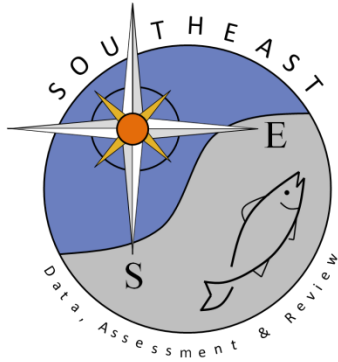


# Evaluation of the Efficacy of the Current Minimum Size Regulation for Selected Reef Fish Based on Release Mortality and Fish Physiology

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**SEDAR41-RD46**

6 August 2014



# **EVALUATION OF THE EFFICACY OF THE CURRENT MINIMUM SIZE REGULATION FOR SELECTED REEF FISH BASED ON RELEASE MORTALITY AND FISH PHYSIOLOGY**



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**Mote Marine Laboratory Technical Report No. 1176**  
**Submitted May 28, 2008**



**MOTE MARINE LABORATORY**

*MARFIN 2008 ~ Evaluation of the Efficacy of Current Minimum Size Regulations for Selected Reef Fish Based on Release Mortality and Fish Physiology*



*This report should be cited as follows: K. M. Burns, N.J. Brown-Peterson, R.M. Overstreet, J. Gannon, P. Simmons, J. Sprinkle and C. Weaver. 2008. Evaluation of the Efficacy of the Current Minimum Size Regulation for Selected Reef Fish Based on Release Mortality and Fish Physiology. Mote Marine Laboratory Technical Report No. 1176 funded by NOAA under MARFIN Grant # NA17FF2010. 75p.*

## ABSTRACT

This project contained multiple objectives. Study objectives included:

1. Test the hypothesis that red grouper are more susceptible to depth-induced mortality than red snapper, based on swim bladder size, thickness, and the number of rete mirabile and gas gland cells in the swim bladder.
2. Test the hypothesis that smaller red grouper (< 30.5 cm) survive rapid decompression better than larger (> 38 cm) fish based on changes in swim bladder structure with fish length between 30.5 – 38 cm.
3. Test the hypothesis that circle hooks greatly reduce release mortality in red snapper.
4. Quantify undersize and legal catch for red grouper, *Epinephelus morio*, gag, *Mycteroperca microlepis*, red snapper, *Lutjanus campechanus*, mangrove snapper, *Lutjanus griseus* and vermilion snapper, *Rhomboplites aurorubens*.
5. Obtain catch and release mortality rates relative to depth and gear for target species.
6. Determine tag shedding rates for red grouper, gag, and red snapper tagged with passive integrated transponder (PIT) tags and single-barbed dart tags.
7. Obtain movement patterns for target species in the Gulf of Mexico and South Atlantic off Florida.

Objectives were accomplished by combining histological examinations to assess general appearance of swim bladders, gas glands, rete mirabile, and hemorrhages with necropsies of red grouper and red snapper acute mortalities to determine cause of death and to examine swim bladder condition. Recaptures of red snapper and red grouper caught on circle and J hooks, tagged, and released were compared to determine release mortality relative to hook type. Red grouper, gag, red snapper, vermilion snapper, and mangrove snapper caught off Florida were quantified and measured aboard headboats, charter, and recreational vessels by Mote Marine Laboratory (MML) staff, student interns, and volunteer taggers. Target species were tagged with Hallprint® single-barbed plastic dart tags and released off recreational-for-hire and recreational vessels by MML staff, interns, and volunteer taggers. Tag number, species, fish length, date, depth (ft), gear type, and fish condition were recorded for each tagging event. Some target species were tagged with both Hallprint® single-barbed plastic dart and PIT tags. MML staff and interns used PIT tag readers to check all target species caught aboard headboats. Recaptures were reported via a dedicated hotline. Data included tag number, date, depth (ft), gear type, fish condition, and whether fish were re-released. A quarterly top tagger prize (\$50), an annual tag lottery (\$100; both tagger and fisher reporting recapture), and a quarterly newsletter were employed to keep anglers informed and provide incentive for tagging and reporting target species. Original tagging and recapture locations were recorded to the nearest 1 minute of latitude and longitude and fish movements were calculated from these data.

Conclusions and Recommendations: 1) Red grouper had larger (in relation to body size), thinner swim bladders than red snapper. Red snapper swim bladder ruptures were smaller than those of

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red grouper. Red grouper > 380 mm FL had an area on the posterior swim bladder ventral wall, absent in red snapper that contained some rete and convoluted tissue that could aid in gas absorption and secretion. Rete area increased significantly with body length (FL) for both species, although the relationship between rete area and fish length was weak, particularly for red snapper. Red snapper, however, had a significantly higher percentage of rete area than red grouper when adjusted for fish length (ANCOVA,  $F_{3,196} = 41.042$ ,  $p < 0.001$  overall,  $p = 0.022$  by species). Gas gland area was not related to FL for red snapper. The relationship was significant, but weak for red grouper. Red grouper had a significantly higher percentage of rete area occupied by gas gland than did red snapper when values were adjusted for fish length (ANCOVA,  $F_{3,200} = 13.258$ ,  $p < 0.001$  overall,  $p = 0.007$  by species). Morphologically, red grouper had a closer association and more connections between gas gland cells and rete than red snapper. Both species had increased rete and swim bladder hemorrhaging with increasing 50 mm length intervals (ANOVA, red snapper rete,  $F_{1,8} = 20.41$ ,  $p = 0.003$ ,  $r^2 = 0.708$ , red snapper gas gland,  $F_{1,8} = 17.38$ ,  $p = 0.004$ ,  $r^2 = 0.672$ , red grouper rete,  $F_{1,11} = 16.21$ ,  $p = 0.002$ ,  $r^2 = 0.580$ , red grouper gas gland,  $F_{1,11} = 19.77$ ,  $p = 0.001$ ,  $r^2 = 0.631$ ). Retal hemorrhaging was significantly higher in red grouper than in red snapper, when adjusted for length (ANCOVA,  $F_{3,21} = 5.96$ ,  $p = 0.026$ ), but there was no significant difference between species in gas gland hemorrhaging ( $p = 0.786$ ). Overall, red snapper survived rapid decompression better than red grouper because of smaller tears in the swim bladder and less tendency to hemorrhage, especially in smaller fish. **2)** Necropsies revealed most red snapper died from hook mortality. **3)** Circle hooks did not increase survival of red snapper but did enhance red grouper survival. Of 3,205 red snapper caught on circle hooks, 8.0% were recaptured; whereas, of the 2,263 red snapper caught on J hooks, 11.4% were recaptured. **4)** Red grouper (473 legal and 5,318 undersized), gag (442 legal and 2,855 undersized), red snapper, (1,154 legal and 5,737 undersized), vermilion snapper (563 legal and 564 undersized), and mangrove snapper (904 legal and 3,138 undersized) were measured during 467 headboat, 90 charter, and 427 recreational fishing trips. **5)** Red grouper ( $n = 4,999$ ), gag ( $n = 2,704$ ), red snapper ( $n = 5,481$ ), vermilion snapper ( $n = 465$ ), and mangrove snapper ( $n = 3,180$ ) were tagged. Recapture rates were 8.7% for red grouper, 11.9% for gag, 9.7% for red snapper, 0.2% for vermilion snapper, and 8.7 % for mangrove snapper. **6)** Six hundred six fish were double tagged (474 red grouper, 3 gag, and 129 red snapper). Most fish were recaptured with both tags present. Tag shedding rates were calculated as 0.2% for red grouper, 33% for gag, and 0% for red snapper; however, since only three gag were double tagged, the tag shedding rate for gag should be considered unreliable **7)** Most fish showed limited movement, but some were recaptured up to 360.3 km (red grouper) and 583.2 km (red snapper) from the original tagging location.

Circle hooks should be used when fishers target red groupers. Circle hooks do not enhance red snapper survival. Although the effects of barotraumas affect both red grouper and red snapper, red grouper begin to experience difficulties at 27.4 m (90 ft) whereas red snapper trauma occurs closer to 42 m (140 ft). Both species benefit from venting, when the procedure is performed on fish from the appropriate depth.

## EXECUTIVE SUMMARY

Reef fish, particularly snappers, support extremely important commercial and recreational fisheries in both the Gulf of Mexico and South Atlantic resulting in the necessity to resolve critical management issues. Although catch quotas, trip limits, and closed seasons are used to regulate the fishery, minimum size limits are the primary tool used to manage reef fish fisheries. Minimum size limits are intended to prevent growth and recruitment overfishing by allowing some portion of fish in a cohort to grow and reproduce at least once before dying of natural or fishing related causes. Minimum size limits apply to both commercial and recreational fishers and require that all undersized fish be released regardless of the location, water depth or fish condition. Survival rates of these undersized fish become critical in determining the efficacy of the minimum size rule.

Depth induced mortality due to injuries sustained during rapid decompression is an important factor that seriously impacts survival of undersized reef fish that are discarded due to minimum size regulations (Render and Wilson 1993; Gitschlag and Renaud 1994; Render and Wilson 1996; Collins et al. 1999). While many reef fish species suffer mortality from rapid decompression injuries, there are differences in mortality among species related to their anatomy, physiology, and behavior. If not allowed to return to an appropriate depth immediately, red grouper (*Epinephelus morio*) are more susceptible to injury and death from rapid decompression at shallower depths than are red snapper (*Lutjanus campechanus*). Although Wilson and Burns (1996) have shown that red grouper, gag, and scamp can potentially survive decompression in sufficient numbers to justify a minimum size rule if fish are rapidly allowed to return to the corresponding habitat depth, differences in morphology influence survival.

The swim bladders of physoclistic fishes (including snappers and groupers) are able to adjust to pressure/depth changes over a relatively rapid period (Jones and Marshall 1953). However, the swim bladders of both red snapper and red grouper rupture with rapid change from two to one atmospheres of pressure (20 - 10 m, or 66 - 33 ft). Data from hyperbaric chamber studies (MARFIN Award NA97FF0349) showed that while both red grouper and red snapper can easily survive rapid decompression from 21m (70 ft) some red grouper suffered trauma at 27 m (90ft) but red snapper did not. There were even greater differences in their ability to tolerate rapid decompression from deeper depths ( $\geq 40$  m, or 140 ft). While some red snapper did suffer mortality or sub-lethal effects during rapid decompression from depths  $\geq 40$  m., many survived at 1 atmosphere pressure when vented. In contrast, red grouper never survived rapid decompression from depths of 40m and greater to 1 atmosphere pressure, even when vented (Burns et al. 2004). Red grouper survival from this depth and deeper occurred only if the red grouper were vented and immediately allowed to return to habitat depth or immediately returned to the capture depth using a rapid release device. This difference in survival demonstrates that morphological and physiological differences between the two species determine the ability to adjust to rapid depressurization.

Red snapper have smaller but thicker swim bladders than red grouper, and swim bladder-ruptures in red snapper tended to be smaller than those in red grouper. Red grouper from 318 to 381 mm

FL developed a secondary vascularized structure on the posterior ventral surface of the swim bladder that was not seen in red snapper; this secondary structure contained convoluted tissue incorporating some rete and a greater number of gas gland cells that probably aided in gas absorption and resorption. Histological analyses of swim bladders from 62 red snapper (123 – 674 mm FL) and 138 red grouper (205 – 766 mm FL) showed rete area significantly ( $p < 0.05$ ) increased with body length (FL) for both species, but gas gland area was not significantly related to FL of red snapper. Red snapper had a significantly higher percentage of rete area in the swim bladder than did red grouper, although there was no difference in the percentage of area occupied by gas gland between species. However, red grouper had a significantly higher percentage of the rete area occupied by gas gland than did red snapper. The percentage of fish showing hemorrhaging in both rete and gas gland increased significantly with FL for both species, but retal hemorrhaging was significantly higher in red grouper when adjusted for fish length. These differences in structures of the swim bladder explain the differences in depth-induced mortality between these two species.

Research results from MARFIN Awards NA97FF0349 and NA87FF0421 have shown that, although all reef fish species can suffer mortality from rapid decompression and hook injuries, there are differences in mortality rates among species related to their anatomy, physiology, and behavior. Knowledge of the number and percent of reef fish that succumb to immediate mortality is important to beneficial management of the species. Necropsies revealed that red snapper were much more susceptible to J-hook mortality than red grouper. Encouraging reports regarding the use of circle hooks to enhance fish survival for other species lead to testing the hypothesis that circle hooks would increase red snapper survival because mortality from hooks had been considered the leading cause of mortality in red snapper. Circle and J hooks were evaluated to determine hook mortality rates for red snapper for fish caught aboard charter vessels, recreational vessels, and headboats. Results, however, did not support the hypothesis that hooks caused extensive mortalities of red snapper and so the hypothesis was rejected. A similar hypothesis for red grouper was accepted because results showed that red grouper survival was significantly enhanced for fish caught on circle hooks.

In addition, recapture rates of 8.7% for red grouper, 11.9% for gag, 9.7% for red snapper, 0.2% for vermilion snapper, and 8.7 % for mangrove snapper were obtained for these targeted species. Most fish showed limited movement, but some were recaptured as far away as 360.3 km (red grouper) and 583.2 km (red snapper) from the original tagging location. Of the 606 double tagged target species (474 red grouper, 3 gag, and 129 red snapper), most individuals were recaptured with both tags present. Tag shedding rates were calculated as 0.2% for red grouper, 33% gag, and 0% for red snapper. The tag shedding rate for gag should be disregarded since it was based on one recapture and not reliable.

## PURPOSE

Survival rates of undersized fish can be easily incorporated into stock assessments, as they have been for Gulf of Mexico red snappers (Goodyear 1995) using well-established methods (Waters and Huntsman 1986). Data from MML's reef fish database have been used by NMFS, for stock assessment for red grouper, red snapper, gag and vermilion snapper. However in addition to providing these data, the purpose of this project was to determine the morphological and physiological reasons causing differences in depth and hook induced mortality between red grouper and red snapper through hypothesis driven experiments.

### A. PROBLEM DESCRIPTION

According to NOAA, by-catch affects at least 149 species or species groups in 159 distinct U.S. fisheries (NOAA 1998). These effects have been documented by Dayton et al. (2002), who showed the grouper/snapper complex is one of the groups affected. Reef fishes, particularly groupers and snappers, support extremely important commercial, recreational-for-hire and recreational fisheries in both the Gulf of Mexico and South Atlantic. Due to minimum size regulations, undersized by-catch occurs in all sectors of the reef fish fishery. Although catch quotas, trip limits, and closed seasons are used by NMFS and fishery management councils to regulate the reef fish fishery, minimum size limits are the primary tools used to manage reef fish fisheries. Minimum size limits result in significant undersized by-catch in the recreational and recreational-for-hire reef fish fisheries off Florida (personal observation). “For decades, by-catches were mostly ignored by scientists working on stock assessment, by fisheries managers, and by environmentalists.” (Hall et al. 2000). According to Dayton et al. 2002, “In many cases, the underestimation of by-catch mortality has lead to overly optimistic estimates of the environmental impact of fishing as a whole”, on the other hand over estimates of by-catch mortality can result in estimates not reflecting true stock levels..

“The ecological ramifications of dumping all of this material -the by-catch and offal- overboard can lead to behavioral changes in resident organisms, particularly among scavenger species” (Camphuysen et al. 1995). The most visible surface scavengers feeding on by-catch are seabirds. Less visible are the numerous sharks and other fish predators that follow fishing vessels to take advantage of the thousands of dead or stunned animals thrown overboard (Dayton et al. 2002). In Florida waters, especially off Panama City, the bottlenose dolphin is a conspicuous predator of headboat by-catch. Limited observations by Mote Marine Laboratory staff aboard headboats have indicated that some dolphin pods may be selective in the species of by-catch they consume. Off Panama City, Florida, pods were observed to consume red snapper, while ignoring red grouper and gag undersized by-catch.

“By-catch problems are notoriously difficult to manage, but this does not diminish the urgent need for solutions. Unfortunately, the difficulty is compounded by a management legacy of poor monitoring and inadequate regulation in the U.S.” (Dayton et al. 2002). To address this matter, NOAA/NMFS has acknowledged this deficiency and has allocated funds for by-catch research. However, traditional means of obtaining by-catch estimates from the recreational and

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recreational-for-hire has many difficulties. During telephone-based surveys and dockside or on-water roving interviews, fishers willing to participate may not remember important information such as the total number of fish caught, fish lengths and the location of where each fish was caught. Unlike commercial fisheries where fishers are required to record catches in logbooks, recreational and recreational-for-hire fishers are under no such obligation. One way to address this difficulty was by including fishers in the data collection process, as suggested by Dayton et al. (2002). “Broader monitoring programs include both fishery-independent and fishery-dependent data-gathering systems that require involvement of the fishers. Fishery-independent systems should emphasize large scale tagging programs that can provide better information on spatial stock dynamics, growth, and fishing mortality rates on both well-known and understudied species.” The inclusion of fishers can be useful for sampling programs for long term monitoring but can also provide a mechanism for testing research hypotheses on undersized by-catch survival, collecting biological samples and testing techniques to enhance discarded undersized by-catch.

Hook mortality and depth induced mortality due to injuries sustained during rapid decompression constitute two factors seriously influencing survival of undersized discards (Render and Wilson, 1993; Gitschlag and Renaud, 1994, personal observation). Research results from ongoing MARFIN Awards NA97FF0349 and NA87FF0421 (Burns et al. 2002) have shown that, although all reef fish species can suffer mortality from hook or rapid decompression injuries, there are differences in mortality among species related to their anatomy, physiology, and behavior. Red snapper are more susceptible to hook mortality while red grouper are more susceptible to injury and death from rapid decompression at shallower depths than are red snapper (Burns and Restrepo 2002.). Although Wilson and Burns (1996) have shown that red grouper, gag, and scamp can potentially survive decompression in sufficient numbers to justify a minimum size rule if fish are returned to habitat depth, depth (rapid changes in pressure) definitely influences survival. However, the anatomy and physiology of an individual fish species can also determine the depth at which serious injuries and death occur (personal observation) and understanding these differences are crucial to fishery management.

## **B. PROJECT OBJECTIVES**

- To test the hypothesis that red grouper are more susceptible to depth induced mortality than red snapper, based not only on swim bladder size and thickness, but also on the number of rete mirabile and gas gland cells in the swim bladder.
- To test the hypothesis that smaller red grouper (<30.5 cm) survive rapid decompression better than larger (>38 cm) red grouper because of changes in swim bladder structure with fish length between 30.5 – 38 cm).
- To test the hypothesis that circle hooks will greatly reduce release mortality in red snapper.
- To quantify undersize and legal catch for red grouper, *Epinephelus morio*, gag,

*Mycteroperca microlepis*, red snapper, *Lutjanus campechanus*, mangrove snapper, *Lutjanus griseus* and vermilion snapper, *Rhomboplites aurorubens*, in the recreational and recreational-for-hire reef fish fishery off Florida

- To obtain catch and release mortality rates relative to depth and gear for red grouper, *E. morio*, gag, *M. microlepis*, red snapper, *L. campechanus*, vermilion snapper, *R. aurorubens*, and mangrove snapper, *L. griseus*.
- To determine tag shedding rates and effects on growth and survival for fish tagged with single barbed dart tags in red grouper, gag, and red snapper.
- To obtain movement patterns for red grouper, gag, red snapper and mangrove snapper in the Gulf of Mexico and South Atlantic off Florida.

## APPROACH

### A. WORK PERFORMED

**Objective 1:** To test the hypothesis that red grouper are more susceptible to depth induced mortality than red snapper, based not only on swim bladder size and thickness, but also on the number of rete mirabile and gas gland cells in the swim bladder.

**Objective 2:** To test the hypothesis that smaller red grouper (<30.5 cm) survive rapid decompression better than larger (>38 cm) red grouper because of changes in swim bladder structure with fish length between 30.5 – 38.0 cm.

### Materials and Methods

#### Swim Bladder Collection and Processing:

Both the laboratory and field work to test the two hypotheses were conducted simultaneously. Laboratory research began with the collection of red grouper and red snapper in an effort to obtain specimens spanning the greatest available size range possible from fish caught off headboat fishing trips at various depths in the Gulf of Mexico and South Atlantic.

Upon arrival at MML, the staff member transporting the specimens logged in the samples to continue documenting the chain of custody. All red snapper and red grouper captured were examined for swim bladder inflation or rupture. Swim bladder gross morphology was examined and compared and the bladders were measured to determine an approximate size ratio of the red grouper and red snapper swim bladders in relation to fish length.

Each fish was measured (FL, mm) and swim bladders from a wide size range of each species were removed and preserved in 10% neutral buffered formalin (NBF). The swim bladder and surrounding body torso were occasionally preserved in 10% NBF. The swim bladders were placed into labeled jars and packed into containers for shipment to the University of Southern Mississippi (USM) campus in Ocean Springs for histological evaluation. Swim bladder and trip data were entered on PC computers using Excel spreadsheets. All data as well as the preserved samples were sent to USM for processing.

A portion of the anterior area of each swim bladder containing rete was placed into cassettes for histological processing. In instances when the rete occupied an area of the swim bladder too large to be placed into a single cassette, multiple sections were placed in separate cassettes such that all rete area was collected for histological processing. Additionally, a portion of the posterior area of each swim bladder was also placed in a cassette. For red grouper, the posterior section corresponded to the secondary swim bladder structure; for red snapper, the area preserved from the posterior portion of the swim bladder corresponded to the same area where the secondary structure occurs in red groupers. Tissues were rinsed overnight in running water, dehydrated in a series of graded ethanols and processed using standard histological techniques. Tissue was embedded in Paraplast® (Fisher Scientific), and blocks were sectioned at 4 µm and stained with Gill's hematoxylin (Statlab) and eosin phloxine (Polyscientific). Swim bladder tissue was inspected microscopically and the occurrence of rete, gas gland, convoluted tissue (posterior sections) and hemorrhaging was recorded. The areas of rete and gas gland were measured microscopically in each section, and the percentage of rete and gas gland was determined for each fish. In instances where several sections were taken from the same fish, total rete and gas gland area was calculated.

### *Statistical Analysis for Histological Comparisons*

Comparisons between red grouper and red snapper for rete area, gas gland area and hemorrhaging were determined using ANCOVA, with fish length as the covariate (SPSS version 11.5, SPSS, Inc., Chicago, IL). Hemorrhaging in rete and gas gland tissues was compared between species by length classes using ANOVA. All percentage data were arcsine square root transformed prior to analysis (Sokal and Rolf 1995). Data were considered significant if  $p \leq 0.05$ .

***Objective 3:*** *To test the hypothesis that circle hooks will greatly reduce release mortality in red snapper.*

### *Fish Tagging:*

Red grouper, gag, red snapper, vermilion snapper and mangrove snapper were tagged by MML staff, student interns and fishers from all sectors of the reef fish fishery, using

single barbed Hallprint® plastic dart tags inserted at an angle next to the anterior portion of the dorsal fin. These tags have already been used successfully in MML's Reef Fish Tagging Program. Information on the tag included tag number; MML's address and the 1-800 dedicated tagging telephone number at Mote. The telephones were answered personally during work hours and calls regarding tag return information were recorded on an answering machine on weekends, holidays and evenings.

Tagging data included date, hook type, gear type, tag number, time of day, species, bait used, water depth, fork length in inches, (transposed to metric units in the Lab), fish condition upon release, amount of time the fish was out of the water, whether or not the fish was vented, capture location to the nearest 5 degrees of latitude and longitude, number of hours fished, and the number of hours of the trip. For double tagged fish (red grouper, gag and red snapper) the PIT tag number was also included. Additionally, the fishing area and fishing sector (charter, headboat or recreational) were recorded for each tagged fish. Areas included the Florida Atlantic Ocean (St. Augustine to Miami), the Florida panhandle (Pensacola to Panama City), the North Florida Gulf of Mexico (Cedar Key to Tarpon Springs), the South Florida Gulf of Mexico (Tampa/Clearwater to Naples) and the Florida Keys.

If a fish was vented before release, abdomen deflation was accomplished by use of the venting device and protocol currently used by MML. The sharpened tube of small diameter (e.g., 18 gauge) needle was inserted at a 45° angle through the body wall of the bloated fish. The venting tool was held in place long enough to allow the expanded gases from the ruptured swim bladder to escape.

Return data included tag number, species, date of capture, gear type, bait type, water depth, fork length in inches, capture location, the overall condition of the fish, the condition of the area around the tag insertion site, and whether the fish was kept or released.

All data were entered on a PC computer using PARADOX software. Before data were transferred to become part of the permanent MML Reef Fish Tagging database, all newly entered data were proofed against the original field data sheet by a second individual to check for and correct any data entry errors. Following proofing, we electronically downloaded the data into the database. Data were transferred to NMFS semi-annually with semi-annual reports and made available for stock assessment workshops.

Tag returns were monitored to obtain estimates of survival. This evaluation of survivorship was accomplished by integrating new data into MML's ongoing long-term Reef Fish Tagging Program, data, which have proved very reliable (Schirripa and Burns 1998).

Returns from fish tagging were used as a means of estimating survival. This evaluation of survivorship was accomplished by integrating the experimental design into MML's

existing and long-term Reef Fish Tagging Program (discussion by Schirripa et al. 1993 and Wilson and Burns 1996).

#### Tag Lottery:

To encourage tag returns, a tag lottery including all fish recaptured each year was held in mid-January of each project year to allow maximum reporting time, especially for fish recaptures near the end of the project year. A cash reward of \$100 was paid to the winner and the original tagger unless he/she was affiliated with MML. MML staff and student interns were ineligible for the prize. The winning tag was chosen at random using a random number generating computer program.

#### Circle vs. J Hook Experiment:

This task was accomplished by asking volunteer taggers participating in this research to alternate their hook type between circle and J hooks during fishing trips. MML staff provided 4/0 and 7/0 zero offset circle hooks (donated by Linda Martin of Eagle Claw®) to participants. Participants were asked to tag and release red grouper and red snapper in approximately the same numbers and were kept informed of preliminary results in the RFSS Reef Fish MARFIN funded newsletter. Data were analyzed with regard to hook type and water depth during the original capture. Included in the evaluation was whether fish were vented or not vented.

**Objective 4:** *To quantify undersize and legal catch for red grouper, *E. morio*, gag, *M. microlepis*, red snapper, *L. campechanus*, mangrove snapper, *L. griseus* and vermilion snapper, *R. aurorubens* in the recreational and recreational-for-hire reef fish fishery off Florida*

#### Quantification of Undersized and Legal Catch:

MML personnel recorded the number and length of as many red grouper, gag, red snapper, vermilion snapper, and mangrove snapper caught during headboat trips as possible off the southwest coast of Florida (Sarasota), the eastern coast of Florida (St. Augustine) and in the northern Gulf, off Panama City, FL. MML personnel recorded the water depth, fish condition, and fate of the fish. Undersized fish in good condition were used in the tagging portion of the study.

Regulations determining legal size of fish varied for some species off the east and west coasts of Florida. Table 1 lists the minimum fish lengths of target species in effect during the course of this project.

Table 1. Minimum size lengths (TL) for target species by region that were in place during the course of this project.

Species	Florida Atlantic		Florida Keys		Florida Gulf	
	in	mm	in	mm	in	mm
Red Grouper	20	508	20	508	20	508
Gag	24	610	24	610	22	559
Red Snapper	20	508	20	508	16	406
Mangrove Snapper	10	254	10	254	10	254
Vermillion Snapper	11	279	11	279	11	279

Additional survival and length/frequency data from recreational-for-hire and recreational vessels were gathered by participating fishers. Information collected included tagging data from dart tagged fish, tag returns, number and length in inches of legal and undersized target species caught/trip, gear type, and number of hours fished.

**Objective 5:** *To obtain catch and release mortality rates relative to depth and gear for target species*

To accomplish this objective, we added gag, vermilion, and mangrove snapper as target species in the fish tagging portion of this study previously described. Tag returns were used to evaluate survivorship.

**Objective 6:** *To determine tag shedding rates and effects on growth and survival for red grouper and red snapper tagged with single barbed dart tags.*

Tag shedding rates were obtained by conducting double tag experiments. Fish were tagged with two tags, a PIT tag and a conventional Hallprint® plastic dart tag. Tag shedding rate was estimated from the frequency at which fish that were originally double tagged were returned with only one tag. PIT (passive integrated transponder) tags measuring 11 mm x 2 mm diameter were injected using modified hypodermic syringe/needles into the musculature directly behind a conventional Hallprint® plastic dart tag. The PIT tags consisted of a microchip contained in a glass capsule about the size and shape of a grain of wild rice, which was read by a reader instrument that energizes the chip by an inductive field. The microchip transmitted a unique digitally coded signal that was picked up by the reader instrument. When a microchip was detected, the reader instrument beeped and provided a digital readout of the tag number. The Hallprint® plastic dart tag was a single dart with an attached reddish orange plastic streamer.

MML staff and student interns aboard headboats scanned all red grouper and red snapper caught in the areas where the double tagging experiment was conducted. Tag shedding rates were estimated using the number of returned PIT/Hallprint® dart tag tagged fish

that shed their dart tags. Growth of double tagged fish was compared to red grouper and red snapper growth rates of fish that were tagged only with Hallprint® dart tags and recaptured.

Return data included tag number, species, date of capture, gear type, bait type, water depth, fork length in inches, capture location, the overall condition of the fish, condition of the area around the tag insertion site and whether the fish was kept or released. All data were entered on a PC computer using PARADOX software. Before data were transferred to become part of the permanent MML Reef Fish Tagging database, all newly entered data were proofed against the original field data sheet by a second individual to check for and correct any data entry errors. Following proofing, data were electronically downloaded into the database. Analysis of recapture data was carried out by standard statistical methods. Survival differences were evaluated by fish length (when possible), geographic location, hook type, and water depth.

***Objective 7: To obtain movement patterns for red grouper, gag, red snapper, and mangrove snapper in the Gulf of Mexico and South Atlantic Ocean***

Red grouper, gag and red and mangrove snapper tag and recapture data were used to examine movement patterns in both the Gulf of Mexico and in the South Atlantic along the east coast of Florida. Data were analyzed for general distributional trends. Red grouper, red snapper and gag movement was mapped by plotting capture and recaptures within a GIS and by calculating distance between points when sufficient data were available. Insufficient data were available for vermilion and mangrove snappers to perform most analyses. The spatial resolution of reporting (one minute of latitude and longitude) imposed restrictions on this analysis, so only moves greater than 3 km were used for analyses. A chi square test was used to determine if fish length was related to movement. Tests were run for both the Gulf and Atlantic.

Length/frequency data for red grouper, red snapper and gag were utilized to examine fish length (cm) with distance (km) from shore at time of tagging in the South Atlantic off Florida and in the eastern Gulf of Mexico. A length/frequency histogram of fish size by distance from shore at original capture was constructed and a graph with regressions of mean fish size (red dots) representing bins of 10 or more fishes and also a regression of individual fish size versus capture distance from shore (km) were superimposed upon the length/frequency histogram. Regressions of the  $r^2$  of the means (red solid line) and the  $r^2$  of each individual fish (cyan dotted line) were drawn. No means were calculated if there were less than 10 fishes in a size category. A line of best fit for the regressions was calculated using the equation  $y = m \cdot x + b$ , where  $y$  = distance from shore (km),  $m$  = slope,  $x$  = fish length (cm) and  $b$  =  $y$  intercept. A linear regression was run on the regression using all fish to determine if the line was significant i.e. the relationship between fish length and distance from shore was significant.

Spatial analyses were conducted using ArcInfo 9.0 (ESRI 2004). Ancillary data, such as bathymetry, were acquired from the state of Florida's Geographic Data Library. Movement data were projected in local UTM NAD 83 coordinate systems (16N and 17N in the Gulf and 18N in the Atlantic). Sigmaplot, Oriana, and GEODISTN (Syrjala 1996) were used to perform statistical analyses.

## **B. PROJECT MANAGEMENT**

**Ms. Karen Burns** (*MML Staff Scientist & Fish Biology Program Manager*) served as Principal Investigator and Project Manager. She provided overall supervision of the project ensuring that all work was completed in accordance with the S.O.W. She served as liaison among MML, NMFS, and USM-GCRL. Ms. Burns was responsible for the supervision of the laboratory and field research, reports, and newsletter publication.

**Ms. Nancy Brown-Peterson** (*Research Associate, USM-GCRL*), Co-Principal Investigator, was responsible for all swim bladder histology work associated with this project, including reading and analyzing each histological slide for swim bladder and gas gland tissues and determining the percentage of each tissue type in each individual. She was responsible for photodocumentation of the swim bladder biology section of the project. Ms. Brown-Peterson participated in report and manuscript preparation and publication as well as scientific presentations of the data. Ms. Brown-Peterson is currently or has been PI or co-PI on three NMFS-sponsored studies (NA04NMF450208, black grouper; NA17FF288, red snapper; NA04NMF4540411, blue marlin) and has over 25 years experience with fish histology and over 50 publications.

**Dr. Robin Overstreet** (*Professor, Department of Coastal Sciences, USM-GCRL*) was a Co-PI and served at no cost to the project. He was responsible for preparation, sectioning and staining of all swim bladder tissues, participated in the analysis of sectioned swim bladder tissues and preparation of reports and publications dealing with the USM-GCRL aspect of the studies. He has been at GCRL since 1969, and although his primary interest has been parasites and diseases, he has been heavily involved with the biology and morphology of many of the host animals including red snapper, groupers, mullets, sciaenids, and cobia. In addition to being Co-PI on two MARFIN studies (NMFS, NA17FF2010 and NA17FF2881), he is a Co-PI of NMFS NA17FU2841 (blue crab), and Principal Investigator on USDA CSREES Grant No. 2002-38808-01381 (shrimp farming), and NSF Award Nos. 0508856, 0529684, and 0608603 (fish digeneans).

**Mr. Nicholas Parnell** (*MML Senior Biologist*) participated in biological sample collection, tag/release, fish measurement, etc. Mr. Parnell was one the MML staff supervisors aboard headboat trips. He was responsible for biological sample data, making sure data were entered into the computer and that all samples were properly shipped to USM-GCRL. He was also responsible for ensuring all field research protocols were followed.

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**Mr. Jay Sprinkel** (*MML Senior Biologist*) served as the data manager for this project. As such, he was responsible for supervising data entry, setting up files, and producing graphs and tables for newsletters, reports, and presentations.

**Mr. Peter Simmons** (*MML Staff Biologist*) was responsible for data entry and ensuring all recaptured information from volunteer taggers were obtained. Mr. Simmons participated in tagging trips aboard various headboats. He tagged and released undersized target species at sea, measured any legal kept fish, and collected swim bladder samples. Mr. Simmons also assisted with final report preparation.

**Mr. Roger DeBruler** (*MML Staff Biologist*) participated in headboat tagging trips. He tagged and released undersized target species at sea as well as collected specimens for swim bladder research. Mr. DeBruler taught interns proper procedures for tagging and sample removal while aboard vessels. He also acted as liaison between headboat captains and MML staff, by setting up trips in various locations around the state of Florida. In addition, Mr. DeBruler was responsible for data QA.

**Ms. Tanya Merkle and Ms. Carolyn Weaver** (*MML Staff Biologists*) were responsible for the layout of the articles about the project in the newsletter and formulated the data sheets. Both kept an inventory of all supplies, and ordered appropriately. They were in charge of answering the tagging hotline and communicating between anglers reporting recaptured fish and MML. Both participated in headboat tagging trips and data entry as well as contacting tag lottery winners and were responsible for ensuring anglers received their cash prizes. Ms. Merkle and Ms. Weaver were also responsible for student intern supervision.

**Ms. Janet Gannon** (*MML GIS Specialist*) carried out the GIS for fish movement analyses for the project. She transferred data into GIS format and mapped sampling locations and recapture sites to plot fish movements.

**MML Volunteers and Student Interns** helped MML get the biological samples ready for shipment, sort and package sampling and research supplies, photocopy datasheets, and help with mobilization and demobilization for fishing trips. Volunteers and interns also participated in tagging trips aboard headboats, data entry, recapture data retrieval, and data QA.

## FINDINGS

### A. ACTUAL ACCOMPLISHMENTS AND FINDINGS

#### Results

***Objective 1:** To test the hypothesis that red grouper are more susceptible to depth induced mortality than red snapper, based not only on swim bladder size and thickness, but also on the number of rete mirabile and gas gland cells in the swim bladder.*

Swim bladders from 62 red snapper (123 – 674 mm FL) and 138 red grouper (205 – 766 mm FL) were examined for this study. Red snapper have smaller (in relation to total body length) and thicker swim bladders than red grouper (Figures 1 and 2), and swim bladder ruptures were smaller in red snapper compared to red grouper (Figures 3 and 4).

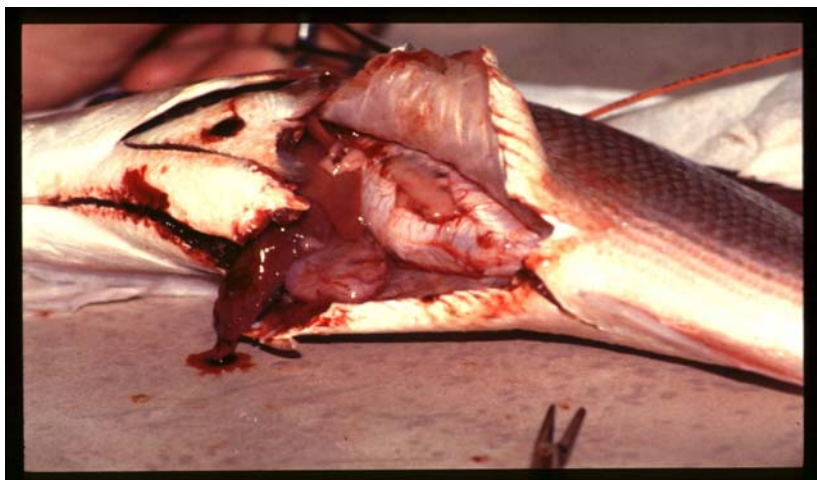


Figure 1. Inflated red snapper swim bladder showing swim bladder size in proportion to total body size.

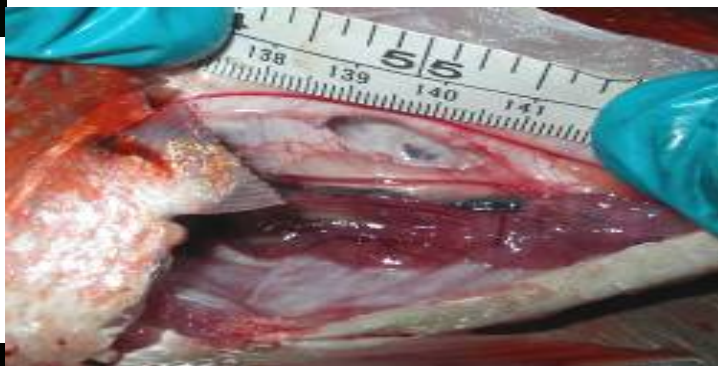


Figure 3. Red snapper swim bladder rupture.



Figure 2. Inflated red grouper swim bladder showing swim bladder size in proportion to total body size.



Figure 4. Red grouper swim bladder rupture.

### *Relationships between Swim Bladder and Fish Length*

Rete area increased significantly with body length (FL) for both red snapper and red grouper (Figure 5) although the relationship between rete area and body length was weak, particularly for red snapper. Red snapper had a significantly higher percentage of rete area in the swim bladder than red grouper when adjusted for FL (ANCOVA,  $F_{3,196} = 41.042$ ,  $p < 0.001$  overall,  $p = 0.022$ , species).

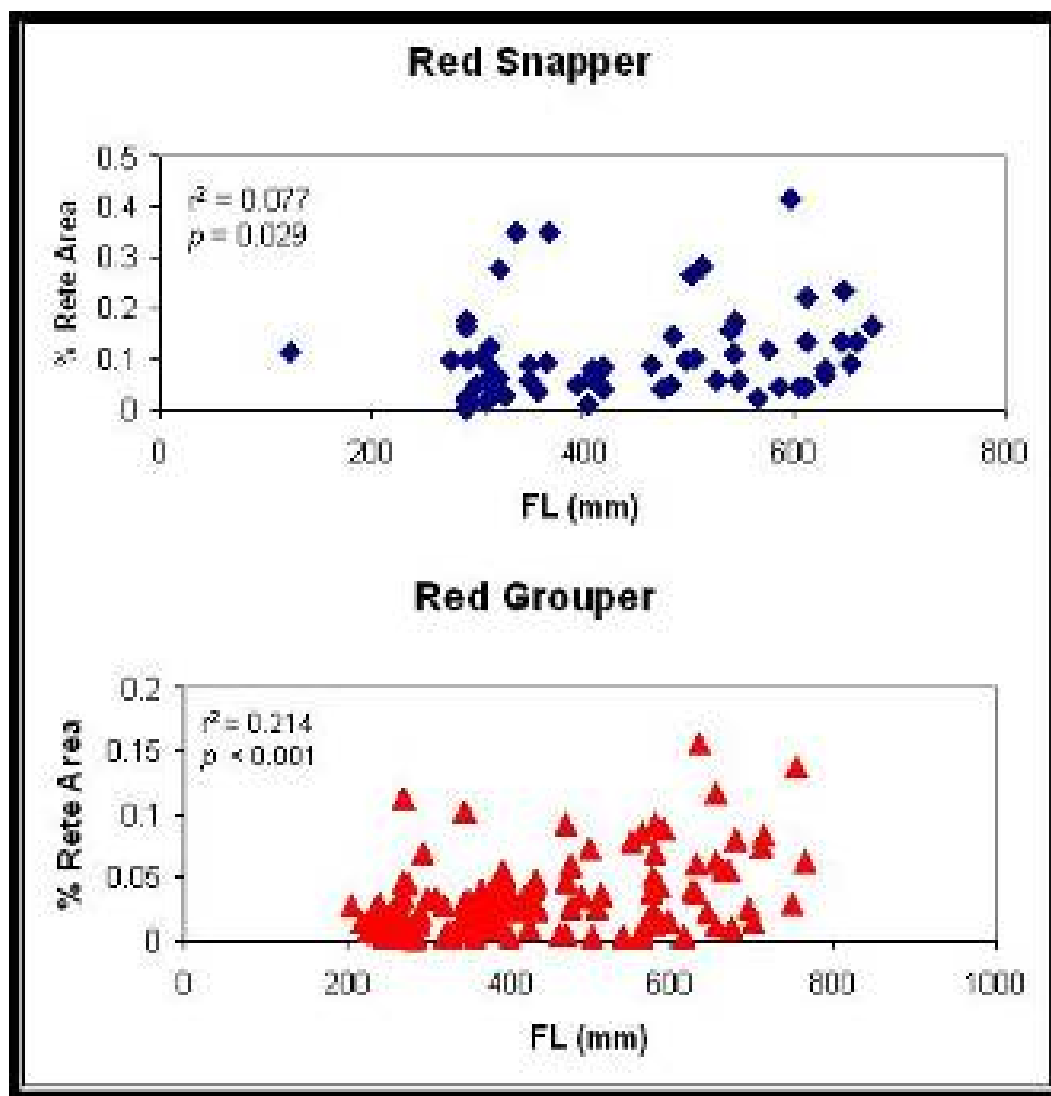


Figure 5. Relationship between the percentage of rete area in swim bladders by fish length (FL) for red snapper and red grouper.

Gas gland area was not related to FL for red snapper; the relationship was significant, but weak, in red grouper (Figure 6). There was no significant difference between species in percentage of gas gland in the swim bladder when adjusted for FL ( $p = 0.155$ ), although red snapper did have a slightly higher mean percentage of gas gland than red grouper.

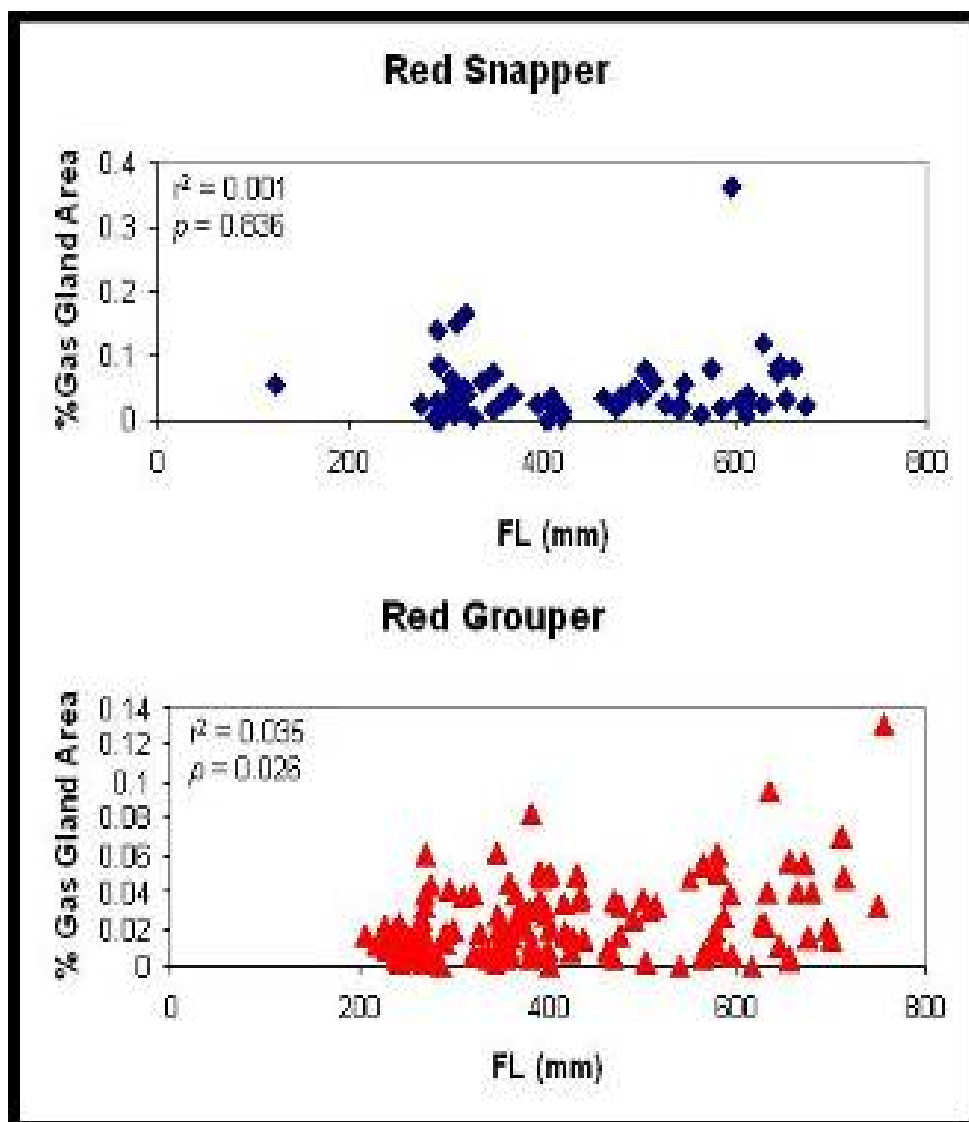


Figure 6. Relationship between the percentage of gas gland area in swim bladders by fish length (FL) for red snapper and red grouper.

### *Relationship between Rete and Gas Gland*

Red grouper had a significantly higher percentage of the rete area occupied by gas gland than did red snapper when adjusted for fish length (ANCOVA,  $F_{3,200} = 13.258$ ,  $p < 0.001$  overall,  $p = 0.007$  for species; Figure 7).

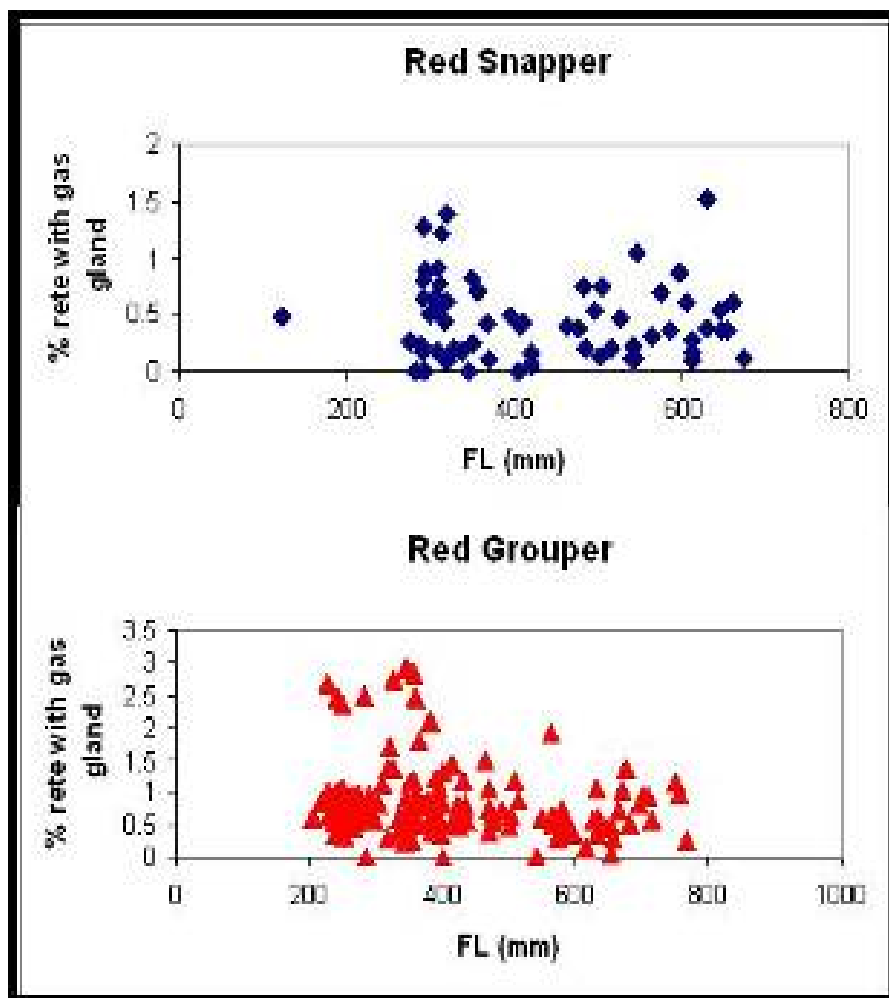


Figure 7. Relationship between the percentage of rete containing gas gland cells and fish length (FL) for red snapper and red grouper.



## Histology

Histological sections showed blood vessels appear to be associated less closely with the rete in red snapper than in red grouper. This is particularly evident in smaller red snapper (Figure 8A). There is an apparent closer association and more connections between gas gland cells and the rete in red grouper than in red snapper (Figure 8B).

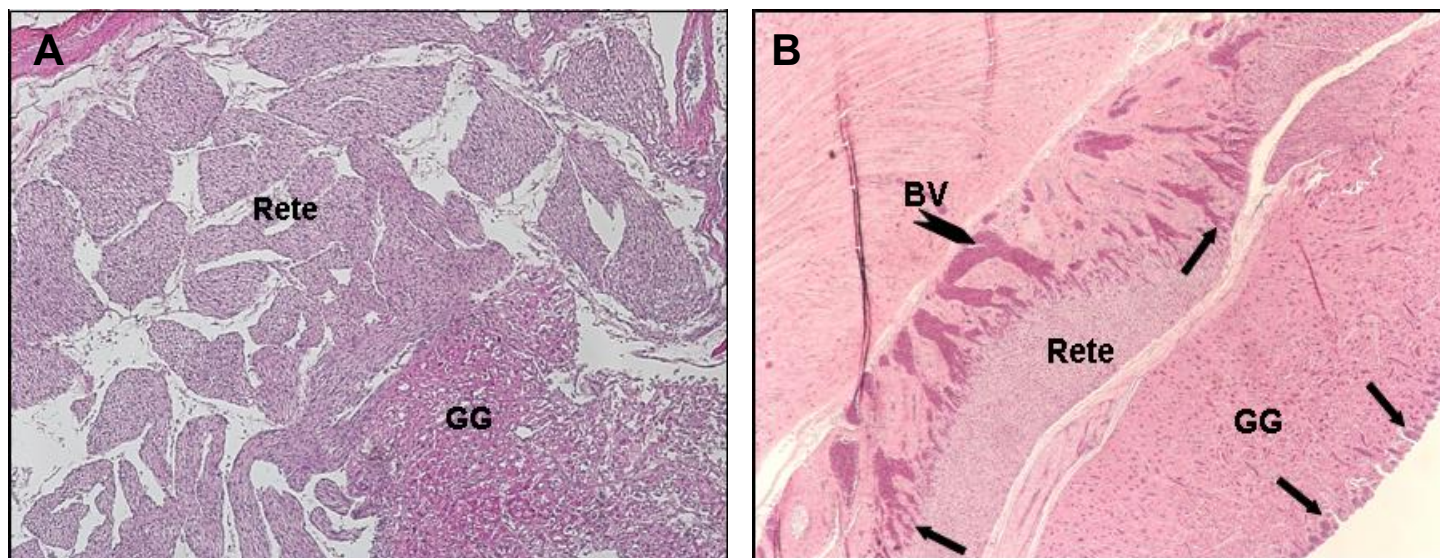


Figure 8. Histological sections of swim bladder tissue showing relationship between rete and gas gland tissues. A. Red snapper, 308 mm FL. Large amount of rete tissue is not associated with gas gland. B. Red grouper, 580 mm FL. Rete is closely associated with gas gland tissue and with blood vessels. BV—blood vessel; GG—gas gland. Arrows indicate areas of hemorrhaging.

### Hemorrhaging in Swim Bladders

The percentage of fish showing hemorrhaging in both the rete and swim bladder increased significantly with increasing 50-mm length classes in both species (ANOVA; red snapper rete,  $F_{1,8} = 20.41$ ,  $p = 0.003$ ,  $r^2 = 0.708$ ; red snapper gas gland,  $F_{1,8} = 17.38$ ,  $p = 0.004$ ,  $r^2 = 0.672$ ; red grouper rete,  $F_{1,11} = 16.21$ ,  $p = 0.002$ ,  $r^2 = 0.580$ ; red grouper gas gland,  $F_{1,11} = 19.77$ ,  $p = 0.001$ ,  $r^2 = 0.631$ ; Figure 9). Hemorrhaging was rare in small red snapper (Figure 8A) compared with large red snapper (Figure 10A). Red grouper in all size classes exhibited hemorrhaging (Figures 8B, 10B). Retal hemorrhaging was significantly higher in red grouper than in red snapper when adjusted for length (ANCOVA,  $F_{3,21} = 5.96$ ,  $p = 0.026$ ); there was no significant difference in gas gland hemorrhaging between species ( $p = 0.786$ ). Histological photos of hemorrhaging in large individuals of both species are shown in Figures 10 A and B. Red grouper showed more hemorrhaging in blood vessels associated with the swim bladder than did red snapper, regardless of the length of the fish.

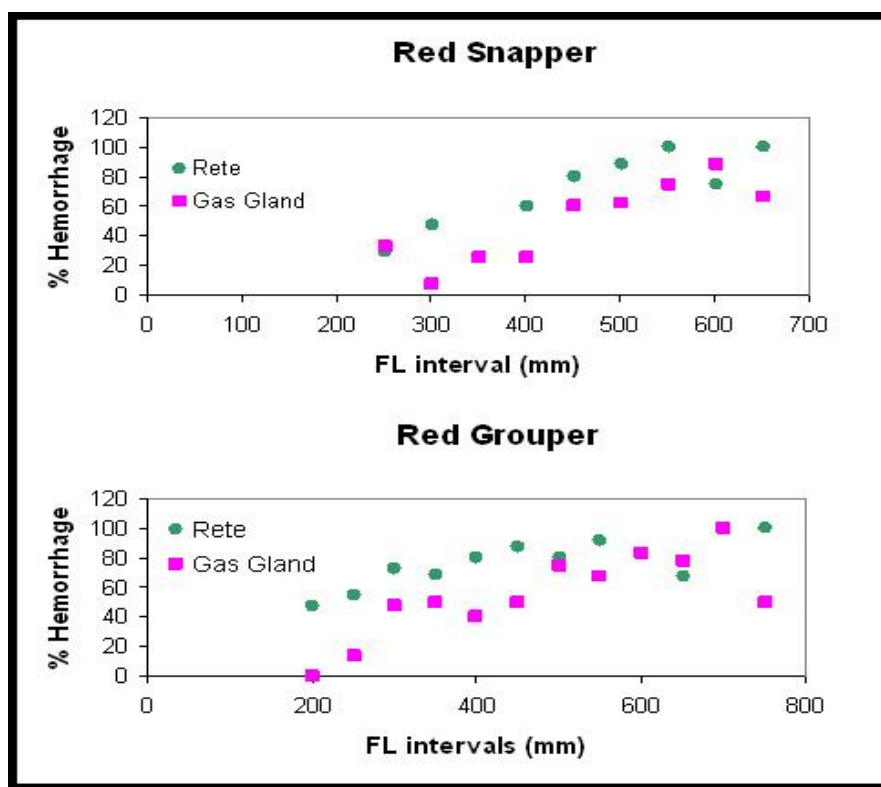


Figure 9. Percentage of red snapper and red grouper in 50 mm FL increments showing retal and gas gland hemorrhaging.

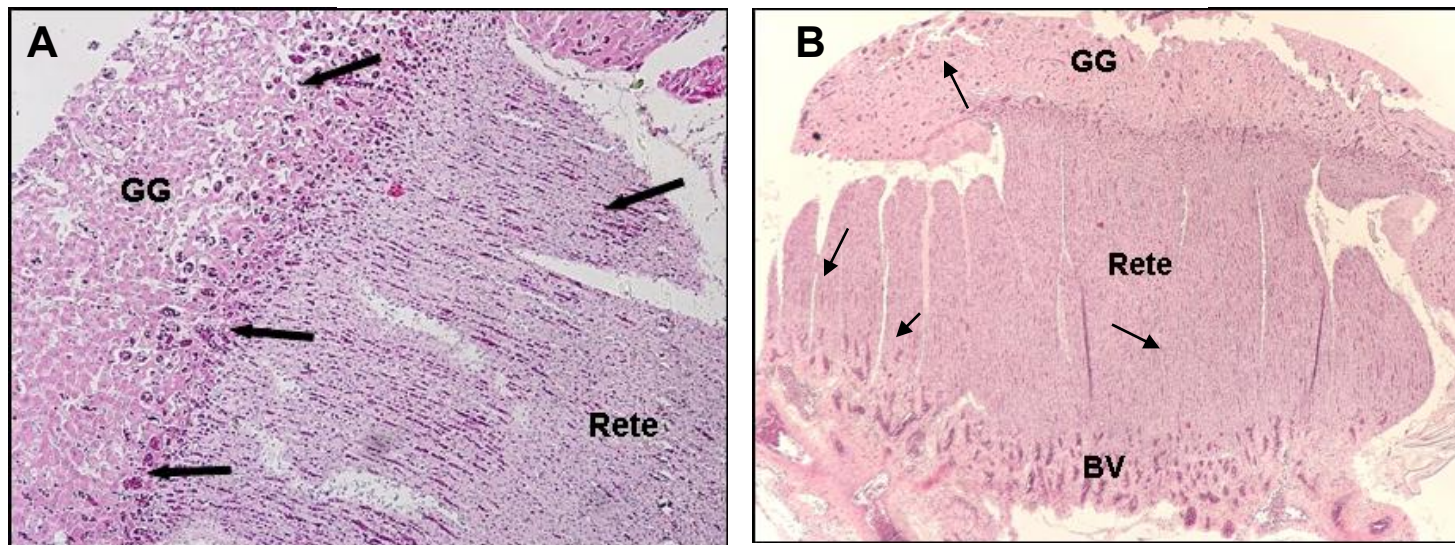


Figure 10. Histological sections of swim bladder tissue showing hemorrhaging. A. Red snapper, 764 mm FL. B. Red grouper, 700 mm FL. BV—blood vessel; GG—gas gland; arrow—hemorrhaging.



Red grouper is deeper bodied and more robust than red snapper, and probably it requires additional assistance with gas exchange as it grows. Thus, the higher percentage of gas gland in the rete of red grouper compared with red snapper at similar lengths is not unexpected given the morphology of the two species. However, additional gas exchange leads to an increased possibility of retal hemorrhaging with rapid decompression in all lengths of red grouper as demonstrated by data presented here. Histological results show overall that red snapper survive rapid decompression better than red grouper, as evidenced by reduced mortality, smaller and less frequent tears in the swim bladder, and less of a tendency to hemorrhage, particularly in smaller fish. The higher percentage of rete area in the swim bladder of red snapper compared with red grouper suggests swim bladder gasses may be exchanged more rapidly in red snapper, allowing greater survival after rapid decompression. Thus, the differences in swim bladder structure documented here are responsible for observed differences in survival from different depths for the two species.

*Field Data – Recaptures of Vented vs Non-vented Red Grouper and Red Snapper by Depth*

Figure 11 compares survival of red grouper and red snapper by depth, determined by tag recaptures. Venting either species obtained from depths  $\leq 27.4$  m is unnecessary. Venting red grouper enhances survival at depths  $> 27.4$  m, consistent with results shown from swim bladder morphology. Red snapper do not require venting until depths  $> 30$  m, depths not included in this project.

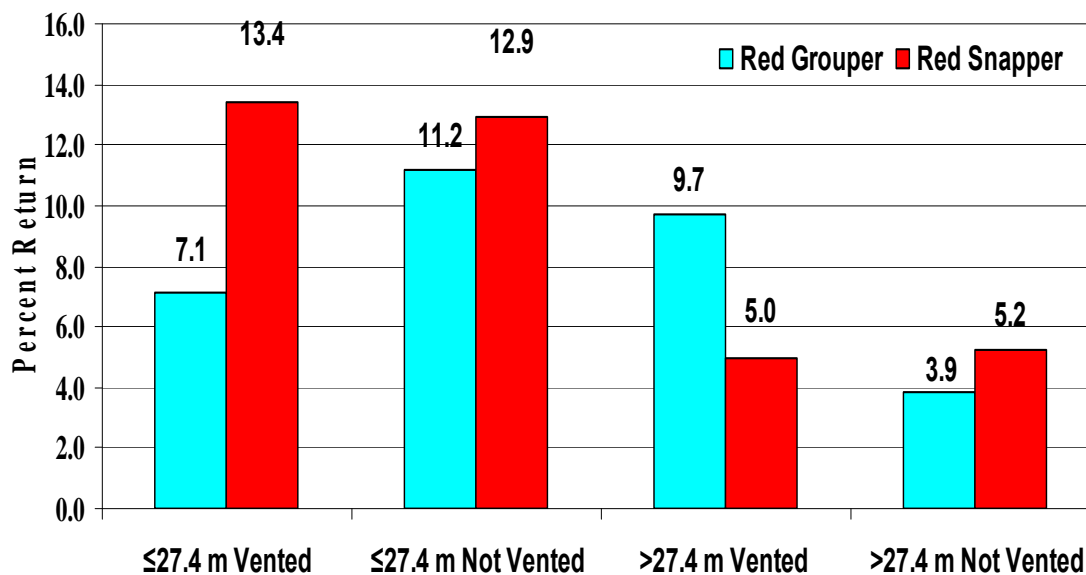


Figure 11. Recapture percentages of vented and non-vented red grouper and red snapper by depth.

These data are consistent with laboratory results from MARFIN Award NA97FF0349 of red grouper and red snapper subjected to depth simulations in fish hyperbaric chambers (Burns et al. 2004). The swim bladders of both red grouper and red snapper ruptured with the change from 1 to 2 atmospheres of pressure (10 - 20 m); however, both species easily survived capture from these depths as well as rapid decompression from 21 m (100% survival). There, however, are marked differences in their ability to tolerate rapid decompression from deeper depths ( $\geq 27$  m). Data from depth simulations of 27.4 m in hyperbaric chambers have shown variable survival due to hemorrhaging in some red grouper, but results for red snapper show 100% survival with no complication. While some red snapper did suffer mortality or sub-lethal effects during rapid decompression from simulated depths  $\leq 42$  m, many survived when held at 1 atmosphere pressure if vented. In contrast, red grouper never survived rapid decompression from simulated depths of  $\leq 42$  m to 1 atmosphere pressure, even when vented (Burns et al. 2004). However, field data have shown that red grouper can survive rapid decompression from depths of 61 m or greater, if the fish were vented and immediately allowed to return to the prior habitat depth (Wilson and Burns 1996; Burns and Robbins 2006), criteria which could not be met in laboratory studies.

***Objective 2:*** To test the hypothesis that smaller red grouper (<30.5 cm) survive rapid decompression better than larger (>38 cm) red grouper because of changes in swim bladder structure with fish length from 30 to 38 cm.

The structure of the gas resorption and secretion areas of the swim bladder differs between the two species (Figure 12). In both species, the anterior, ventral area contains the rete, and is involved in gas absorption, secretion and resorption. However, the general size and structure of the anterior rete area differs between red snapper and red grouper. Red grouper also have a secondary structure on the posterior ventral wall of the swim bladder that red snapper lack. This area can be observed under a dissecting microscope to initiate vascularization and change at 305 mm FL and appeared to be completed in red grouper  $\geq 380$  mm FL. Histological sections of the posterior secondary structure show that small red grouper (251 mm FL; Figure 13A) have some vascularized tissue, as represented by blood vessels and capillaries but no organized gas resorption/secretion area. In contrast to the lack of this convoluted tissue in small red grouper, histological sections from larger red grouper (550 mm FL, Figure 13B) have obvious areas of convoluted tissue that is probably involved with gas resorption/secretion as well as large blood vessel and duct areas.

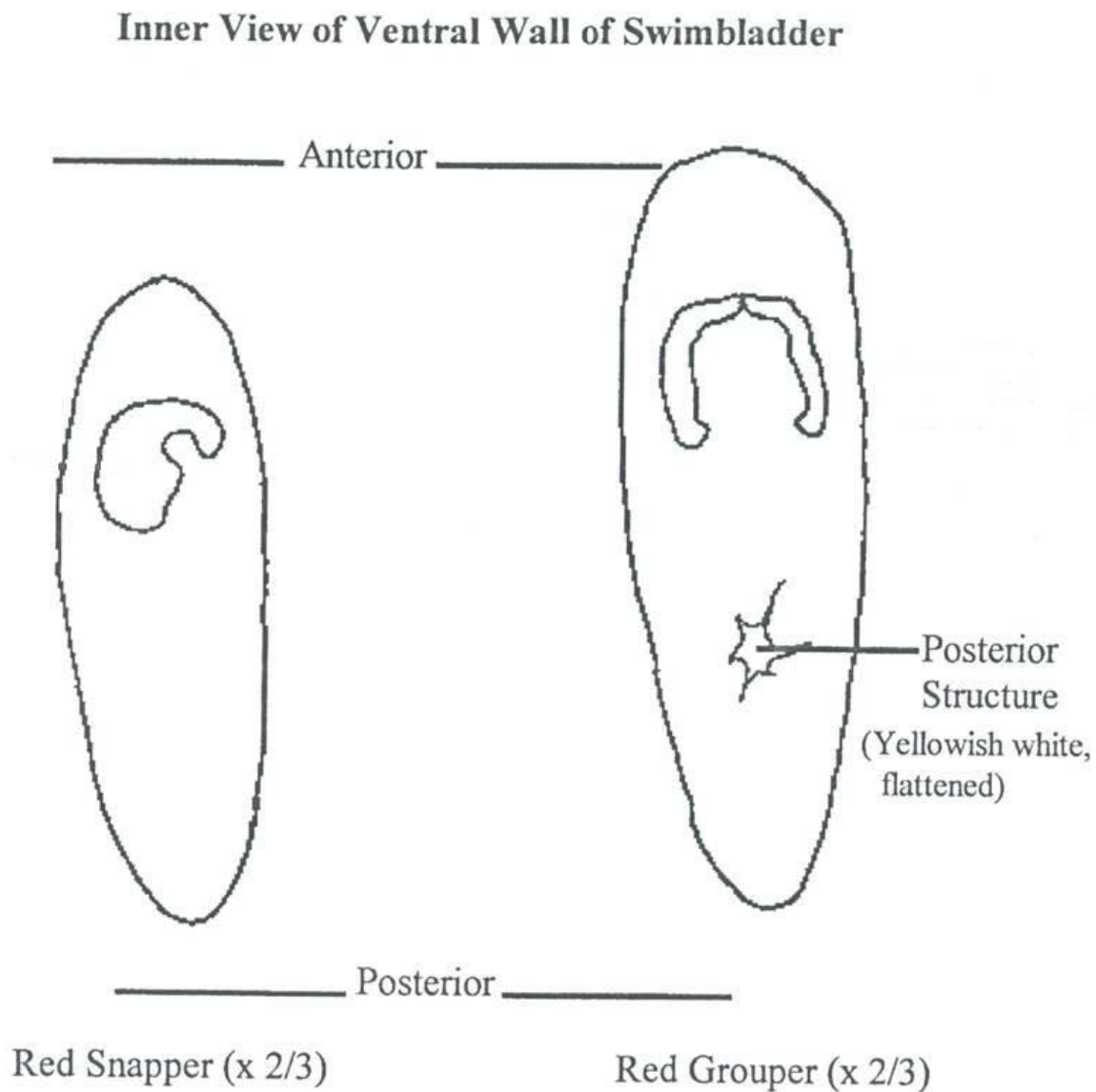


Figure 12. Comparison of ventral wall of swim bladder in red snapper and red grouper showing the secondary structure.

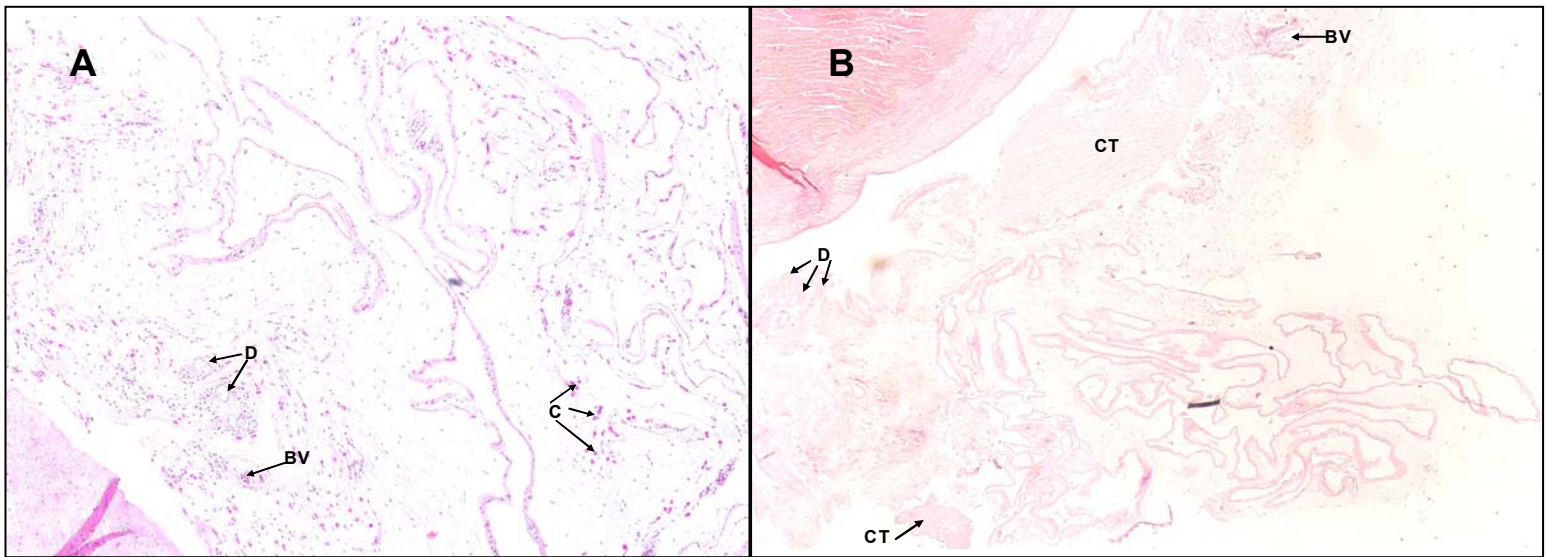


Figure 13. Histological sections from posterior portion of red grouper swim bladder. A. Small (251 mm FL) red grouper. 100X magnification. B. Large (550 mm FL) red grouper showing areas of convoluted tissue. 40X magnification. BV—blood vessel; C—capillaries; CT—convoluted tissue; D—duct.

In spite of demonstrated differences between the two species in the ability to tolerate rapid decompression with respect to barotrauma, both species exhibited the same trend in survival from depth with respect to fish length. Analysis of recapture data by fish length showed that if recaptures are used as a measure of survival, shorter fish (< 381 mm FL) of both species survive rapid decompression better than larger ( $\geq 381$  mm FL) fish. The proportion of recaptured small (< 381 mm FL) to larger red grouper ( $\geq 381$  mm FL) was compared using a log-likelihood G test. The results were highly significant ( $p = 9.7 \times 10^{-19}$ ). Similar results were found when comparing recaptured small red snapper (< 381 mm FL) to larger red snapper ( $\geq 381$  mm FL) with  $p = 9.5 \times 10^{-6}$ . There was no significant difference ( $p=0.2788$ ) between recaptured fish when a log-likelihood G test was run comparing recaptures for red grouper and red snapper > 381 mm FL. This was not the case when the same test was run comparing recaptures of red grouper and red snapper  $\leq 381$  mm FL ( $p=0.00023$ ). Table 2 lists the results of the G tests by fish length.

Table 2. Results of G tests showing affect of fish length on survival of red snapper and red grouper. Data based on tag recaptures.

Test Group	Parameter tested	G test result (p value)
Red Grouper	Length ( $\leq 381$ mm vs > 381 mm)	$9.7 \times 10^{-19}$
Red Snapper	Length ( $\leq 381$ mm vs > 381 mm)	$1.08 \times 10^{-9}$
Length ( $\leq 381$ mm)	Red Grouper vs Red Snapper	$2.3 \times 10^{-4}$
Length (> 381 mm)	Red Grouper vs Red Snapper	0.2788

## Discussion/Summary/Recommendations Regarding Swim Bladder Findings

Red grouper are deeper bodied and bulkier than red snapper, and thus probably require additional assistance with gas exchange as individuals grow. Thus, the higher percentage of gas gland in the rete of red grouper compared to red snapper at similar lengths is not unexpected given the morphology of the two species and the larger swim bladder observed in red grouper. However, additional gas exchange leads to an increased possibility of retal hemorrhaging with rapid decompression in all sizes of red grouper as demonstrated by data presented here. Furthermore, the presence of an additional gas secretion/absorption area in the posterior of large (>380 mm FL) red grouper can potentially increase the amount of gas in red grouper and also contribute to the increased hemorrhaging observed in this species.

Overall, red snapper survive rapid decompression better than red grouper, as evidenced by reduced mortality, smaller tears in the swim bladder, and less of a tendency to hemorrhage, particularly in smaller fish. These conclusions are supported by tagging data showing significantly greater survival of large (>381 mm FL) red snapper compared to red grouper. Because of its anatomy and relatively larger swim bladder, the red grouper has a greater quantity of swim bladder gases trapped within its body cavity compared with the quantity in the red snapper. The higher percentage of rete area in the swim bladder of red snapper compared to red grouper suggests swim bladder gasses may be exchanged more rapidly in red snapper, allowing greater survival after rapid decompression. The differences in swim bladder morphology documented can help explain why venting red snapper caught at depths  $\leq 27.4$  m is not necessary, as shown in both this and previous studies (MARFIN project NA17FF2881; Render and Wilson 1996; Gitschlag and Renaud 1994). Similarly, the morphological explanation and histological verification for increased gas in the swim bladder of red grouper strongly supports the importance of venting red grouper captured at depths > 20 m. The morphological/histological findings reported here may also be applicable to black sea bass, *Centropristis striata*, another serranid that significantly benefits from venting prior to release (Collins et al. 1999). Thus, the distinguishing features in swim bladder structure documented here explain differences in depth-induced mortality, and these data can assist managers in providing recommendations for maximum survival when releasing undersized fish of these two species.

**Objective 3:** *To test the hypothesis that circle hooks will greatly reduce release mortality in red snapper.*

Necropsies of acute mortalities of red snapper, red grouper, and vermilion snapper caught from headboats using a J hook showed that the red snapper exhibited much greater acute and latent hook trauma than either of the two other species. Hook trauma was the leading cause of death for red snapper. Circle hooks have been found to be effective in reducing hook mortality in a number of fish species, but not all (Cooke et al. 2003a; Cooke et al. 2003b; Cooke et al. 2003c; Cooke and Suski 2004). To test the hypothesis that circle hooks greatly reduced hook mortality, we conducted a log likelihood G-test comparing the number of red snapper tagged and the number recaptured by hook type.

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Tag recaptures for red snapper caught on J (n=2,145) and circle (n=3,172) hooks resulted in a 12.5 % recapture rate for fish originally hooked on J hooks versus a 8.1% recapture rate for red snapper originally caught on circle hooks (Table 3; Figure 14). Results of the log likelihood G – test were highly significant ( $p= 2.34 \times 10^{-6}$ ), showing a higher recapture rate for J hooked fish (Table 4). Results comparing recaptures from all fishing sectors combined showed no benefits in using circle hooks for red snapper, in spite of 1,027 more red snapper being caught on circle hooks than on J hooks. Since one headboat tagged and recaptured a large number of red snapper, a G-test of circle versus J hook restricted to recreationally caught red snapper was conducted. Results were the same as for all fishing sectors combined and again showed no benefit from using circle hooks.

Table 3. Number of target species tagged and recaptured by hook type

Species	J hook tagged	J hook recaps	% J hook recaps	Circle hook tagged	Circle hook recaps	% Circle hook recaps
Red Snapper	2145	269	12.5	3172	258	8.1
Red Grouper	3935	287	7.3	863	121	14.0
Gag	1833	240	13.1	780	77	9.9
Mangrove Snapper	3013	272	9.0	122	3	2.5
Vermilion Snapper	436	1	0.2	29	0	0.0

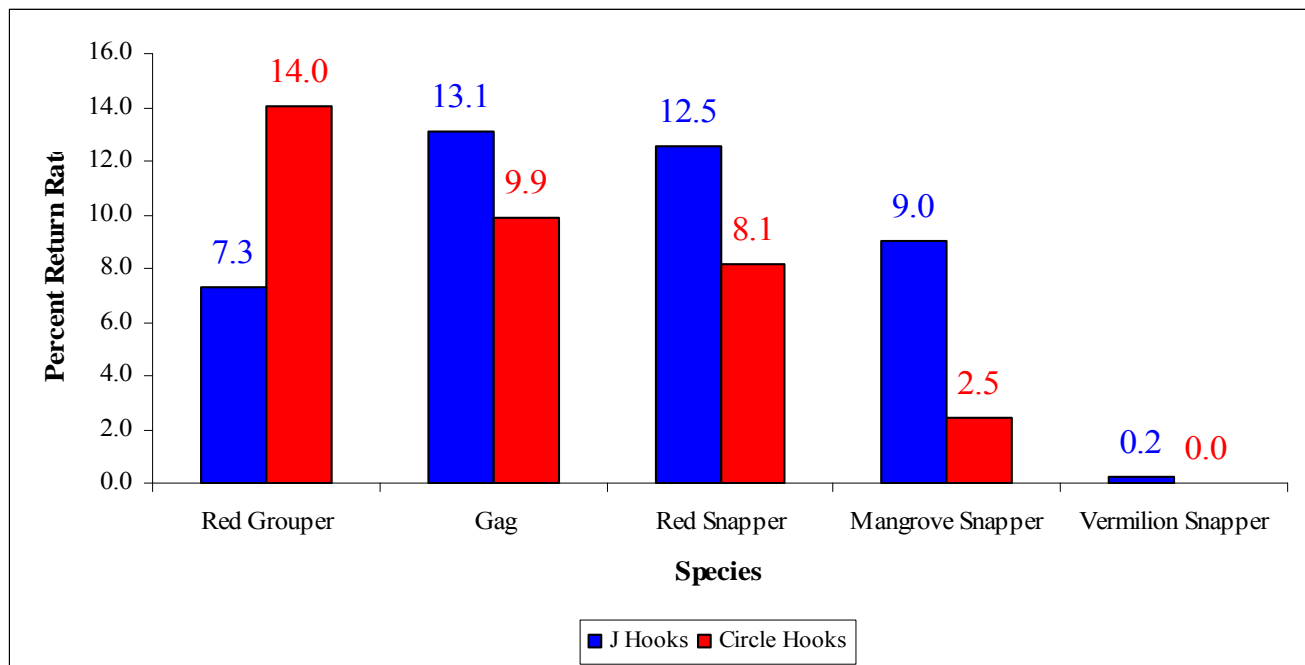


Figure 14. Number of target species recaptured by hook type

Table 4. Differences in recaptures, and assumed mortality, between target species captured on J hooks and circle hooks. n = number of recaptured fish

Species	J hook recaptures		Circle hook recaptures		G test <i>p</i> values
	n	%	n	%	
Red snapper	269	12.5	258	8.1	$2.3 \times 10^{-6}$
Red grouper	287	7.3	121	14.0	$4.49 \times 10^{-8}$
Gag	240	13.1	77	9.9	0.036939
Mangrove snapper	272	9.0	3	2.5	0.006531

Results of recaptures for different hook types for red snapper were compared with those for other target species (Table 3; Figure 14), and G-tests were determined (Table 4). Results for red grouper were also highly significant ( $p = 5.78 \times 10^{-8}$ ), but they contrasted with those for red snapper. With twice as many recaptures of red grouper originally caught on circle hooks (14%) than on J hooks (7.3%), red grouper clearly benefited from the use of circle hooks ( $p = 5.78 \times 10^{-8}$ ). However, neither gag nor mangrove snapper appeared to benefit from the use of circle hooks (Table 4). It is not surprising that results for these species were more similar to those for red snapper than those for red grouper based on the morphological features and ecological habitats.

Based on the results, the hypothesis was rejected. Using circle rather than J hooks does not appear to increase release-survival of red snapper. However, red grouper clearly benefit from the use of circle hooks.

**Objective 4:** To quantify undersize and legal catch for red grouper, *E. morio*, gag, *M. microlepis*, red snapper, *L. campechanus*, mangrove snapper, *L. griseus* and vermillion snapper, *R. aurorubens* in the recreational and recreational-for-hire reef fish fishery off Florida

#### *Quantification of Undersized and Legal Catch:*

Of the 23,660 target species captured and measured during this project, 7,934 fish (1,333 red grouper, 2,744 red snapper, 1,144 gag, 905 mangrove snapper and 1,808 vermillion snapper.) were measured but not tagged. The number of target species measured but not tagged are broken down by species, area, and the percentage of undersized fish captured (Table 5) as well as by species, fishing sector, and percentage of undersized fish captured (Table 6). Table 7 shows the total number of target species by area, while Table 8 lists the total number of legal target species caught aboard recreational and recreational-for-hire vessels during the project as well as the percentage of fish that were undersize. Not unexpectedly, the majority of red grouper, gag, and red snapper captured by fishers in all fishing sectors were undersized. However, only a small percentage of mangrove snapper captured by recreational-for-hire fishers (i.e., charter and headboats) were undersize while most mangrove snapper caught by recreational fishers were

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undersized since most were caught inshore in shallow water while fishing from piers. In contrast, the majority of vermillion snapper caught by charter boat fishers were undersized, while a much higher percentage of vermillion snapper captured by head boat and recreational fishers were legal size fish.

Table 5. Total number of target species that were measured but not tagged in each fishing area of Florida. n = total number/area; %U = percentage of undersized fish/area based on size limits during this project.

Species	Atlantic		Panhandle		North Gulf		South Gulf		Florida Keys	
	n	%U	n	%U	n	%U	n	%U	n	%U
Red Grouper (n = 1,332)	83	11.2	153	59.5	36	50.0	678	56.1	382	47.2
Gag (n = 1,107)	239	21.4	273	61.7	92	14.1	494	41.3	9	55.5
Red Snapper (n = 2,726)	915	35.6	1704	50.2	3	66.6	81	12.1	23	34.5
Mangrove Snapper (n = 933)	503	0.59	62	0.0	164	65.2	199	54.8	5	60.0
Vermillion Snapper (n = 1,755)	480	18.1	927	28.2	8	37.5	330	28.8	10	90.0
<b>TOTAL</b>	<b>2220</b>	<b>21.8</b>	<b>3119</b>	<b>41.4</b>	<b>1030</b>	<b>13.9</b>	<b>1782</b>	<b>44.9</b>	<b>429</b>	<b>48.0</b>

Table 6. Total number of target species that were measured but not tagged by fishing sector in Florida waters. All fishing areas are combined. n = total number/sector; %U = percentage of undersized fish/sector.

Species	Charter Boats		Head Boats		Recreational Boats	
	n	%U	n	%U	n	%U
Red Grouper (n = 1,332)	444	50.9	641	48.8	227	64.3
Gag (n = 1,107)	159	29.6	500	26.4	508	41.9
Red Snapper (n = 2,726)	234	46.6	1490	42.6	1002	45.7
Mangrove Snapper (n = 933)	21	0.0	574	0.7	311	77.8
Vermillion Snapper (n = 1,755)	47	65.9	1541	21.6	167	36.5
<b>TOTAL</b>	<b>905</b>	<b>45.6</b>	<b>4,746</b>	<b>30.5</b>	<b>2,215</b>	<b>51.0</b>



Table 7. All target species sampled by Florida fishing area. Data include fish that were measured only as well as fish tagged and released.

Species	Atlantic	Panhandle	North Gulf	South Gulf	Florida Keys
Red Grouper (n = 6,329)	343	535	250	4382	819
Gag (n = 3,824)	1022	837	478	1406	81
Red Snapper (n = 8,207)	3514	4557	11	96	29
Mangrove Snapper (n = 5,056)	534	226	2018	2199	79
Vermillion Snapper (n = 2,219)	820	947	8	434	10
<b>TOTAL</b>	<b>6233</b>	<b>7102</b>	<b>2765</b>	<b>8517</b>	<b>1018</b>

Table 8. All legal and undersized target species by fishing sector. Data include fish that were measured only as well as fish that were tagged and released. n =total number of fish; %U =percentage of undersized fish/sector.

Species	Charter Boats		Headboats		Recreational Boats	
	n	%U	n	%U	n	%U
Red Grouper (n=6,249)	962	75.6	3801	91.1	1486	90.4
Gag (n=3,870)	457	74.8	1567	76.2	1846	81.9
Red Snapper (n=8,220)	396	66.4	4258	79.5	3566	79.8
Mangrove Snapper (n=4,086)	23	0	1634	2.2	3429	92.1
Vermillion Snapper (n = 2,217)	84	80.9	1966	38.0	167	36.5
<b>TOTAL</b>	<b>1922</b>	<b>72.8</b>	<b>13226</b>	<b>66.6</b>	<b>10494</b>	<b>85.3</b>

#### *Length/Frequency of Target Species from All Areas Sampled*

Figures 15 –19 show the size ranges of all fish captured (measured only and tagged and released) during the course of the project, with the legal size limit indicated by a colored line at the size limit for each species. The majority of fish captured were under the legal limit for all species

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except vermillion snapper (Figure 19). However, most red snapper captured in the Gulf were legal sized (Figure 17), which was not the case for red grouper (Figure 15). This difference in legal size between the Atlantic and the Gulf for red snapper and gag (Figure 16) resulted in more undersize fish of both species captured in the Atlantic. The broadest size range of fish captured was seen with gag, while vermillion snapper had the narrowest size range.

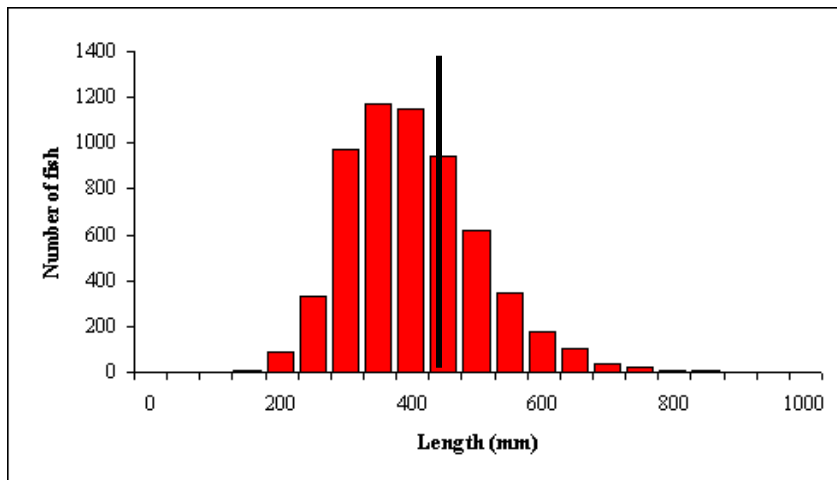


Figure 15. Length frequency histogram of all red grouper captured. Legal size limit indicated by black line at 508 mm TL.

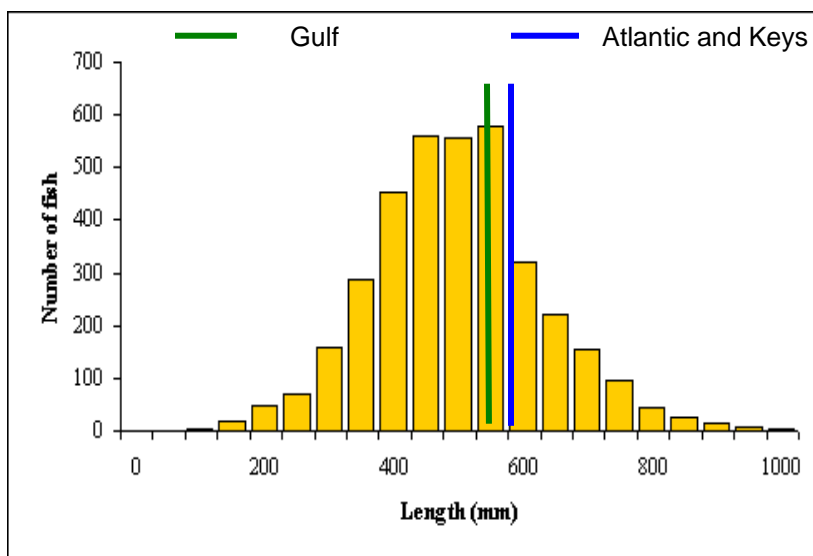


Figure 16. Length frequency histogram of all gag captured. Legal size limit in the Gulf (559 mm TL) and Atlantic and Florida Keys (610 mm TL) indicated by solid lines.

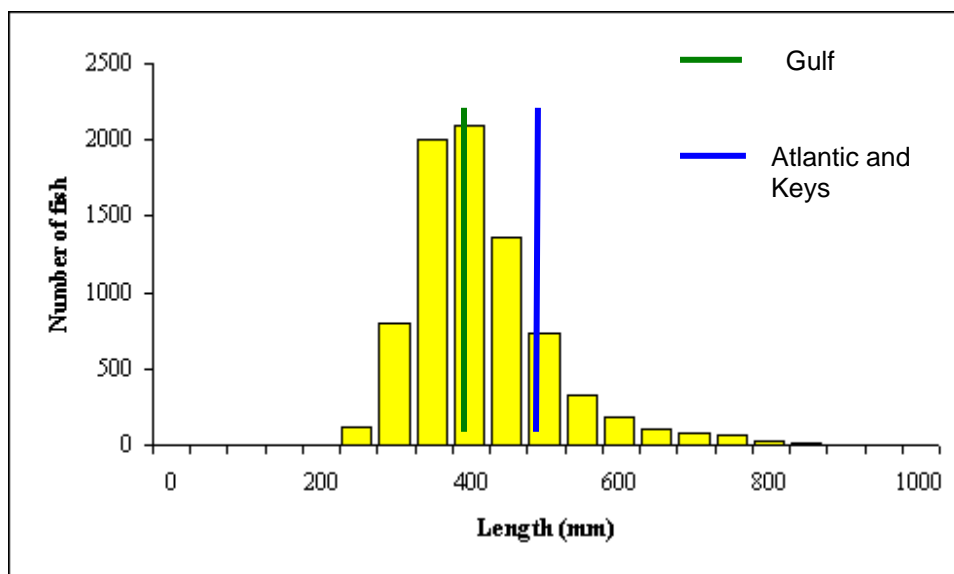


Figure 17. Length frequency histogram of all red snapper captured. Legal size limit in the Gulf (406 mm TL) and Atlantic and Florida Keys (508 mm TL) indicated by solid lines.

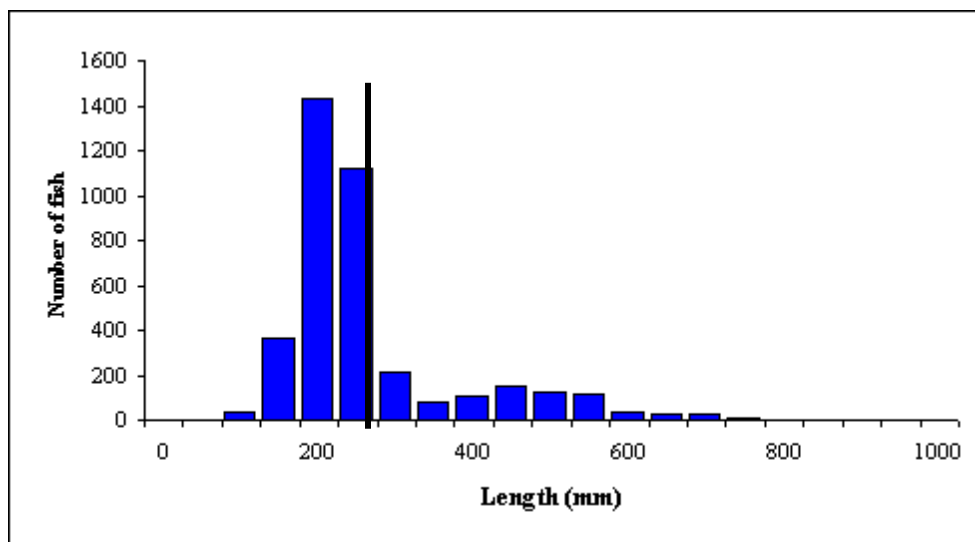


Figure 18. Length frequency histogram of all mangrove snapper captured. Legal size limit indicated by black line at 254 mm TL.

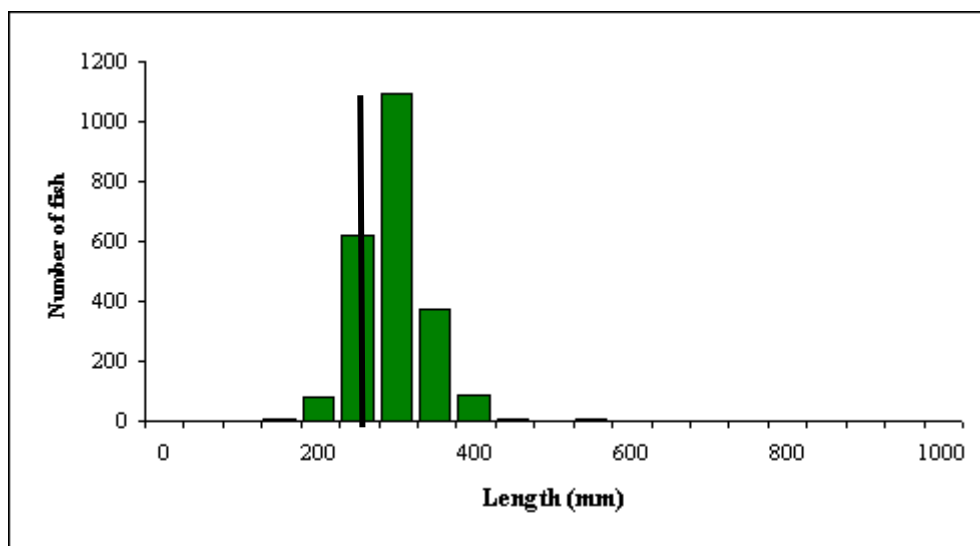


Figure 19. Length frequency histogram of all vermilion snapper captured. Legal size limit indicated by black line at 279 mm TL.

**Objective 5:** *To obtain catch and release mortality rates relative to depth and gear for red grouper, gag, red snapper, mangrove snapper, and vermilion snapper.*

#### *Fish Tagging:*

A total of 15,263 target species were tagged and released. Most were tagged and released in the eastern Gulf of Mexico including the Florida Keys ( $n = 12,814$ ) with the remainder ( $n = 4,009$ ) released in the South Atlantic. Of these fish, 29.7% were red grouper, 16.1% were gag, 32.6% were red snapper, 18.9% were mangrove snapper and 2.8% were vermilion snapper. These data include fish previously tagged if they were recaptured during this project. Table 9 shows the number of target species tagged and recaptured in each fishing area of Florida. The greatest tagging and recapture effort was in the south Gulf area, while the fewest tags and recaptures occurred in the Florida Keys. Red grouper were predominately captured in the south Gulf area, gag in the Atlantic and south Gulf areas, red snapper in the Atlantic and panhandle areas, mangrove snapper in the south Gulf area and vermilion snapper in the Atlantic area. Mangrove snapper were captured in a relatively high percentage (59.9%) in the north Gulf area compared to all other species, while red grouper were the dominant target species caught in the Florida Keys (74.2%). Red snapper dominated the total catch in both the Atlantic (64.7%) and the Florida panhandle (71.6%).

Table 9. Total number of target species tagged and recaptured in each fishing area of Florida

<b>Species</b>	<b>Atlantic</b>	<b>Panhandle</b>	<b>North Gulf</b>	<b>South Gulf</b>	<b>Florida Keys</b>
Red Grouper (n = 4,997)	260	382	214	3704	437
Gag (n = 2,717)	783	564	386	912	72
Red Snapper (n = 5,483)	2599	2853	8	17	6
Mangrove Snapper (n = 3,180)	31	164	910	2001	74
Vermillion Snapper (n = 465)	341	20	0	104	0
<b>TOTAL</b>	<b>4014</b>	<b>3983</b>	<b>1518</b>	<b>6738</b>	<b>589</b>

Targeted species tagged during this project by fishing sector included 1,016 (6.0%) off charter boats, 7,483 (44.5%) off headboats, and 8,311 (49.4%) off recreational vessels. Although most target species were tagged and recaptured by a combination of recreational and recreational-for-hire fishers, vermilion snappers were exclusively tagged and recaptured by fishers aboard head and charter boats, while recreational fishers dominated mangrove snapper tagging and recaptures. Table 10 shows the number of target species tagged by fishing sector for all areas combined. Fishers in charter boats predominately tagged red grouper (51%), while head boat fishers most commonly tagged red grouper (42.2%) and red snapper (37%). Mangrove snapper was the species most frequently tagged by recreational fishers (37.5%), followed by red snapper (30.6%). Most of the mangrove snappers tagged were caught from shore off seawalls and fishing piers.

Table 10. Total number of target species tagged by fishing sector in Florida waters. All fishing areas are combined.

<b>Species</b>	<b>Charter Boats</b>	<b>Head Boats</b>	<b>Recreational Boats</b>
Red Grouper (n = 4,987)	518	3160	1309
Gag (n = 2,703)	298	1067	1338
Red Snapper (n = 5,476)	162	2768	2546
Mangrove Snapper (n = 3,179)	1	60	3118
Vermillion Snapper (n = 465)	37	428	0
<b>TOTAL</b>	<b>1016</b>	<b>7483</b>	<b>8311</b>

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## Recaptures

A total of 1,561 recapture events occurred during this project. Red snapper were recaptured most frequently, followed by red grouper. Recaptures were distributed fairly evenly between the Atlantic and the southern Gulf. The total number of recaptures by species and area is shown in Table 11. Too few individuals of the vermillion snapper were recaptured to include in this analysis.

Table 11. Total number of target species recaptures in each Florida fishing area

Species	Atlantic	Panhandle	North Gulf	South Gulf	Florida Keys
Red Grouper (n = 434)	52	85	21	241	34
Gag (n = 323)	136	64	50	69	4
Red Snapper (n = 529)	299	229	1	0	0
Mangrove Snapper (n = 277)	0	6	106	161	3
<b>TOTAL</b>	<b>487</b>	<b>384</b>	<b>178</b>	<b>471</b>	<b>41</b>

Table 12 shows the percentage of each target species tagged/released and recaptured by fishing sector. Red grouper, gag and red snapper were predominately recaptured by head boat fishers, while recreational fishers almost exclusively recaptured mangrove snapper. Charter boat fishers accounted for few recaptures for all species.

Table 12. Percentage of each tagged target species recaptured by fishing sector.

Species	Charter	Head Boat	Recreational
Red Grouper (n = 434)	10.2%	53.5%	36.5%
Gag (n = 323)	8.6%	47.4%	43%
Red Snapper (n = 529)	1.1%	56.9%	41%
Mangrove Snapper (n = 277)	0%	0.4%	99.6%

Some target species were recaptured multiple times. Although there were 1,401 fish tagged, there were 1,561 release events due to multiple recapture and release of previously tagged fish. The number of multiple releases varied by species with red grouper having the most individual fish recaptured multiple times (Figure 20). The greatest number of multiple recaptures and releases occurred in the combined areas of the Gulf of Mexico (95 fish) followed by the Atlantic region

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(60 fish) (Table 13). Multiple recaptures of red grouper were most common in the combined Gulf, while multiple recaptures of mangrove snapper occurred exclusively in the northern and southern Gulf (Table 13).

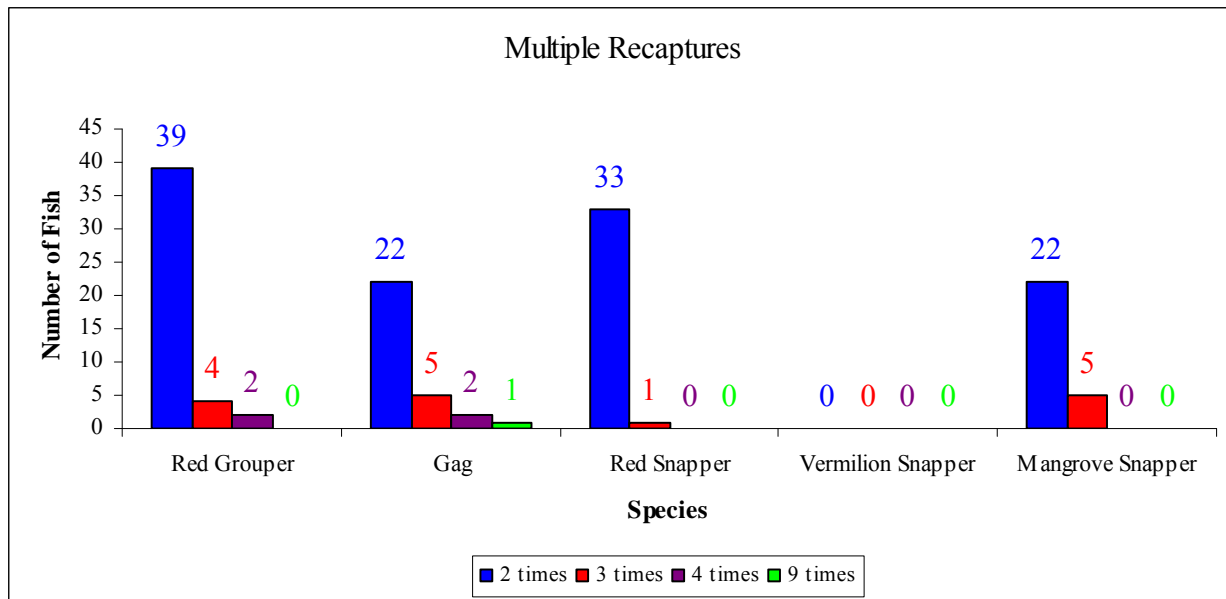


Figure 20. Multiple recaptures of target species in Florida waters during this project. All areas and fishing sectors combined.

Table 13. Total number of multiple recaptures of target species in each Florida fishing area. Individual fish were recaptured between one and nine times; each recapture is counted as a recapture event.

Species	Atlantic	Panhandle	North Gulf	South Gulf	Florida Keys
Red Grouper (n = 50)	11	16	3	15	5
Gag (n = 44)	23	10	7	3	1
Red Snapper (n = 35)	26	9	0	0	0
Mangrove Snapper (n = 32)	0	0	20	12	0
<b>TOTAL</b>	<b>60</b>	<b>35</b>	<b>30</b>	<b>30</b>	<b>6</b>

## Depth

Fish were recaptured at a variety of depths, ranging from 1 m to > 61 m (Figure 21). In general, fish tended to be recaptured at the depths at which they were originally caught. Most fish (41.1%) were recaptured between 21-25 m and 19.6% at depths <6 m. Red grouper, red snapper and gag were all predominately recaptured at 21-25 m, while mangrove snapper were almost exclusively recaptured at depths < 6 m (Figures 22 a-d). Both red grouper and gag were recaptured at depths > 61 m, although gag recaptures occurred more commonly at greater depths (> 36 m) than did red grouper recaptures. Few red snapper were recaptured at depths > 45 m.

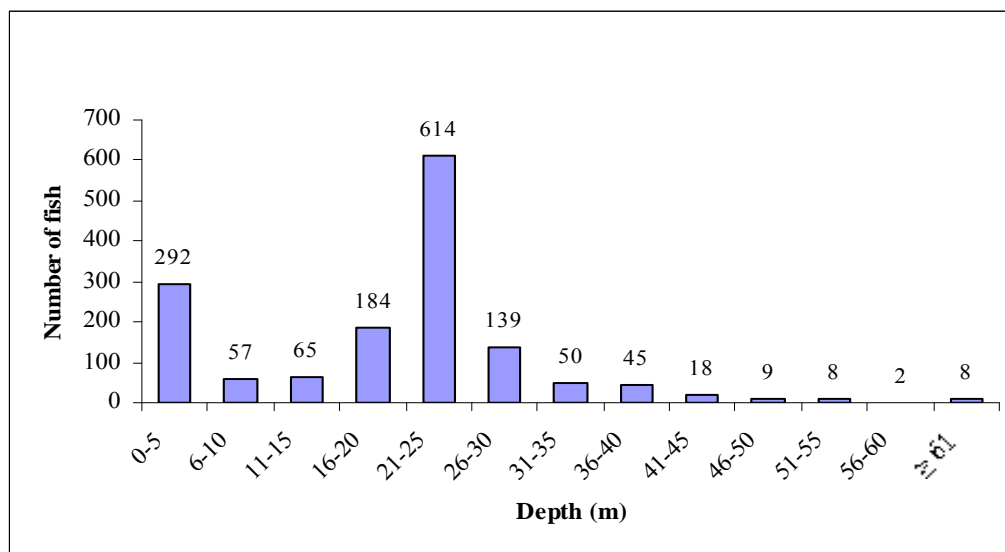


Figure 21. Depth distribution of recaptures for all species combined.

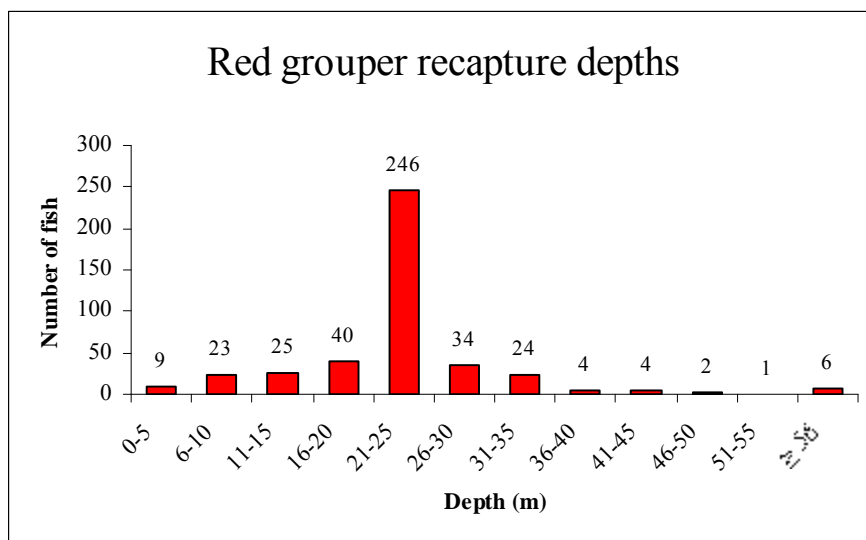


Figure 22a. Depth distribution of recaptured red grouper.

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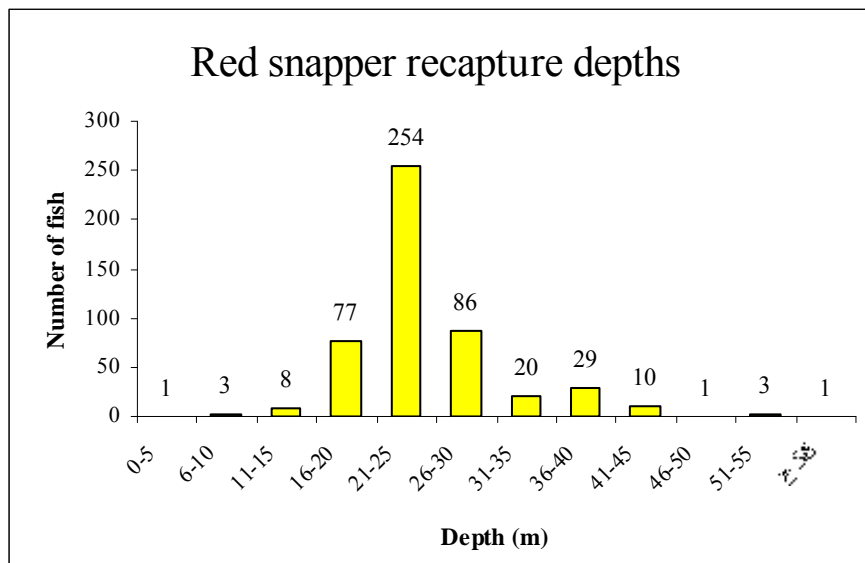


Figure 22b. Depth distribution of recaptured red snapper.

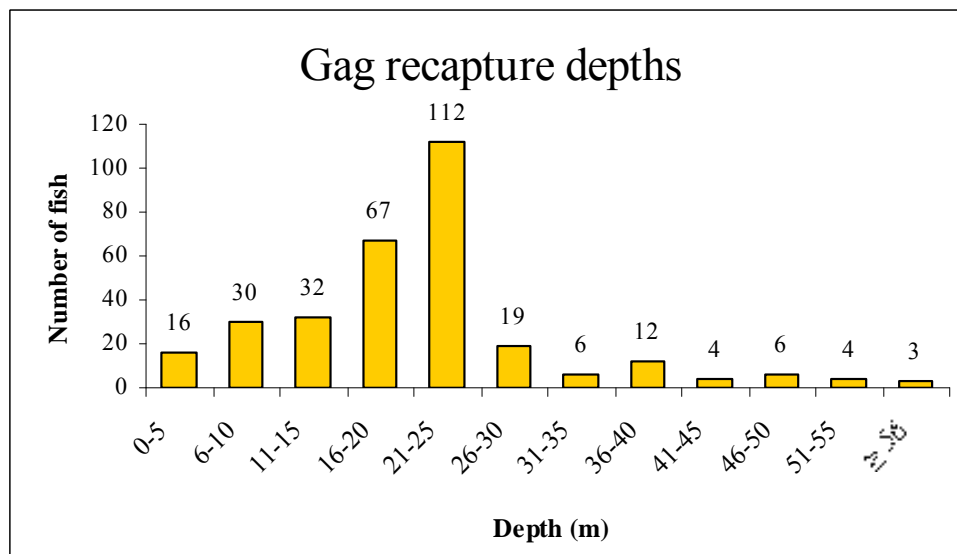


Figure 22c. Depth distribution of recaptured gag.

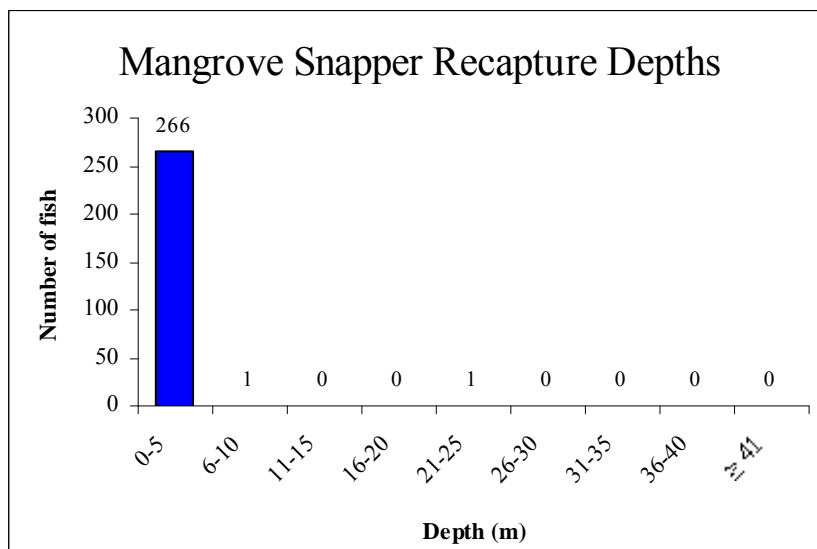


Figure 22d. Depth distribution of recaptured for mangrove snapper.

From both laboratory depth simulations and field studies, it appears that rapid decompression from depths  $\geq 21$ -25 m are readily survivable by those target species where recaptures were reported. As expected, mortality increased with increasing depth.

**Objective 6:** *To determine tag shedding rates and effects on growth and survival for fish tagged with single barbed dart tags for red grouper, gag, and red snapper.*

#### *Tag Retention*

A total of 626 fish (489 red grouper, 3 gag and 134 red snapper) caught off headboats were double tagged with PIT and Hallprint® plastic dart tags during the project. Of these, 36 fish (26 red grouper, 1 gag and 9 red snapper) were recaptured (Table 14). Most double tagged fish (34 = 94%) were recaptured with both tags. Only 2 (6%) recaptured fish, one gag and one red grouper, shed the external Hallprint® plastic dart tag. These data suggest most Hallprint® plastic dart tag tagged red grouper and red snapper retain their tags, and thus statistics calculated from these species of recaptured fish tagged with Hallprint® plastic dart tags need not take tag shedding into account. Since only three gag were double tagged the 33.3% tag shedding rate should be disregarded.

Table 14. Target species double tagged with Hallprint ® plastic dart tags and PIT tags. All fish tagged and recaptured from headboats.

Species	# Tagged	# Recaptured	% Recaptured	% Shed tag
Red Grouper	489	26	5.3	0.2
Red Snapper	134	9	6.7	0
Gag	3	1	33.3	33.3

### *Growth Rate Comparisons*

Growth of tagged fish was calculated as a ratio of growth in inches for the time interval between tagging and recapture by the number of days fish were at liberty. Growth was calculated for both double and single tagged red grouper, red snapper, and gag. Single tagged red grouper and red snapper grew faster than double tagged fish, while double tagged gag grew slightly faster than single tagged fish (Table 15). Overall, red grouper exhibited the slowest growth while red snapper had the highest growth rate. However, the small sample size (n=36) for double tagged fish coupled with the variation of growth in different regions (Lombardi et. al) suggests these data are not reliable and additional studies should be undertaken.

Table 15. Growth rates of tagged and recaptured target species. Growth rate presented as % change in length/days of freedom. Double tagged fish were tagged with PIT tags and Hallprint ® plastic dart tags; single tagged fish were tagged with Hallprint ® plastic dart tags.

Species	N double tagged	Growth rate (%)	N single tagged	Growth rate (%)
Red Grouper	26	0.56	388	0.92
Red Snapper	9	0.92	512	1.91
Gag	1	2.05	310	1.78

### *Objective 7: To obtain movement patterns for red grouper, red snapper, gag and mangrove snapper in the Gulf of Mexico and South Atlantic*

Movement data from this project were combined with other data already in the MML database from other MARFIN and CRP projects to create the movement figures for red grouper, red snapper, and gag. Only one vermilion snapper was recaptured so the species could not be included in any of the analyses. Most individuals (> 69%) within the target species were site faithful exhibiting little or no movement (Figure 23). However, there were some exceptions as some red grouper, red snapper and gag did travel long distances, from 50 to  $\geq 101$  km (Figures 24a-c). Mangrove snapper were never recaptured at distances >25 m from their original tagging location (Figure 24d). The spatial resolution of reporting (one minute of latitude and longitude) imposed restrictions on this analysis, so only moves greater than 3 km were counted as showing any movement.

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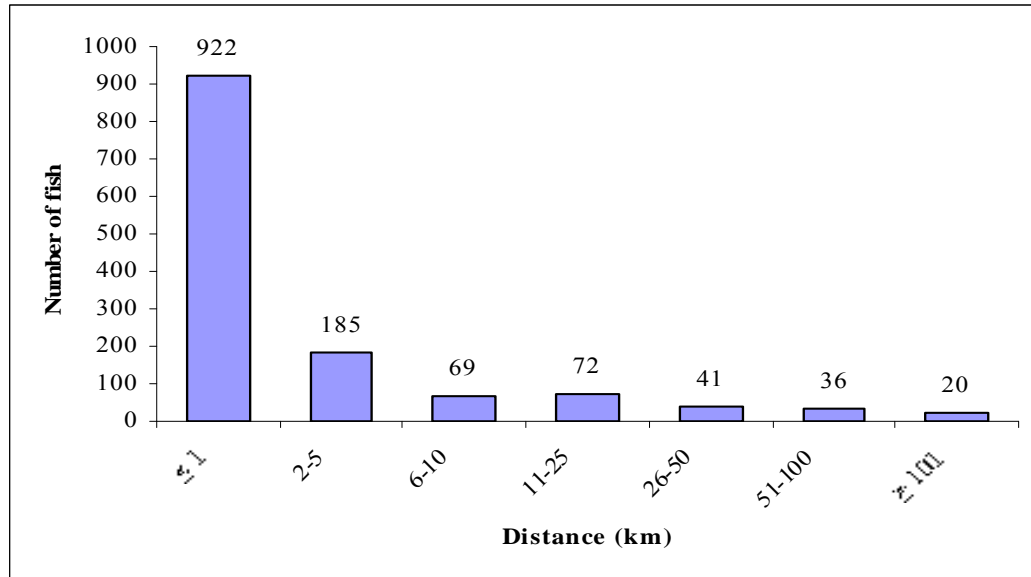


Figure 23. Distance moved between release and recapture sites by all target species combined.

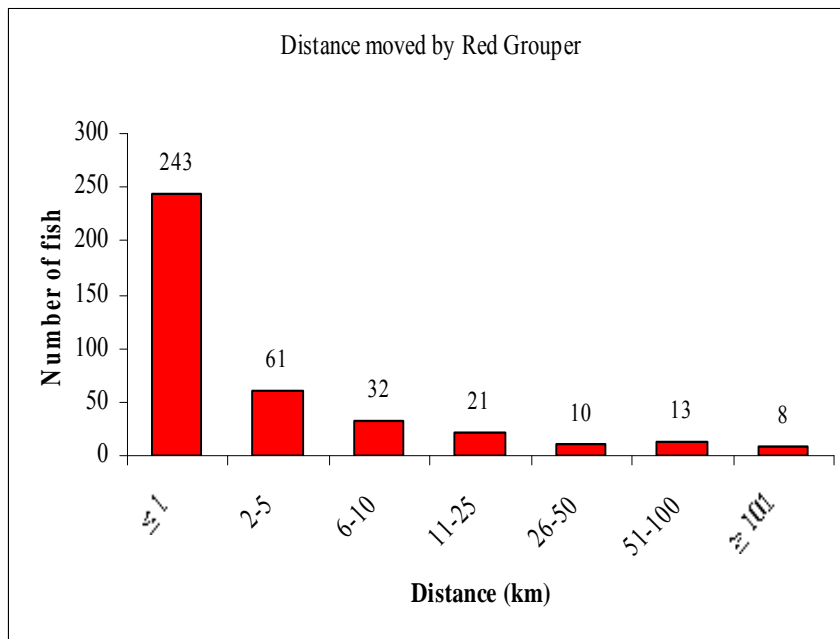


Figure 24a. Distances moved between release and recapture sites by individual red groupers.

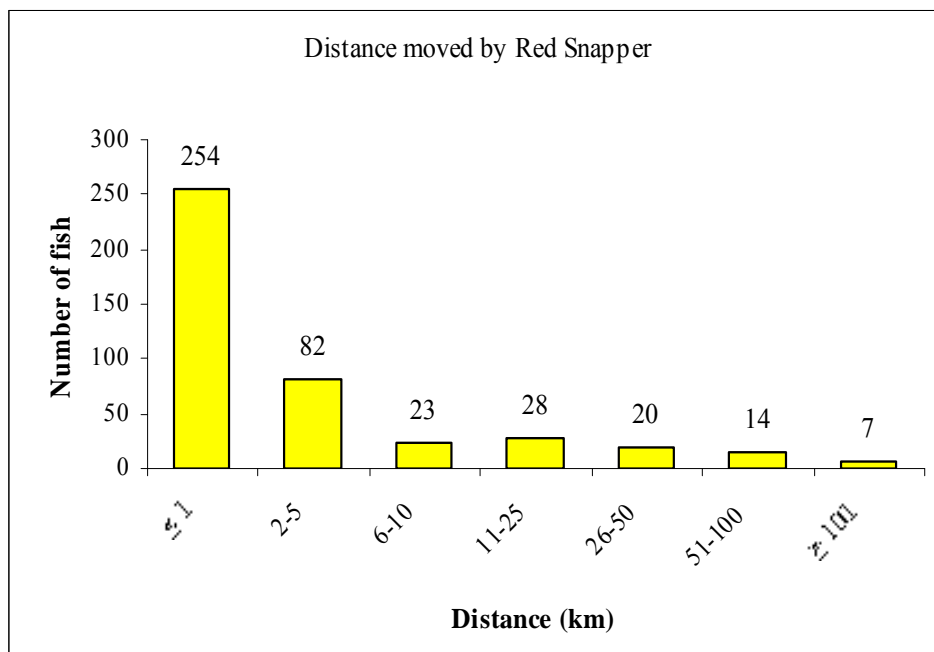


Figure 24b. Distances moved between release and recapture sites by individual red snappers.

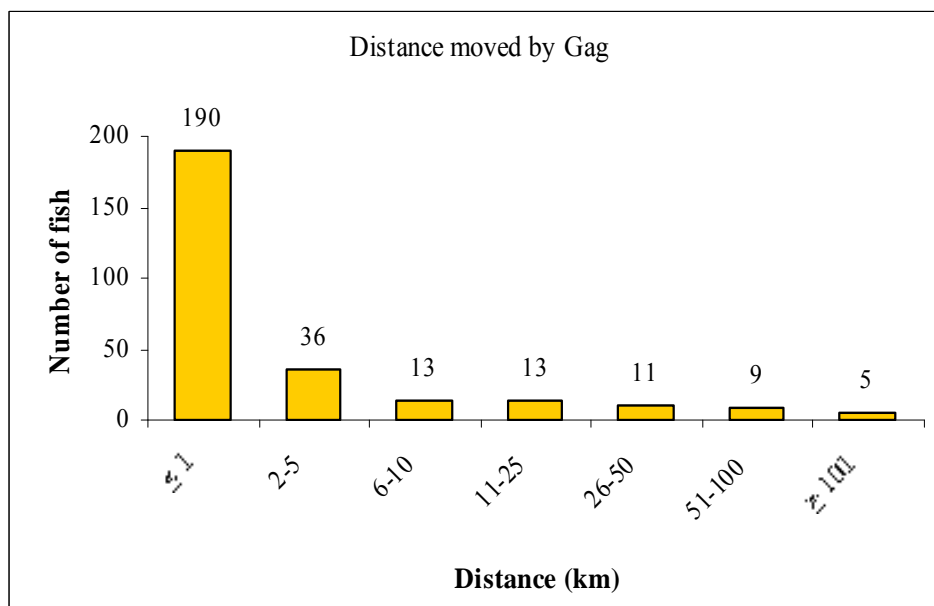


Figure 24c. Distances moved between release and recapture sites by individual gags.

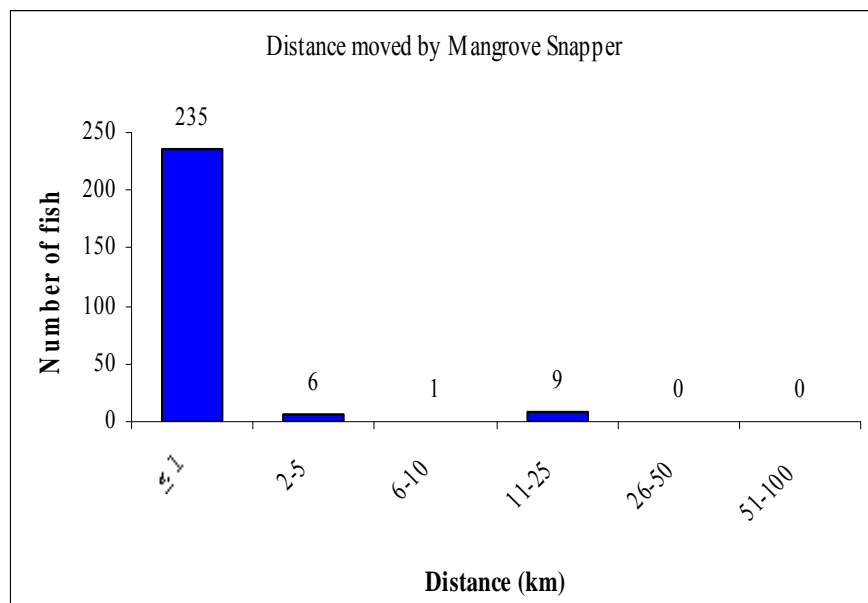


Figure 24d. Distances moved between release and recapture sites by individual mangrove snappers.

### *Red Grouper*

Of the 434 specimens of red grouper recaptured during this project, 46 recaptured individuals were reported that lacked usable recapture locations. Most fish with complete recapture data (89 % = 243 fish) exhibited zero – 1.9 km of movement. Thirty-two percent (124 fish) of recaptured red grouper were recaptured within 50 km of the release site and only 5.4 % (21 fish) were recaptured at distances greater than 50 km from their original capture site (Figure 24a).

Off the coast of southwest Florida small red grouper commonly occur in shallow water (3-18 m). When they become larger, they are captured years later by commercial fishers at depths greater than 36 m (Bullock and Smith 1991). Data from this study and other studies conducted by the staff of the Fish Biology Program at Mote Marine Laboratory support this finding. Table 16 lists the tag numbers of small red grouper from the MML database initially caught tagged and released inshore and recaptured 43-1200 days later offshore by commercial fishers. By plotting fish length at first capture by distance from shore, the trend shows that overall fish length increased with distance from shore for red grouper (Figure 25). When the regression for all fish was tested for significance, the two variables (fish length and distance from shore) were significantly correlated ( $p=0.001$  for both Atlantic and the eastern Gulf of Mexico). See appendix.

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Table 16. Red grouper originally tagged inshore as sub-adults by recreational and recreational-for-hire fishers and recaptured by commercial fishers after moving offshore. Gear Type: LL = commercial long-line; ERR = commercial electric reel; NA = data not available

Tag #	Tag Date	Data Type	Tag depth (m)	Tag Size (cm)	Recap Date	Distance Traveled (km)	Recap Depth (m)	Recap Gear	Recap Size (cm)	DOF
10493	07/16/92	RECREATIONAL	6.4	31.8	09/29/93	88.5	43.9	LL	36.3	440
10546	07/10/92	RECREATIONAL	6.1	43.2	06/15/94	106.2	43.9	LL	49.5	705
25904	08/25/00	HEADBOAT	21.0	45.7	01/10/03	92.2	84.1	LL	NA	868
2789	02/06/91	RECREATIONAL	24.9	45.7	06/03/91	51.5	43.9	LL	45.7	117
42807	06/06/03	HEADBOAT	18.3	40.6	04/20/05	69.4	47.5	LL	58.4	684
4394	09/15/91	RECREATIONAL	20.7	45.7	12/28/94	NA	NA	LL	63.5	1200
45399	07/02/03	HEADBOAT	29.0	40.6	09/27/05	99.3	82.3	LL	56.4	818
46177	07/30/03	HEADBOAT	27.4	44.5	08/20/05	NA	NA	LL	50.8	752
46214	07/09/03	HEADBOAT	25.6	47.0	01/30/05	134.4	85.3	LL	58.4	571
47199	05/28/04	HEADBOAT	21.3	43.2	08/24/05	69.2	45.7	LL	63.5	453
49283	07/13/04	HEADBOAT	21.6	43.2	08/01/05	212.3	82.3	LL	55.9	384
50655	07/27/05	HEADBOAT	27.4	37.5	07/16/06	2.3	29.0	ERR	48.3	354
50656	07/27/05	HEADBOAT	27.4	48.9	09/08/05	NA	NA	LL	50.8	43
52122	06/16/04	HEADBOAT	48.8	53.3	03/18/05	80.5	63.4	LL	61.0	275
5555	04/08/92	HEADBOAT	36.6	33.0	07/14/93	57.9	36.6	ERR	51.3	462
699	08/03/91	CHARTER	24.4	45.7	07/06/92	48.3	39.6	LL	50.8	338
7964	12/03/91	CHARTER	3.7	26.0	09/06/92	65.7	25.9	ERR	41.9	278

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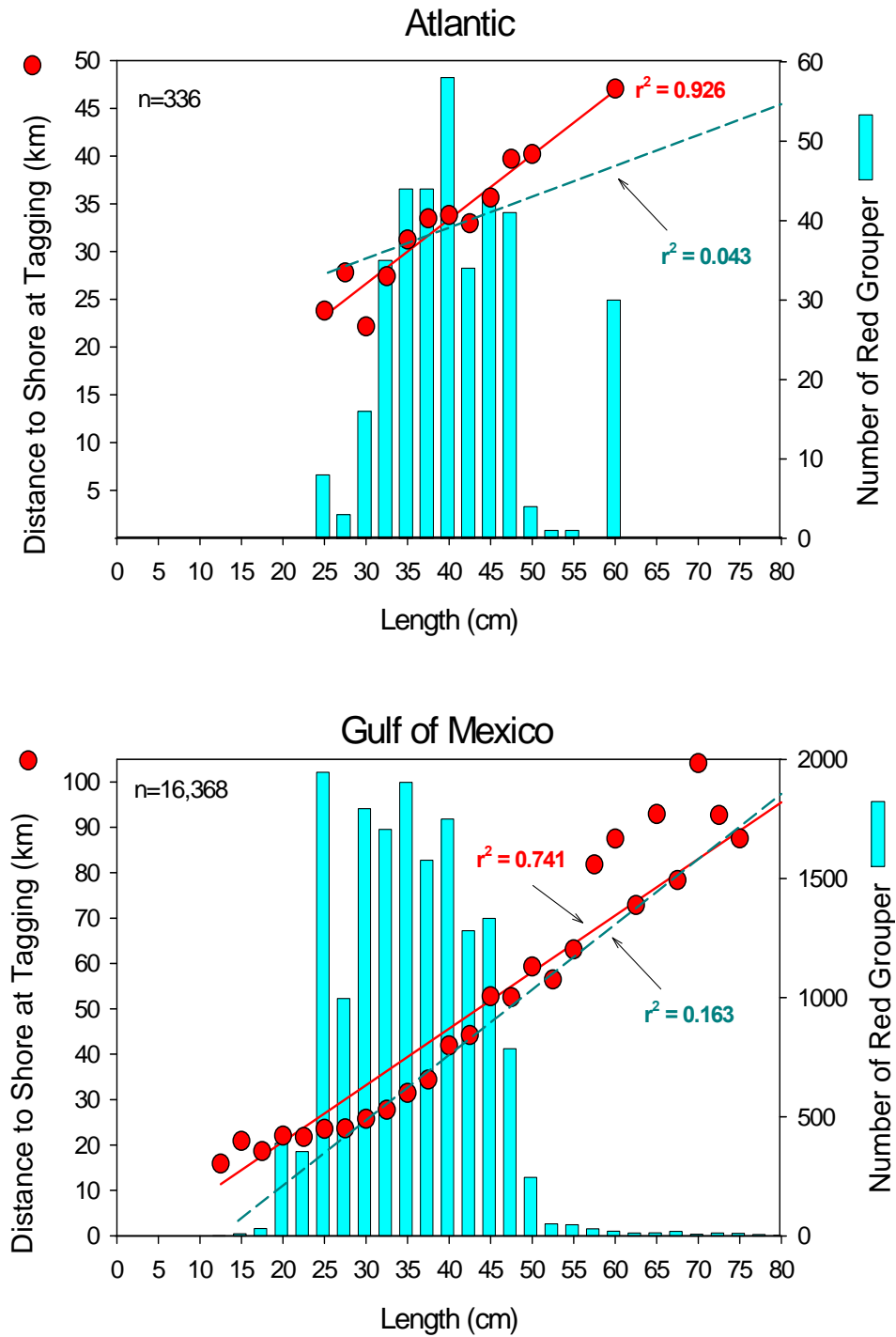


Figure 25. Red grouper length (cm) vs. distance from shore when originally captured and tagged. Regressions of mean lengths (red dots) and the length of each individual fish (cyan dotted line) vs. distance to shore are shown.



Figure 26 shows tag and recapture locations for red grouper in the MML database. Some onshore/offshore movements have been attributed to seasonality and inshore feeding migrations (SEDAR 12). Hurricanes have also been documented as influencing fish movement. Franks (2003) documented the presence of juvenile and adult red grouper on artificial reefs and petroleum platforms off Mississippi in the aftermath of Hurricane Lili in 2002.

Although most red grouper were site faithful, some fish did exhibit long distance movements (Figure 26). The maximum verifiable distance traveled was 375 km. Four red groupers were reported tagged off the Florida west coast and recaptured in the South Atlantic, but these recaptures could not be verified. However, it is plausible that mixing at some level does occur as genetic studies have not discovered significant differences in Florida east and west coast red groupers (Zatcoff et al.2004). Other groupers have also been documented to exhibit long distance movements; a yellowfin grouper travelling from the Marquesas, Florida, to the Berry Islands in the Bahamas has been a verified recapture in the MML reef fish tagging database.

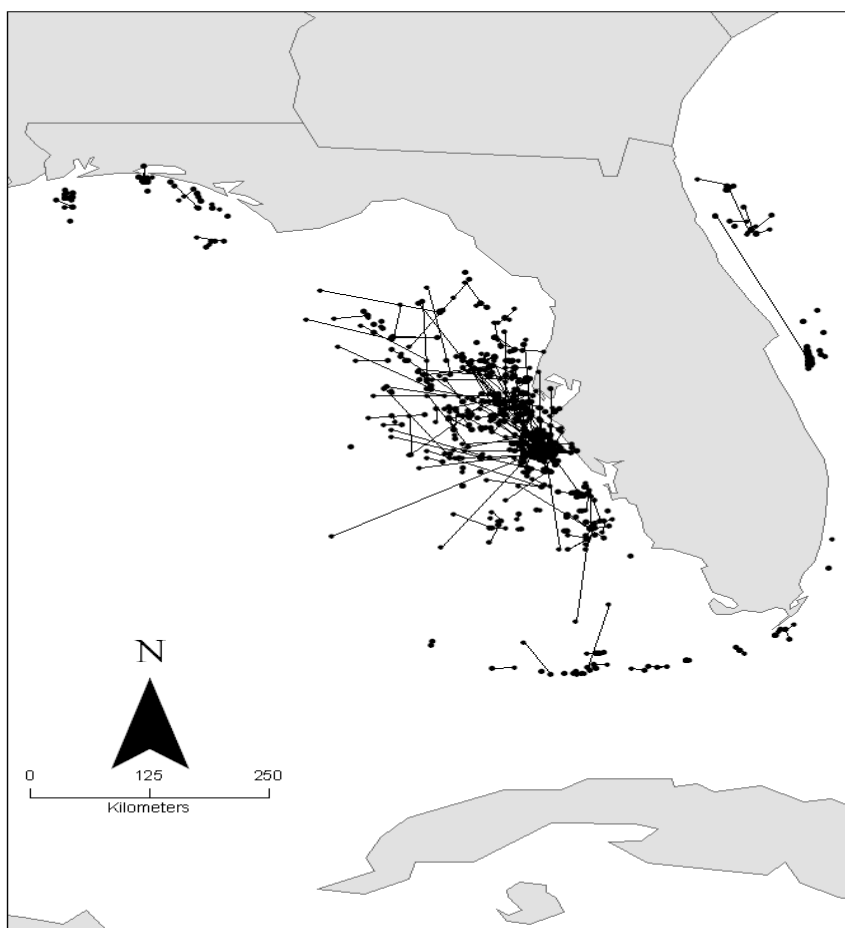


Figure 26. Long distance movements of tagged red grouper.

A number of these long distance movements, particularly those demonstrating movement offshore towards deeper water, can be attributed to ontogeny (Figure 27). According to Moe (1966), red grouper larvae move shoreward to settle inshore on hard bottom nursery grounds. At around age 5, when they become sexually mature, they move offshore to inhabit the continental shelf and shelf edge.

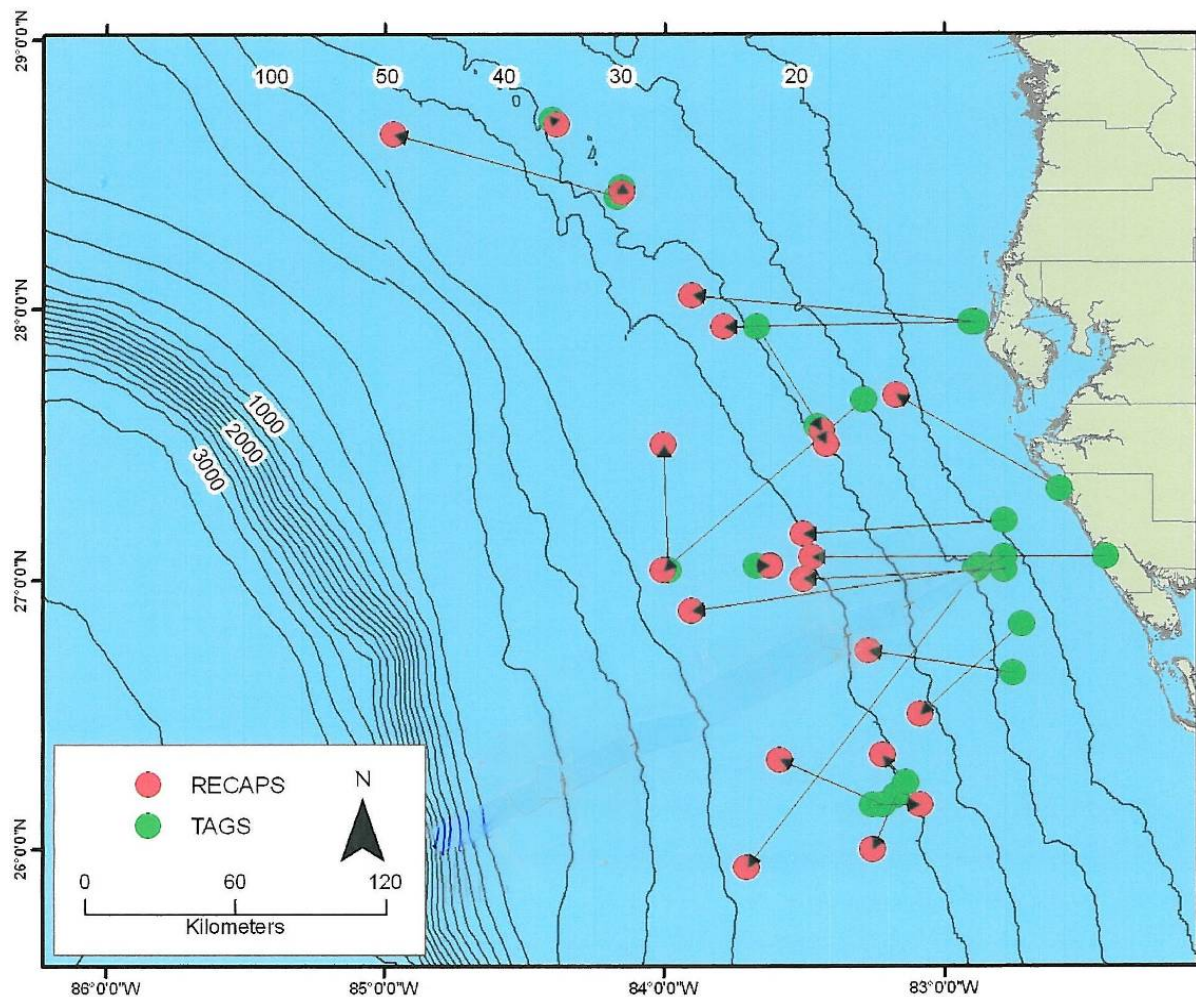


Figure 27. Ontogenetic movement of red grouper from inshore to offshore locations with fish age and growth.

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## Red Snapper

Of the 529 red snappers recaptured during this project, 28.7 % (152 fish) exhibited zero movement. Of those that moved greater than 3 km from the original tagging site, 15.1% (80 fish) were recaptured within 10 km of the release site and only 3.8% (20 fish) were recaptured at distances greater than 50 km (Figure 24b). Some red snappers originally captured and tagged off recreational or recreational-for-hire vessels were later recaptured by commercial fishers (Table 17).

Table 17. Red snappers originally tagged by recreational and recreational-for-hire fishers and recaptured by commercial fishers. Gear Type: LL = commercial long-line; ERR = commercial electric reel; NA = data not available

Tag #	Tag Date	Data Type	Tag Depth (m)	Tagging Length (cm)	Recapture Date	Distance Traveled (km)	Recapture Depth (m)	Recapture Gear	Recapture Size (cm)	Days of Freedom
25494	07/21/99	HEADBOAT	31.7	35.6	7/29/2000	NA	30.5	ERR	45.7	374
25495	07/21/99	HEADBOAT	31.7	40.0	6/30/2000	0	30.5	ERR	49.5	345
27504	07/21/99	HEADBOAT	31.7	33.7	7/29/2000	NA	30.5	ERR	44.5	374
29495	09/23/99	HEADBOAT	29.9	34.3	6/30/2000	NA	30.5	ERR	40.6	281
29749	04/26/00	NA	29.0	33.0	9/15/2000	NA	30.5	ERR	38.1	142
34507	08/16/00	HEADBOAT	31.7	36.8	9/12/2000	NA	30.5	ERR	45.7	27
58150	05/09/06	RECREATIONAL	27.4	35.6	12/04/2006	NA	NA	LL	40.6	209

As seen in red grouper, larger red snapper tend to occur offshore in deeper water. The only exceptions were red snapper caught by charter captains in the Dry Tortugas (Figure 28). When the regression for all fish was tested for significance, the two variables (fish length and distance from shore) were significantly correlated ( $p=0.001$  for both Atlantic and the eastern Gulf of Mexico). See appendix. These data support findings by Allman et al. (2002) who reported that larger, older red snapper were found offshore in deeper water and most commonly landed by the commercial reef fish sector.

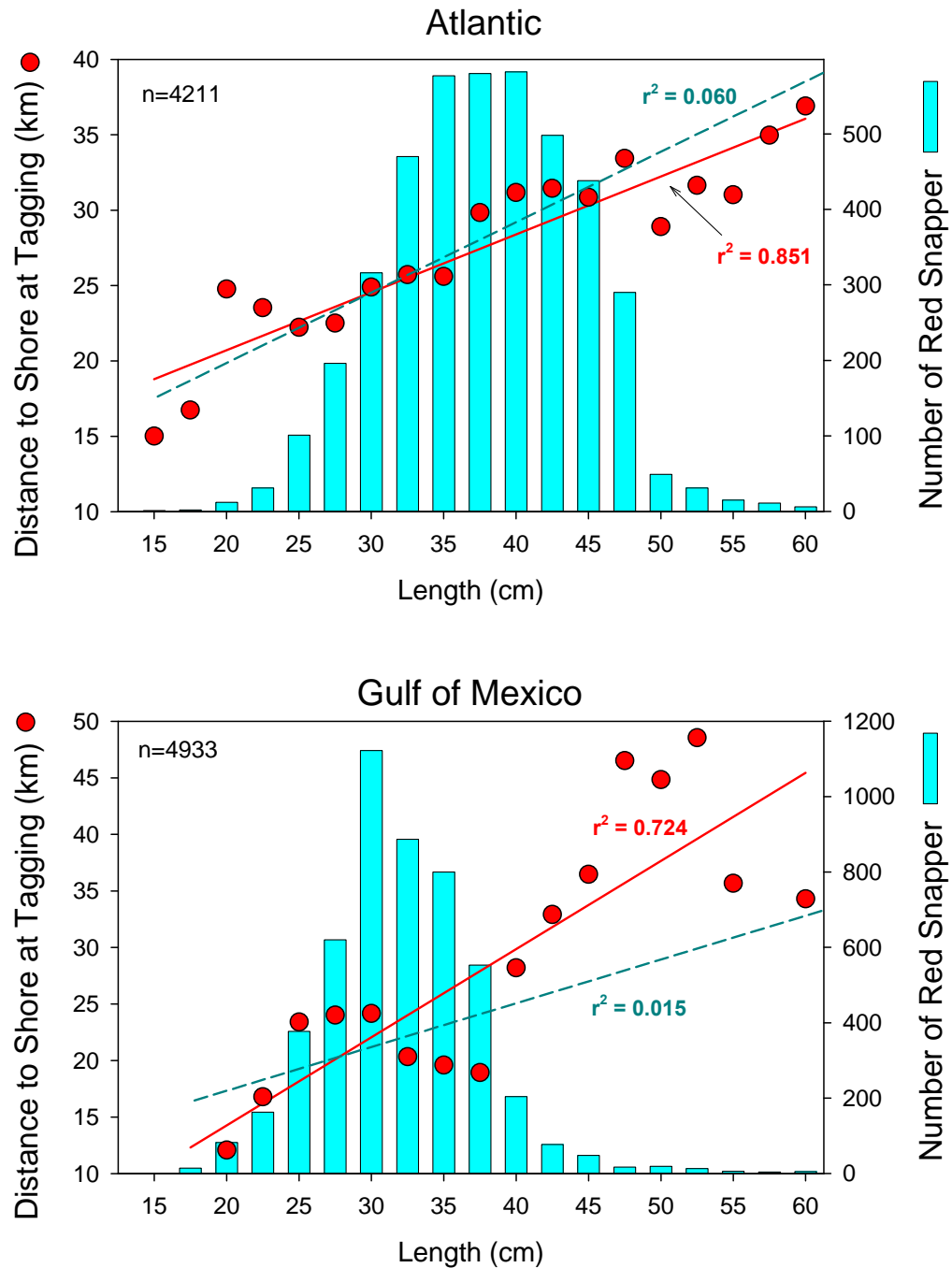


Figure 28. Red snapper size (cm) vs. distance to shore when originally captured and tagged. Red circles represent mean fish length for each 5 cm length increment. Regressions of the mean length (red solid line) and the length of each individual fish (cyan dotted line) vs. distance to shore are shown.

Plotting red snapper tag and recapture location showed the majority of the fish were site faithful, although some made great movements (Figure 29). The largest movement distance recorded was 362.4 km or 225 miles over a period of 48 months. Additionally, tag recaptures from the MML database show a red snapper traveling from off Pensacola (Florida panhandle) to St. Augustine (Atlantic coast). Movement on the east coast tended to be more often from north to south and vice versa due to the narrow shelf whereas west coast fish could expand over the much broader shelf. The general direction of movement, determined by fish tagged in the Florida Panhandle, was to the east and southeastern in the Gulf of Mexico (Figure 30). This is consistent with a clockwise direction along the Gulf coast reported for red snapper in previous studies (Ingram and Patterson 2001, Patterson et al 2001). This may indicate that re-recruitment to the southwest Florida coast may have come from the northern Gulf of Mexico.

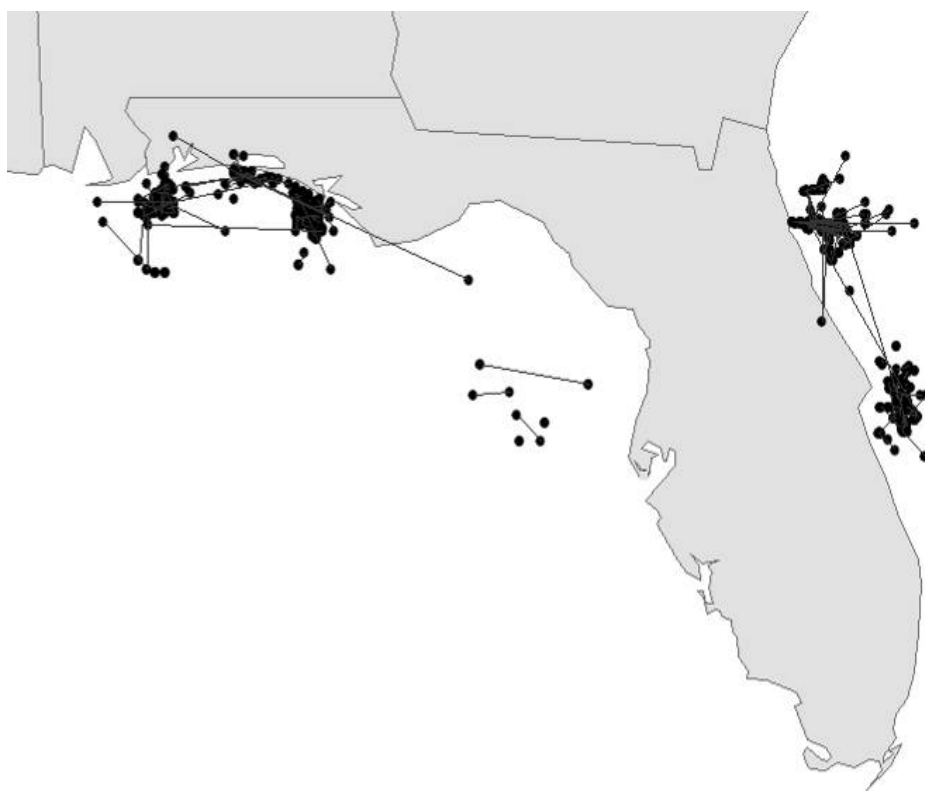


Figure 29. Movements of red snapper determined through tag and recapture data.

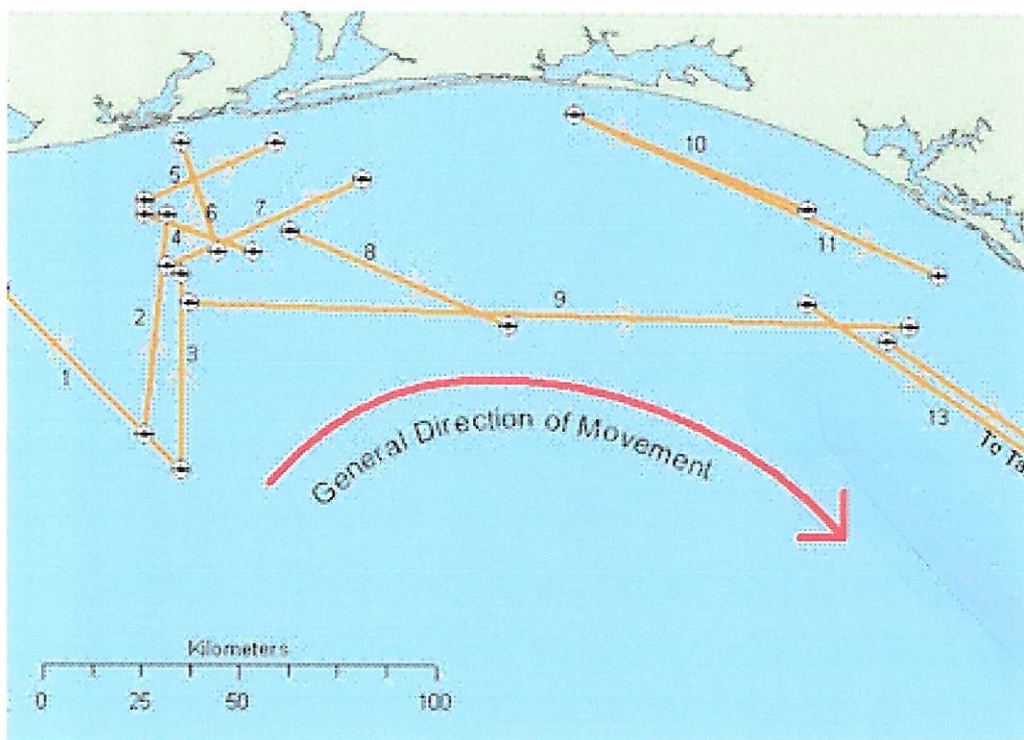


Figure 30. General direction of movement of red snapper in the Gulf of Mexico as determined from tag recaptures.

In the early 1990's, when red snapper were being caught as by-catch in the Florida Middle Grounds and off Venice, Florida and southward, they were undersized fish. Over time, red snapper have been spreading out and reclaiming more and more of their historical habitat off southwest Florida (personal communication with reef fish fishermen). Now that these fish have grown to legal size and have entered the commercial, recreational-for-hire and recreational reef fish fisheries in increasing numbers, it is not only important to determine the age and reproductive state of these fish, but also to obtain habitat information for southwest Florida fish. Habitat requirements for various life stages of red snapper in the northern Gulf have been described (Ouzts and Szedlmayer 2003; Szedlmayer and Lee 2004). Evidence from NMFS long-line surveys suggests that larger older red snapper do not appear to require the structured habitats described in the northern Gulf (Allman et al. 2002). This agrees with data reported by Futch and Bruger (1976) from the report by Adams and Kendall (1891), which states that off southwest Florida, 8 of 12 sites where snapper were caught were sandy with shell or muddy bottoms, not reef or rocky areas exhibiting a vertical relief.

## *Gag*

Figure 31 shows fish length versus distance from shore. The slope of the regression lines for both the means and for individual fish is flatter in the South Atlantic off the Florida east coast than for the Gulf. It is also flatter than the slope for both red grouper and red snapper off the east coast of Florida. When the regression for all fish was tested for significance, the two variables (fish length and distance from shore) were significantly correlated ( $p=0.001$  for both Atlantic and the eastern Gulf of Mexico). See appendix.

Of the 323 gag recaptured during this project, 37.8 % (122 fish) exhibited zero movement. For those gag that moved greater than 3 km from the original tagging site 12.1 % (39 fish) were recaptured within 10 km of the release site and only 3.4 % (11 fish) were recaptured at distances greater than 50 km. Beaumariage (1969) also working in the Gulf, reported that most gag tagged in his study were recaptured at or near the original tagging site. Figures 32 and 33 show gag tag/release and recapture sites in both the South Atlantic and the Gulf. Like movements for red grouper and red snapper, gag movements in the Gulf extend farther offshore utilizing the broad southwestern shelf and are more restricted off the narrow Florida east coast shelf.



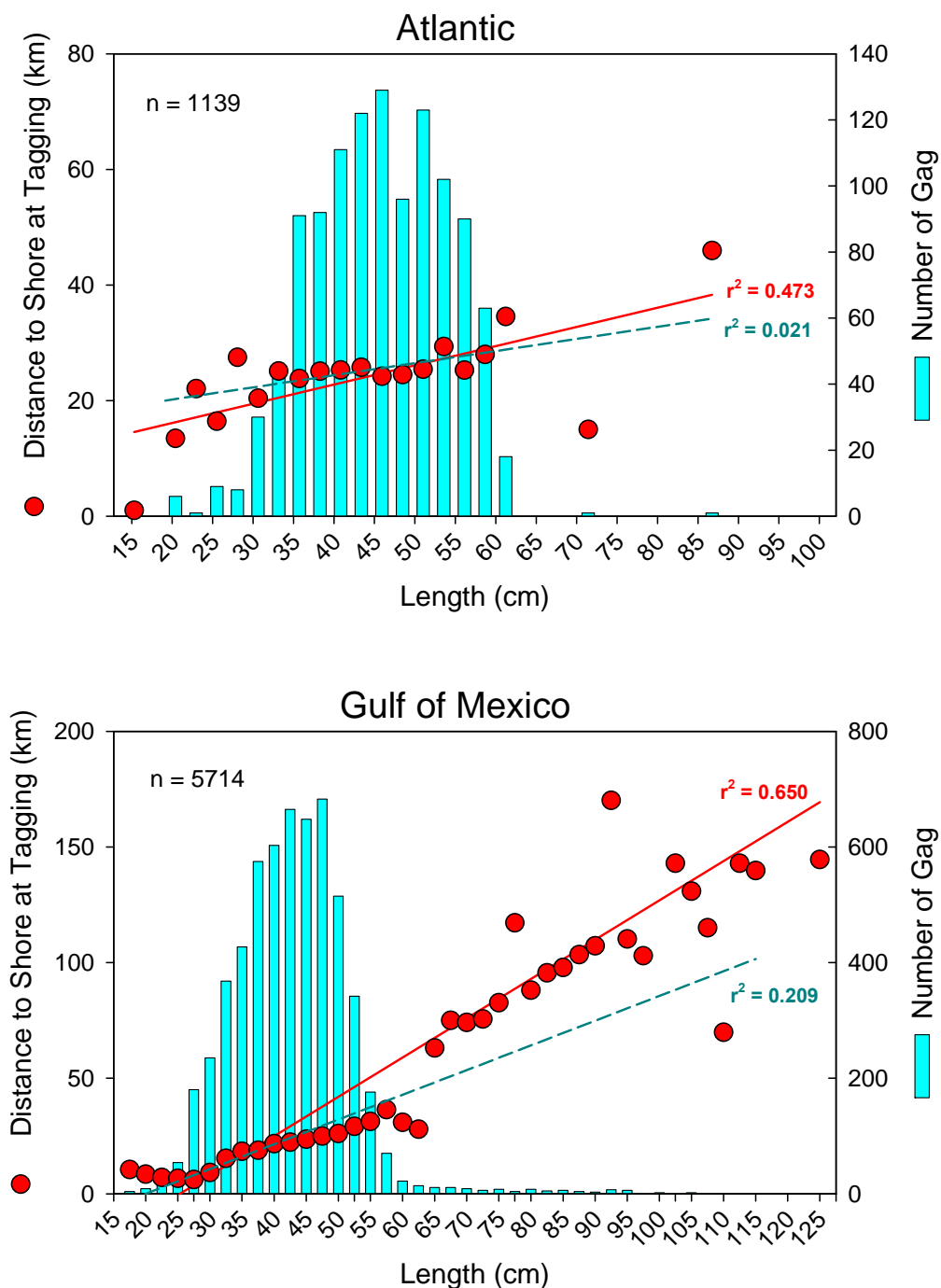


Figure 31. Gag length (cm) vs. distance from shore when originally captured and tagged. Red circles represent mean fish length for each 5 cm length increment. Regressions of the mean length (red solid line) and the length of each individual fish (cyan dotted line) vs. distance to shore are shown.



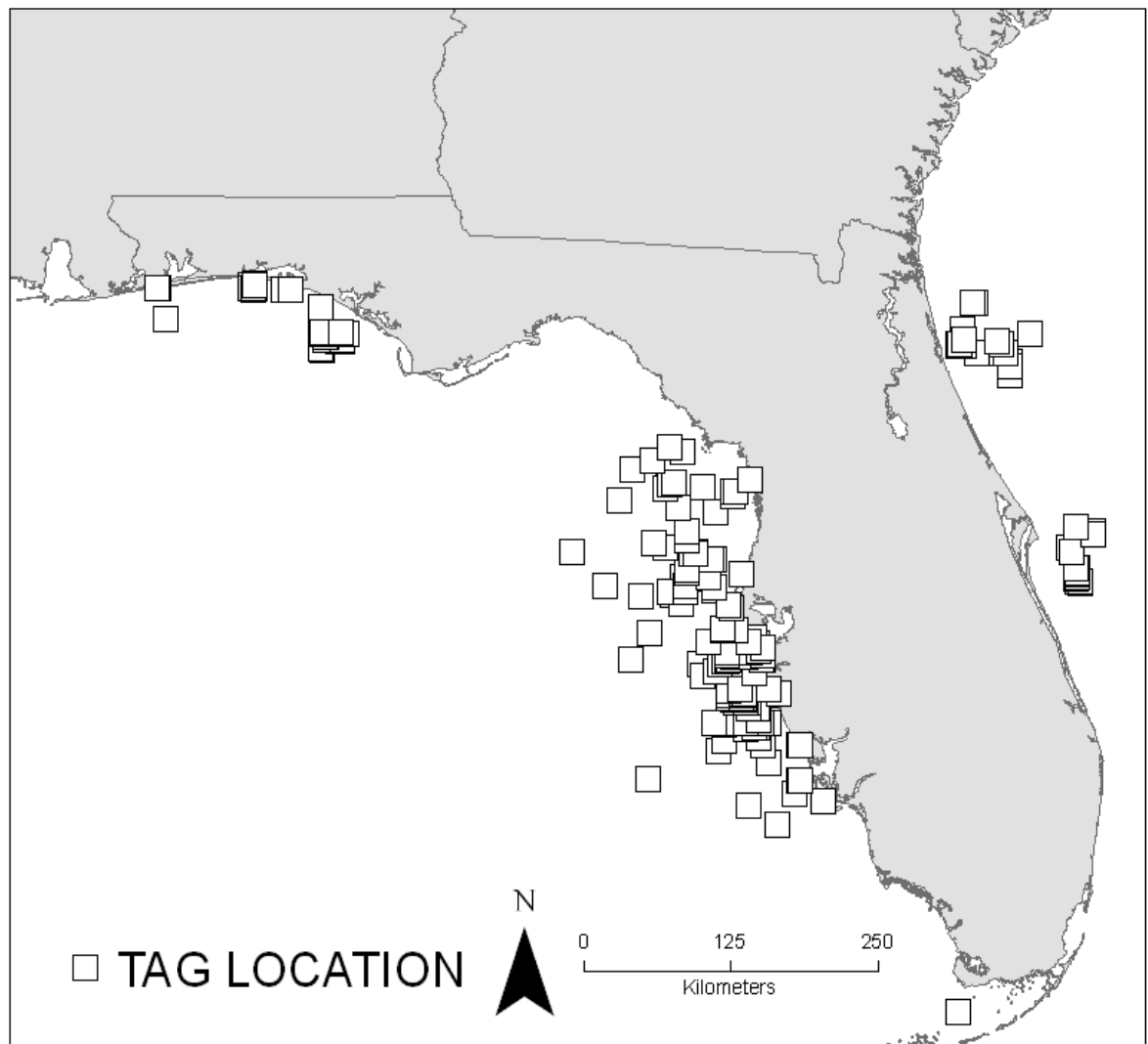


Figure 32. Gag tagging locations off the west and east coasts of Florida.

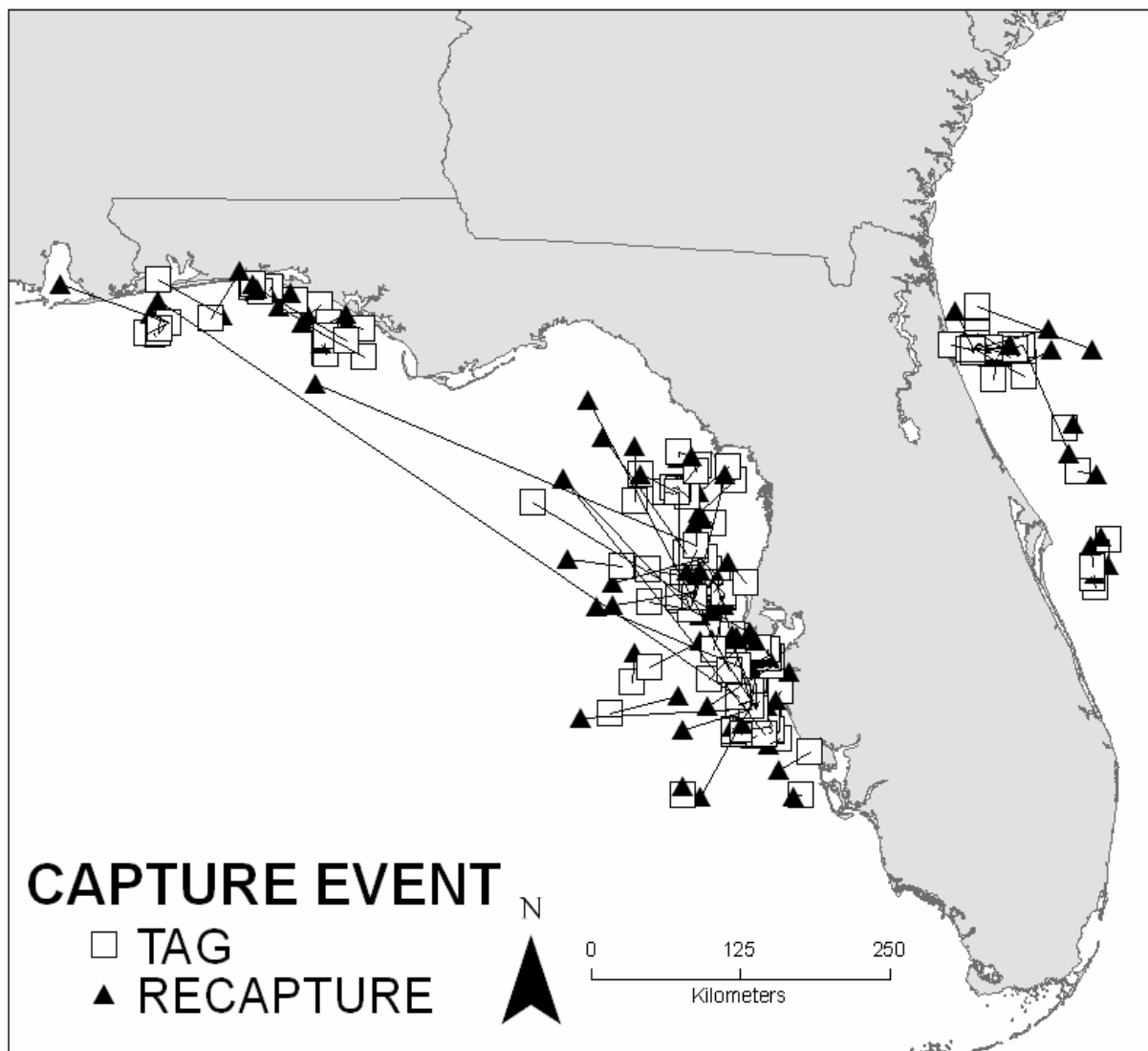


Figure 33. Gag tag and recapture sites.

These results differ from gag from the South Atlantic, which have been reported to move from South Carolina to the Florida east coast, Florida Keys, and the Gulf of Mexico (McGovern et al. 2005). However, Gulf of Mexico gag do exhibit ontogenic movement as juveniles migrate from bay grass beds to reef associated structures to deeper water (Coleman et al. 1996). This finding was supported by our limited recapture data (Table 18).

Table 18. Gag originally tagged inshore as sub-adults by recreational-for-hire fishers and recaptured by commercial fishers after moving offshore. Gear Type: LL = commercial long-line; ERR = commercial electric reel

Tag #	Tag Date	Data Type	Tag depth (m)	Tagging Length (cm)	Recapture Date	Distance Traveled (km)	Recapture Depth (m)	Recapture Gear	Recapture Size (cm)	Days of Freedom
22792	04/20/00	HEADBOAT	20.4	44.5	11/20/03	117.5	69.5	LL	73.7	1309
26620	05/29/99	CHARTER	12.2	45.7	05/07/03	111.0	47.5	ERR	142.2	1439
38561	06/26/01	HEADBOAT	19.8	58.4	11/15/04	89.3	50.3	LL	90.8	1238

### *Mangrove Snapper*

Of the 277 mangrove snapper recaptured during this project, 74.4 % (206 fish) exhibited zero movement (Figure 24d). Of those that were recaptured greater than 3 km from the original tagging site 1.6 % (5 fish) were recaptured within 10 km of the release site and none were recaptured at distances greater than 50 km. Bortone and Williams (1986) reported strong site fidelity for mangrove snapper, which appears to be confirmed by the data gathered during this project.

### *Vermilion Snapper*

There were insufficient data for analyses.

## **SIGNIFICANT PROBLEMS**

Although all project objectives were met, more data were collected for some of the target species than others because red grouper, gag, and red snapper were targeted by more fishers. The greatest problem was weather. Many sampling trips had to be cut short or repeatedly cancelled because of the numerous hurricanes, which affected both coasts during the project duration. The damage done to the University of Southern Mississippi's Gulf Coast Research Laboratory by Hurricane Katrina was extensive and no-cost extensions were required to have sufficient time to complete the work.

## **NEED FOR ADDITIONAL RESEARCH**

The results of this project suggest that there are differences in depth-induced mortality of red snapper and red grouper that are related to swim bladder morphology and physiology. However, it is unknown if these are family-specific differences that would apply to many snapper and grouper species or just represent species-specific differences. Additionally, the enhanced survival of small individuals compared to larger individuals documented in this project may also be

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species-specific rather than a family trait. Therefore, additional research should be conducted on depth-induced mortality and swim bladder morphology, including the area of rete mirabile and gas gland cells, in a variety of reef fish species in the families Lutjanidae and Serranidae.

## **EVALUATION**

### **A. EXTENT TO WHICH THE PROJECT GOALS AND OBJECTIVES WERE ATTAINED**

Project goals and objectives were fully completed in spite of two years of major hurricanes affecting all of Florida, decimating some parts of Florida, and causing massive damage at the University of Southern Mississippi's Gulf Coast Research Laboratory campus (Hurricane Katrina). The presence of numerous hurricanes and tropical storms radically influenced the time required to collect the number of samples needed and delayed the time processing required to complete the project.

Despite set-backs from weather, a large quantity of data was collected and reported. Important information on swim bladder morphology has been documented which will aid in further understanding depth-induced mortality of both red grouper and red snapper. Data collected will also be useful for stock assessment analyses of the targeted species. Furthermore, new information regarding the efficacy of circle hooks for survival of red grouper was shown through this project.

### **B. DISSEMINATION OF PROJECT RESULTS**

Early results of this research were used at the 2004 Red Snapper SEDAR Meeting held in New Orleans, LA, the 2006 Gag SEDAR Meeting held in January in Charleston, SC, and the 2006 Red Grouper SEDAR Meeting held in July in St. Petersburg, FL. A poster presentation of the swim bladder results was presented at the 2006 Joint Meeting of Ichthyologists and Herpetologists held in July in New Orleans, LA (Brown-Peterson et al. 2006, see Appendix). Some data were put in the RFSS MARFIN funded reef fish newsletter, which was sent to MML volunteer taggers, fishery biologists and managers, members of the Gulf and South Atlantic Councils, and newspaper and fishing magazine writers. Results were also presented at the 2007 MARFIN Conference held in St. Petersburg, FL (abstract in Appendix). Project results will also be disseminated through sending a copy of this report not only to NOAA, but also to the Gulf and South Atlantic Council, and the state of Florida. Some project results will be used as part of the doctoral dissertation of the Project Manager, Karen Burns, for completion of her Doctorate in Biological Oceanography at the University of South Florida's College of Marine Science. In addition some of the results of this research will be presented as part of an invited speaker presentation at the 2008 American Fisheries Society Annual Meeting Sea Grant sponsored Special Symposium entitled "Research, Education, and Management Needs Regarding Barotrauma in Fish", this August in Ottawa, Canada (See abstract in Appendix)

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Finally, results will be published as papers in peer reviewed scientific journals. Papers in progress include “Red snapper distribution, size, and movement patterns in the eastern Gulf of Mexico and Atlantic Ocean off Florida” by K. M. Burns, J. G. Gannon, J. M. Sprinkel, and B. D. Robbins, which was submitted to the *North Atlantic Journal of Fishery Management* and is in revision and “Swim bladder morphology in red snapper, *Lutjanus campechanus* and red grouper, *Epinephalus morio*: insights into survival of rapid decompression”, a manuscript in preparation by Nancy Brown-Peterson, Karen Burns, and Robin Overstreet.

## ACKNOWLEDGEMENTS

The authors wish to thank NOAA/NMFS MARFIN for funding this research. Thanks are also extended to MML’s Fish Biology Program staff, student interns and volunteers for all their hard work on all aspects of this research. Special thanks are extended to Nicholas Parnell for his help with necropsies and photography and Roger DeBruler for leading much of the fieldwork for fish tagging and sample collection. Special thanks to all the recreational-for-hire vessel owners, captains and crew who allowed MML staff to conduct this research aboard their vessels. The authors are very grateful to all the volunteer taggers who measured/tagged/released red snapper and reported recaptures. We greatly appreciate the assistance of Kim Lamey, USM/GCRL, who did all the histological processing, sectioning, and staining of swim bladder tissues and Rusty Holms (Mote) for her help with the production of this report..

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## APPENDIX



Abstract submitted to Joint Meetings of Ichthyologists and Herpetologists (ASIH) for poster presentation at July 2006 meeting, New Orleans Louisiana

## Swim bladder morphology differences in red snapper and red grouper

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Depth-induced mortality due to injuries sustained during rapid decompression is common in red grouper (*Epinephelus morio*) and red snapper (*Lutjanus campechanus*) and may be related to morphological and physiological aspects of the swim bladder. Red snapper have smaller but thicker swim bladders than red grouper, and swim bladder-ruptures tend to be smaller and less frequent. Red grouper between 318 and 381 mm FL develop a secondary vascularized structure on the dorsal surface of the swim bladder that is not seen in red snapper; this secondary structure contains rete but no gas gland. Histological analyses of swim bladders from 62 red snapper (123 – 674 mm FL) and 138 red grouper (205 – 766 mm FL) showed rete area significantly ( $p < 0.05$ ) increased with body length (FL) for both species, but gas gland area was not significantly related to FL in red snapper. Red snapper had a significantly higher percentage of rete area in the swim bladder than did red grouper, although there were no differences between species in the percentage of gas gland area. However, red grouper had a significantly higher percentage of the rete area occupied by gas gland than did red snapper. The percentage of fish showing hemorrhaging in both rete and gas gland increased significantly with FL for both species, but retal hemorrhaging was significantly higher in red grouper when adjusted for fish length. These differences in structures of the swim bladder may have implications in depth-induced mortality. Funded in part by NMFS MARFIN Award No. NA17FF2010.



Abstract from 2007 MARFIN meeting, St. Petersburg, FL

**Evaluation of the Efficacy of Current Minimum Size Regulation for Selected Reef Fish Based on Release Mortality and Fish Based on Release Mortality and Fish Physiology**

**MARFIN Grant No. NA17FF2010**

Funding Amount: \$359,804 Federal (\$133,852 non-Federal)

Karen M. Burns, Nancy Brown-Peterson,  
Robin Overstreet, PhD

Mote Marine Laboratory and University of Southern Mississippi

**Study Objectives:** **1)** Test the hypothesis that red grouper are more susceptible to depth-induced mortality than red snapper, based on swim bladder size, thickness, and the number of rete mirabile and gas gland cells in the swim bladder. **2)** Test the hypothesis that smaller red grouper (< 30.5 cm) survive rapid decompression better than larger (> 38 cm) fish based on changes in swim bladder structure with size between 30.5 – 38 cm. **3)** Test the hypothesis that circle hooks greatly reduce release mortality in red snapper. **4)** Quantify undersize and legal catch for red grouper, *Epinephelus morio*, gag, *Mycteroperca microlepis*, red snapper, *Lutjanus campechanus*, vermilion snapper, *Rhomboplites aurorubens*, and mangrove snapper, *Lutjanus synagris*. **5)** Obtain catch and release mortality rates relative to depth and gear for target species. **6)** Obtain movement patterns for target species in the Gulf of Mexico and South Atlantic off Florida. **7)** Determine tag shedding rates for red grouper, gag, and red snapper tagged with passive integrated transponder (PIT) tags and single-barbed dart tags.

**Methods and Materials:** **1)** Histological examinations were conducted to assess general appearance of swim bladders, gas glands, rete mirabile, and hemorrhages. Representative samples embedded in paraffin were sectioned at 4 µm and stained in hematoxylin and eosin to compare and photograph secretory and resorption structures. **2)** Necropsies were performed on acute mortalities to determine cause of death and to examine swim bladder condition. **3)** Red snapper and red grouper were caught on circle and J hooks, tagged, and released. Recaptures were compared to determine release mortality relative to hook type. **4)** Red grouper, gag, red snapper, vermilion snapper, and mangrove snapper caught off Florida were quantified and measured aboard headboats, charter, and recreational vessels by Mote Marine Laboratory (MML) staff, student interns, and volunteer taggers. **5)** Target species were tagged with Hallprint® single-barbed plastic dart tags and released off recreational-for-hire and recreational vessels by MML staff, interns, and volunteer taggers. Tag number, species, size, date, depth (ft), gear type, and fish condition were recorded for each tagging event. Recaptures were reported via a dedicated hotline. Data included tag number, date, depth (ft), gear type, fish condition, and whether fish were re-released. A quarterly top tagger prize (\$50), an annual tag lottery (\$100; both tagger and fisher reporting recapture), and a quarterly newsletter were employed to keep anglers informed and provide incentive for tagging and reporting target species. **6)** Original tagging and recapture

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locations were recorded to the nearest 1' of latitude and longitude. Distance for recaptured fish. Red snapper had a significantly higher percentage of rete area than red grouper when adjusted for size (ANCOVA,  $F_{3,196} = 41.042$ ,  $p < 0.001$  overall,  $p = 0.022$  by species). Gas gland area was not related to FL for red snapper. The relationship was significant, but weak for red grouper were calculated. 7) Some target species were tagged with both Hallprint® single-barbed plastic dart and PIT tags. MML staff and interns used PIT tag readers to check all target species caught aboard headboats.

**Conclusions and Recommendations:** 1) Red grouper have larger (in relation to body size), thinner swim bladders than red snapper. Red snapper swim bladder ruptures are smaller than those of red grouper. Red grouper > 380 mm FL have an area on the posterior swim bladder ventral wall, absent in red snapper that contains some rete and convoluted tissue that may aid in gas absorption and secretion. Rete area increased significantly with body size (FL) for both species, although the relationship between rete area and body size was weak, particularly for red snapper. Red grouper had a significantly higher percentage of rete area occupied by gas gland than did red snapper when adjusted for fish size (ANCOVA,  $F_{3,200} = 13.258$ ,  $p < 0.001$  overall,  $p = 0.007$  by species) and a closer association and more connections between gas gland cells and rete. Both species had increased rete and swim bladder hemorrhaging at 50 mm (ANOVA, red snapper rete,  $F_{1,8} = 20.41$ ,  $p = 0.003$ ,  $r^2 = 0.708$ , red snapper gas gland,  $F_{1,8} = 17.38$ ,  $p = 0.004$ ,  $r^2 = 0.672$ , red grouper rete,  $F_{1,11} = 16.21$ ,  $p = 0.002$ ,  $r^2 = 0.580$ , red grouper gas gland,  $F_{1,11} = 19.77$ ,  $p = 0.001$ ,  $r^2 = 0.631$ ). Retal hemorrhaging was significantly higher in red grouper than red snapper, when adjusted for length (ANCOVA,  $F_{3,21} = 5.96$ ,  $p = 0.026$ ), but no significant difference between species in gas gland hemorrhaging ( $p = 0.786$ ). Overall, red snapper survive rapid decompression better than red grouper because of smaller tears in the swim bladder and less tendency to hemorrhage, especially in smaller fish. 2) Necropsies revealed most red snapper died from hook mortality. 3) Circle hooks did not increase survival of red snapper, but did enhance red grouper survival. Of 3,205 red snapper caught on circle hooks, 8.0% were recaptured, whereas 2,263 red snapper were caught on J hooks and 11.4% were recaptured. 4) Red grouper (473 legal and 5,318 undersized), gag (442 legal and 2,855 undersized), red snapper, (1,154 legal and 5,737 undersized), vermilion snapper (563 legal and 564 undersized), and mangrove snapper (904 legal and 3,138 undersized) were measured during 467 headboat, 90 charter, and 427 recreational fishing trips. 5) Red grouper ( $n = 4,997$ ), gag ( $n = 2,704$ ), red snapper ( $n = 5,481$ ), vermilion snapper ( $n = 465$ ), and mangrove snapper ( $n = 3,180$ ) were tagged. Recapture rates were 8.7% for red grouper, 11.9% for gag, 9.7% for red snapper, 0.2% for vermilion snapper, and 8.7 % for mangrove snapper. 6) Most fish showed limited movement, but some were recaptured up to 360.3 km (red grouper) and 583.2 km (red snapper) from the original tagging location. 7) Six hundred six fish were double tagged (474 red grouper, 3 gag, and 129 red snapper). Most fish were recaptured with both tags present. Tag shedding rates were calculated as 0.2% for red grouper, 0% gag, and 0% for red snapper.

Circle hooks should be used when fishers target red groupers. Circle hooks do not enhance red snapper survival. Although the effects of barotraumas affect both red grouper and red snapper, red grouper begin to experience difficulties at 27.4 m (90 ft) whereas red snapper trauma occurs closer to 42 m (140 ft). Both species benefit from venting at the appropriate depth.

Abstract submitted to 2008 Annual Meeting of the American Fisheries Society Sea Grant Special Symposium Research, Education, and Management Needs Regarding Barotrauma in Fish for plenary session, Ottawa, Canada, August 21, 2008

## Laboratory and field studies on the effects and survival of red grouper and red snapper following rapid decompression from depth

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Depth-induced mortality resulting from injuries sustained during rapid decompression from depth is an important factor that seriously impacts survival of undersized reef fish returned to the sea in compliance with minimum size regulations. Laboratory and field studies were conducted to compare the effects of barotraumas on red grouper (*Epinephelus morio*) and red snapper (*Lutjanus campechanus*) under various conditions. In the laboratory, necropsies were conducted on acute mortalities from headboats to quantify and evaluate injuries between the two species from fish caught at various depths. Fish hyperbaric chambers were used to compare survival rates and depth related injuries between the two species at simulated depths of 21, 27, 43, and 70 m. Histological studies on swim bladders from 138 red grouper (205-766 mm FL) and 62 red snapper (123-674 mm FL) were performed in conjunction with scientists at the University of Southern Mississippi to investigate differences in swim bladder morphology. Comparisons revealed red grouper have a larger swim bladder in relation to total body size than red snapper. The red grouper swim bladder membrane is thinner and swim bladder ruptures are larger than those of red snapper however rupture healing rate in survivors is identical. Swim bladders heal sufficiently to hold gas within 2 days. Gas resorption and secretion areas are different leading to difference in percent of swim bladder hemorrhaging. Data from laboratory studies were interpreted in light of field results obtained from a 17 year tagging study conducted off both coasts of Florida. Tag recaptures were used as a proxy for survival. Fish were tagged by researchers, recreational, recreational-for-hire, and commercial fishers at depths ranging 6-82 m. Gear types included J and circle hooks, commercial long-lines and commercial reef fish traps. Treatments included venting and not venting tagged fish before release at all depths fished. Fish at shallow depths (less than 10 m) were also treated as a control to determine if venting was in itself harmful to fish due to injury from the venting tool or injection of pathogens. Although venting enhanced survival in both species, morphological and physiological differences between the two species determined the depth at which venting was necessary and the fish's ability to survive rapid decompression.





### RED GROUPER ATLANTIC Linear Regression

$$\text{Col 1} = 19.585 + (0.323 * \text{Col 2})$$

N = 332

R = 0.208      Rsqr = 0.0434      Adj Rsqr = 0.0405

Standard Error of Estimate = 11.194

	<b>Coefficient</b>	<b>Std. Error</b>	<b>t</b>	<b>P</b>
Constant	19.585	3.469	5.645	<0.001
Col 2	0.323	0.0835	3.868	<0.001

Analysis of Variance:

	<b>DF</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>P</b>
Regression	1	1874.935	1874.935	14.963	<0.001
Residual	330	41351.601	125.308		
Total	331	43226.536	130.594		

Normality Test: Failed (P = <0.001)

Constant Variance Test: Failed (P = <0.001)

### RED GROUPER GULF Linear Regression

$$\text{Col 1} = -18.275 + (1.457 * \text{Col 2})$$

N = 15985

R = 0.407      Rsqr = 0.165      Adj Rsqr = 0.165

Standard Error of Estimate = 25.890

	<b>Coefficient</b>	<b>Std. Error</b>	<b>t</b>	<b>P</b>
Constant	-18.275	0.966	-18.925	<0.001
Col 2	1.457	0.0259	56.270	<0.001

Analysis of Variance:

	<b>DF</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>P</b>
Regression	1	2122324.057	2122324.057	3166.297	<0.001
Residual	15983	10713177.005	670.286		
Total	15984	12835501.063	803.022		

Normality Test: Failed (P = <0.001)

Constant Variance Test: Failed (P = <0.001)

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### RED SNAPPER ATLANTIC Linear Regression

$$\text{Col 1} = 10.517 + (0.467 * \text{Col 2})$$

N = 4211

R = 0.246      Rsqr = 0.0604      Adj Rsqr = 0.0602

Standard Error of Estimate = 12.281

	<b>Coefficient</b>	<b>Std. Error</b>	<b>t</b>	<b>P</b>
Constant	10.517	1.117	9.417	<0.001
Col 2	0.467	0.0284	16.450	<0.001

Analysis of Variance:

	<b>DF</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>P</b>
Regression	1	40813.251	40813.251	270.595	<0.001
Residual	4209	634834.045	150.828		
Total	4210	675647.296	160.486		

Normality Test: Failed (P = <0.001)

Constant Variance Test: Passed (P = 0.197)

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### RED SNAPPER GULF Linear Regression

$$\text{Col 1} = 9.583 + (0.387 * \text{Col 2})$$

N = 4920      Missing Observations = 13

R = 0.123      Rsqr = 0.0152      Adj Rsqr = 0.0150

Standard Error of Estimate = 16.832

	<b>Coefficient</b>	<b>Std. Error</b>	<b>t</b>	<b>P</b>
Constant	9.583	1.480	6.475	<0.001
Col 2	0.387	0.0444	8.709	<0.001

Analysis of Variance:

	<b>DF</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>P</b>
Regression	1	21490.236	21490.236	75.855	<0.001
Residual	4918	1393301.891	283.307		
Total	4919	1414792.127	287.618		

Normality Test: Failed (P = <0.001)

Constant Variance Test: Failed (P = <0.001)

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### GAG ATLANTIC Linear Regression

$$y = 16.006 + (0.534 * x)$$

N = 1134

R = 0.146      Rsqr = 0.0215      Adj Rsqr = 0.0206

Standard Error of Estimate = 11.605

	<b>Coefficient</b>	<b>Std. Error</b>	<b>t</b>	<b>P</b>
Constant	16.006	1.955	8.187	<0.001
x	0.534	0.107	4.982	<0.001

Analysis of Variance:

	<b>DF</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>P</b>
Regression	1	3342.566	3342.566	24.820	<0.001
Residual	1132	152447.777	134.671		
Total	1133	155790.343	137.503		

Normality Test: Failed (P = <0.001)

Constant Variance Test: Failed (P = 0.012)

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### GAG GULF Linear Regression

$$y = -18.656 + (2.672 * x)$$

N = 4936

R = 0.458      Rsqr = 0.209      Adj Rsqr = 0.209

Standard Error of Estimate = 17.976

	<b>Coefficient</b>	<b>Std. Error</b>	<b>t</b>	<b>P</b>
Constant	-18.656	1.217	-15.331	<0.001
x	2.672	0.0739	36.141	<0.001

Analysis of Variance:

	<b>DF</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>P</b>
Regression	1	422093.880	422093.880	1306.181	<0.001
Residual	4934	1594427.402	323.151		
Total	4935	2016521.282	408.616		

Normality Test: Failed (P = <0.001)

Constant Variance Test: Failed (P = <0.001)

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