. Sound production by red hind (*Epinephelus guttatus*) on spatially segregated spawning aggregation sites. *Aquatic Biology* 10:49-154.

Nemeth, R.S., J. Blondeau, S. Herzlieb, and E. Kadison. 2007. Spatial and temporal patterns of movement and migration at spawning aggregations of red hind, *Epinephelus guttatus*, in the U.S. Virgin Islands. *Environmental Biology of Fishes* **78**:365–381.

Ojeda-Serrano, E., R.S. Appeldoorn, and I. Ruíz-Valentín. 2007. Reef fish spawning aggregations of the Puerto Rican shelf. Proceedings of the Gulf and Caribbean Fisheries Institute 59:467-474.

Rowell, T.J., R.S. Appeldoorn, J.A. Rivera, D.A. Mann, T. Kellison, M. Nemeth, and M. Schärer-Umpierre. 2011. Use of passive acoustics to map grouper spawning aggregations, with emphasis on red hind, Epinephelus guttatus, off western Puerto Rico. Proceedings of the Gulf and Caribbean Fisheries Institute 63:139-142.

Rowell, T.J., M.T. Schärer, R.S. Appeldoorn, M.I. Nemeth, D.A. Mann, and J.A. Rivera. 2012. Sound production as an indicator of red hind (*Epinephelus guttatus*) density at a spawning aggregation. *Marine Ecology Progress Series* **462**:241–250.

Schärer, M.T., M.I. Nemeth and R.S. Appeldoorn. 2010. Protecting a multi-species spawning-aggregation at Mona Island, Puerto Rico. Proceedings of the Gulf and Caribbean Fisheries Institute 62:252-259.

Schärer, M.T., M.I. Nemeth, D. Mann, J. Locascio, R.S. Appeldoorn and T.J. Rowell. 2012a. Sound production and reproductive behavior of yellowfin grouper, *Mycteroperca venenosa* (Serranidae) at a spawning aggregation. *Copeia* 2012:135–144.

Schärer, M.T., T.J. Rowell, M.I. Nemeth, and R.S. Appeldoorn. 2012b. Sound production associated with reproductive behavior of Nassau grouper *Epinephelus striatus* at spawning aggregations. *Endangered Species Research* 2012:29–38.