

National Oceanic and Atmospheric Administration

MARFIN Project FINAL Report

Population density, demographics, and predation effects of adult goliath grouper

Project NA05NMF4540045 (FSU Project No. 016604)

October 2006 - September 2009



Submitted:

by

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EXECUTIVE SUMMARY

Over the past several years we have conducted studies on the protected reef fish, goliath grouper (*Epinephelus itajara*). Goliath grouper, a species indigenous to the southeastern U.S., the Caribbean, Brazil, and the west coast of Africa has been overfished to the extent that the IUCN (World Conservation Union) has classified it as 'critically endangered'. The species was protected in the southeastern U.S. in 1990 through action by Gulf of Mexico Fishery Management Council and the South Atlantic Fishery Management Council and the National Marine Fisheries Service (NMFS). Since that time there has been a steady recovery. The primary purpose of our goliath grouper studies was to provide an understanding of adult ecology and behavior so that the population could be managed sustainably. Investigations involved: 1. Distribution throughout the southeastern U.S., 2. Regional densities in Florida, 3. Habitat characteristics, 4. Movement patterns, 5. Regional size distributions, 6. Diet and trophic patterns, and 7. Spawning biology. We found that the vast majority of the goliath grouper population of the southeastern U.S. is restricted to Florida waters, mostly in south Florida. Population recovery began off southwest Florida in the area of abundant juvenile habitat, the Ten Thousand Islands, and expanded to the north and east to southeast Florida, especially off Palm Beach County where spawning aggregations now exist. Goliath grouper prefer high relief habitat with extensive holes and caves large enough to accommodate adults, except in coral reef habitat where population densities are relatively sparse. Except for spawning migrations they display strong site fidelity. Goliath grouper feed mostly on crabs (over 60% of the diet) and especially mud crabs and swimming crabs. Fishes dominating the diet are mostly sedentary species such as toadfish, boxfish, and burrfish. They are not apex predators, but instead feed at a relatively low trophic level, similar to that of pinfish (*Lagodon rhomboides*). According to our bioenergetics model adult goliath grouper food consumption per day is very low, but additional data are needed to reduce uncertainty. Spawning occurs in aggregations during the late summer through early fall (August through October). Night-time sounds produced by goliath grouper during this period (and no other) appear directly related to spawning and may be useful in determining spawning areas, spawning season, and frequency. We confirmed night-time spawning by collecting goliath grouper eggs (verified genetically) in the late evening and early morning downstream from spawning aggregations during the last quarter moon in the Atlantic and during the new moon in the Gulf. Our quantitative reef survey data on the regional population densities of goliath grouper in Florida was concordant with semi-quantitative Reef Environmental Education Foundation (REEF) data; this result will reduce uncertainty about the use of REEF's population density data for future stock assessments.

INTRODUCTION

Goliath grouper (*Epinephelus itajara*) is the largest grouper in the western hemisphere, reaching lengths of over 3 m and weights of 800 – 1,000 pounds. The species ranges from North Carolina (Francesconi and Schwartz 2000) on the Atlantic seaboard to Texas in the Gulf of Mexico, throughout the Caribbean, and south to southeastern Brazil. While they are reported in the eastern hemisphere along the west coast of Africa, Craig et al. (2009) suggest that the African population may have gone extinct because none have been observed or captured in the fishery for over 10 years. Recently, the population in the Pacific, which ranges from the Gulf of California to Peru, has been determined to be a distinct species, *E. quinquefasciatus* (Craig et al. 2009).

In the United States, goliath grouper briefly supported both commercial and recreational fisheries, primarily in the Gulf of Mexico off Florida (Appendix, Tables A1 and A2). Populations were rapidly overexploited and the fisheries closed in 1990 (Sadovy and Eklund 1999). Today, they are considered critically endangered throughout their range globally (IUCN 2008), and continue to be listed as overfished in the United States (NMFS 2009). The actual status of the adult population remains unknown, making it difficult for NMFS to develop management measures aimed at either rebuilding the fishery, ending overfishing, or both, as required by the Sustainable Fishery's Act. Because the population appears to be increasing off the Gulf and Atlantic coasts of Florida, NMFS is being asked by commercial and recreational fishers to consider reopening the fishery at some level, while being asked concomitantly by conservation groups to continue protection while establishing a rebuilding plan.

The intent of this study was to estimate the regional population densities of goliath grouper managed by the Gulf of Mexico Fishery Management Council and the South Atlantic Fishery Management Council in the southeastern United States, emphasizing those populations occurring on the West Florida Shelf and the Atlantic coast of Florida. Five major questions we addressed were:

- (1) What is the current distribution of goliath grouper in the southeastern United States and the current regional population densities along Florida's coasts?
- (2) What are the regional demographic patterns of the recovering goliath grouper population?
- (3) What habitat constitutes essential habitat for adult goliath grouper?

(4) What are the spawning characteristics of goliath grouper?

(5) What are the dominant prey species of goliath grouper and what is the direct impact of the recovering goliath grouper population on those species?

We evaluate the distribution using several data sets, one derived from surveys conducted by the Reef Environmental Education Foundation (REEF), one from our own surveys and those sponsored by the state of Florida. We compared our quantitative survey data with REEF's semi-quantitative data to determine if the two data sets were concordant. We also conducted tagging surveys so that movement patterns could be discerned in both adult and maturing juvenile populations. In the process, we evaluated habitat use and describe essential habitat for adults. To determine the impact of goliath grouper on other species, we conducted trophic and bioenergetic studies, defining the diet through stomach content and isotope analyses and development of a bioenergetic model. Lastly we evaluated the timing of spawning both seasonally and daily using passive and active acoustic methods, then using the results of those studies, collected goliath grouper eggs on two spawning aggregations, one in the Atlantic and one in the Gulf of Mexico.

PART I. DISTRIBUTION, REGIONAL DENSITY, HABITAT AND MOVEMENTS

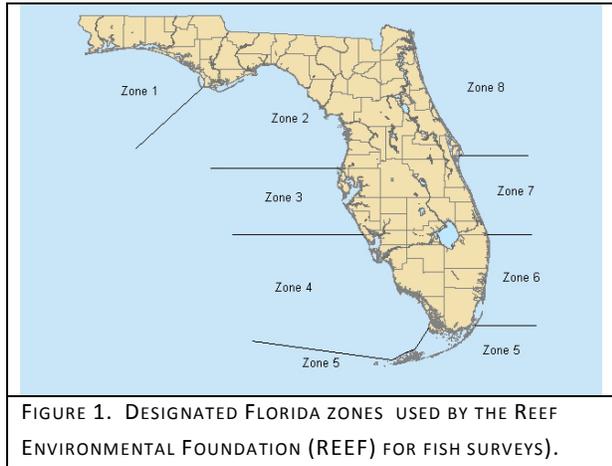
MATERIALS AND METHODS

We started tagging goliath grouper in 1996, tagging adults *in situ* using a modified spear with a point designed to drive the tag head no more than 5 cm beneath the skin of the fish (a method we have used effectively to tag over 2200 fish) and tagging juveniles by capture in cages or with hook and line (N = 2100) (Koenig et al. 2007). Our intent was to get information on their distribution and movement patterns in Florida waters by having fishermen and divers report sightings of tagged fish to an 800-number hotline maintained by the Florida Fish and Wildlife Conservation Commission's Florida Fish and Wildlife Marine Research Institute (FWMRI). To encourage reporting, we distributed posters to dive shops and marinas throughout the state stating the purpose of the study, a request for information on the location (latitude, longitude, depth) and size (length or weight) of the fish, and the FWMRI 800 number (Appendix, Figure A1). The FWMRI staff sent the reports to us so that we could contact responders for additional information about the location of capture and details about the fish. We entered responses into an ArcView database and produced maps plotting distances from initial capture to point of recapture. We also communicated with natural resource agencies from Texas to North Carolina to obtain information on goliath grouper sightings. We continue to update distribution information gathered through cooperative websites we have developed with state agencies.

Other sources of information on goliath grouper distribution came from The Reef Environmental Education Foundation (REEF) and through our own systematic surveys conducted throughout Florida waters for this project. REEF provided us with their raw data from volunteer reef surveys around Florida from 1994 to 2008 on goliath grouper sightings¹ and from other states for comparative purposes. Our observations and anecdotal information gleaned from fishermen over the past 18 years suggest that the center of abundance of goliath grouper populations is southwest Florida (Koenig et al. 2007) and that adult goliath grouper primarily associate with reef structure of relatively high relief. Whenever they appeared over unstructured bottom, it was always in close proximity to structure. We used these observations to develop a survey strategy throughout eight zones in Florida coastal and

¹ We used all of their goliath grouper data on the assumption that misidentifications of goliath grouper would be rare.

marine waters that corresponded as closely as possible to zones used by REEF (Figure 1). Zone 1 includes the western panhandle from Cape San Blas to the Florida-Alabama state line; zone 2 extends from Cape San Blas to the Pasco-Pinellas county line; zone 3 extends from there to the Sarasota-Charlotte county line; zone 4 includes the remainder of peninsular southwest Florida; zone 5 includes



the Florida Keys and Florida

Bay; zone 6 extends from Key Biscayne National Park to Jupiter Inlet; zone 7 extends from Jupiter Inlet to Cape Canaveral; and Zone 8 extends north from Canaveral to the St. Mary's River or the Florida-Georgia state line. Using the REEF zones allowed us to make comparisons between REEF's semi-quantitative data and our quantitative data to determine the degree of concordance between these two data sets, a high degree of concordance

indicating that the REEF data provided a reasonable alternative to quantitative surveys for the purposes of future stock assessments.

Our initial quantitative survey effort focused on reefs, both natural and artificial, from each of the 8 zones, with reefs stratified into four categories: low relief and high relief, artificial reefs and natural reefs, and pre-selecting reefs prior to conducting surveys. Locations of artificial reef sites were obtained from the FFWCC and locations of natural reef sites were obtained from fishermen. The intent was to randomly (i.e., without prior knowledge of goliath grouper presence) select high and low relief reefs (artificial and natural) within each zone, and estimate the population size of goliath grouper on each. We had to modify the pre-selection aspect for two primary reasons: (1) the relief of reefs differed significantly from expectations (many reefs had far less relief than recorded in fisherman log books, apparently due to silting); and (2) fishermen's willingness to provide reef locations was highly variable, with some opening their logbooks while others only provided published artificial reefs. The result is that the number of natural reefs surveyed differed among zones. We ended up selecting reefs blind and using fishermen's knowledge when it was forthcoming. We did not stratify by depth because within depths sampled (10 to 50 m) there were no discernible differences in population densities—differences were predominantly due to type and size of structure. Further, data from technical (trimix)

divers suggest that goliath grouper in southwest Florida rarely occur on high-relief sites deeper than about 60 m (Michael Barnette, Association of Underwater Explorers, personal communication).

Density estimates: On randomly selected reefs, we used a combination of roving diver visual surveys for densities of less than 10 individuals per site and catch-effort and Peterson mark-sight methods for densities greater than 10 individuals per site based on our preliminary sampling exercises (see Appendix, Figure A2, Table A3). Using this approach, we developed a mean population density within each reef type for each zone. We also recorded whether or not there were holes, caves and/or crevasses large enough for a goliath grouper to occupy. We then calculated the mean population density for a zone for each stratum. We also used goliath grouper abundance data from Florida-sponsored artificial reef surveys and used their habitat descriptions as a basis for assigning reef type.

We used a catch-effort (= tag-effort) method and the Petersen mark-resight (= mark-recapture) method to estimate population size of goliath grouper on individual sites. The catch-effort method (modified for goliath grouper) involves tagging the fish *in situ* with the modified spear point and recording the time-to-tag for each fish tagged. This method requires a skilled tagger who can apply effort evenly throughout the tagging period, and requires that a large proportion of the population is tagged. From the tag-effort data, we obtain a relationship between tags per unit effort (TPUE) and accumulated tagged fish (e.g., Appendix, Figure A2). If this relationship is linear (as estimated from regression methods) then we can assume equal catchability (= equal tag-ability). Ricker (1975) suggests adjustments to the estimates if the relationship is not linear.

The Petersen method for population estimation is also modified for our purposes. One day post tagging², three divers swam random transects over the fish to estimate the relative proportion of tagged and untagged fish. Divers swam above the fish so that the tag was visible regardless of which side of the fish was tagged, and so that fish were not alarmed by diver presence. Resighting fish multiple times

² We found that tagged fish and untagged fish formed separate groups immediately following tagging, thus violating the assumption of equal sightability. They assorted randomly the following day, which explains our conducting re-sighting the day following tagging.

by this method is equivalent to sampling with replacement. The population size (N) is determined by the following equation:

$$N=T(C+1)/(R+1),$$

where T = the number tagged, C = the number of resights, and R = the total number of tagged fish observed on the day after initial tagging. If the population estimate derived from the Petersen method does not differ significantly from the estimate derived from the tag-resight method, then the assumptions of a closed population and random assortment of the sampled population can be assumed valid. Tests of the combined methods on populations of small non-spawning aggregations and large spawning aggregations (Appendix, Table A3) indicate that these are appropriate for estimating population size under a variety of conditions. The tag-effort method resulted in linear regressions (Appendix, Figure A2) and the estimates derived from the Petersen method were very similar to those of the tag-resight method. Also in Appendix, table A3, see that visual censuses were similar to tagging estimates with small population sizes, but considerably underestimated population size at larger population sizes, as represented in the spawning aggregations. Clearly, the methods proposed worked well and provided accurate and precise estimates of population size on the reefs that were not affected by varying visibility or habitat conditions. The only drawback of the combined method was the cost of returning to the tagging site a second day, so we only used it on populations sizes > 10 individuals.

The REEF data categorized goliath grouper abundances as single (one goliath grouper seen), few (2-10), many (11-100), and abundant (>100). We eliminated the “abundant” category because there were no data for it, and truncated the “many” category to 65, the greatest density of fish we encountered. We then used the median number from the remaining categories as the abundance to compare with our data, resulting in the following conversions: none = 0, single = 1, few = 6, and many = 38. Using these values from REEF’s diver surveys, we estimated mean goliath grouper densities for each zone, habitat type, and year. For our purposes, we grouped REEF data into three 5-year intervals: 1994-1998, 1999-2003, and 2004-2008 to provide a sufficient number of observations for comparison.

HABITAT AND ASSOCIATED SPECIES. --We obtained habitat data from two sources: REEF and our own systematic surveys, using the habitat strata described above). REEF categorized habitat into 11 categories as follows: (1) mixed = multiple habitat types covered, (2) high profile = coral reef structure with relief >4 ft., (3) low profile = coral reef structure with relief < 4 ft, (4) sloping drop-off = bottom slopes to deeper water, (5) wall = shear drop-off of over 25 ft, (6) ledge = single or few sharp drops in

bottom topography >3 ft, (7) grass = seagrass dominant, (8) Sand = sand dominant, (9) rubble = broken coral, rock, boulders and/or gravel, (10) artificial = includes ship wrecks, platforms, dumped debris or other artificially created habitats, (11) open Water = deep water, bottom not visible. Of these categories, only 2, 3, 5, 6, 9, and 10 were relevant to goliath grouper surveys in Florida.

We described the reef structure (including reef height and the presence of caves large enough to accommodate adult goliath grouper) and community structure (recording fish and invertebrates presents) using video cameras mounted with a parallel laser system to measure goliath grouper and as many associated species as possible. All video recordings are archived at Florida State University Coastal and Marine Laboratory (FSUCML).

We identified goliath grouper preferred habitat as the habitat with the highest density of adults. We compared densities within each habitat type within each zone. We then compared densities in the preferred habitat among zones, and compared densities in recent (2004-2008) REEF data with those from our surveys for similarities in the density patterns. We also compared REEF data among zones at three different time intervals to determine whether a pattern of recovery emerged. Finally, we used regression analysis to evaluate the impact of high densities of adult goliath grouper on associated fish communities, particularly on the diversity of associated fish species and the abundance of economically important reef fish species.

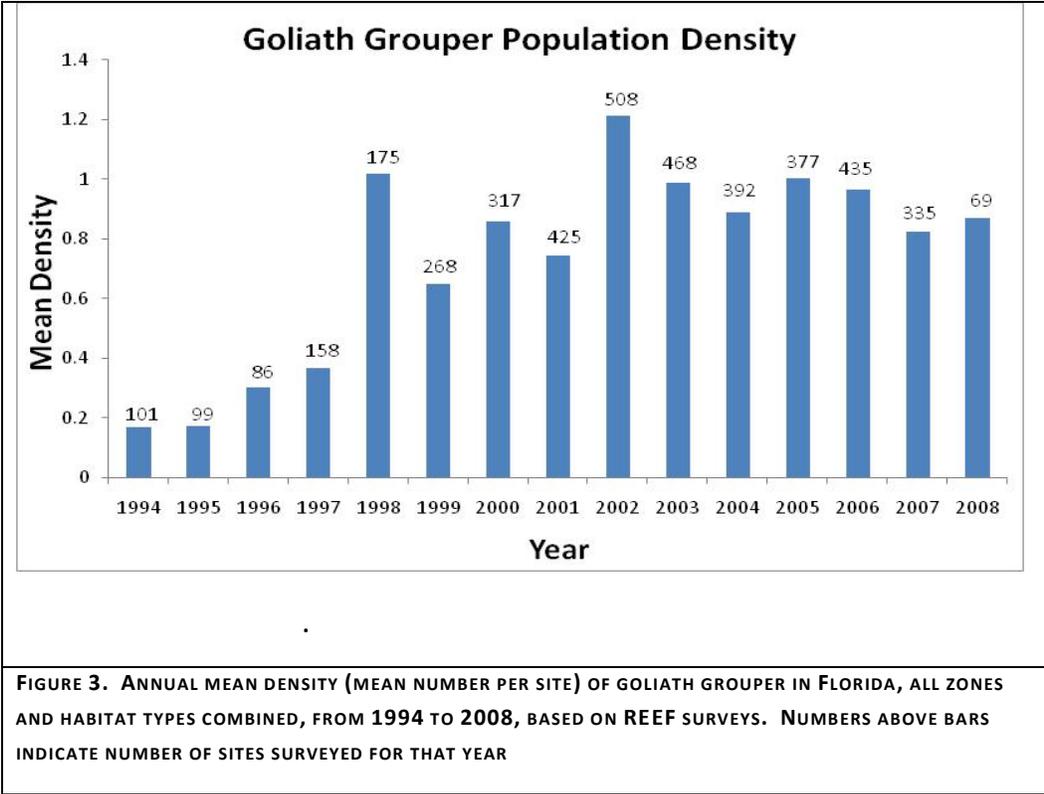
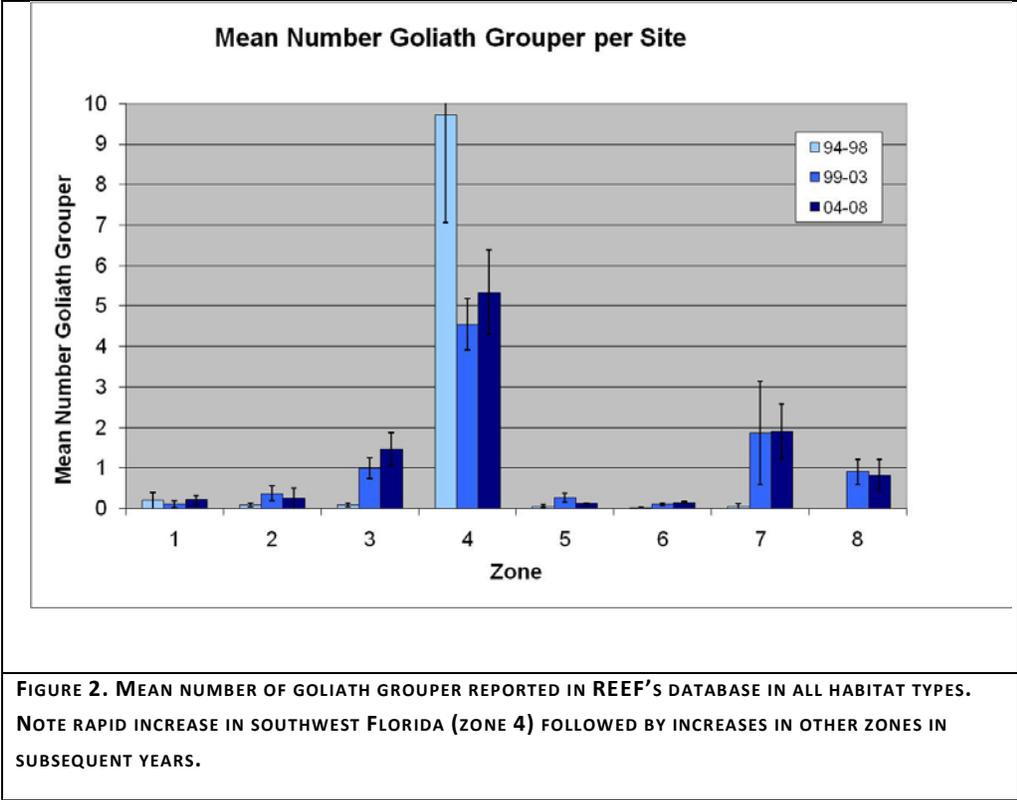
RESULTS

DISTRIBUTION AND RECOVERY.—Goliath grouper abundance is by far greater in Florida (mostly South Florida) than neighboring states in the southeastern U.S., according to REEF data. In Florida during the 5-y interval 2004 to 2008, 9.6% of the surveys and 23.6% of the surveyed sites reported goliath grouper. Texas, Georgia, and South Carolina were the only other states reporting sightings of goliath grouper (Table 1), although some states (AL and MS) did not submit reports.

Table 1. Relative abundance of goliath grouper reported to REEF for the period 2004-2008 for each state in the southeastern U.S.

STATE	Number of Surveys	Number of Sites Surveyed	Number of Surveys with Goliath Grouper	Number of Sites with Goliath Grouper	% Surveys with Goliath Grouper	% Sites with Goliath Grouper
Florida	9488	1000	912	236	9.6%	23.6%
Texas	639	24	3	3	0.5%	12.5%
Georgia	196	59	5	2	2.6%	3.4%
South Carolina	115	18	1	1	0.9%	5.6%
North Carolina	69	21	0	0	0.0%	0.0%
Louisiana	2	1	0	0	0.0%	0.0%
Alabama	0	0	0	0	0.0%	0.0%
Mississippi	0	0	0	0	0.0%	0.0%

The pattern of recovery for adult goliath grouper following the fishery closure in 1990 is apparent from REEF's survey data. Mean densities initially expanded rapidly off southwest Florida from 1994 to 1998 in zone 4 (Figure 2), and then increased in other parts of the state during the last 10 years. Mean density increased gradually through 1997 (Figure 3, Figure 4), then rapidly in 1998 and remained at an overall mean density of around one goliath grouper per site through 2008. The general pattern, then, was one of rapid recovery of the adult population off the Ten Thousand Islands-- the dominant juvenile habitat in the S. E. U.S. (Koenig et al. 2007)--followed by relatively slow recovery in other areas of the state.



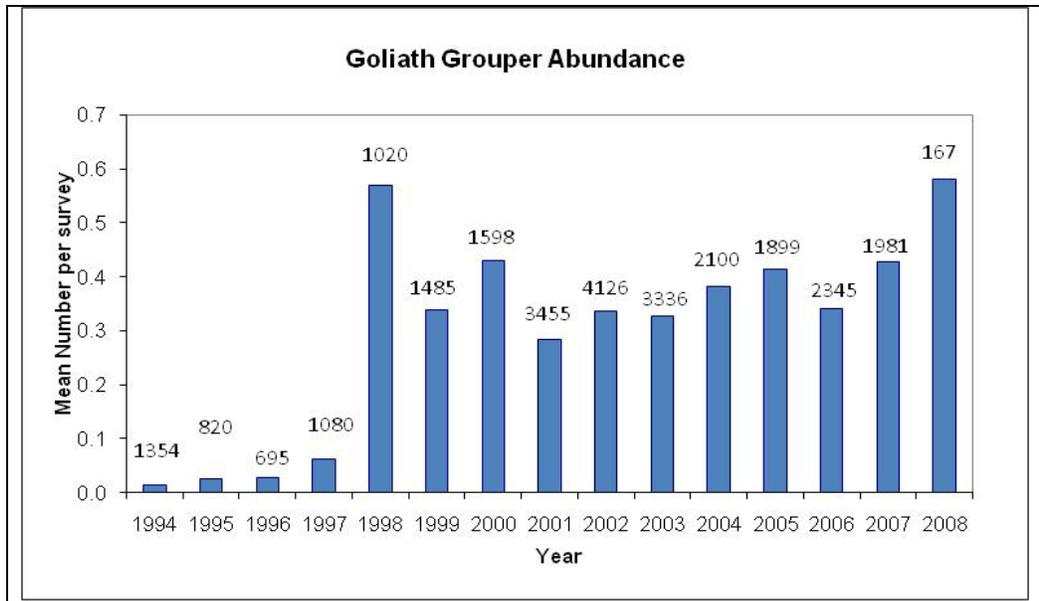
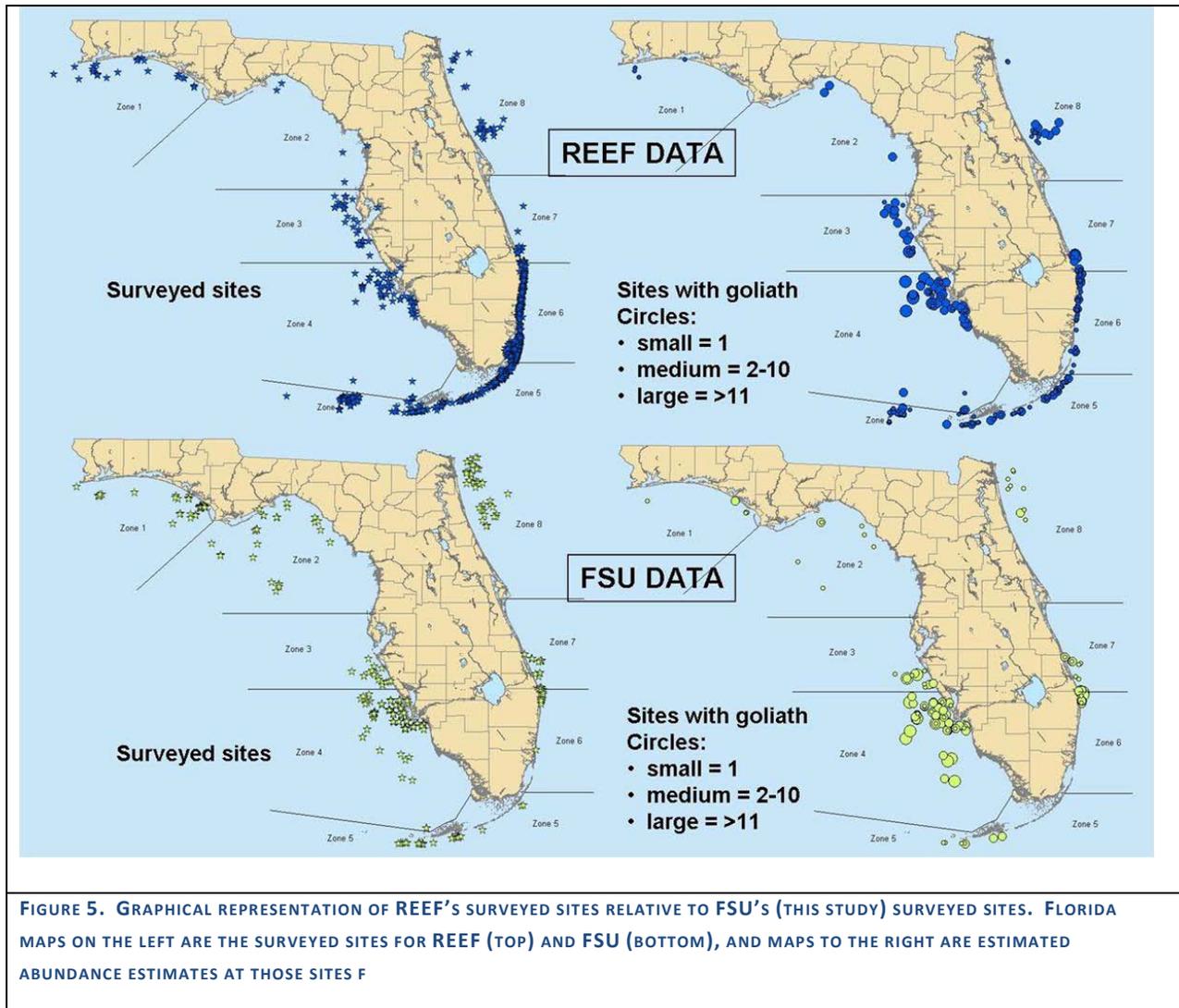


FIGURE 4. ANNUAL MEAN NUMBER OF GOLIATH GROUPEER PER SURVEY FROM FLORIDA REEF DATA, ALL ZONES AND HABITAT TYPES, FROM 1994 TO 2008. NUMBERS ABOVE BARS ARE NUMBER OF SURVEYS FOR THAT YEAR.

HABITAT AND REGIONAL DENSITIES.--FSU- and REEF-sampled sites (2004-2008) and respective population densities for the 8 zones of Florida indicate that goliath grouper densities are greatest in southwest Florida (zone 4 and the southern part of zone 3) (Figure 5). Zone 3 in the Gulf and zone 6 in the Atlantic represent transition zones where high densities grade into relatively low densities within the same zone.

Despite the numerous surveys of coral reefs in the REEF database (zones 5 and 6, including both low and high relief habitat), only low densities of goliath grouper are reported. In our surveys, based on known preferences of goliath grouper, we targeted high-relief reef types, including coral habitat in the Keys Reef Tract, natural ledges, and artificial reefs throughout Florida. A comparison of these high relief sites indicated that goliath grouper preferred high relief ledges and artificial reefs over high relief coral habitat (Figure 6). A similar pattern can be seen in habitat comparisons in REEF's data (Figure 7), where high relief artificial reefs and ledges have the densest populations while high- and low-relief coral and low-relief rubble have the lowest populations. "Wall" habitat, which occurs only in the lower Keys of Florida, had relatively high abundances.



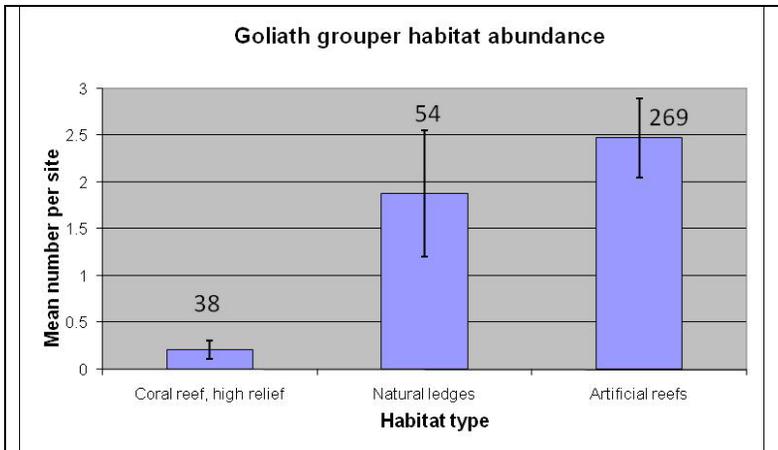


FIGURE 6. COMPARISON OF HIGH-RELIEF CORAL REEF HABITAT WITH HIGH-RELIEF NATURAL LEDGES AND ARTIFICIAL REEFS IN ALL ZONES OF FLORIDA USING FSU'S SURVEY DATA. NUMBER OF SURVEYED REEFS INDICATED ABOVE BARS. ERROR BARS ARE SE.

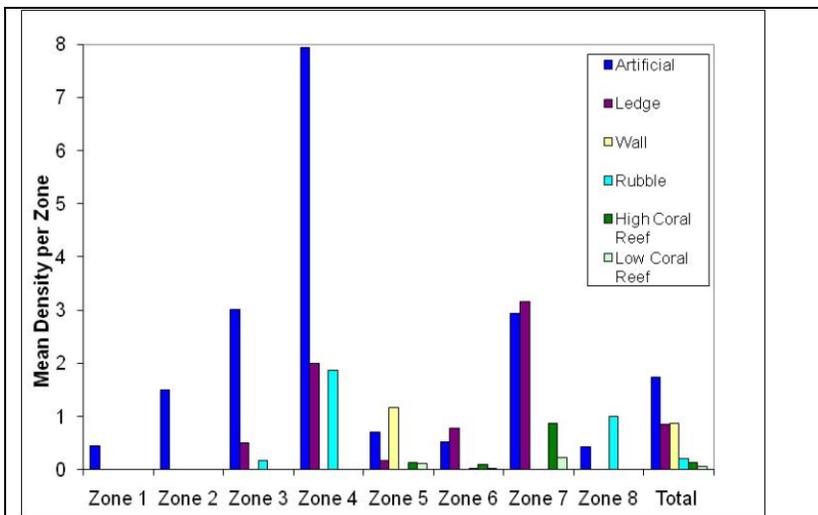
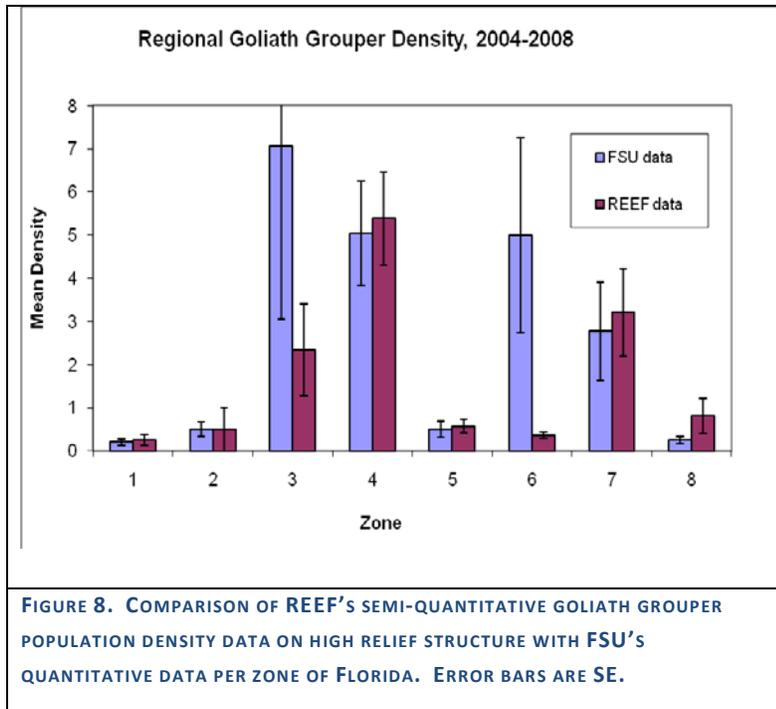


FIGURE 7. COMPARISON OF GOLIATH GROUPER DENSITIES ON REEF'S HABITAT TYPES IN THE EIGHT ZONES OF FLORIDA USING SURVEYS TAKEN OVER THE LAST FIVE YEARS (2004-2008). "TOTAL" REFERS TO THE OVERALL AVERAGE DENSITY IN EACH HABITAT TYPE FOR ALL ZONES.

Comparisons of densities on high relief sites among Florida zones using FSU’s data only (Figure 8) shows significantly higher densities in zones 3, 4, 6, and 7; and low densities in the other four zones. Importantly, spawning aggregations are known only from the high density zones. Comparison of goliath grouper densities on high and low relief reefs in zone 4 (where density is highest) show clearly that high relief is preferred (Figure 7). The fish particularly prefer reefs and wrecks that have an abundance of holes and caves sufficiently large to accommodate an adult goliath grouper.

The cover provided by structure is extremely important to the vast majority of reef fish. The larger the fish, the larger the cover required. Typically in high relief areas, there are high densities of fish. The only exception to this pattern for goliath grouper occurred in the Florida Keys, where, despite



there being ample high relief on the reef edge with numerous large holes and caves, there were few goliath grouper. This pattern extended to adjacent wrecks and artificial reefs, sites normally quite attractive to goliath grouper. It would appear that reef height is not the primary attractant to these fish in the Florida Keys.

We found high concordance between our quantitative data and REEF’s semi-quantitative data for adult goliath grouper population

density in 8 Florida zones (Figure 8, Table 2), despite the fact that REEF data collection involved divers whose expertise ranged from expert to inexperienced. Indeed, we found no significant difference in the data for most of the zones, specifically zones 1, 2, 4, 5, 7, and 8. Zones 3 and 6 stand because of the distinct differences between our data and that collected by REEF. In both cases, our estimates of population density exceed those derived from REEF’s data. Why this pattern exists is evident in Figure 5: our data came from the southern part of zone 3 where densities were higher, whereas REEF’s data came mostly from the northern part of zone 3 where densities are lower. Similarly, in zone 6, our data

Table 2. Regional sample sizes for REEF and FSU data on goliath grouper density on high-relief sites.

Zones	1	2	3	4	5	6	7	8
FSU sites	36	36	17	76	43	11	18	43
FSU surveys	38	44	17	161	45	11	19	43
REEF sites	15	6	36	55	63	190	25	20
REEF surveys	38	8	111	167	714	943	216	56

came from the higher density northern part of that zone, whereas REEF's data mostly came from coral reef habitat in the southern part of zone 6. As is clear, densities are relatively low in all coral reef habitat (Figure 6, Figure 7). These results provide support for our assumption that REEF's semi-quantitative data can be used to great effect in future goliath grouper stock assessments.

HABITAT ASSOCIATIONS.—Our evaluation of the composition and abundance of fish species associated with goliath grouper obtained from video surveys revealed a statistically significant ($P < 0.01$) positive relationship between the number of fish species (fish species richness) relative to the density of goliath grouper (Figure 9) on high relief sites in zone 4 (where goliath grouper densities were highest). We also found a significant ($P < 0.01$) positive relationship between the density of snappers (total number of all species present) and the density of goliath groupers (Figure 10). However, there was no significant relationship between the density of goliath grouper and the density of other groupers.

The species of snapper and the total number observed with goliath grouper were: gray snapper *Lutjanus griseus*, 1244; yellowtail snapper (*Ocyurus chrysurus*), 527; lane snapper (*Lutjanus synagris*),

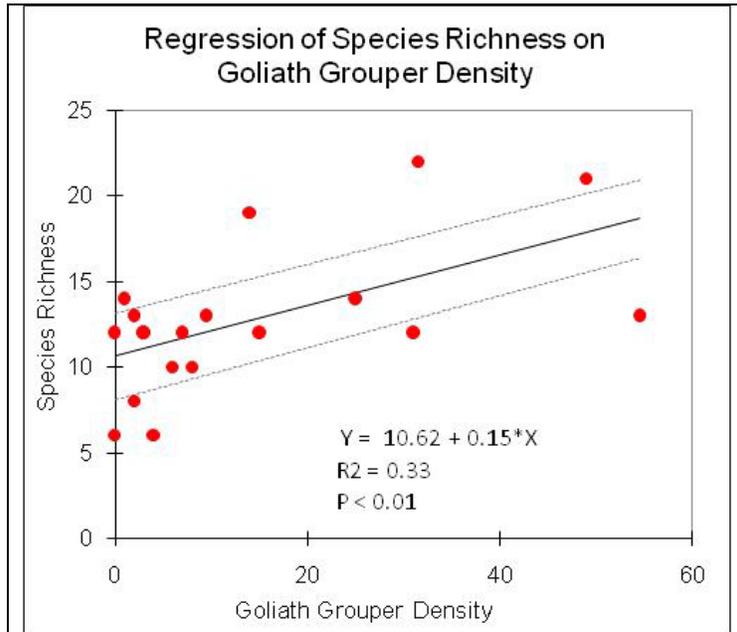


FIGURE 9. REGRESSION OF SPECIES RICHNESS (ALL REEF FISH) ON GOLIATH GROUPE DENSITY (NO. PER REEF SITE) IN FLORIDA ZONE 4 ON HIGH RELIEF STRUCTURE. DASHED LINES DENOTE 95% CONFIDENCE LIMIT.

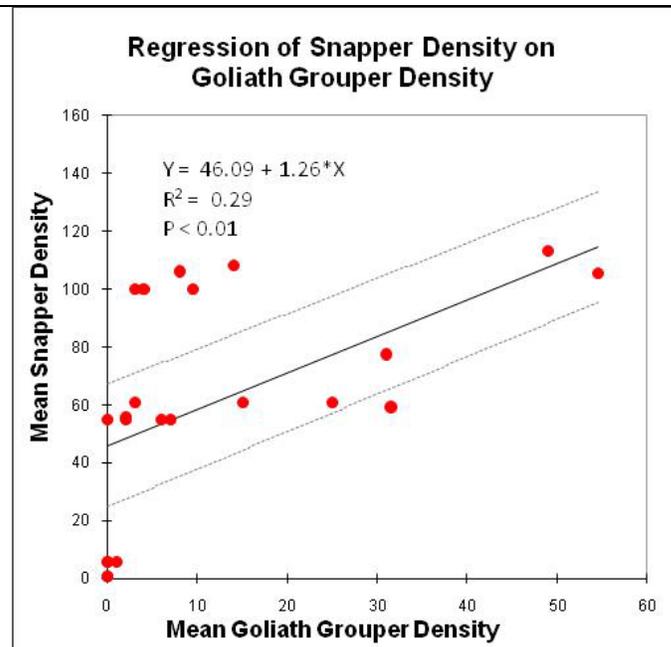
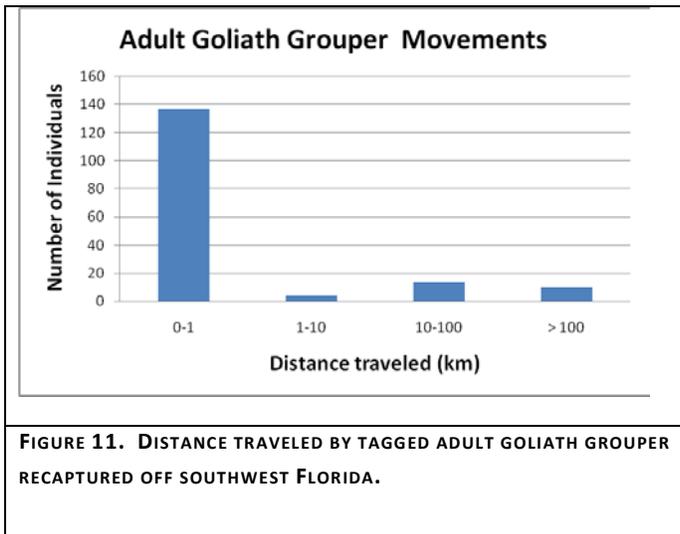
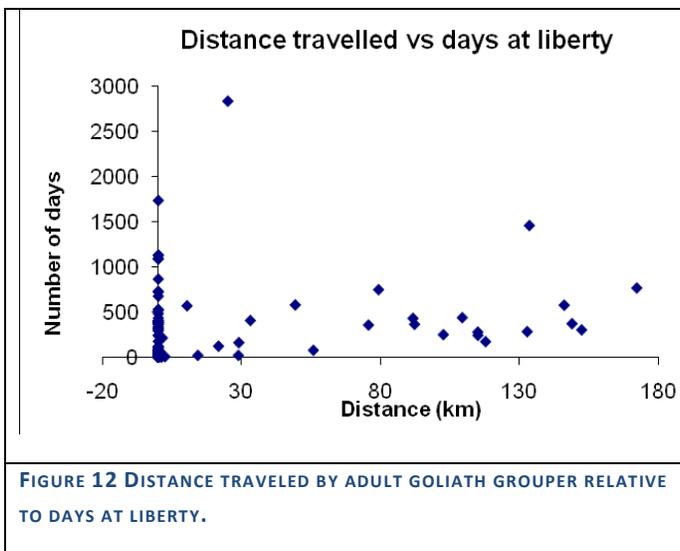


FIGURE 10. REGRESSION OF SNAPPER POPULATION DENSITY (NO. OF SNAPPERS PER REEF SITE) ON GOLIATH GROUPE DENSITY (NO. PER REEF SITE) IN FLORIDA ZONE 4 ON HIGH RELIEF STRUCTURE. DASHED LINES = 95% CONFIDENCE LIMITS.

247; red snapper (*Lutjanus campechanus*), 141; and schoolmaster (*Lutjanus apodus*), 36. The species of grouper and the total number observed with goliath grouper were: scamp (*Mycteroperca phenax*), 219; graysby (*Epinephelus cruentatus*), 121; red grouper (*Epinephelus morio*), 30; and gag (*Mycteroperca microlepis*), 25.



MOVEMENTS.--Goliath grouper adults move about very little. Eighty-two percent of recaptured adults moved less than one km in their time at liberty (Figure 11). Also, there is no clear pattern when comparing time at liberty to distance traveled (Figure 12). Some fish recaptured after years at liberty were either on the same site or not far from it. The maximum distance traveled by an adult was 175 km. Movements greater than about 1 km appear to be related mostly to migrations to and from spawning aggregations, but may also occur in relation to feeding aggregations. Some adult fish appear to move from spawning sites back to home sites many km away while others tagged in the mouth of Charlotte Harbor also showed travel distances of over 50 km (spawning site) (Figure 13).



Over the years we recorded recaptures adult goliath grouper that were tagged as juveniles in the mangrove habitat of the Ten Thousand Islands during our study in that habitat

(Koenig et al. 2007). Those fish displayed a range of movement patterns. One moved northwest to an area off Tampa, over 200 km away from the nursery. Most moved west or south, but one moved

around the tip of the Florida peninsula and was recaptured on the east coast near Indian River Lagoon (Figure 14).

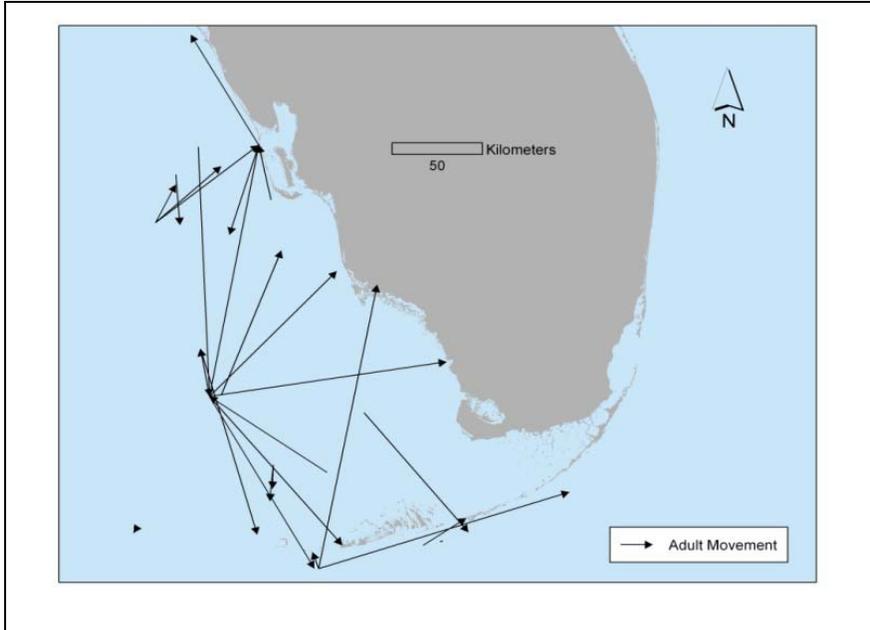


FIGURE 13. LONG-RANGE MOVEMENTS OF ADULT GOLIATH GROUPEE IN THE VICINITY OF A SPAWNING SITE (SW OF CHARLOTTE HARBOR) AND A FEEDING SITE IN CHARLOTTE HARBOR OFF SOUTHWEST FLORIDA.

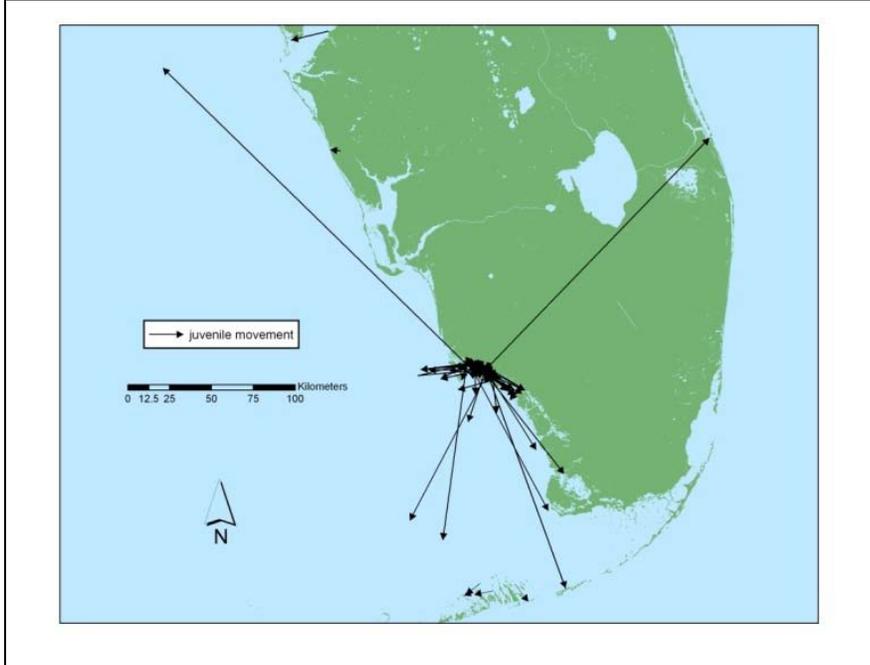


FIGURE 14. MOVEMENT OF JUVENILE GOLIATH GROUPEE FROM THE TEN THOUSAND ISLAND AREA TO OFFSHORE ADULT LOCATIONS.

DISCUSSION OF DISTRIBUTION, REGIONAL DENSITY, MOVEMENTS, AND HABITAT PREFERENCE

In the southeastern U.S., the dominant goliath grouper population occurs off Florida. All the data presented here indicate that population densities are quite low in north Florida on both Gulf and Atlantic coasts (zones 1, 2, and 8; Florida Panhandle, Big Bend and off Jacksonville, respectively) and very sparse in other states. Over time, we suspect that the population may expand to the northern parts of Florida due to recovery, but there are no records of historical population sizes in the region prior to intense fishing, and a primary factor inhibiting expansion is low winter temperatures. Goliath grouper avoid temperatures below 15° C (Sadovy and Eklund 1999), and in the Panhandle, inshore segments of the goliath grouper population move to deeper reefs when inshore water temperatures decline (Koenig personal observation). Thus, northern populations may never achieve densities as high as those in the south except in the face of climate change.

Goliath grouper recovery started in southwest Florida around the Ten Thousand Islands, an area of prime juvenile habitat (Koenig et al. 2007). There was a pulse in the juvenile goliath grouper population in southwest Florida between 1994 and 1997, four to seven years after the fishery closure, based on recreational fishing data from the Everglades National Park (Cass-Calay and Schmidt 2008). Following that, there was a clear and dramatic increase in the adult goliath grouper population off south Florida by 1998, based on REEF's recreational scuba surveys. The vast majority of juveniles exceeded 400 mm TL, a size at which most fish were likely 4 to 6 year olds and nearing time of migration from mangrove to offshore reef sites (Koenig et al. 2007). It is highly likely that this pulse of juveniles accounts for the increased observations of adults several years later.

HABITAT.--Adult goliath grouper prefer habitat that provides shelter like holes, caves, and other large structure (Sadovy and Eklund 1999), so adults are commonly found around high-relief ledges and wrecks (Smith 1976). Data in this study confirm that, but also show that this characteristic does not extend to coral reef habitat, even when it provides ample high-relief shelter. Apparently, structural features are not sufficient to support high densities. Although we have no tested observation, we suspect that low densities of goliath grouper on coral reefs may be related to low availability of food resources. For example, it is possible that the primary food of goliath grouper, crabs, are low in abundance in coral reef habitat and/or inaccessible because of the extreme rugosity of coral reef habitat.

Adult goliath grouper typically occur in water depths < 40 m, although there are reports of individuals as deep as 70 to 90 m (Sadovy and Eklund 1999, Kevin Rademacher, NMFS, Pascagoula, personal observation). In Florida, the vast majority of adults occur at depths < 60 m deep (Michael Barnette, Association of Underwater Explorers, personal observation). Juveniles smaller than 1 m are typically restricted to the shallow productive waters of estuaries (primarily mangroves) (Koenig et al. 2007), and are rarely seen on the shelf.

Goliath grouper are known to alter habitat by excavating sediment-smothered ledges and artificial reefs (Felix-Hackradt and Hackradt 2008, C Koepfer, Lee Co. FL Artificial Reef Program pers.

comm, W Stearns Underwater J pers. comm.). This behavior has not been studied in detail, but it is clearly beneficial to goliath grouper and other species occupying the habitat by providing enhanced architecture, i.e., holes and caves, and the cover it provides. Another excavating species, red grouper (*Epinephelus morio*), also excavates rocky habitat (Coleman et al. 2009). It is possible the these two congeneric and sympatric species potentiate each other's positive effects on exposing reefs buried by storms and thereby provide habitat for myriad other species requiring high-relief rocky reefs.

ASSOCIATED SPECIES.—Because goliath grouper adults are indigenous, large, and can dominate the biomass of the reef fauna, they likely play a dominant role in shaping reef communities within their range. We found a significant positive relationship between goliath grouper density and the biological diversity of other fish species using data from our video surveys. It may be that their excavating activities enhance this positive effect on biodiversity, as Coleman et al. (2009) have demonstrated for red grouper, from shallow bay nursery habitat to shelf-edge spawning habitat. The driving force may well be the increase in the spatial complexity of the habitat. Further, we found a significant positive relationship between the density of a number of different snapper species and the density of goliath grouper. This is born out again in our diet studies (section III), wherein we show that goliath grouper are not top level predators but, rather, feed mainly on crabs and other crustaceans.

DENSITY DATA.—REEF semi-quantitative goliath grouper population density data provides a convenient measure of mean regional density of adult goliath grouper in Florida for future stock assessments. We found no reason to suspect the quality of the REEF data related to goliath grouper for several reasons. First, whether someone is an expert or inexperienced diver, the probability of misidentifying a goliath grouper is very low. Second, we compared the density estimates we made using REEF data to those we obtained ourselves—data that was carefully and systematically collected by a single team of professionally trained divers using statistically sound and standardized methods, and found a high degree of agreement between the two data sets. The two data sets are concordant in all regions except in the zones where both low and high densities exist (zones 3 and 6), but the differences in those zones can be explained by differences in geographical effort.

MOVEMENTS.—Goliath grouper move very little from their home reefs. Indeed, 82% of our recaptures moved less than 1 km. As juveniles, goliath grouper show even less movement and can occupy the same 160 m of mangrove shoreline for several years (Koenig et al. 2007). The only significant long-range movement patterns we observed were (1) movements of juveniles from their nursery habitat to offshore adult habitat after reaching 5 to 6 years of age, (2) movement of adults to feeding locations in inlets, and (3) movements of adults to and from spawning sites. Adults apparently also respond to density-dependent factors, moving from high density areas, such as those in southwest Florida adjacent to juvenile mangrove habitat, to lower density areas elsewhere in the state. Recovery would therefore depend on emigration from regions of productive nursery to regions where their population is sparse, such as north Florida. It has been suggested that the goliath grouper population should be “thinned” in areas of high density, such as southwest Florida by many fishers, but this would

likely retard recovery in other areas of the southeastern U.S. by slowing emigration from high density areas. A much better idea is to allow recovery of the goliath grouper population throughout its former range and to restore mangrove habitat where it has deteriorated to allow expansion of functional nursery for goliath grouper and provide habitat for the myriad other species that depend on it.

PART II: DEMOGRAPHICS & ECOLOGICAL CHARACTERISTICS



FIGURE 15. GOLIATH GROUPEE ON STRETCHER ABOVE BOAT GUNWALE AND BELOW TWO DAVITS. THE FISH'S EYES ARE PROTECTED FROM DIRECT SUNLIGHT AND A GILL IRRIGATION HOSE IS IN ITS MOUTH.



FIGURE 16. GOLIATH GROUPEE BEING RELEASED FROM AN INFLATABLE BOAT AFTER SAMPLING AND TAGGING.

MATERIALS AND METHODS

DEMOGRAPHIC METHODS

Demographic features of interest for goliath grouper are individual size, age, sex ratio, and reproductive condition; however we obtained mostly regional size information because the other demographic data required capture of the fish and this proved more challenging than anticipated. Nevertheless, we have developed successful methods of capture and holding goliath grouper during the course of this project.

Capture required using large circle hooks, 600 lb test monofilament leader, a 1.0 cm diam. braided nylon hand line, and cut or live bait. We attached a large (60 cm diameter) float to the hand line to suspend bait just off the bottom. The float submerged when a fish hit the hook, and popped to the surface repeatedly until the fish tired. We typically waited from 3 to 5 min before bringing the fish to the surface to ensure exhaustion. This is important because an active fish can exert forces that are dangerous to themselves, to researchers, and to equipment onboard the vessel. Fish are vented before being brought onboard. Venting occurs either at the

surface or, if the fish is caught in water depths exceeding 25 m, at about 10 m depths, thereby minimizing the sometimes lethal effects of barotrauma, or expansion of swimbladder gas caused by rapid pressure changes. At the surface, the fish is guided onto a stretcher, strapped in place with Velcro straps, and then hoisted above the gunwale with two davits (Figure 15). Gills are irrigated with seawater supplied by an overboard bilge pump connected to a hose inserted into the fish's mouth. The fish's eyes are covered to protect them from direct sunlight. When no davits are available, we haul fish into a 3.5 m inflatable boat that is half filled with seawater. Either method provides a means of sampling, tagging, and releasing fish (Figure 16).

We determined size (cm total length, TL) of individual goliath grouper either directly from captured fish or indirectly from digital videos taken underwater. The latter technique provided a way to measure fish harmlessly and rapidly, and was especially useful for measuring fish in spawning aggregations. It required use of a video camera mounted with a laser device consisting of two green lasers adjusted to



FIGURE 17. GOLIATH GROUPER UNDERWATER WITH GREEN LASERS DOTS PROJECTED ONTO SIDE. LASERS ARE ARRANGED IN PARALLEL AND 10 CM APART.

produce parallel beams 10 cm apart when projected on the fish's lateral side (Figure 17). We used green lasers because they penetrate further in the water and are brighter than red lasers.

Currently, the most common method of aging marine fish is to use otoliths or "ear stones" from the otic region of the skull, requiring that the fish is killed. For protected fish species such as goliath grouper, this lethal aging process is contradictory to the aim to conserve these populations. Dorsal fin elements provide an accurate non-lethal alternative to using otoliths in goliath grouper; dorsal fin spines are useful for juveniles

(Brusher and Schull 2009) and dorsal fin rays provide accurate age estimates in adults (Murie et al. 2009) (Figure 18).

Dorsal fin rays 2 and 3 were cut at the base of the fin and stored in the freezer. In the laboratory the fin rays were cleaned of any soft tissue and fat and allowed to air dry. The dried rays were then embedded in epoxy resin and thin-sectioned (~0.5-0.8 mm thick). Sections were mounted on glass slides within Flo-texx liquid cover slip medium and examined at 100 x under a compound microscope with images projected by camera onto a video monitor to facilitate resolution of readings.

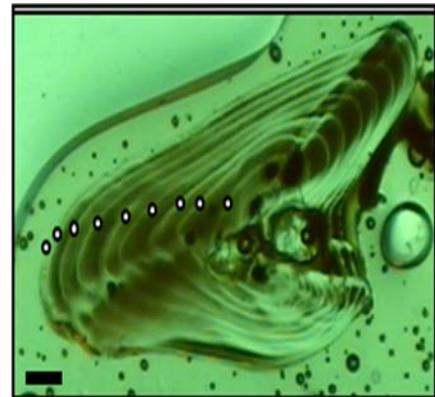


FIGURE 18. A CROSS SECTION OF THE DORSAL FIN RAY OF A 9 YEAR-OLD GOLIATH GROUPER (MURIE ET AL. 2009).



FIGURE 19. RESEARCHER SAMPLING STOMACH CONTENTS OF A CAPTURED GOLIATH GROUPER BY REACHING A GLOVED HAND DIRECTLY INTO THE STOMACH.

TROPHIC METHODS

Stomach content samples were collected from hook-and-line-captured fish in south Florida. Juvenile stomach contents were sampled by gastric lavage, pumping water into the stomach through a 2-cm diameter PVC tube connected to a small bilge pump, and collecting stomach contents flowing out of the mouth with a 3-mm mesh net placed under the head. Adult stomachs were sampled directly. The mouth was held open by inserting a large (15-cm diameter x 35-cm long) aluminum tube between the jaws, allowing the researcher to reach through the tube to remove stomach contents by hand. The stomach contents of each individual were bagged and treated with ~ 50 ml of 10% formalin to arrest digestion and preserve contents. In the laboratory, contents were rinsed, blotted dry, weighed to the nearest 0.1 gm, measured, identified to the lowest possible taxon, and enumerated. We also estimated the intact size of predigested food items (primarily blue crabs, mud crabs and shrimp) from measurements of whole specimens of the same species. Non-dietary items such as algae, shells and debris, while noted, were excluded from diet analyses.

In addition to stomach content analyses, which provide a list of regional prey species and their relative proportion in the diet, we analyzed stable isotope patterns in muscle tissue to estimate long-term trophic status. Stable isotopes (particularly carbon, C, and nitrogen, N) are used to trace food linkages within and between ecological systems (Peterson and Fry 1987, Wada et al. 1991). Isotopes, inherent parts of all biological material, vary proportionally in biological systems due to metabolic fractionation which occurs during carbon fixation in plants and because plants take up inorganic constituents from different reservoirs that vary in isotopic composition. While ^{13}C reflects diet after multiple trophic transfers, ^{15}N shows a 3‰ increase per trophic level up the food web, so information about the trophic level of the predator and prey can be used to interpret trophic dynamics and trace carbon flow. Thus, the ratios of C and N isotopes give valuable information about trophic position of the fish in the food web.

We ran ^{13}C and ^{15}N analyses on muscle tissue removed from the base of spines and rays of goliath grouper collected for aging purposes, and from stomach contents collected for diet studies and preserved in formalin. (For preserved crustacean samples, we removed a small square of exoskeleton and attached tissue either from the claw or dorsal side of the carapace, depending on size and tissue availability, and allowed the formalin to evaporate under a fume hood for at least 24 hours.)

Prior to analysis, tissue samples were dried at 50° C. All tissue samples were then dried in an oven for at least 24 hours, homogenized using an electric grinder, and placed in micro centrifuge tubes before storing in a freezer for subsequent analysis. Subsamples (500 mg) were removed from centrifuge tubes with a clean spatula, placed in tin cups, and then analyzed for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values using a Finnigan Delta Plus mass spectrometer connected to a CHN elemental analyzer³. Isotope ratios are expressed in delta (δ) notation, defined as the parts per thousand (‰ or “per mil”) deviation from a standard material. For nitrogen, the standard is atmospheric nitrogen, set at 0‰, such that:

$$\delta^{15}\text{N} = ([R_{\text{sample}}/R_{\text{standard}}] - 1) * 1000,$$

where R is $^{15}\text{N}/^{14}\text{N}$. In the case of carbon, the standard is PeeDee Belemnite;

$$\delta^{13}\text{C} = ([R_{\text{sample}}/R_{\text{standard}}] - 1) * 1000,$$

where R is $^{13}\text{C}/^{12}\text{C}$. The standard error for our repeated measurements was ± 0.1 ‰.

Specimens preserved in formalin exhibited ^{13}C value depletions of approximately 1.1‰, whereas ^{15}N values were enriched by 2‰ (Edwards et al. 2002, Kelly et al. 2006). Therefore, we adjusted the mass spectrometer values to account for these changes.

BIOENERGETICS MODEL

Dr. Paul Richards (NMFS, Miami) developed a bioenergetics model from our existing dietary data (n = 242 juvenile and adult stomachs, sampled from south and southwest Florida). The model includes: (1) dietary mass and composition, (2) feeding patterns, and (3) overall energetic needs for growth and reproduction. Such a bioenergetics model in combination with adult population size estimates will allow development of estimates of the impact of the goliath grouper population on forage species.

Richards used a generalized bioenergetic model (e. g., Kitchell et al. 1977) such that:

$$\frac{dB}{Bdt} = C - (E + U + M) \quad \text{Eq. 1}$$

where B is body mass, dB/Bdt is the specific growth rate in body mass in units of body mass change mass⁻¹ (t⁻¹), C is total specific consumption rate in mass of all prey, E is mass lost to egestion, U is mass lost to excretion, and M is mass lost to metabolic costs. Basic physiologic rates (e.g., E, U, and M) of goliath grouper are not currently available, so we used known values for length and body-mass-to-length estimates (Bullock et al. 1992) to estimate growth rate, and then calculated the sum of E, U, and M using eq. 1. Determining this value at different ages and sizes allows us to develop a general

³ All isotopic analyses were conducted at the Florida State University High Magnetic Laboratory

allometric relationship between age (or size) and E, U, and M combined, and a consumption rate (C) for any size, or a growth rate (dB/Bdt) for any consumption rate.

We also estimated total consumption rate (C) using maximum and minimum boundaries for E, U, and M from the literature on other groupers or other similar fish. Total C could be estimated for each region, broken down by region j and prey species (or functional group) i such that:

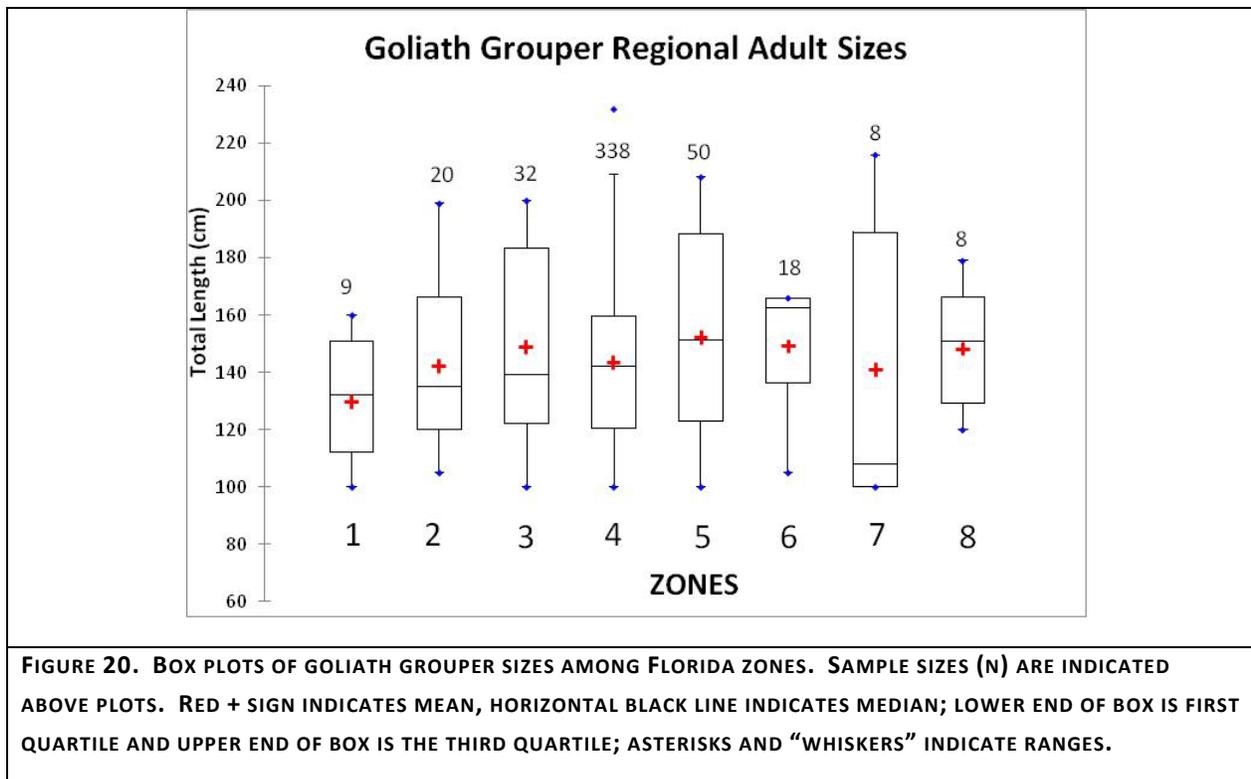
$$C_j = C_j \sum_i e_{ji} \tag{Eq. 2}$$

where e_{ji} is the proportion of grouper diet by region j and prey functional group i, and C_j is total consumption by region j.

RESULTS

DEMOGRAPHIC RESULTS

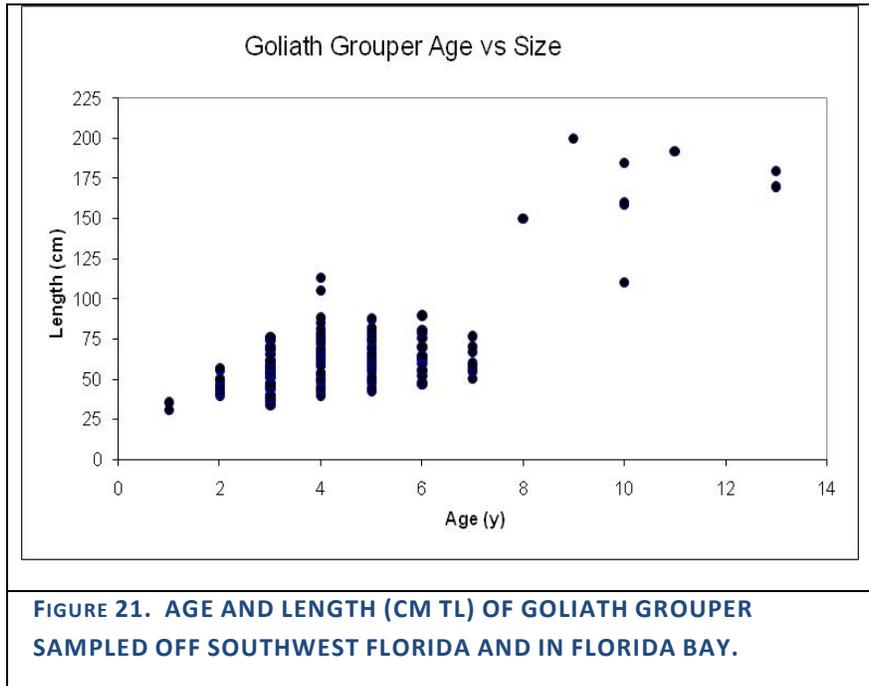
Regional size distributions of goliath grouper (Figure 20) determined from our laser-video and



capture surveys indicate similar mean sizes throughout all regions (Kruskal-Wallis, $p > 0.05$). Size range was greatest in region 4, the region containing the Ten Thousand Islands area and considered the goliath grouper center of abundance (Koenig et al. 2007). This likely results from frequent recruitment

of juveniles to the offshore adult population and large breeders remaining in the area where we observed the most spawning aggregations, but it could also be the result of a large sample size.

We found significant variation in the in size at age in juveniles (<100 cm TL) collected in the Florida Bay mangrove habitat in this study (Figure 21), mimicking our previous results for the Ten Thousand Islands (Koenig et al. 2007).



TROPIC ANALYSIS

We conducted trophic analyses on adults captured in south Florida from both the east and west coasts (area 1; N=22, 12 with stomach contents), and juveniles captured in Florida Bay (area 2; N = 224, 179 with stomach contents).

Goliath grouper juveniles predominately fed on invertebrates (~69 % of stomach content weight, ~80 % of the occurrences) (Table

3) and are significantly less dependent on fishes as part of their diet (~31 % of the total weight, 20% of the occurrences) (Table 4). Xanthid crabs (mud crabs) predominated in the diet, making up 35% of the prey items by weight (51% of the invertebrates) and about 34 % of occurrences (44 % of the invertebrates). Of the vertebrates, the gulf toadfish *Opsanus beta*—a slow moving, bottom-associated fish—occurred most frequently (4 %) whereas grunts (Family Haemulidae, several species) made up the largest percentage of fish by weight (13 %).

A similar pattern occurs in adults which feed primarily on invertebrates (~70% of the total weight of prey items, 72 % of the total occurrences). Where mud crabs predominated in juvenile diets, swimming crabs (Portunidae) were most prevalent in the adult diet (36 % by weight, 43 % by frequency of occurrence). When just investigating the invertebrate component, a slightly different pattern emerges, wherein box crabs (Calappidae) are most prevalent by weight (52%) and swimming crabs by occurrence (60 %). Fishes comprised approximately 30 % of total weight and 28 % of total occurrences (Table 5).

Table 3. Frequency of occurrence and mass of invertebrate prey found in the stomachs of juvenile goliath grouper (*Epinephelus itajara*) collected from Florida Bay. Prey items are categorized by family and common name.

Family, Inverts	Common Name	Occurrence	%		
			Occurrence	Mass (g)	% Mass
Xanthidae	mud crabs	107	34.19	2074	34.7928
Portunidae	swimming crabs	74	23.64	1649.1	27.6648
Penaeidae	pink shrimp	29	9.27	34.9	0.5855
Majidae	spider crabs	27	8.63	225.1	3.7762
Calappidae	box crabs	6	1.92	76.9	1.2901
Grapsidae	grapsid crabs	2	0.64	4.1	0.0688
Alpheidae	snapping shrimp	1	0.32	0.1	0.0017
Loliginidae	squid	1	0.32	<0.1	<0.001
Marginellidae	snail	1	0.32	0.1	0.0017
Palinuridae	spiny lobster	1	0.32	56.1	0.9411
Porcellanidae	porcellin crab	1	0.32	10.4	0.1745
Potamididae	snail	1	0.32	0.4	0.0067
Total		251	80.21	4131.2	69.30

We found no clear differences between adult and juvenile diets when considering broad prey categories, although this could be biased by our small sample size for adults. When considering both goliath grouper life stages combined (juveniles plus adults), the composition of the diet was 72% invertebrates, primarily crabs (63%). The next largest prey category was unidentified fish (13%) (Figure 22). Appendix table A4 gives the scientific names of all identified stomach contents.

Table 4. Frequency of occurrence and mass of fish prey found within the stomachs of juvenile goliath grouper (*Epinephelus itajara*) collected from Florida Bay. Prey items are categorized by family and common name.

Family, fish	Common Name	Occurrence	% Occurrence	Mass (g)	% Mass
Batrachoididae	toadfish	11	3.51	178.8	2.9995
Lutjanidae	snappers	8	2.56	342.9	5.7524
Cyprinodontidae	killifish	7	2.24	50.6	0.8489
Diodontidae	burrfish	6	1.92	35.1	0.5888
Haemulidae	grunts	6	1.92	773.5	12.976
Mugilidae	mullet	6	1.92	273.4	4.5865
Ostraciidae	boxfish	5	1.59	33.1	0.5553
Monacanthidae	filefish	4	1.28	0.1	0.00168
Sparidae	porgies	4	1.28	137.7	2.31
Sphraenidae	barracuda	2	0.64	1.4	0.0235
Atherinopsidae	silversides	1	0.32	0.6	0.0101
Clupeidae	herring	1	0.32	2.6	0.0436
Ogcocephalidae	batfish	1	0.32	0	0
Total		62	19.82	1829.8	30.69

Table 5: Occurrence and mass of fish and invertebrate prey in stomachs of adult goliath grouper. Prey items categorized by family.

Family	Occurrence	% Occurrence	Mass (g)	% Mass
Calappidae (box crab)	4	28.5	304.5	33.60
Clupeidae (herring)	1	7.1	< 1	< 1
Dasyatidae (ray)	1	7.1	< 1	< 1
Diodontidae (burrfish)	2	14.2	275.6	30.41
Portunidae (swimming crabs)	6	42.8	326.1	35.98

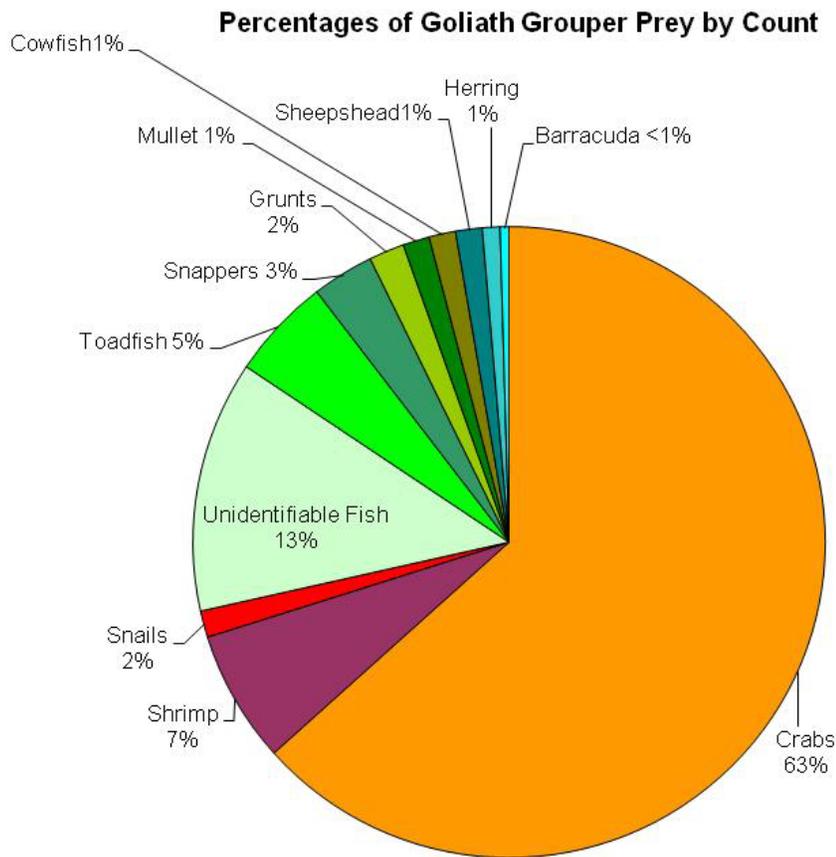


FIGURE 22. PIE DIAGRAM SHOWING MAIN PREY ITEMS OF GOLIATH GROUPEr DIET, ADULT AND JUVENILES COMBINED.

Stable isotopes: Goliath grouper muscle tissue ^{13}C values ranged from -13 ‰ to -8.9 ‰ (mean = -10.66 ‰), while the ^{13}C values for their prey ranged from -19.3 ‰ to -12.3 ‰ (mean = -16.4 ‰), after adjusting for the effects of the formalin preservation. Goliath grouper ^{15}N values ranged from 9.5 ‰ to 11.7 ‰ (Mean = 11 ‰), while the ^{15}N values for their prey ranged from 2.3 ‰ to 6.6 ‰ (mean = 5.3 ‰) (Table 6, **Figure 23**).

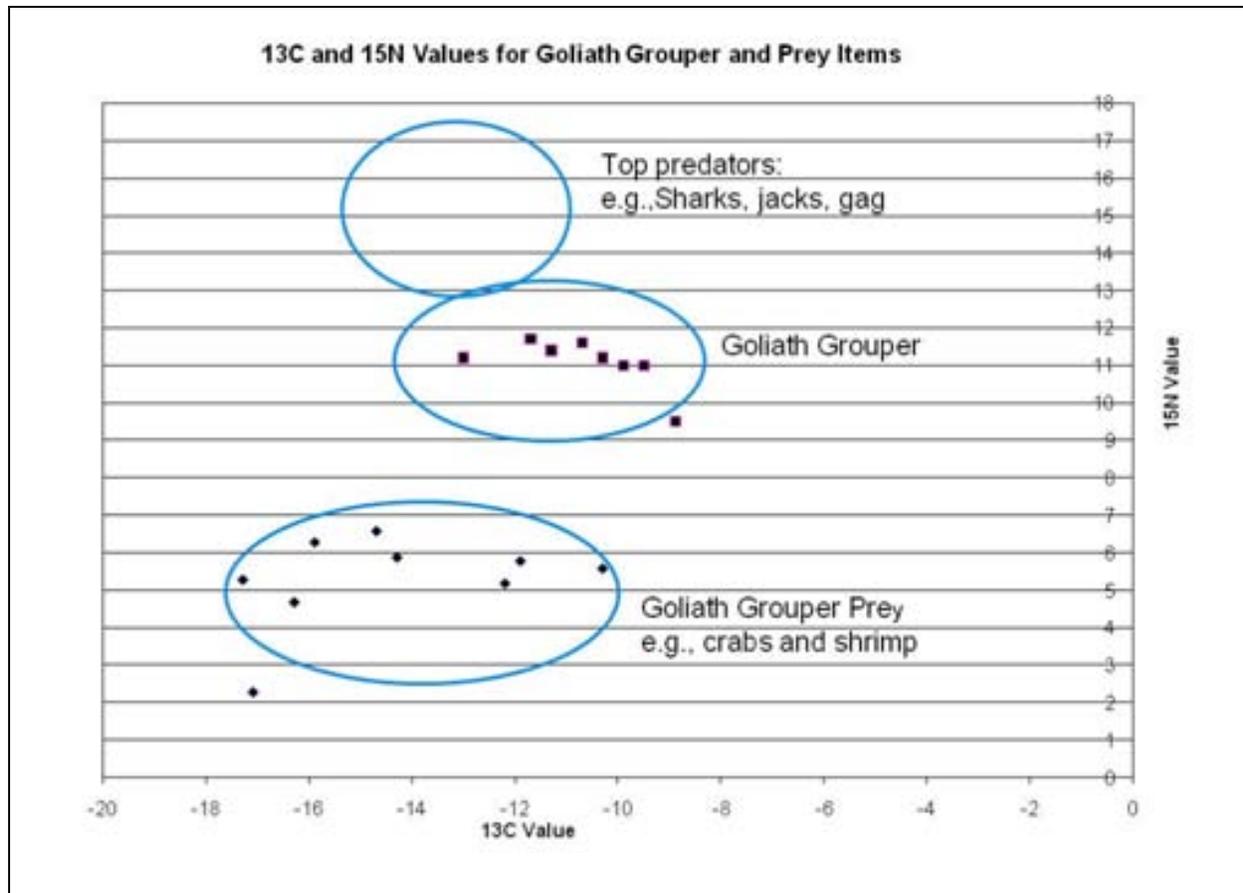


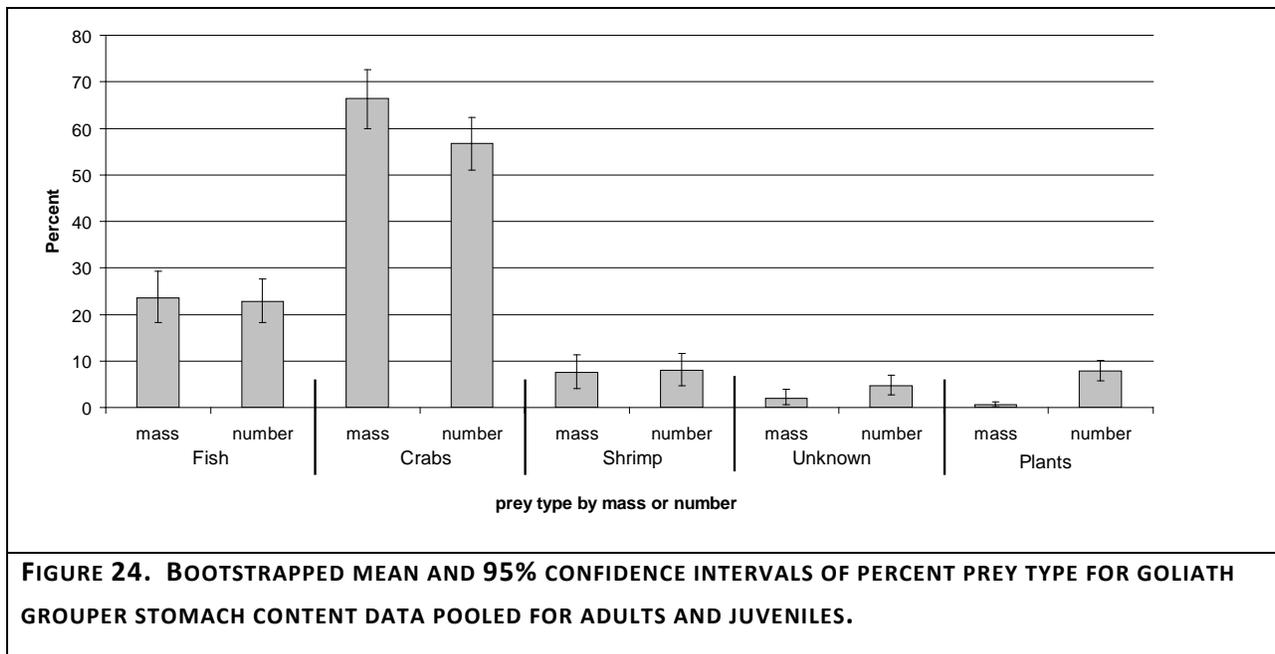
FIGURE 23. STABLE ISOTOPE VALUES OF ^{15}N AND ^{13}C OBTAINED FOR GOLIATH GROUPER (*EPINEPHELUS ITAJARA*) MUSCLE TISSUE (MEAN ^{13}C VALUE = -10.7 ‰, MEAN ^{15}N = 11.0 ‰) AND TISSUE TAKEN FROM PREY ITEMS FOUND WITHIN STOMACH CONTENTS (MEAN ^{13}C VALUE = -16.4 ‰, MEAN ^{15}N = 5.3 ‰). THE THIRD CATEGORY SHOWS THE TYPICAL VALUES FOR TOP-LEVEL PREDATORS (MEAN ^{13}C VALUE = -12.0 - 17.0 ‰, MEAN ^{15}N = 12.0 – 15.0 ‰) IN FLORIDA BAY; GOLIATH GROUPER FEEDS AT A TROPHIC LEVEL SIMILAR TO PINFISH OR GRAY SNAPPER (CHASAR ET AL. 2005).

Table 6: Stable isotope (^{15}N and ^{13}C) values for goliath grouper muscle tissue and tissue taken from prey species in stomach contents. Values given in ‰.

Species of Prey	Common Name	15N	13C	Goliath Grouper	15N	13C
<i>Eurypanopeus depressus</i>	Mud crab	4.7	-18.3	1	11.2	-13
<i>Eurypanopeus depressus</i>	Mud crab	6.3	-17.9	2	11.4	-11.3
<i>Eurypanopeus depressus</i>	Mud crab	6.6	-16.7	3	11.7	-11.7
<i>Callinectes sapidus</i>	Blue crab	5.2	-14.2	4	9.5	-8.9
<i>Callinectes sapidus</i>	Blue crab	5.8	-13.9	5	11.2	-10.3
<i>Mithrax spinosissimus</i>	Spider crab	2.3	-19.1	6	11.6	-10.7
<i>Mithrax spinosissimus</i>	Spider crab	5.3	-19.3	7	11	-9.5
<i>Mercenaria mercenaria</i>	Stone crab	5.9	-16.3	8	11	-9.9
<i>Panulirus argus</i>	Spiny lobster	5.6	-12.3	-	-	-
Mean		5.3	-16.4		11.1	-10.6

We created five prey-type categories--fish, crabs, shrimp, other animals, and plants. Proportion by mass and proportion by number of prey items were calculated per stomach. Bootstrapped or transformed group means were estimated from only those stomachs that contained prey.

For the two groups with stomach contents--adults in area 1 (Florida south east and west coasts; N=12), and juveniles in area 2 (Florida Bay; N=173) proportion data were arcsine square root transformed (following Zar 1984) and compared using general linear models by prey type, and proportion mass and number. The groups were significantly different in the proportions of “other animals” for both mass ($F = 12.32, p < 0.01$) and number ($F = 3.90, p = 0.0497$), and plants for proportion by mass only ($F = 13.65, p < 0.01$). No other prey types and proportions exhibited significant differences between adults and juveniles (or areas). Based on this mixed result, we evaluate the adult and juvenile stomach data both separately and pooled in the consumption model (described below). Adult, juvenile, and pooled means, medians, and confidence intervals were determined from a bootstrap ($n = 10,000$) of the per stomach data for unadjusted percent mass and number (Figure 24, Table 7, pooled data only).



A power analysis (glm power) suggests that a sample size of > 350 stomachs would be needed to find a significant difference ($\alpha = 0.05, \text{power} > 0.7$) between adult and juveniles, given the observed standard deviation. This type of power analysis is probably inappropriate because we used the prospective power routine of SAS intended for study design, and this retrospective use of power calculations is losing favor (Hoenig and Heisey 2001).

In addition to recommending the use retrospective power analysis in fish diet studies, Ferry and Cailliet (1996) suggested examining the cumulative prey curves, postulating that sufficient sample size to characterize diet is achieved when the cumulative prey curve reaches an asymptote. Cumulative prey curves for the adult and juvenile do not quite reach an asymptote when prey are categorized at the family taxonomic level (Figure 25). These curves were developed from the stomachs in the order they were numbered in the database, not randomized as suggested by Ferry and Cailliet (1996). We suggest that randomization would have little effect on the overall pattern.

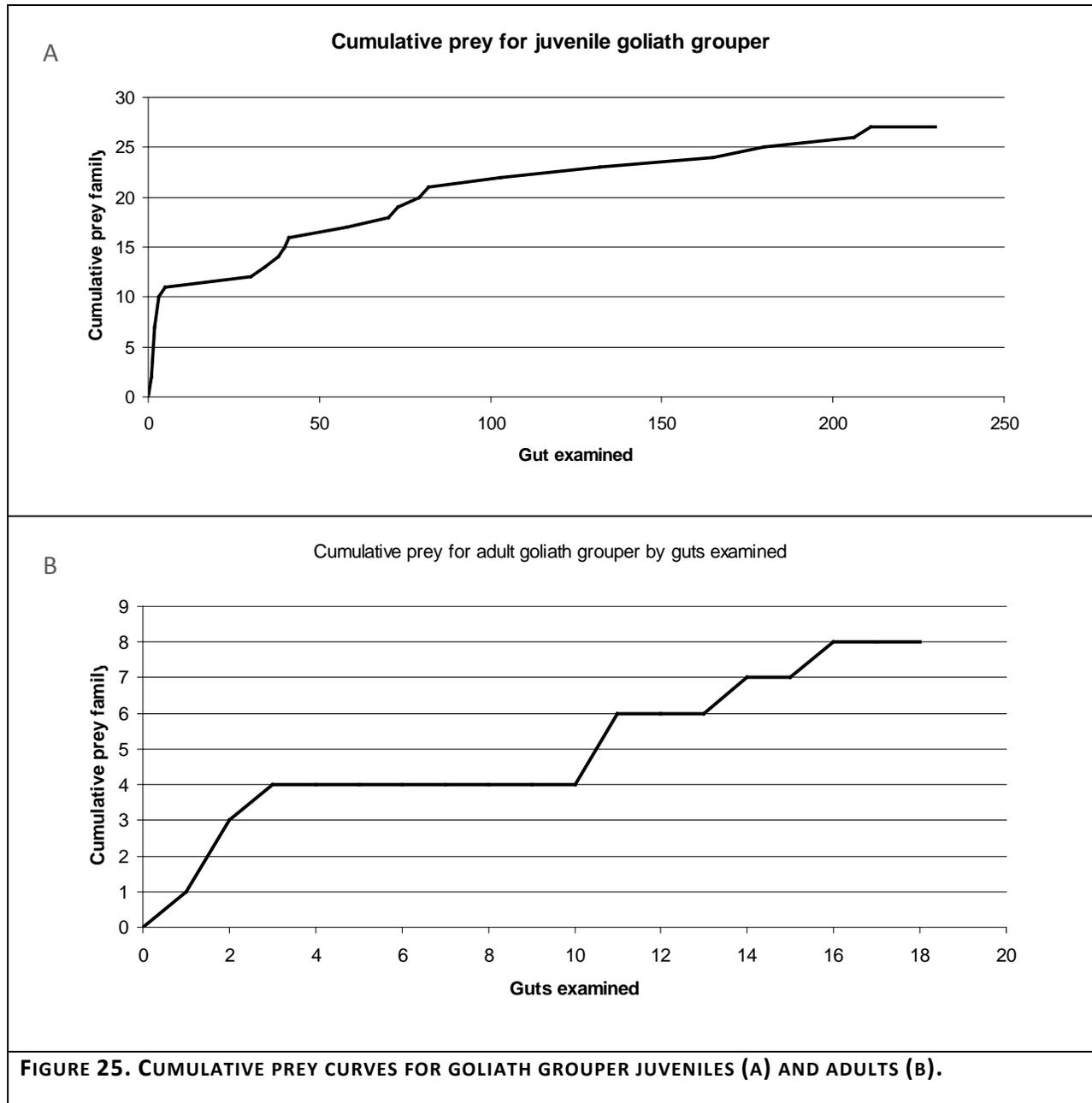
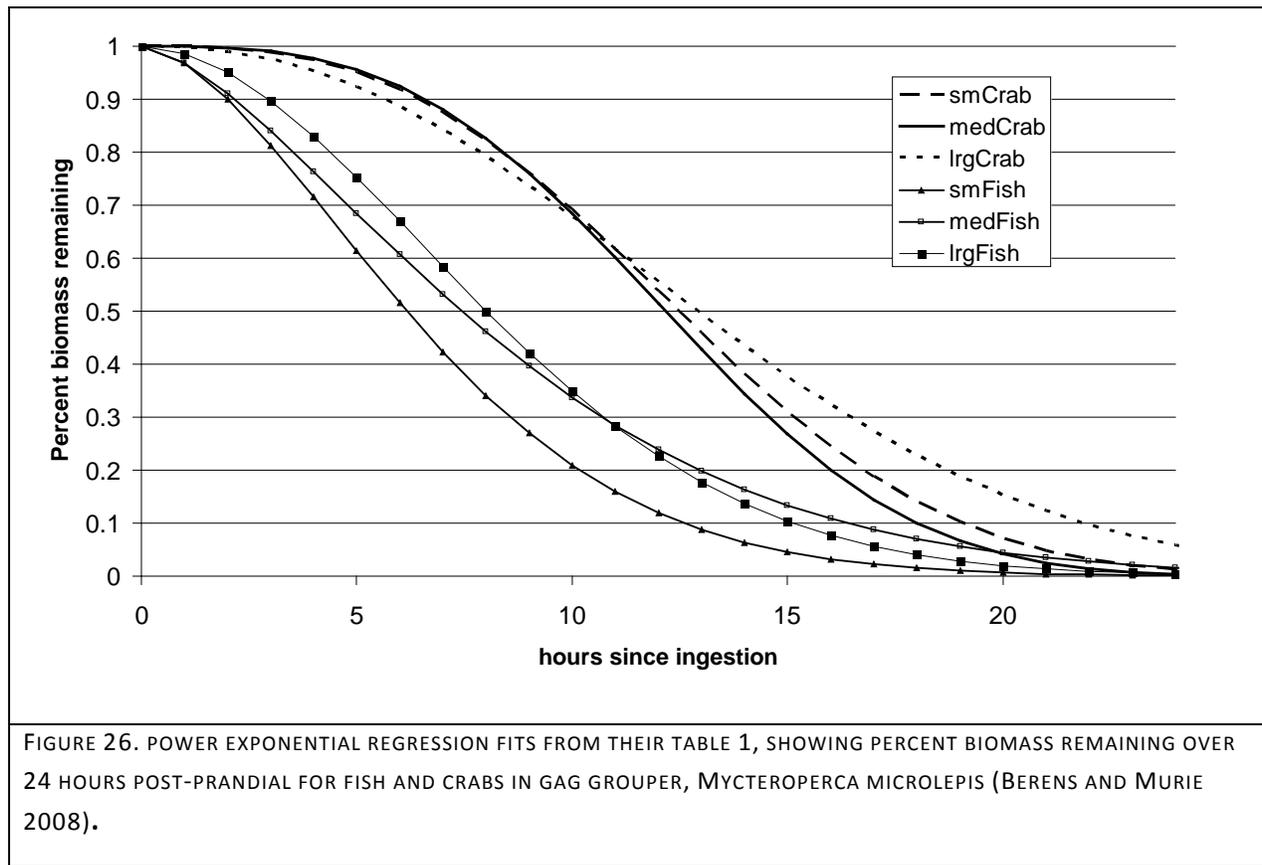


Table 7: Bootstrapped mean, median, and lower (LCI) and upper (UCI) 95% confidence intervals for adults, juveniles, and pooled data, percent by mass and count of prey type.

Prey Type	Adult				Juveniles				Pooled			
	mean	media	LCI	UCI	mean	median	LCI	UCI	mean	median	LCI	UCI
fish	17.3	16.7	3.3	36.7	23.9	23.9	18.3	29.9	23.6	23.6	18.2	29.3
crabs	65.4	65.8	41.7	87.5	66.5	66.6	59.9	73.0	66.3	66.3	59.9	72.6
shrimp	0.0	0.0	0.0	0.0	8.1	8.0	4.5	12.0	7.5	7.4	4.1	11.2
other	12.7	11.7	3.3	25.0	1.3	1.2	0.1	3.0	2.0	2.0	0.6	4.0
plants	4.5	4.2	0.0	12.5	0.2	0.2	0.1	0.3	0.5	0.5	0.1	1.2
Percent by count												
fish	17.5	16.7	3.3	36.7	23.1	23.1	18.5	28.0	22.8	22.8	18.3	27.5
crabs	65.4	65.8	40.8	87.5	56.2	56.2	50.4	62.1	56.7	56.7	5.0	62.4
shrimp	0.0	0.0	0.0	0.0	8.6	8.5	5.1	12.4	8.0	7.9	4.7	11.6
other	12.6	11.7	3.3	25.0	4.0	4.0	2.1	6.3	4.6	4.6	2.7	6.9
plants	4.5	4.2	0.0	12.5	8.0	8.0	5.9	10.4	7.9	7.8	5.7	10.2

The results of the sample size analyses are not clear. The sensitivity analysis suggests that as variation in the other parameters is effectively reduced (by reducing the estimated standard errors), the relative importance of variance in proportion diet data will increase, affecting model output (predicted total mass of prey consumed). As sample size increases--for adults in particular--the variance in diet proportions by categories proposed should be reduced, but it is uncertain how much. The juvenile and pooled data suggest little reduction in variation.

Differential gut passage rates: Although different passage rates of fish and crabs have been found for gag (*Mycteroperca microlepis*) at intermediate times (Berens and Murie 2008), these data have not been completely incorporated into this model. Fish and crabs passed the gut of gag in about the same amount of time, and remained identifiable for the same amount of time. Fish and crab prey types do differ in the biomass remaining at intermediate times. This could affect the mass-based diet proportion estimates but will not affect the prey type by number estimates because they remain identifiable for the same time period. We incorporated this as a single parameter that adjusts mass-based percent fish relative to crabs. We assumed that shrimp, other, and plants are similar to fish relative to crabs.



The adjustment parameter was estimated from the regression parameters for the power exponential given in Table 1 of Berens and Murie (2008) (Figure 26) by taking the average proportional difference predicted between all crab and fish types for all hours post-prandial (1 to 24, mean = 0.197), hours that seem to have a big difference (3 to 18hrs, mean = 0.273), and the maximum difference (mean = 0.390), converted these to relative crabs mass units, and recalculated percent diet by stomach. This results in “crab per gram” adjustment factor of between 1.0 and 1.67, (nominal estimate of 1.25) for non-crab prey types occurring in mixed prey guts. Depending on parameter distribution, this will have a large effect on the proportion diet data for some individual guts, but the group means are little affected.

Consumption model

The goliath grouper consumption model was designed to answer the question, “What biomass (in kg) of fish, crabs and other prey, does the population of goliath grouper consume?” The consumption model (e. g., Kitchell et al. 1977) is a 3 parameter function (commonly used negative exponential) of goliath grouper wet weight biomass:

$$C(B) = P \cdot a_1 \cdot (B)^{b_1}$$

where: B = individual biomass (kg)

a₁ = “intercept” of the maximum consumption to biomass relationship (as B ⇒ age-0)

C(B) = a₁ at optimal conditions, really a₁ is scaled so that age-0 intercept approx 25% (range 10 to 40%) or a₁ = 0.25/(B₀)^{b₁}.

b₁ = allometric scaling parameter

P = aggregate proportional adjustment parameter that integrates the proportional adjustment parameter, optimal temperature, minimum temperature and Q₁₀ for the relationship between temperature and maximum consumptions.

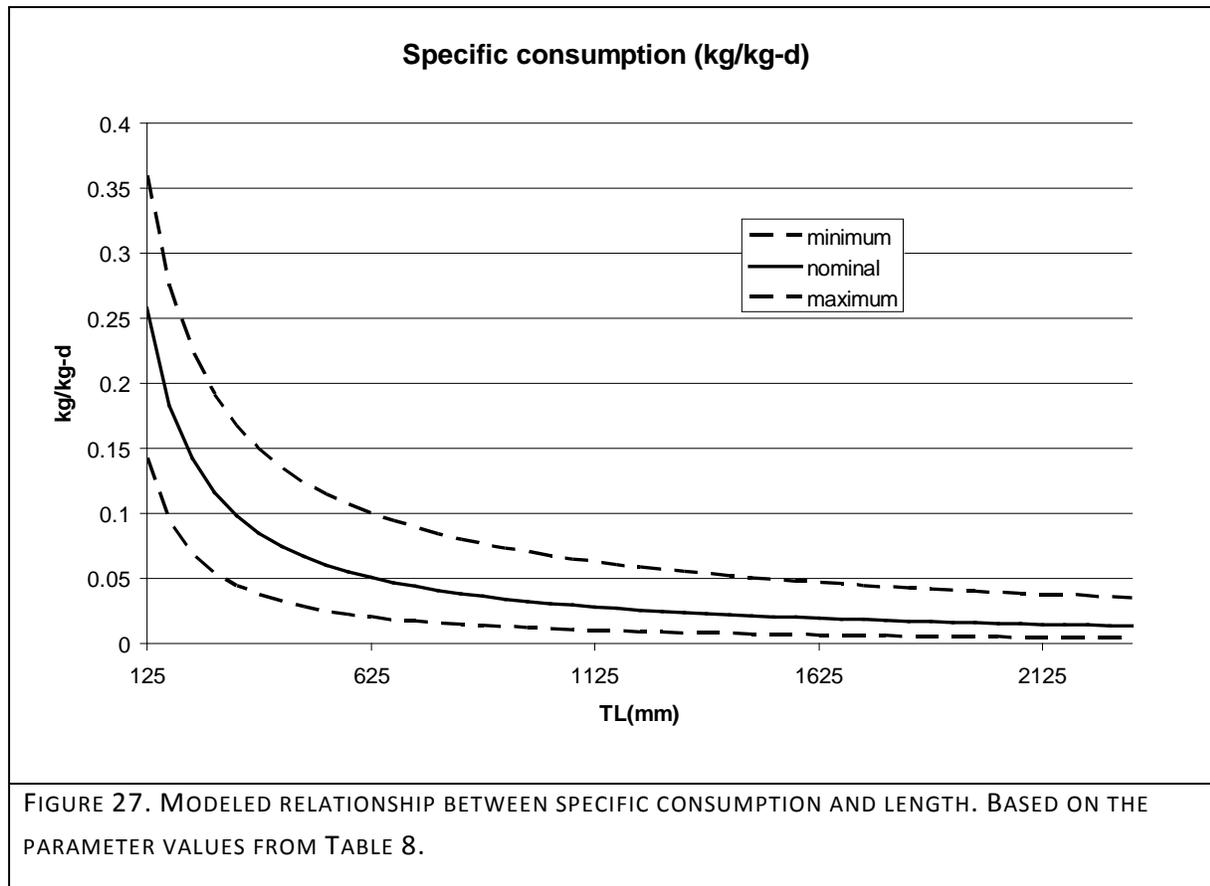
What is missing from this model is the prey and activity-dependent consumption, which has been shown to have a large impact on consumption rates in fish, including walleye pollock (Paul et al. 1998), and any increase in consumption that might be required for reproduction.

There are no suitable species from which to borrow parameters for a full bioenergetic model or even the consumption portion. Instead, we bounded the consumption model with maximum consumption rates observed across published fish bioenergetic studies. This leads to a maximum specific consumption rate of about 0.36 kg/kg-day for age-0, to about 0.035 kg/kg-day for adults, with large uncertainty around these point estimates, minima at 0.14 kg/kg-day for age-0, and 0.003 kg/kg-day for largest adults (Figure 27). These estimates are for non-reproductive fish. We assume that the maintenance diet for adult goliath grouper could be extremely low, given their sedentary behavior.

Total length to mass: Total length (TL) was converted to mass using the two parameter regression from Bullock et al. (1992) (Table 8, Figure 28). Error about the conversion was estimated by adjusting the range of the regression parameters to yield about a 10% deviation from the best estimate for TL from 100 to 2260 mm. This approximates the reported $R^2 = 0.96$. Integration of the consumption function with this TL to mass function allows predictions of total daily consumption as a function of TL (**Figure 29**). In the model, total daily consumption per individual or per size class was then partitioned into proportion fish, crab, shrimp, other, and plant using either the mean values from the diet analysis with the nominal parameter values (for a point estimate), or in a mixed Monte Carlo parameter selection routine with the bootstrapped diet data (for output distribution).

Table 8: Parameter values, nominal and max/min boundaries, for predicting total consumption by goliath grouper.

Parameter	Nominal	Range	Description
a1	0.12	(0.09, 0.15)	"intercept" for consumption model
b1	-0.33	(-0.4, -0.26)	exponent of consumption model
P	0.7	(0.4, 1.0)	aggregate parameter, proportional adjustment to consumption as function of the regression and temperature effect
a2	1.31	(1.30, 1.32)	TL to mass regression parameter ($\times 10^{-8}$)
b2	3.056	(3.05, 3.062)	TL to mass regression exponent
AJ	1050	(950, 1150)	Adult to Juvenile classification cutoff (mm)



Juvenile and Adult determination from TL: The cutoff parameter between adults and juveniles appeared between 95 cm and 115 cm. This cutoff and its parameter range were used to partition the total daily consumption into that consumed by adults and juveniles.

Sensitivity analysis: All parameters were examined using both a uniform distribution and a truncated normal distribution. The truncated normal was examined at three levels determined by scaling the distribution by substituting the standard error term with the parameter range divided by either 2, 4, or 8. For all distributions, this resulted in the following qualitative descriptions: 2 = approximate uniform distribution, 4 = approximate triangular distribution, and 8 = approximate normal distribution. These distribution types had some effect on parameter rankings, in particular as the parameter distributions become more centralized, the importance of the bootstrapped diet data rose in the rankings.

We examined three artificial input populations: the first population based on a negative exponential of abundance by 50 mm size class increments from 10 cm to 235 cm, the second based on a single individual per size class (for testing), and the third based on the observed TL by individual. All

preliminary sensitivity analyses were unaffected by input population. The preliminary sensitivities show that the consumption proportional adjustment parameter (P), and other consumption parameters were ranked consistently the highest, with the exception that the bootstrapped proportion diet data (BD) moved up in the rankings, eventually to the highest ranking as parameters distributions increased in central tendency (Table 8) and was the highest ranked in the adult consumption output when using unpooled data. Sensitivity also showed that TL-to-mass regression parameters had little effect (Table 9, and Table 10 for CV magnitudes). This preliminary sensitivity is also consistent with other bioenergetic model sensitivity analyses (e. g., Bartell et al. 1986).

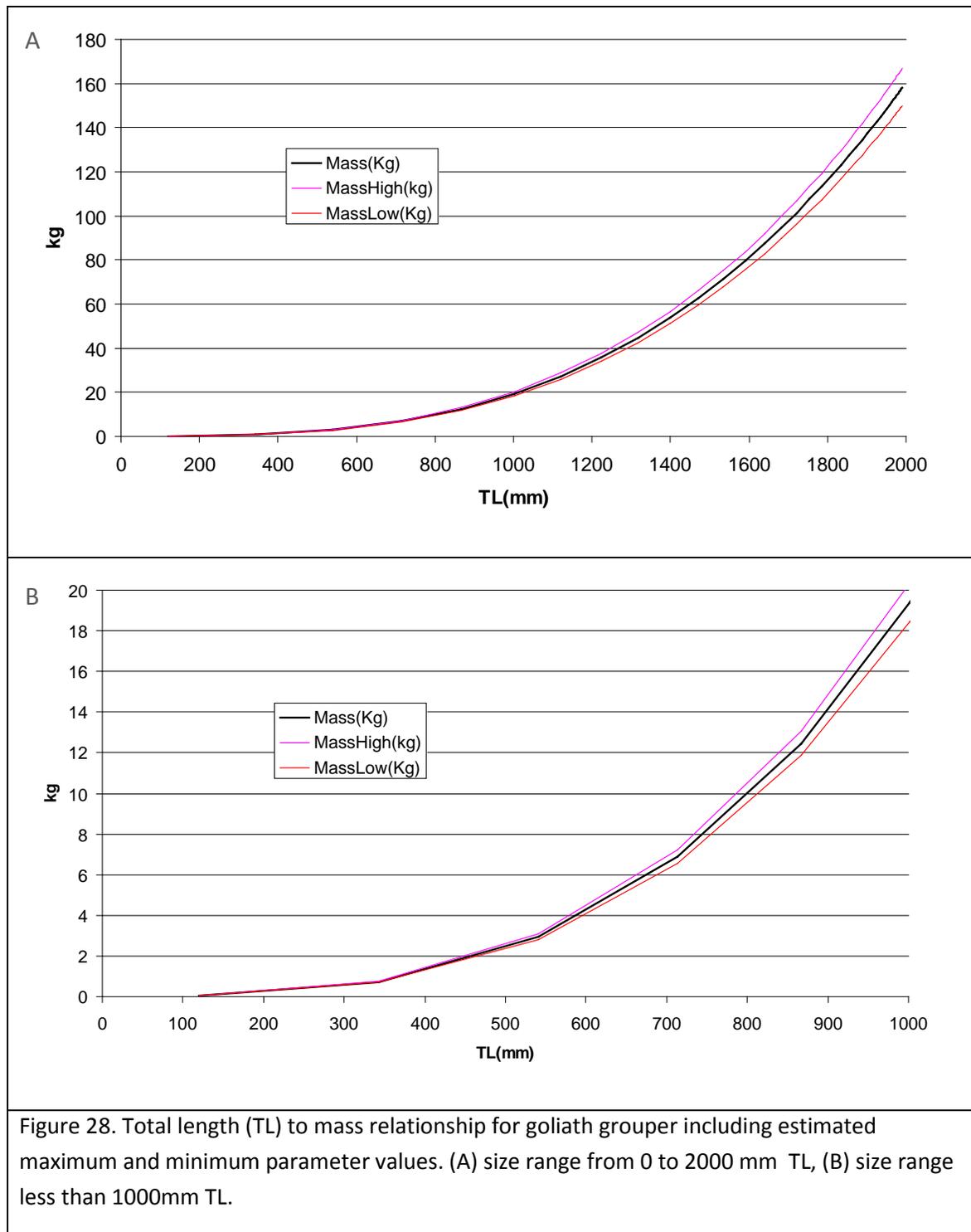
Table 9: Sensitivity analysis rankings for adult and juvenile predicted total fish consumed with proportion diet data pooled and unpooled. Sensitivity compares the individual effect of each parameter in Table 8 across their hypothesized ranges, and the effect of bootstrapped proportion diet data (BD). Parameter distribution code as U=uniform, TN2=Truncated normal with SE = Range/2, TN4= Truncated normal with SE = Range/4, TN8=Truncated normal with SE = Range/8. Input population was a size structured population ($N \sim 4,500$, TL from 10 to 235 cm, in 5.0 cm size classes) distributed as a negative exponential ($N_0 = 500$).

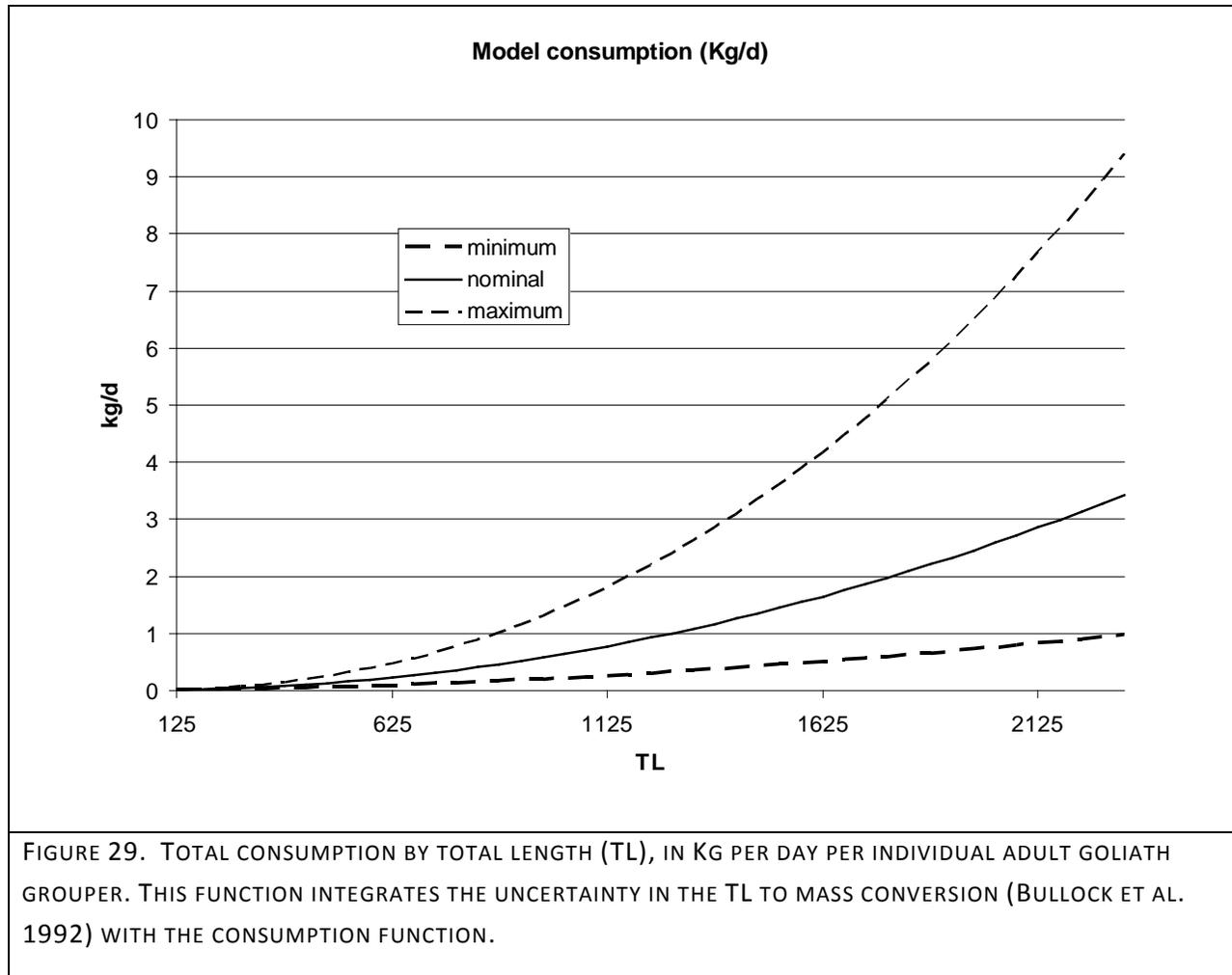
Rank	U	TN2	TN4	TN8	U	TN2	TN4	TN8	
Adult fish consumption, unpooled					Juvenile fish consumption, unpooled				
1	BD	BD	BD	BD	P	P	P	BD	
2	P	P	P	P	a1	a1	BD	P	
3	b1	b1	B1	b1	BD	BD	a1	a1	
4	a1	a1	A1	a1	AJ	AJ	AJ	AJ	
5	AJ	AJ	AJ	AJ	b1	b1	b1	b1	
6	b2	b2	B2	b2	b2	b2	b2	b2	
7	a2	a2	A2	a2	a2	a2	a2	a2	
Adult fish consumption, pooled					Juvenile fish consumption, pooled				
1	P	P	P	BD	P	P	P	BD	
2	b1	b1	b1	P	a1	a1	BD	P	
3	a1	a1	BD	b1	BD	BD	a1	a1	
4	BD	BD	a1	a1	AJ	AJ	AJ	AJ	
5	AJ	AJ	AJ	AJ	b1	b1	b1	b1	
6	b2	b2	b2	b2	b2	b2	b2	b2	
7	a2	a2	a2	a2	a2	a2	a2	a2	

Table 10

Sensitivity analysis, showing the values of output mean (kg/day fish consumed) and associated CV due to individual parameter variation. This is the uniform distribution from the above sensitivity Table 7. CV's show the magnitude of the parameters effect on output uncertainty.

Parameter	Mean fish consumed (kg/day)	CV	Parameter	Mean fish consumed (kg/day)	CV
Adult fish consumption, unpooled			Juvenile fish consumption, unpooled		
BD	134.01	51.97	P	182.12	24.91
P	179.15	24.91	a1	133.83	14.41
b1	141.58	17.04	BD	123.66	12.66
a1	112.34	14.41	AJ	133.91	9.34
AJ	132.06	7.19	b1	166.61	6.22
b2	132.37	1.86	b2	134.56	1.64
a2	133.02	0.32	a2	135.22	0.32
Adult fish consumption, pooled			Juvenile fish consumption, pooled		
P	232.37	24.91	P	179.27	24.91
b1	183.64	17.04	a1	131.73	14.41
a1	145.72	14.41	BD	121.74	12.00
BD	181.10	12.00	AJ	131.81	9.34
AJ	171.29	7.19	b1	164.00	6.22
b2	171.69	1.86	b2	132.45	1.64
a2	172.53	0.32	a2	133.10	0.32





Input-output model operation: All the codes for the model are in Matlab input data and are in ASCII text files, output data are in Matlab figures and ASCII text files. Input data, TL and number of individuals, were presented in Total Length either in size classes or by individual. Data in size classes should be the geometric mean of the boundaries of the size class (due to the non linear relationship between consumption and mass). Output data are the distributions of total kg consumed of a diet type, either by the total population or by juveniles and adults for the entire population using either pooled diet data or individual group data. Means, medians, variance, and the upper and lower 95% boundaries of the output and graphics of the output distribution are depicted in **Figure 30** and **Figure 31**. Diet data type can be switched between proportions dependent upon mass or counts. Sensitivity analyses were run separately and produced rankings of parameters based on their effect on the coefficient of variation (CV) of the output (predicted biomass of consumed fish) when all other parameters are held constant.

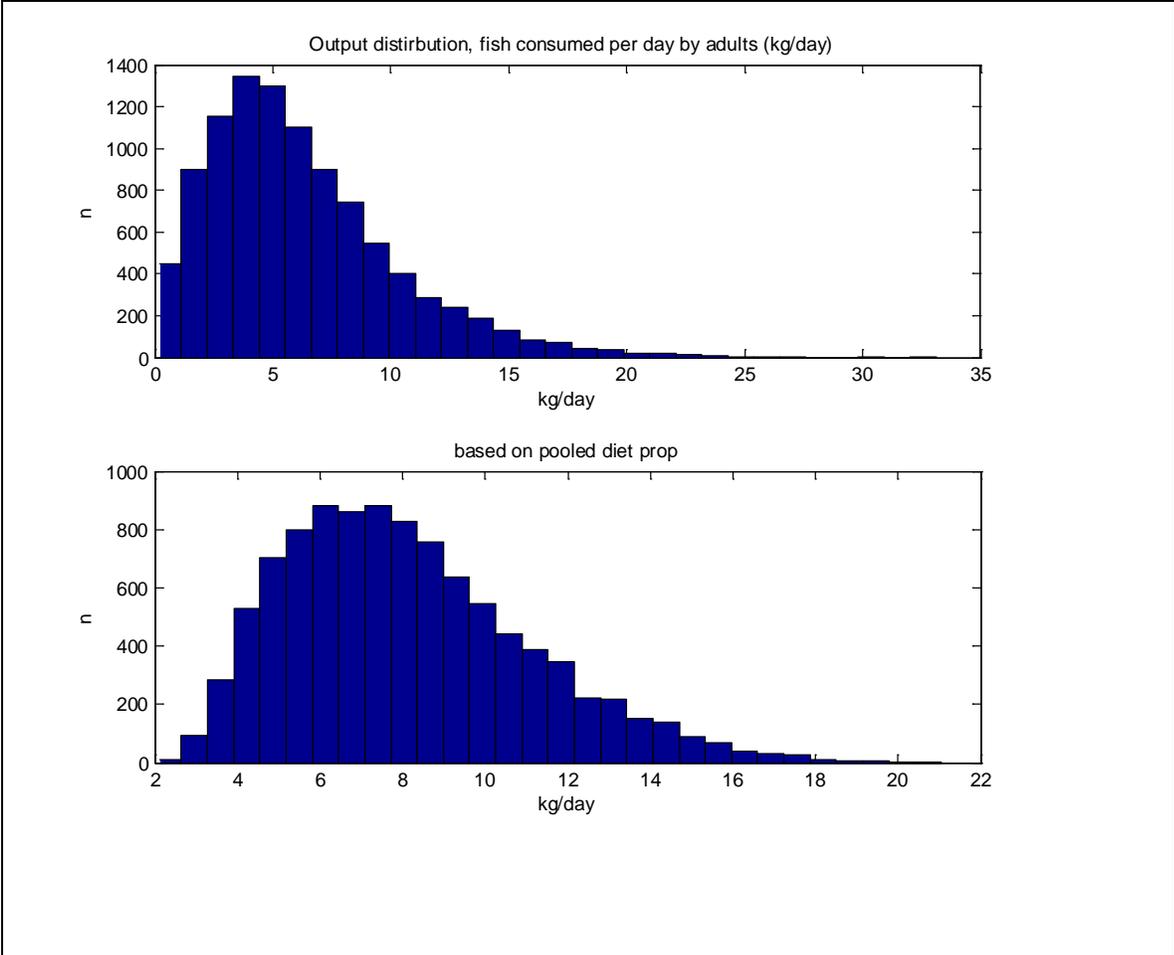


FIGURE 30. DISTRIBUTION OF OUTPUT TOTAL FISH CONSUMED BY ALL (18) ADULTS IN ADULT GOLIATH GROUPER SAMPLED BY FSU RESEARCHERS (I.E., BASED ON UNPOOLED OR POOLED PROPORTIONAL DIET DATA).

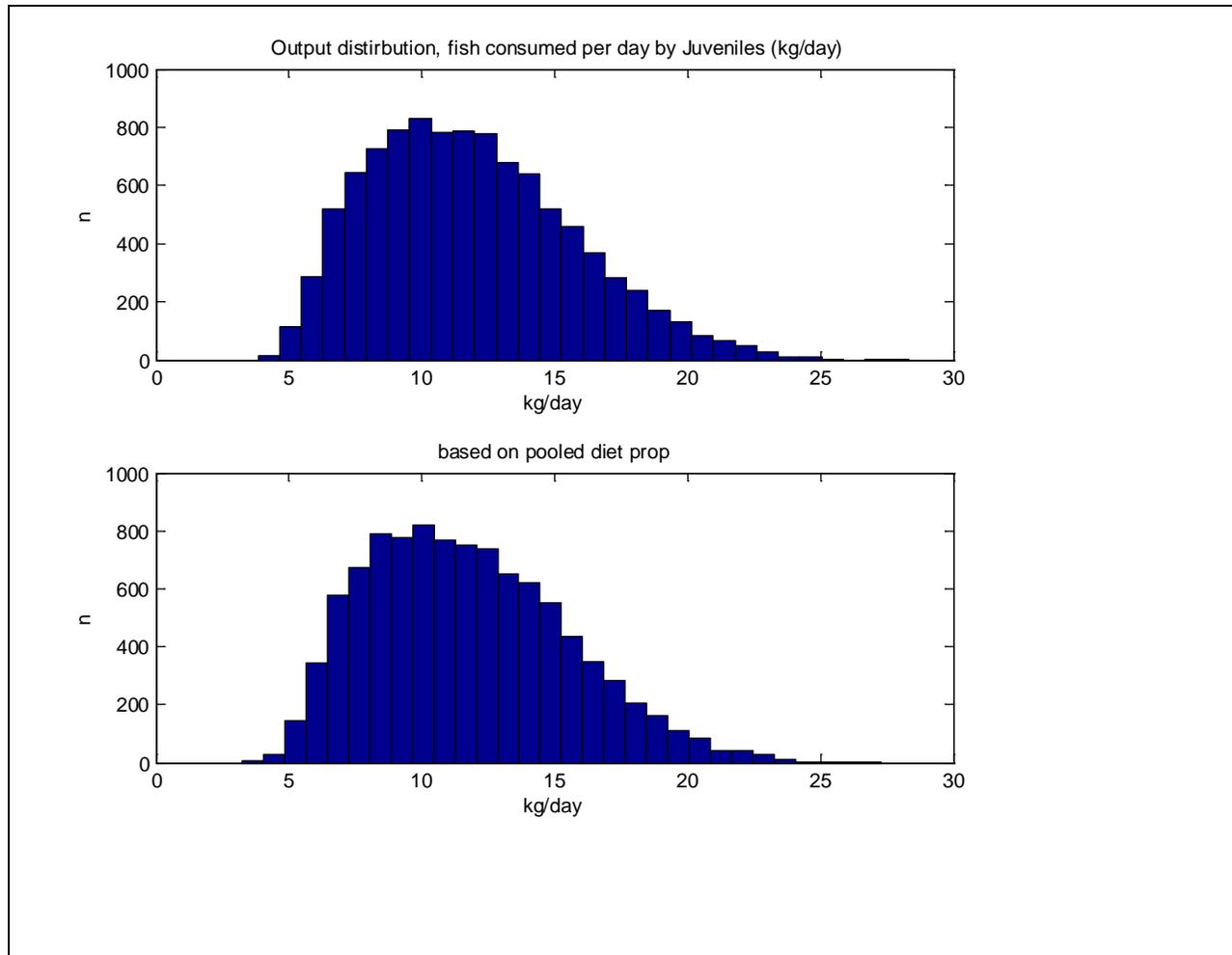


FIGURE 31. DISTRIBUTION OF OUTPUT TOTAL FISH CONSUMED BY ALL (224) JUVENILE GOLIATH GROUPEE IN THE SAMPLE COLLECTED BY FSU RESEARCHERS, BASED ON UNPOOLED OR POOLED PROPORTIONAL DIET DATA.

DISCUSSION OF DEMOGRAPHICS AND TROPHODYNAMICS

We observed no significant regional differences in size among adult goliath grouper (Figure 6). The largest individuals occurred in region 4, just off the Ten Thousand Islands, where most of the spawning aggregations occur. Juveniles exhibited considerable variation in size at age, as great in Florida Bay as it was in the Ten Thousand Islands (Koenig et al. 2007). This pattern makes it clear that determining age structure is critically important.

Determining ages on live goliath grouper required direct sampling of dorsal spines and rays. This non-lethal method has been used successfully in juvenile goliath grouper, validating ages in three ways: (1) by comparison with adjacent spines taken from recaptured fish, (2) by oxytetracycline marking of spines, and (3) by sacrificing fish and comparing spine counts with otolith counts (Brusher and Schull 2009). The spines of juveniles (0-5 years old) provided accurate estimates of age. However, the central portion of the bony spines becomes more and more vascularized as fish grow, so that the

spines of adults may lose the inner-most annuli. Soft rays are preferable to spines because they are less vascularized, cartilaginous, and bipartite (bilateral components) so that the earliest annuli are not lost to core vascularization.

We compared dorsal fin ray ages in adult goliath grouper to corresponding otolith ages on “fish of opportunity,” that is mortalities due to cold or red tide and enforcement confiscations (Murie et al. 2009). We found a very good relationship, indicating that fin rays provided ages nearly equivalent to those following from otoliths in terms of accuracy and precision. We are currently evaluating how our ability to age fish, in terms of accuracy and precision, changes with fish age, older fish being more difficult to age because of the accumulation of annuli in the fin-ray edge. The age at which this accumulation occurs is species-specific, however, and may not occur within the range of age determination required for management. For example, lingcod are routinely and reliably aged by their

Table 11. Examples of fish species that have been aged using sections of fin structures, along with their maximum observed age. FW = freshwater; and M = marine.

Common Name and Species	Locality of Study	Maximum Age (Yrs)	Reference
Albacore tuna (<i>Thunus alalunga</i>)	M	12	Beamish (1981)
Arctic grayling (<i>Thymallis arcticus</i>)	FW	11	Sikstrom (1983)
Chum salmon (<i>Oncorhynchus keta</i>)	M	5	Bilton and Jenkinson (1969)
Gag grouper (<i>Mycteroperca microlepis</i>)	M	17	Debicella (2005)
Goliath grouper (<i>Epinephelus striatus</i>)	M	17	Murie et al (2009)
Kelp greenling (<i>Hexagrammos decagrammus</i>)	M	16	Murie & Parkyn (unpub. data)
Lake sturgeon (<i>Acipenser fulvescens</i>)	FW	152	(Anonymous 1954)
Lingcod (<i>Ophiodon elongatus</i>)	M	21	Chilton and Beamish (1982)
Pacific cod (<i>Gadus macrocephalus</i>)	M	8	Beamish (1981)
Sockeye salmon (<i>Oncorhynchus nerka</i>)	FW & M	4	Bilton and Jenkinson (1969)
Walleye pollock (<i>Theragra chalcogramma</i>)	M	9	Beamish (1981)
White grunt (<i>Haemulon plumieri</i>)	M	18	Murie and Parkyn (1999)
White sturgeon (<i>Acipenser transmontanus</i>)	FW & M	104	Rienand Beamesderfer (1994)
White sucker (<i>Catostomus commersoni</i>)	FW	14	Beamish and Harvey (1969)

fin rays up to ~20 years before the accumulation of annuli on the edge interferes with accurate age estimates. But because there are very few lingcod >10 years taken in the fishery (McFarlane and

Leaman 1993), the method is appropriate for stock assessment. Fin rays have been used to age a diverse group of long-lived fish species, including warm temperate gag grouper and white grunt (Table 11). For gag and goliath grouper, fin-ray ages were unbiased up to age 17, the maximum age available in our samples (Debicella 2005, Murie et al. 2009).

Diet, Trophic Pattern and Bioenergetics

Stomach content analysis shows that goliath grouper prey mostly on crustaceans, with fish prey consisting primarily of slow-moving, bottom-associated species. These data are consistent with historical observations of their diet (Table 12). Adult stomach contents contained fish from only three families: burrfishes (Diodontidae), herring (Clupeidae), and stingrays (Dasyatidae)—although stingrays in the stomach contents were, in most instances, likely bait. Our field observations support the view that goliath grouper do not typically eat other groupers, snappers or other fast and/or maneuverable fish. When surrounded by young gray snappers (*Lutjanus griseus*), lane snapper (*Lutjanus synagrus*), yellowtail snapper (*Ocyurus chrysurus*), and amberjack (*Seriola dumerili*), they make no attempt to prey upon them. That is not to say, however, that they will not attack an injured or tethered fish, including one on a hook at the end of a fishing line or one on a spear.

Based on literature suggesting that goliath grouper prey heavily on spiny lobsters (*Panulirus argus*) in the Virgin Islands (Randall 1967), we fully expected to find many of them in the diet of goliath grouper sampled in Florida Bay where spiny lobsters are relatively plentiful and support an important fishery. However, we only found a single small spiny lobster in all 224 juvenile goliath groupers sampled. The most plausible explanation for this is that the daily activity patterns of both species are diametrically opposed; spiny lobster are nocturnal and goliath grouper are diurnal. If there is sufficient day-time refuge for the lobsters, they may be largely inaccessible to the grouper. Dr. Jack Randall's (University of Hawaii, personal communication) ecological observations have some bearing on this issue. He told us that lobsters were very common in the area in the Virgin Islands where he captured goliath grouper because there was no significant lobster fishery. Apparently, under high lobster densities or limited lobster cover, all predators on lobsters would prey heavily on their local population, including goliath grouper. But it is not valid to extrapolate from another place, time, and ecological condition (1960s in the Virgin Islands with an unfished lobster population) to the present conditions (2008 in the Florida Keys with a heavily fished lobster population) to evaluate diet of any species. It is important to make a distinction between what an animal can eat and what it does eat in its ecological context. The former is irrelevant, but finding out the latter requires stomach content analyses.

Observations by us and by Bullock and Smith (1991) (Table 12) that goliath grouper prey upon small (< 10 cm) pelagic round herring (*Etrumeus teres*) seem implausible at first glance. However, closer observation reveals that herring will pack in tightly around goliath grouper when the little fish are under attack by schools of little tunny (*Euthynnus alletteratus*). They apparently perceive the goliath

Table 12. Historical Accounts of Goliath Grouper Diet. N = sample size

Life stage (N)	Location	Mollusks	Crustaceans	Fishes	Reptiles	References
Juvenile (1)	Caribbean	-----		parrotfish		Beebe and Tee van (1928)
Adult (1)	Caribbean	-----	spiny lobster			Erdman (1956)
Adults (9)	Caribbean	-----	spiny lobsters, slipper lobster	sting ray, porcupine fish	Sea turtle	Randall (1967)
Adults (32)	Gulf of Mexico	Tulip shell, octopus	spiny lobster, slipper lobster, box crabs, stone crabs, blue crabs, swimming crabs	spadefish, cowfish, porcupine fish, round herring, striped burrfish		Bullock and Smith (1979)

grouper as refuge, crowding in so tightly that they enter the grouper's mouth. Many times we observed goliath grouper vomiting round herring when disturbed.

As the stomach content analysis shows goliath grouper feed primarily on lower trophic level prey, many species of which have no direct economic value. The results of the stable isotope analysis support the finding that goliath grouper are mid-trophic-level consumers rather than top-level predators. We base this on their having a mean ^{15}N value of 11.0, which is much closer to that of mid to upper trophic level consumers (mean ^{15}N , ~ 11.4 ‰) than to top-level predators (mean ^{15}N , ~ 15 ‰) (Chasar et al. 2005). This result suggests that anecdotal information is unfounded of adult goliath grouper causing predatory depletion of grouper and snapper populations on reefs.

Both blue crabs (*Callinectes sapidus*) and pink shrimp (*Farfantepenaeus duorarum*) have ^{13}C values around -19 ‰ (Chasar et al. 2005) while the stomach contents of goliath grouper (primarily invertebrates) had a mean ^{13}C value of -16.4 ‰, indicating a depletion of approximately 6 ‰ when compared with the muscle tissue of the goliath grouper (mean = -10.7‰). The difference between stomach content and muscle tissue ^{13}C values may result from either digestion or formalin fixation.

Behavior and morphology are to great extent predictors of feeding patterns. Fish such as goliath grouper that exhibit cryptic coloration, large, rounded caudal fins, and stout dorsal fin spines are relatively slow swimmers that tend to feed on slow moving prey. They are capable of powerful swimming bursts, especially when hooked, but rarely if ever chase down their prey. Their large mouths with small teeth are adapted for suction feeding (Moyle and Cech 2000). However, goliath grouper and other large predators are opportunistic, feeding on easily captured prey such as the sick or moribund or prey tethered at the end of a fishing line or spear. The observations of fishers, that goliath grouper attack hooked fish, may have given rise to the assumption that those species are part of their normal diet. But again a distinction must be made between what goliath grouper can eat and what they do eat in their ecological context. They can probably eat anything smaller than their gape, but, as our stomach content data and stable isotope data show, they prey mostly on crabs and slow-moving fish species with little direct economic value.

Bioenergetics model: Our primary question related to the goliath grouper diet is, "What biomass of prey (e.g., fish, crabs and other types of organisms) does the population of goliath grouper consume?" We developed a consumption-only three parameter function model (commonly used negative exponential) based on wet weight biomass, growth, and maintenance (disregarding reproduction) rather than the more complex (20+ parameter) bioenergetic model we originally proposed (based on Kitchell et al. (1977)). The primary reason for this switch was that we could not find a suitable species from which to borrow parameters for the full model. Thus, we proposed bounding the problem with maximum consumption rates observed across most fish bioenergetic studies and the minimum as some function of the observed consumptions in goliath grouper. This works out to a specific maximum rate of about 0.25 kg/kg-day for age-0, to about 0.05 kg/kg-day for adults, with large uncertainty around these point estimates from nearly zero to these maxima. However, it is likely that

the maintenance diet for adult goliath grouper is extremely low. Additional adult dietary data will allow us to refine this consumption estimate further.

The consumption model we use lacks temperature dependence. It can be expanded to include temperature dependence by the addition of three more parameters--optimal temperature, minimum temperature, and a Q10, or slope of the relationship between temperature and consumption rate—once additional data become available from other more northern regions of Florida. However, this addition may not be necessary, given that goliath grouper have a relatively narrow temperature tolerance that limits out at minimum temperature of 15.5° C (Sadovy and Ecklund 1999), and that in north Florida, goliath grouper avoid winter low temperatures in shallow water by migrating offshore (personal observations).

Our next studies will involve integrating estimates of consumption rates with present and projected population estimates to estimate impacts of a recovered goliath grouper population on prey species. Population estimate will have to be determined regionally and so will dietary data. At this point we only have goliath grouper dietary data from south Florida.

PART III. SPAWNING BIOLOGY

MATERIALS AND METHODS

Location of spawning sites.--We relied heavily on the knowledge of commercial fishermen, recreational scuba divers, and, more recently, our own surveys of suitable habitat to locate goliath grouper spawning sites off the Gulf of Mexico and Atlantic coasts of Florida. In this study, our intent was to verify that sites having > 10 individual adult goliath grouper aggregating (an arbitrary but reasonable quantitative indicator) during the spawning season were in fact spawning sites. The primary indicators that we used to make this determination were: (1) the seasonality with which the aggregations occurred (consistent with known spawning periods of July through October); (2) the presence of ripe gonads; and (3) the presence of fertilized eggs verified as those of goliath grouper.

Timing of spawning.--We used a combination of passive acoustics (sounds produced directly by fish logged on a Long-term Acoustic Recording System (LARS) Loggerhead Instruments receiver) and active acoustics (sounds produced by a depth-sensing acoustic transmitter (Vemco V13P-S256) placed dorsally on one fish and logged on a Vemco VR2 receiver) to identify the timing of spawning on several presumptive spawning sites associated with ship wrecks off the southwest coast of Florida during the spawning season (Mann et al. 2009). Passive and active recordings were made on *The Cathy Wreck* (N24° 45.9', W082° 59.4'; depth = 47 m, off the northern border of the Tortugas Ecological Reserve North) in September and October 2005, while only passive acoustic recordings were made at *The Fantastico Wreck* (N 26° 17.7', W 082° 50'; depth = 34 m) from July 29 to December 21, 2007.

The LARS (custom built by co-investigator Mann, USF) consists of a Toshiba Pocket PC in an underwater housing connected to a hydrophone (HTI96-min; High-Tech; sensitivity -164 dBV/ μ Pa, 2 Hz to 37 kHz), which samples acoustic data at 10,000 Hz for ten continuous seconds every 10 minutes and records to 32 GB Secure Digital flash memory. With this recording schedule, the devices ran for three months. Data were downloaded to a PC and sounds were compared to known goliath grouper sounds using MATLAB (Mathworks) signal processing to evaluate frequency range and pulse duration

For active acoustics at *The Cathy*, we attached a transmitter tag to a large (~ 2 m) adult goliath grouper (sex could not be determined) underwater using a speargun-delivered dart tag (Floy tag BFIM-96), assuming, based on past experience tagging over 2,000 fish, that this method of tagging minimized stress relative to that produced by capture. We monitored the fish from 9 September 2005 through 6 November 2005. The transmitter had a random output between 5 and 15 s, allowing us to detect short-

duration vertical movements, such as rapid spawning ascents (common among reef fish, as indicated by Colin and Clavijo (1988), Powell and Tucker (1992), and Colin et al. (1996).

Determining spawning times with egg and sound sampling.--To directly verify that goliath grouper spawned, we sampled the water column for the presence of fertilized eggs. To verify the timing of spawning, we evaluated sound production logs. Both these events occurred at two sites: a natural ledge, *Hole-in-the-Wall*, off the Atlantic coast of Florida (N26° 56.0' W79° 59.4'; depth: 33 m; see Figure 1), and the shipwreck *The Fantastico*. Eggs were collected at night by deploying two mooring lines 200 m downstream from presumptive spawning sites (allowing sufficient drift time for eggs to become fertilized), and attaching to each mooring a series of 8 plankton nets (30 cm diameter opening, 0.5 mm mesh) distributed from the surface to the bottom at even intervals. *The Caves* were sampled on 21-22 September 2008, on the last quarter moon (moon rise = 2310 EST), deploying nets by 2000 EST (and the LARS soon after). *The Fantastico* was sampled on 27-28 September 2008, one day before the new moon (moon rise = 0430 EST), deploying the nets by 2230 EST. Nets and the LARS were retrieved soon after daybreak. The contents were then sieved (33 mm mesh) and examined microscopically for grouper eggs (0.9 - 1.0 mm diameter spheres, according to Colin *et al.* (1996). Collected eggs were staged and sent to Dr. Matthew Craig (University of Hawaii) for genetic verification of species identity, given his expertise

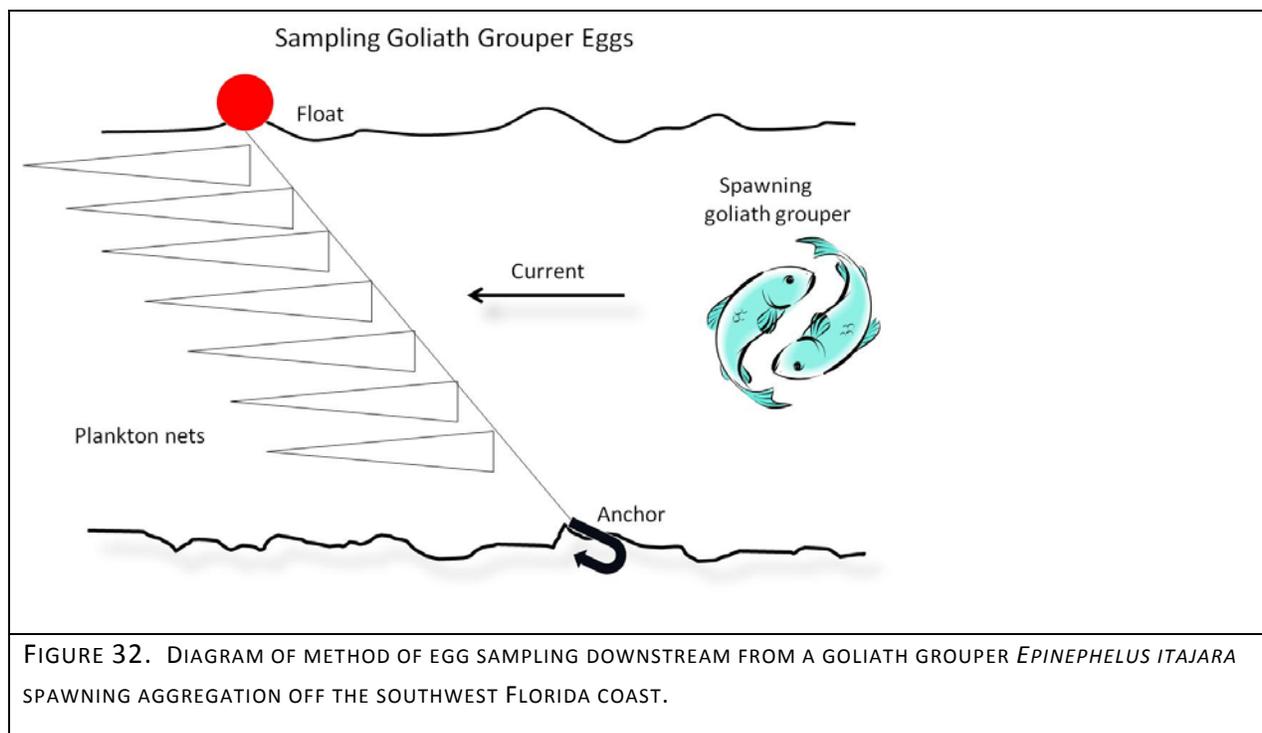


FIGURE 32. DIAGRAM OF METHOD OF EGG SAMPLING DOWNSTREAM FROM A GOLIATH GROUPER *EPINEPHELUS ITAJARA* SPAWNING AGGREGATION OFF THE SOUTHWEST FLORIDA COAST.

in grouper genetics (Craig and Hastings 2007, Craig et al. 2009).

Sounds were recorded using a LARS receiver deployed directly on each aggregation site.

Spawning site fidelity.--We examined spawning site fidelity on *The Californian Wreck* (N25° 21.6' W082° 31.9') off southwest Florida using active acoustic methods. We tagged 17 adult fish (presumed spawners) with acoustic transmitter tags (Eklund and Schull 2001, and Eklund and Schull Unpublished data) and placed VR1 receivers on the wreck through two spawning seasons so that we could evaluate spawning site fidelity.

RESULTS

Location of Spawning Sites.-- The only confirmed spawning sites for goliath grouper in the Gulf of Mexico occurred off southwest Florida (**Figure 33**). These were confirmed either by the presence of ripe gonads or the presence of fertilized goliath grouper eggs. In a number of sites, we recorded unique night-time chorusing by goliath grouper that appears to occur during the spawning season and at no other time. We suspect that these sounds can be used as a proxy for spawning activity, which strongly suggests that passive acoustics can be used to evaluate the temporal and spatial distribution of spawning activity. While there were a number of sites throughout the Gulf that contained > 10 individuals, those located near inlets (e.g., at Boca Grande) appear to be feeding aggregation sites, not spawning sites, based on observations of intense feeding activity by the group at variable times, including non-spawning times.

Sound production.-- Goliath groupers produced predominantly single-pulse calls (pulse duration with peak received levels > 124 dB re: 1 μ Pa = 132.0 ± 30.1 ms; mean \pm SD; n = 507), with a low dominant frequency (= 60.5 ± 9.2 Hz; mean \pm SD; n = 1065) (**Figure 34**), consistent with sounds produced by large fish and by goliath grouper as experienced by all divers on this project. Because the majority of acoustic energy was < 100 Hz, the sound pressure level in the 0 to 100 Hz frequency band was used as a measure of overall goliath grouper sound production.

We isolated a total of 1065 individual pulses from five days of recordings using automatic detection algorithm. This algorithm consisted of the following steps: (1) low-pass filter at 100 Hz, (2) high-pass filter at 10 Hz, (3) rectification of the signal, (4) smoothing of the signal with a 25 millisecond (ms) moving average, and (5) threshold detection set at 110 dB re: 1 μ Pa. Once signals were identified,

they were manually inspected with a spectrogram to confirm that they were goliath grouper sounds. Power spectra of individual pulses were calculated using Fast Fourier Transforms (FFT), where the

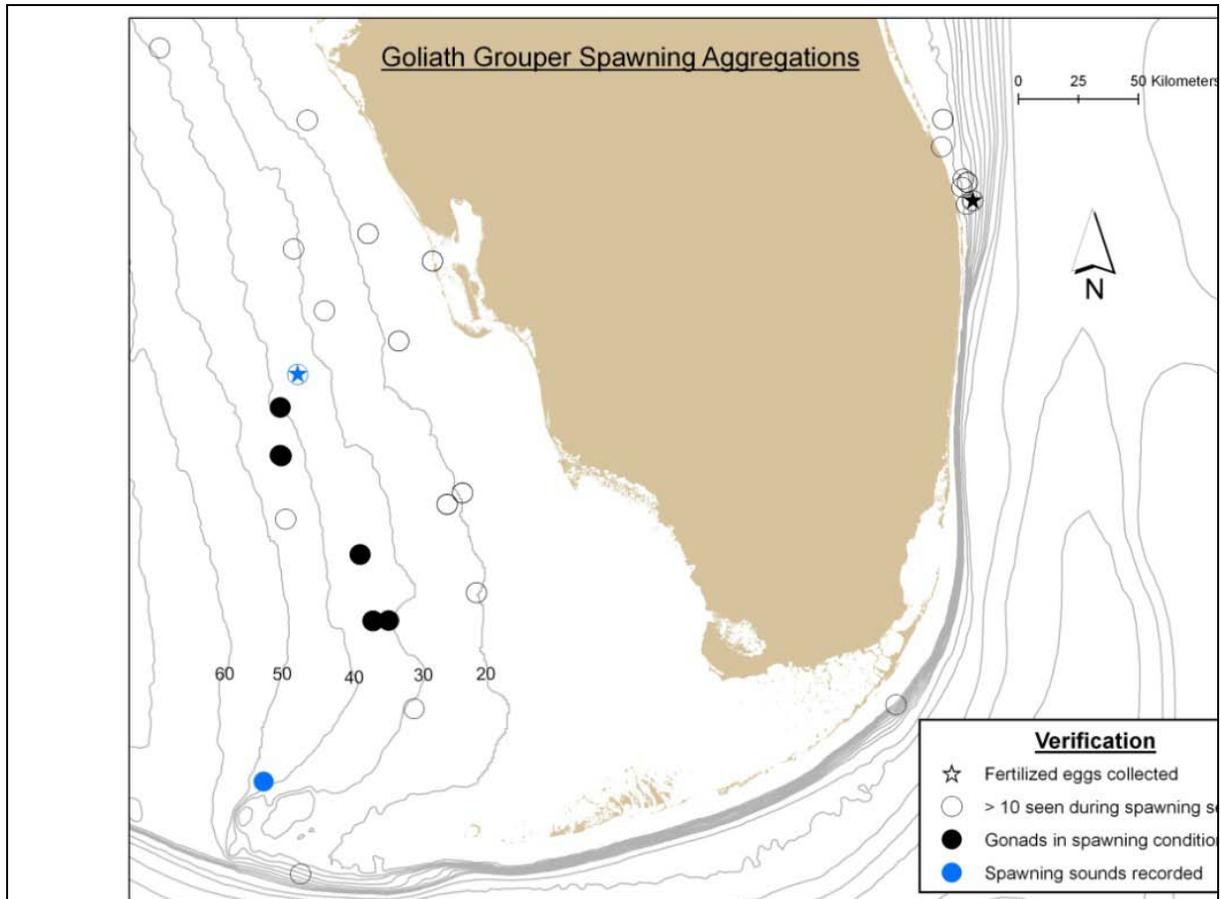


FIGURE 33. GOLIATH GROUPER AGGREGATION SITES OFF SOUTH FLORIDA (EACH HAVING > 10 INDIVIDUALS ON SITE DURING THE SPAWNING SEASON, JULY - OCTOBER); STARRED SITES = CONFIRMED SPAWNING SITES (FERTILIZED EGGS COLLECTED) ON *THE FANTASTICO WRECK* (GULF SIDE) AND *THE CAVES* (ATLANTIC SIDE); SOLID CIRCLES: BLACK = CONFIRMED BY RIPE GONADS; GREY = SUSPECTED BY SOUND PRODUCTION ON *THE CATHY WRECK*.

number of points in the FFT was equal to the number of samples in the signal. For example, a sound 100 ms in duration sampled at 11 025 Hz would consist of 1102 data points and its FFT would yield a 10 Hz frequency resolution. The dominant frequency (frequency with most energy) for each sound was calculated from the power spectrum of the original recorded signal without filtering. At lower signal-to-noise ratios, a significant portion of the signal can be hidden in the noise floor. Therefore, only the loudest signals (>124 dB re: 1 μ Papeak) were used to calculate sound duration (n = 507). Time series of goliath grouper sound production were analyzed by calculating band sound pressure levels in the 0 to 100 Hz frequency band for each 10 s recording using QLOGGER (available at www.qlogger.com).

marine.usf.edu/bio/fishlab/). The time of maximum band sound pressure level for each day was calculated for each dataset using MATLAB and presented in a histogram using 1 h resolution.

Calls from multiple individuals were obvious in most recordings, based on differences in sound level and overlap of sounds. Multiple single-pulse sounds of the type seen during night-time chorusing can be seen in **Figure 35**.

Off the southwest Florida coast, there were ~25 goliath grouper at the *The Cathy* and ~40 goliath grouper at *The Fantastico*. Sound production was strongly diel, peaking between 01:00 and 02:00 at *The Cathy* (Mann et al. 2009) and between 01:00 and 04:00 at *The Fantastico* (Figure 5). Each time series also showed a strong lunar periodicity in sound levels, with decreased levels of sound production on and around the full moon (**Figure 36**) for 1 to 7 d. The pattern of sound production observed from August through September on *The Fantastico* ceased after October 16, 2007, until recording ended on December 21, suggesting that goliath grouper night-time chorusing could be used to delineate the spawning season as well as identify spawning aggregations.

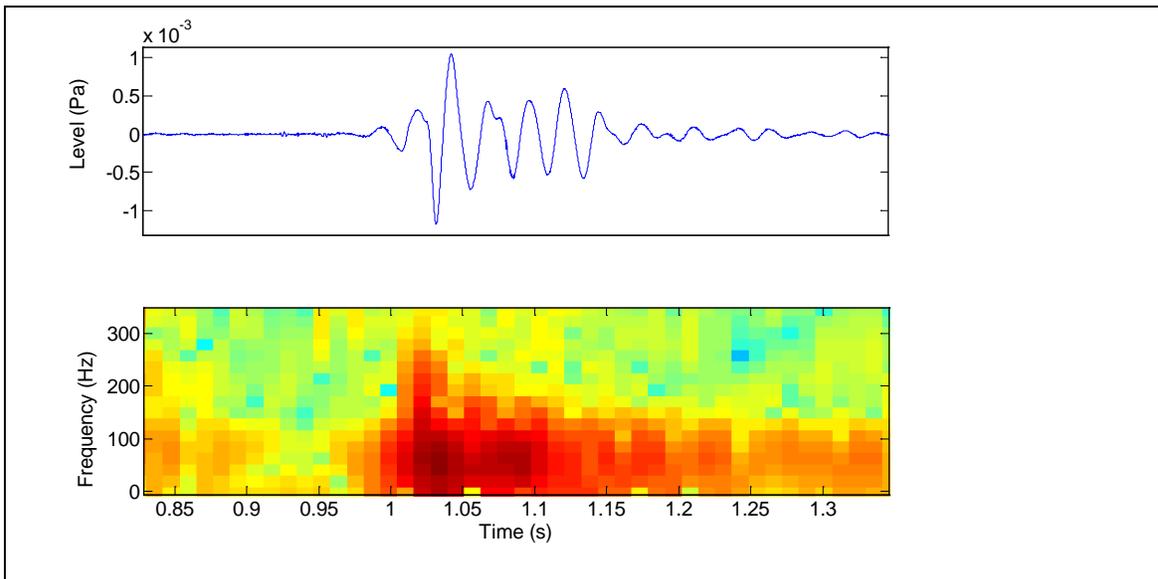
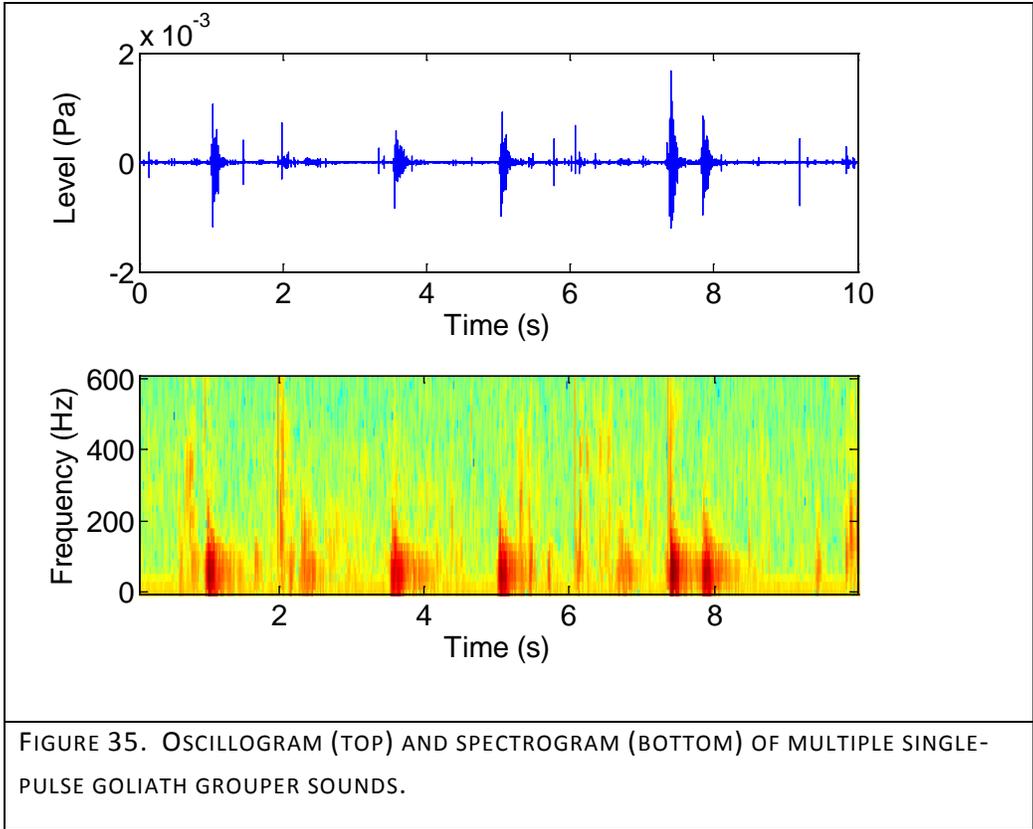


FIGURE 34. SINGLE PULSE CALL OF A GOLIATH GROUPEER ON A PUTATIVE SPAWNING AGGREGATION OFF SOUTHWEST FLORIDA. OSCILLOGRAM (TOP) AND SPECTROGRAM (BOTTOM). THE DOMINANT FREQUENCY OF THIS PULSE IS ABOUT 60 HZ.



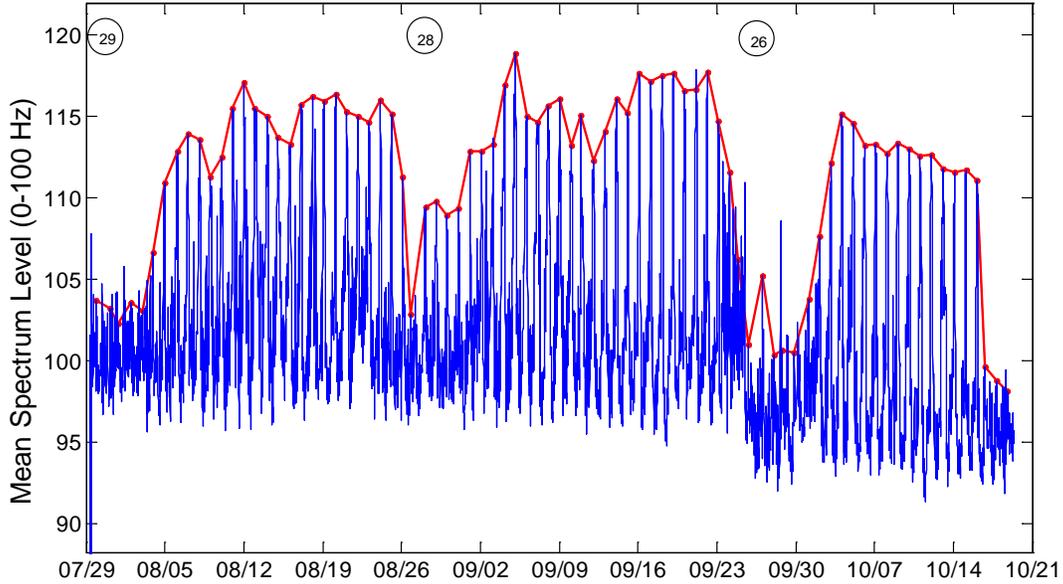


FIGURE 36. TIME SERIES OF SOUND PRODUCTION (PEAKS AROUND MIDNIGHT) IN THE 0-100 HZ FREQUENCY BAND OVER A THREE MONTH SPAWNING PERIOD, RECORDED AT *THE FANTASTICO*, A GULF OF MEXICO GOLIATH GROUPER SPAWNING SITE. DATA WERE SMOOTHED WITH 6-POINT (1 HOUR) MOVING AVERAGE. THE CIRCLES AND CONTAINED NUMBERS INDICATE THE DATES OF THE FULL MOON. TICK MARKS ARE LOCATED AT MIDNIGHT.

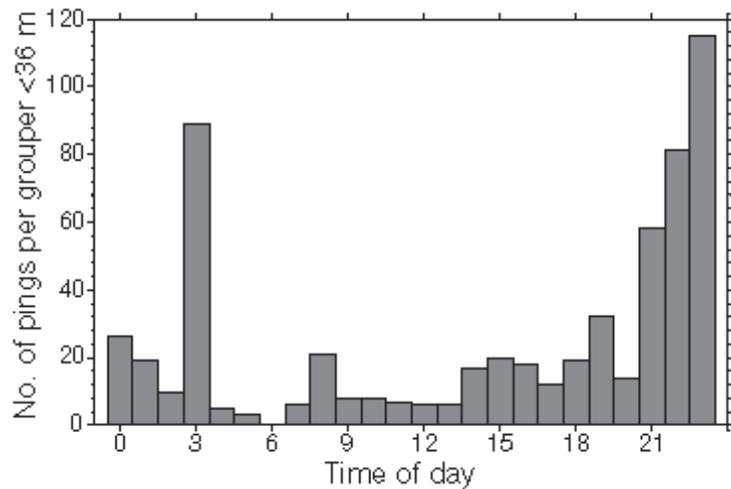


FIGURE 37. TIME OF DAY WHEN PINGER-IMPLANTED GOLIATH GROUPER HAD ASCENDED TO DEPTHS SHALLOWER THAN 36M (BOTTOM DEPTH = 47M).

With the exception of one day (October 24 to 25), the pinger-tagged fish at *The Cathy* was always present at the study site (N = 306,282 pings during the study period). The fish was located near the bottom at depths > 40 m most of the time, making occasional short (< 2 minute) forays to shallower depths (up to 20 m off the bottom), the majority of which took place at night from 21:00 to 00:00 h and 03:00 to 04:00 h (Figure 36).

Egg sampling—spawning verification.—We verified the presence of goliath grouper spawning aggregations (N > 20 individuals) at *The Caves* by diving the site prior to deploying the plankton net array. We did not dive on *The Fantastico* and so we are not sure how many goliaths were present there, although past surveys over the last two years indicated N = 40 - 50 individuals during the spawning season. At *The Fantastico* (depth = 33 m, top of the wreck = 22 m), it was evident that spawning occurred during moonless periods of the night, at which time (2330 EST) we saw goliath grouper rolling at the surface, apparently in a spawning run. Moonrise did not occur until 0430 EST. Although we saw no rolling at the surface at *The Caves*, the first spawning probably took place soon after we set up our plankton nets, based on the developmental stage of the eggs.

At each site we collected hundreds of eggs fitting our search image (spherical diameters between 0.9 and 1.0 mm) for *Epinephelus* grouper eggs from both study sites. A random sample from each site was preserved in 10% formalin and another random sample was preserved in 95% ethyl alcohol (to preserve DNA for genetic verification). Sampling was more effective in the Atlantic site where there was a constant current (about 1 kt). Without a similar current at the Gulf site, it was difficult to determine the downstream direction. Therefore, after deploying the net arrays, we

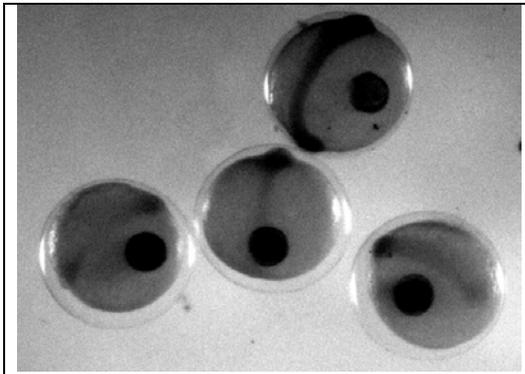


FIGURE 38. GOLIATH GROUPER EMBRYOS (= 1 MM DIAMETER) IN THE NEURULA STAGE COLLECTED FROM AN ATLANTIC SPAWNING AGGREGATION.

actively towed a single one meter diameter plankton net (0.3 mm mesh through that site for several hours, from about 2300 to 0300 EST.

The eggs were confirmed unequivocally as goliath grouper eggs. The eggs collected from the Atlantic site consisted of two developmentally different stages: neurula stage (Figure 37) and early gastrula stage. This suggests that there had been two separate and distinct spawns that night, probably several hours apart. We cannot verify the exact timing because there are no descriptions of developmental rates (or the influence of temperature on developmental rates) in the literature for

goliath grouper. Moonrise at the Atlantic spawning site (20-21 September 2008) occurred at 2310 EST,

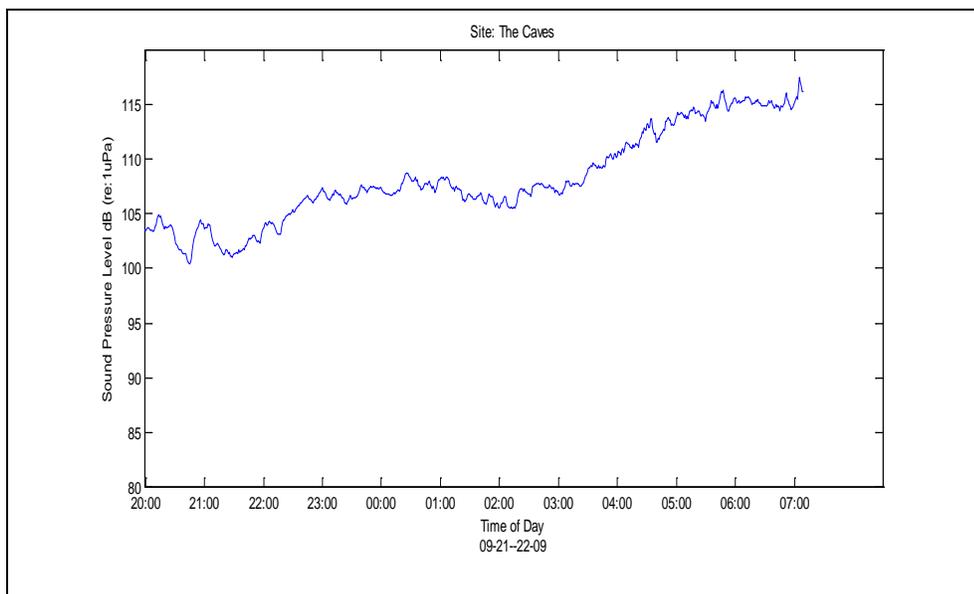


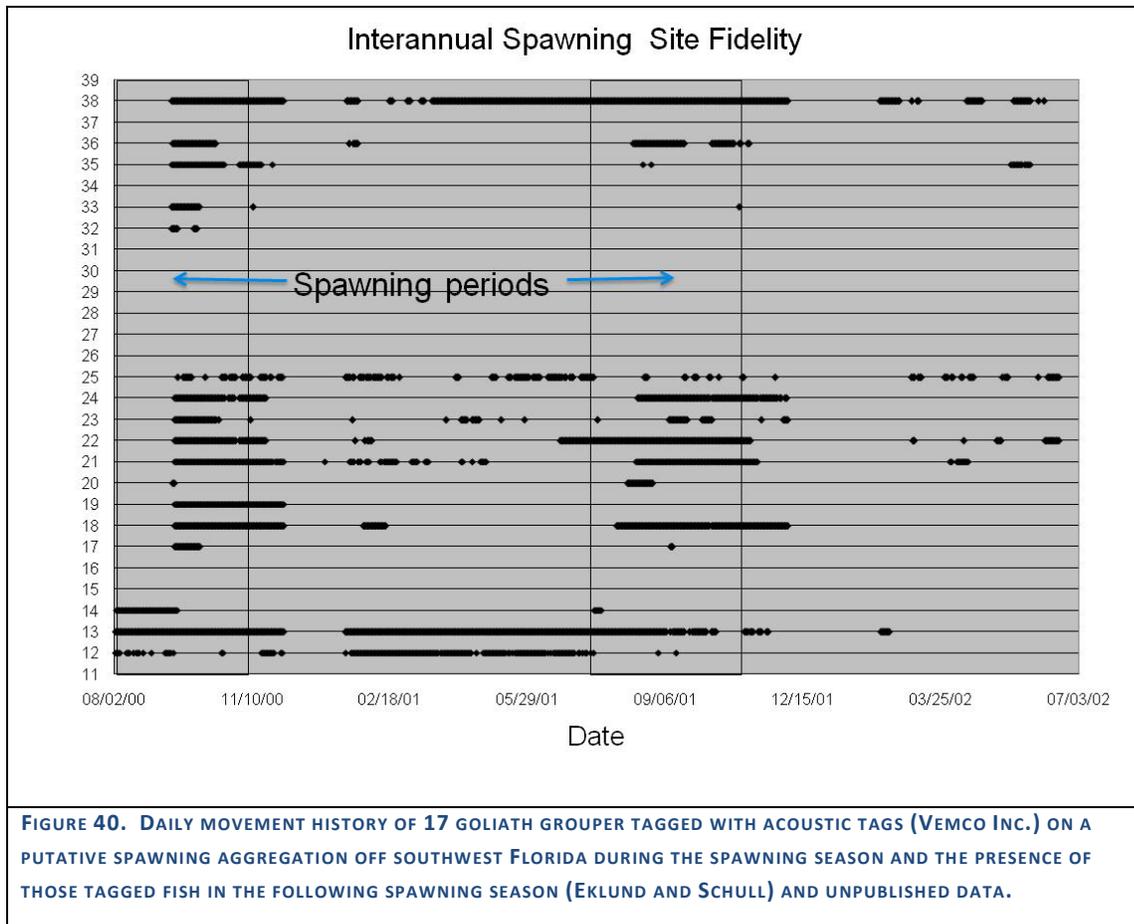
FIGURE 39. TIME SERIES OF ACOUSTIC RECORDINGS MADE AT THE ATLANTIC GOLIATH GROUPER AGGREGATION. THE MAJORITY OF ACOUSTIC ENERGY IN THESE RECORDINGS WAS PRODUCED BY GOLIATH GROUPER, HOWEVER THEY WERE MADE DURING LATE EVENING AND EARLY MORNING HOURS ONLY AND SO DO NOT SHOW THE DIEL PATTERN DOCUMENTED BY MANN ET AL. (2008)

so the second spawning event probably took place after moonrise, based on the stage of egg development.

The sound recordings from *The Caves* (Figure 39) indicated that the output produced by the goliaths increased throughout the night to 0700, when the receiver was

retrieved. If we had left the receiver in place for 24 hrs, we would have likely seen a drop in sound levels throughout the daytime period. No sounds were recorded from *The Fantastico* because the receiver failed.

Spawning site fidelity.-- Adult goliath grouper tagged with acoustic tags showed considerable site fidelity in the Gulf of Mexico (**Figure 40**). Fifteen of seventeen tagged fish were detected on the



same aggregation site on two consecutive years. Some appearances were quite brief and some periodically visited the site during non-spawning times. One of the fish we tagged during this period appeared the next day on another aggregation site 3.5 nm away, suggesting that some individuals at least move rather rapidly among spawning sites.

Four spawning sites (four of the five sites in black on **Figure 33**) were monitored from the 1980s through 2004. All monitoring prior to 1990 was conducted by commercial fisherman, Don DeMaria. We used his direct count methods as a first approximation for all monitoring to ensure that the data were comparable, and represented the data as the mean number of goliath groupers at each site. It is clear

that the number of fish on spawning sites (**Figure 41**) has increased since the 1990 closure. Anecdotal information strongly suggests that the number of spawning sites has also increased.

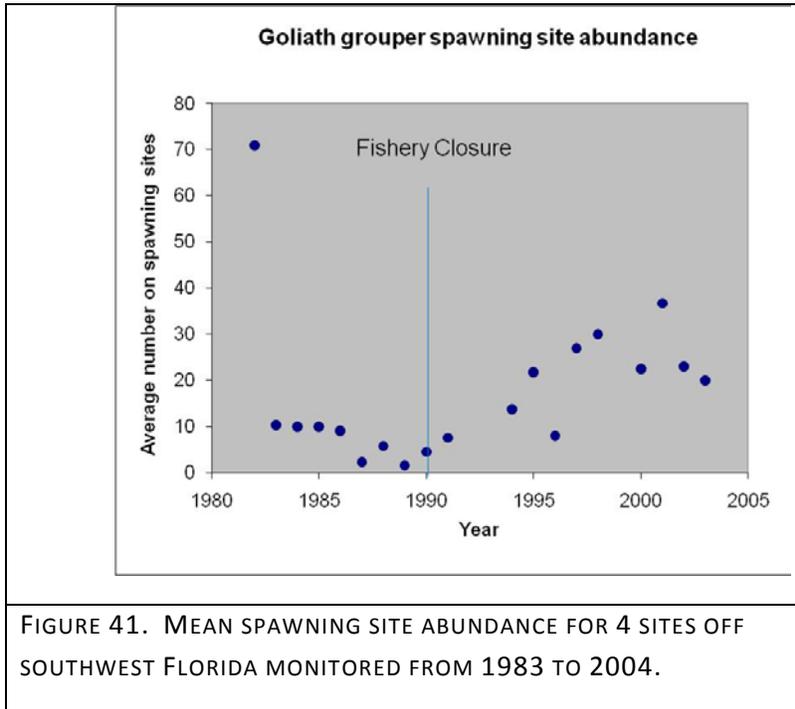


FIGURE 41. MEAN SPAWNING SITE ABUNDANCE FOR 4 SITES OFF SOUTHWEST FLORIDA MONITORED FROM 1983 TO 2004.

DISCUSSION

Goliath grouper are managed based on an SPR (spawning potential ratio) of 50%. Current information on reproductive parameters includes studies by Bullock et al. (1992) and Colin (1990), and information included in the review by Sadovy and Eklund (1999). Based on gonad histology, Bullock et al. (1992) showed that goliath grouper from the eastern Gulf of Mexico spawn in the late summer and fall. Lara et al. (2009) showed from back-calculated daily rings of early juveniles from the Ten Thousand Islands, FL that spawning occurs from July through October. The ovarian structure and oogenesis pattern of goliath grouper has not yet been described (Sadovy and Eklund 1999), and no estimates of fecundity have been made, although they are likely batch spawners like other groupers. If they also have indeterminate fecundity, then fecundity estimates will need to be based on size/age at first maturity, batch fecundity, spawning frequency, and seasonal spawning duration (Hunter and Macewicz 1985). It is possible that size at first spawning (= functional maturity), batch fecundity, spawning season duration, and spawning frequency can be measured directly on the spawning sites using some of the methods we describe in this report.

The LARS permanent recording devices have allowed us to monitor these soniferous aggregations continuously for prolonged periods of time. Such recordings provide detailed information on diel and seasonal periodicity. This is important because most marine fishes show a diurnal pattern of spawning that can range from sunrise to evening (Holt et al. 1985). Thus, identifying the diel periodicity of peak spawning is necessary to collect gravid females or eggs for fecundity estimates (Hunter and Macewicz 1985, Peebles 2002), to field-age POFs (Alheit et al. 1984), and to detect and map spawning aggregations (Samoilys 1997). Sampling times must be synchronized with spawning times for reproductive parameters to be accurate. It is possible that we can measure all these parameters non-destructively on the spawning aggregations using the methods we have used in the past through acoustic monitoring, sampling of fertilized eggs, and gonad biopsies.

Sexual pattern (hermaphroditic or gonochoristic) can also affect how vulnerable a species is to fishing pressure. Sequential hermaphrodites may be more vulnerable to overfishing than gonochorists if there is sex-specific fishing mortality rates (Coleman et al. 1996). Whether or not goliath grouper are hermaphroditic (protogynous) is still inconclusive. Goliath grouper testes have been reported as having a lumen and peripheral, sperm-collecting sinuses like the males of most protogynous hermaphrodites (Smith 1971) and at least one testes has been reported to have a few regressed oocytes (Bullock and Smith 1991). However, Bullock et al. (1992) collected males and females with substantially overlapping age compositions (males 3-26 years and females from 0-36 years) and a nearly 1:1 sex ratio. In addition, they did not find any sexual differences in growth pattern. Lastly, they report that males matured at slightly smaller and younger ages than females. None of these patterns are what would be expected if goliath grouper are in fact demonstrating protogeny.

Assessing the reproductive capacity of fish populations is important to proper long-term management of these resources (Goodyear 1993, Myers and Barrowman 1996). Current management practices in United States domestic marine fisheries require that managers determine what level of fishing pressure leads to recruitment overfishing (i.e., the adult stock fished beyond the point where it can replace itself under natural spawning conditions) (Powers 1996). However, this often necessitates an in-depth knowledge of a stock's reproductive biology at a time when the stock is perceived as being at great risk. Although the US stock of goliath grouper is undergoing a recovery from the time of the harvest ban in 1990 (Porch et al. 2006), the current lack of data on the goliath grouper's life history parameters makes it difficult to determine both the level of the stock's recovery and the level of harvest it can sustain, if any.

We believe that many methods are available for non-lethal estimation of reproductive parameters. Biopsy methods within and outside of the spawning season are appropriate to determine maturity, sex ratios and oogenesis patterns. Other methods have been developed to determine sex ratios during the spawning season (Heppell and Sullivan 2000) which involve vitellogenin assays from small amounts of tissue. Collection of fertilized eggs on spawning sites together with acoustic methods may be appropriate in defining not only diel patterns of spawning but also fecundity, spawning frequency, reproductive seasons, and lead to maps of spawning sites which may be slated by fishery management for special protection.

Sound production.--Goliath grouper were prolific producers of single-pulse low-frequency (60 Hz) sounds. The mechanism of sound production is likely to be similar to that for Nassau grouper (*Epinephelus striatus*), which involves contraction of sonic muscles connected to the vertebrae and swimbladder (Hazlett and Winn 1962). Based on the structure of the waveform, which begins with a rapid onset followed by a rapid decay (Figure 3), the swimbladder system appears to be highly damped, like that of the toadfish (Fine et al. 2001). There is often an inverse relationship between size and dominant frequency (Myrberg et al. 1993, Lobel and Mann 1995) of fish calls, and goliath grouper sounds are among the lowest frequency fish sounds that have been recorded to date. Sound production occurred on a diel basis, with the highest levels between 01:00 and 03:00 h. This pattern was consistent between the 2 separate aggregation sites in the Gulf, with recordings made 2 yr apart. There was also a strong lunar pattern at both sites, with reduced sound production around the full moon. Recordings were made at both aggregation sites during the peak of the spawning season and show high levels of sound production nightly, resembling patterns documented for sciaenids (Locascio and Mann 2005, Locascio and Mann 2008) except that they tended to be later in the night than most sciaenid species. The acoustic telemetry data, while limited to 1 individual, showed that the fish was resident at the aggregation site for 2 mo (the duration of the record), traveling out of the range of the receiver for less than 1 d. Goliath groupers show strong site fidelity in the adult and juvenile stages (Koenig et al. 2007, Koenig and Coleman this report).

The purpose of using the depth encoded tag was to attempt to document spawning ascents that have been observed in other reef fish species (Colin and Clavijo 1988). Thus, the ping rate was set high to capture such events. A number of ascents were detected in the depth record, with the majority occurring near midnight and 03:00 h. The forays at 03:00 h occurred just after peaks in sound production. The excursions recorded with the acoustic telemetry likely represent spawning ascents. This is supported by their high amplitude and short duration. It is also supported by collections of goliath

grouper eggs on two spawning aggregations and by our night-time (2230 EST) observation of adult goliath grouper rolling at the surface at *The Fantastico* spawning aggregation (33 m deep).

Goliath grouper eggs: We know of no other researchers who have collected goliath grouper eggs in the wild. This of course requires a knowledge of diel spawning times, which we determined through the use of active and passive acoustics. Because the acoustic signals recorded on spawning aggregations suggest that spawning does not take place on full-moon nights, and because we collected goliath grouper eggs on last quarter moon and new moon nights, we assume that goliath grouper avoid spawning on bright nights. However, we intend to conduct future studies to test that assumption by conducting a series of experimental egg samplings throughout a full lunar cycle while measuring ambient light levels at depth. Only through these types of experimental studies can we directly determine diel spawning times in goliath grouper so that the assumptions of some researchers based on indirect evidence (e. g., Colin 1992, Lara et al. 2009) can be resolved. Egg sampling on spawning aggregations can also be used to precisely resolve the issue of spawning season as opposed to traditional studies (e.g., Bullock et al. 1992) which are based on gonad histology.

Overall, our results suggest that a combination of active and passive acoustic methods and the collection of eggs with passive down-stream nets can be used to monitor spawning activity. It is possible to determine spawning frequency for individual females on spawning site. Eventually sound may be used as a proxy for spawning activity. These types of studies can augment traditional studies and allow mapping of goliath grouper aggregation sites over large spatial areas at relatively low cost. Passive acoustics can be used to gain knowledge of goliath grouper spawning aggregations and aid in their conservation and management through long-term monitoring, which would otherwise be difficult given their location relatively far from shore.

Our ultimate goal is to determine when and under what conditions goliath grouper are spawning (moon phase, ambient light, water temperature, season, etc.) and to determine reproductive parameters (spawning season, fecundity, spawning frequency, etc.) so that the data can be used: (1) in physical oceanographic models to predict larval transport and recruitment patterns, and (2) in stock assessment models to manage goliath grouper stocks sustainably.

Changing Seas, PBS-Miami , accompanied us on our research cruises involving the spawning biology of goliath grouper and has produced a special episode on goliath grouper recovery in Florida. The special can be seen at (<http://www.changingseas.tv/episode101.html>).

ACKNOWLEDGMENTS

We thank Don DeMaria, a commercial spear fisherman and conservationist (Summerland Key, Florida); recreational fishermen, Rich Johnson, Jim Fyfe, Charlie Sobczak, and Tony Grogan; commercial fishermen, Christian Boniface, Danny Tankersley and David Sauls; Chris Koepfer, (Lee Co. Artificial Reef Program) ; Christy Semmens (REEF Program); David Mann, Jim Locacio (University of South Florida); Kelly Kingon, Jimmy Nelson, Chris Peters, Nancy Hyams, Justin Lewis, Dennis Swanson (Florida State University); Jeff Ueland (Bemidji State University); Paul Richards (for developing the bioenergetics model and for serving as technical monitor, NOAA Fisheries, Miami). This project was funded by the National Oceanic and Atmospheric Administration (NA05NMF4540045; FSU Project No. 016604). All research was conducted in accordance with institutional and national guidelines concerning the use of animals in research, and was approved by the Institutional Animal Care and Use Committee (IACUC) of The Florida State University (Protocol #9902) and under a permit from the Florida Fish and Wildlife Conservation Commission (FWC 05-SRP-690A).

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APPENDIX

Table A1. Goliath grouper commercial landings in thousands of pounds, excluding foreign catch for each gulf state; percent catch by gear type from 1972-1987 (Sadovy and Eklund 1999)

State	Year															
	1972	1973	1974	1975	1976	1977	1978	1979	1980	1981	1982	1983	1984	1985	1986	1987
Florida	178		190	219	218	236	227	189	238	213	185	206	105	120	108	100
Alabama	57	48	25	19	14	17	4	3	2	6	13	14	7	13	1	0
Mississippi	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
Louisiana	0	6	0	0	0	0	0	0	0	0	0	1	0	0	0	1
Texas	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gear type																
	1972	1973	1974	1975	1976	1977	1978	1979	1980	1981	1982	1983	1984	1985	1986	1987
entangling net	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
handline/bandit	97	89	93	85	92	93	92	87	88	81	71	66	69	46	40	40
longline/buoy	0	0	0	0	0	0	0	0	7	11	20	25	19	28	20	14
fish trap	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
shrimp trawls	3	11	7	15	7	6	6	7	7	1	1	1	2	0	3	2
fish trawls																
spears etc.						1	1	5	4	6	8	8	11	26	47	45
Total landings	235	245	215	238	232	253	231	192	240	219	199	221	112	133	109	101
Average no. fish	980	860	952	928	1012	924	768	960	876	796	884	448	532	436	404	

Table A2. Goliath grouper recreational landings for the U.S. Gulf of Mexico (Sadovy and Eklund 1999).

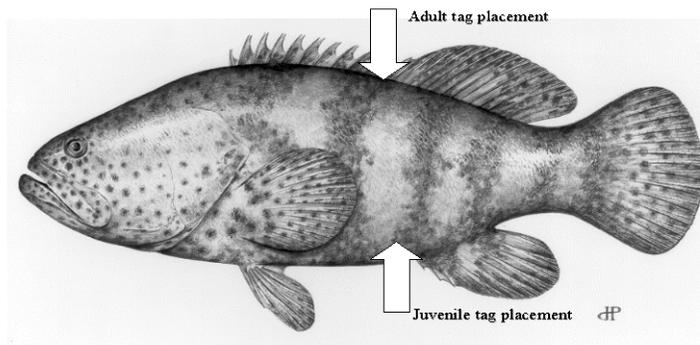
Recreational percentag distribution in numbers of fish recreational landed (A + B1) by species, state, year in Gulf

	1979	1980	1981	1982	1983	1984	1985	1986	1987	1979-1987
Florida	77.9	90.4	100	86.3	0	0	100	10.9	97.8	80.5
Alabama	0	0	0	0	0	0	0	0	0	
Mississippi										
Louisiana		9.6		13.7		100		89.1	2.2	18.2
Texas	22.1									1.2
Number	3823	16904	14330	10175	0	2456	10651	7963	3039	69341

About equal numbers of rec fish taken in state and federal waters, primarily by private boat anglers.

	1979	1980	1981	1982	1983	1984	1985	1986	1987	1988	1989	1990
shore	22.1	0									53.85	
party/chart headboat	0	35.4				100			39.7	100	1.71	0.32
private/ren	77.9	64.6	100	100		100	100	100	60.3		44.47	99.68
number	3823	16904	14330	10175	0	2456	10651	7963	3039	746	7138	1849
average	6589.5											

NOTICE TO FISHERMEN AND DIVERS



JUVENILE TAG



ADULT TAG



* See above for tag placement

HAVE YOU SEEN THIS FISH?

**We need your help in locating juvenile or adult
GOLIATH GROUPE**
(FORMERLY KNOWN AS JEW FISH)

If you see a tagged or untagged fish, please email:

coleman@bio.fsu.edu

Or write:

Dr. Felicia Coleman, The Institute for Fishery Resource Ecology, Department of Biological Science,
Florida State University Tallahassee, FL 32306-1100

When you contact us, we need the following information:

- your name, address, phone number, and e-mail
- the fish's location (prefer latitude and longitude), depth, and date seen
- the fish's estimated total length, weight
- the tag number and a photo if possible

DO NOT REMOVE TAG!

REMEMBER GOLIATH GROUPE ARE PROTECTED BY LAW.



DO NOT ATTEMPT TO HARVEST.

YOUR HELP IS GREATLY APPRECIATED



Figure A1. Poster sent to dive shops, fishing tackle shops, and marinas for advertizing our need for goliath grouper siting data.

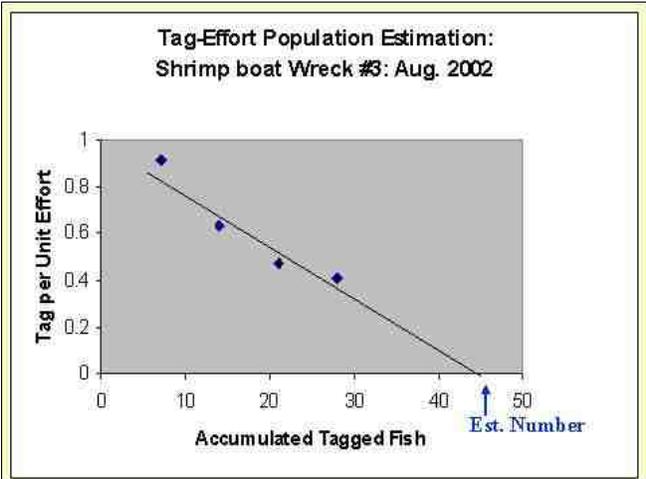


Figure A2. Example of tag-effort method of abundance estimation.

Table A3. Preliminary surveys of goliath grouper abundance using visual estimation (Roving Diver Survey), Tag-effort, and Petersen mark-resight estimation on low and high abundance sites.

Location	Type	Total Tagged	% Tagged	Pop., Visual	Pop., Tag-Eff. (95% CL)	Pop., Petersen (95% CL)
SW Fla wreck 1	Non-agg	6	75	7	5 (4-6)	8 (6-14)
SW Fla wreck 2	Non-agg	4	80	5	---	5 (4-7)
SW Fla Tower	Non-agg	3	100	3	3 (---)	---
SW Fla Calif. wreck	Agg site	36	57	40	67 (35-100)	63 (53-77)
SW Fla wreck 3	Agg site	28	67	30	45 (33-66)	42 (36-53)
SE Fla Natural	Agg site	24	52	30	---	46 (33-75)

Table A4. Fish and invertebrates found in the stomach contents of goliath grouper sampled in south Florida.

Fish	
Family	Species
Atherinopsidae	<i>Menidia sp.</i>
Balistidae	Species unidentifiable
Batrachoididae	<i>Opsanus beta</i>
Clupeidae	Species unidentifiable
Cyprinodontidae	Species unidentifiable
Cyprinodontidae	<i>Floridichthys carpio</i>
Dasyatidae	<i>Dasyatis americana</i>
Dasyatidae	<i>Dasyatis sp.</i>
Diodontidae	<i>Chilomycterus antillarum or schoepfi</i>
Diodontidae	<i>Chilomycterus reticulatus</i>
Diodontidae	<i>Chilomycterus spp.</i>
Haemulidae	<i>Haemulon macrostomum</i>
Haemulidae	<i>Haemulon plumieri</i>
Haemulidae	<i>Haemulon spp.</i>
Lutjanidae	<i>Lutjanus griseus</i>
Lutjanidae	<i>Lutjanus spp.</i>
Monacanthidae	Species unidentifiable
Mugilidae	<i>Mugil cephalus</i>
Mugilidae	<i>Mugil sp.</i>
Ogcocephalidae	Batfish species unidentifiable
Ostraciidae	<i>Acanthoscracion quadracornis</i>
Sparidae	<i>Archosargus probatocephelus</i>
Sparidae	<i>Archosargus rhomboidalis</i>
Sparidae	<i>Lagodon rhomboides</i>
Sphraenidae	<i>Sphyaena barracuda</i>
Invertebrates	
Family	Species
Calappidae	<i>Calappa flammea</i>
Calappidae	<i>Calappa sp.</i>
Calappidae	<i>Hepatus epheliticus</i>
Grapsidae	<i>Araus pisonii</i>
Grapsidae	Species unidentifiable
Majidae	<i>Libinia dubia</i>
Majidae	<i>Mithrax spinosissimus</i>
Majidae	<i>Pitho anisodon</i>
Majidae	<i>Pitho quadridentata</i>
Majidae	<i>Pitho sp.</i>
Marginellidae	<i>Prunum apicinum</i>
Palinuridae	<i>Panulirus argus</i>
Penaeidae	<i>Farfantepenaeus duorarum</i>
Penaeidae	<i>Farfantepenaeus spp.</i>
Porcellanidae	<i>Petrolisthes galathinus</i>
Portunidae	<i>Callinectes sapidus</i>
Portunidae	<i>Callinectes sp.</i>

Portunidae	<i>Portunus depressifrons</i>
Portunidae	<i>Portunus gibbesii</i>
Portunidae	<i>Portunus sayi</i>
Portunidae	<i>Portunus sp.</i>
Xanthidae	<i>Eurypanopeus depressus</i>
Xanthidae	<i>Eurytium limosum</i> or <i>Eurypanopeus abbreviatus</i>
Xanthidae	<i>Menippe mercenaria</i>
Xanthidae	<i>Panopeus americanus</i>
Xanthidae	<i>Panopeus sp.</i>
Xanthidae	Species unidentifiable