

Monitoring of the Gonad of Rabbitfish *Siganus guttatus* in an Oil Spill -Affected and Unaffected Area

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Abstract

Gonad development of rabbitfish *Siganus guttatus* from an oil spill affected (Taklong Island) and unaffected (Ave Maria Sanctuary) area in Guimaras province were monitored from November 2007 to December 2008. There were instances wherein samples could not be collected in both sites at the same period since tidal change was not abrupt so that the fish did not migrate to the seagrass bed where they fed and eventually get caught in the barrier net. Male rabbitfishes sampled from both area had 23-400 g body weight (BW) and 6.15-29.00 cm total length (TL) while females had 25-753 g BW and 6.50-31 cm TL. The fish were dissected and gonads were preserved in Bouin's solution. The left lobe of the gonad was used for histological procedures. Due to limitations in measuring the gonad weight in the site, the gonads were weighed after being preserved for several months. The range of the gonadosomatic index (GSI) of the males in Taklong Is. was 0.02-8.39 and 0.04-3.25 in the Ave Maria Sanctuary. Male GSI in Taklong was lower in May and June, higher from July to November but the same in December. For the females, GSI range was 0.1-7.78 in Taklong Is. and 0.03-1.68 in Ave Maria Sanctuary. Female GSI in Taklong was lower in June and December, the same in July but was higher in August and November. Results in the GSI did not seem to show a clear difference between the oil spill affected and unaffected area. A comparison of the maturity stages of the testes and ovaries of sampled fish from both areas also showed no distinct difference between the oil spill affected and unaffected area. These results indicate that more than a year after the oil spill incident, gonad development of *S. guttatus* were already not affected.

Motor Tanker (MT) Solar I carrying 2 million liters of oil in unusually rough waters sank last 9 August 2006, about 24km southwest of the Province of Guimaras, central Philippines. It is known that the Province of Guimaras is considered a tourist area having pristine white sand beaches, diverse marine sanctuaries and unspoiled coral reefs and mangrove forest. Oil slick affected nearly 200 km of coastline covering 10 barangays in the municipality of Nueva Valencia.¹⁾

The Solar I Oil spill affected very much the resources of the Guimaras islands particularly those sensitive to oil pollution. Some of these were the coral reefs and seagrass beds which are sometimes exposed from water during low tide. Oil contamination may have lethal or sub-lethal effects on the flora and fauna and could cause alteration of the natural habitats. To minimize the effect of oil covering the coastal area including mangrove forests in the affected sites of Guimaras, dispersant was applied. The marine ecosystem is a complex environment

and is susceptible to sudden changes. Oil slicks drift towards the shore into nearshore niches and estuaries that serve as an important spawning and nursing grounds of many aquatic species. At present, there have reports that dispersed oil elicits higher toxicity to marine organisms than oil or dispersants alone.^{2,3)}

A review on the biology of rabbitfish or siganid showed that juveniles and adults occupy very shallow water habitats including coral reefs, sandy and rocky bottoms with or without vegetation, lagoons and river mouths and mangrove swamps.³⁾ These species are primarily herbivores. Seagrass beds, which is one of their feeding grounds was very much affected by the oil and dispersed oil during the Solar I Oil spill incident. Thus, it is of ecological importance to know if such exposure to mixture of oil, seawater, and dispersant to a complex multi-phase system can affect aquatic environment and the reproductive biology of some marine species.

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In this study, the gonad development of rabbitfish *Siganus guttatus* (Bloch) caught from an oil spill affected area were monitored in comparison with those in the unaffected area in Guimaras province (more than a year after the incident).

Materials and Methods

Experimental Fish

Rabbitfish *S. guttatus* were collected from November 2007 to December 2008 in the oil spill affected area (Taklong Island, Nueva Valencia) and unaffected area (Ave Ma. Sanctuary, Jordan) at Guimaras province (Fig. 1). Barrier net was used to collect samples by surrounding the nearshore area of the two sampling sites. The period of collection was based on the highest high tide (approx. 0.3 – 0.5 m) and lowest low tide (approx. -0.1 – -0.3 m) of the month. However, there were instances wherein samples were not collected in the two sites at the same period. The rabbitfish were anaesthetized using 100 ppm 2-phenoxy ethanol. Sex was determined by gently pressing the abdomen if milt will be released. If no milt is released,

the fish was cannulated using a Clay Adams PE 60 cannula. Then the total length (TL) and bodyweight (BW) of the fish were measured and then dissected to get the gonads.

Histological Procedures

Gonads were preserved in Bouin's solution. The left lobe was used for histological procedure, while the right lobe was used to measure the gonad weight (GW) after several months in the solution. The Gonadosomatic Index (GSI) was calculated as: $GSI = (\text{gonad weight/body weight}) \times 100$. Gonads were dehydrated in alcohol series and embedded in paraffin. Representative 5- μm cross sections were stained with Delafield's hematoxylin and eosin and observed under a light microscope.

Statistical Analysis

The GSI of fish from the affected and unaffected area were compared when possible using Student's t-test to determine their statistical difference.

Results

The range of BW of fish collected were 23 – 400 g for the males and 25 – 753 g for the females while the TL were 6.15 – 29.00 cm and 6.5 – 31 cm, respectively. Figure 2 shows the GSI of male and female siganids collected from Taklong Is. and Ave Maria Sanctuary. The range of the GSI of the males in Taklong Is. was 0.02 – 8.39 and 0.04 – 3.25 in the Ave Maria. Male GSI in Taklong was lower in May and June, higher from July to November but the same in December. Only the GSI of the males collected in July 2008 collected from Taklong Is. and Ave Maria were compared which showed significant difference ($P < 0.05$). For the females, GSI range was 0.1 – 7.78 in Taklong Is. and 0.03–1.68 in Ave Maria Sanctuary. Female GSI in Taklong Is. was lower in June and December, the same in July but was higher in August and November. Statistical comparison was made in the GSI of females collected from the two sites in June, July, and December 2008 which showed no significant difference between the two sites ($P > 0.05$). Results in the GSI did not seem to show a clear differences between the oil spill affected and unaffected area in both male and female.

Photomicrographs of representative gonad sections from male and female rabbitfish collected from the two sites are shown in Fig. 3. In general, we observed that the maturity stages of the testes and ovaries of sampled fish from both ar-

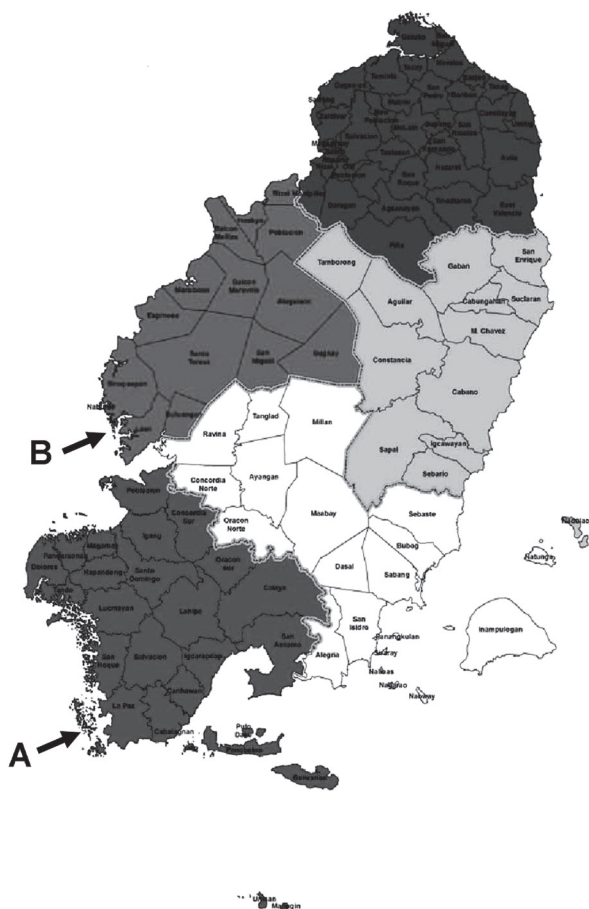


Fig. 1. Map showing the sampling sites of the oil spill affected area at Taklong Island (A) and unaffected at Ave Maria Sanctuary area (B) in Guimaras Province.

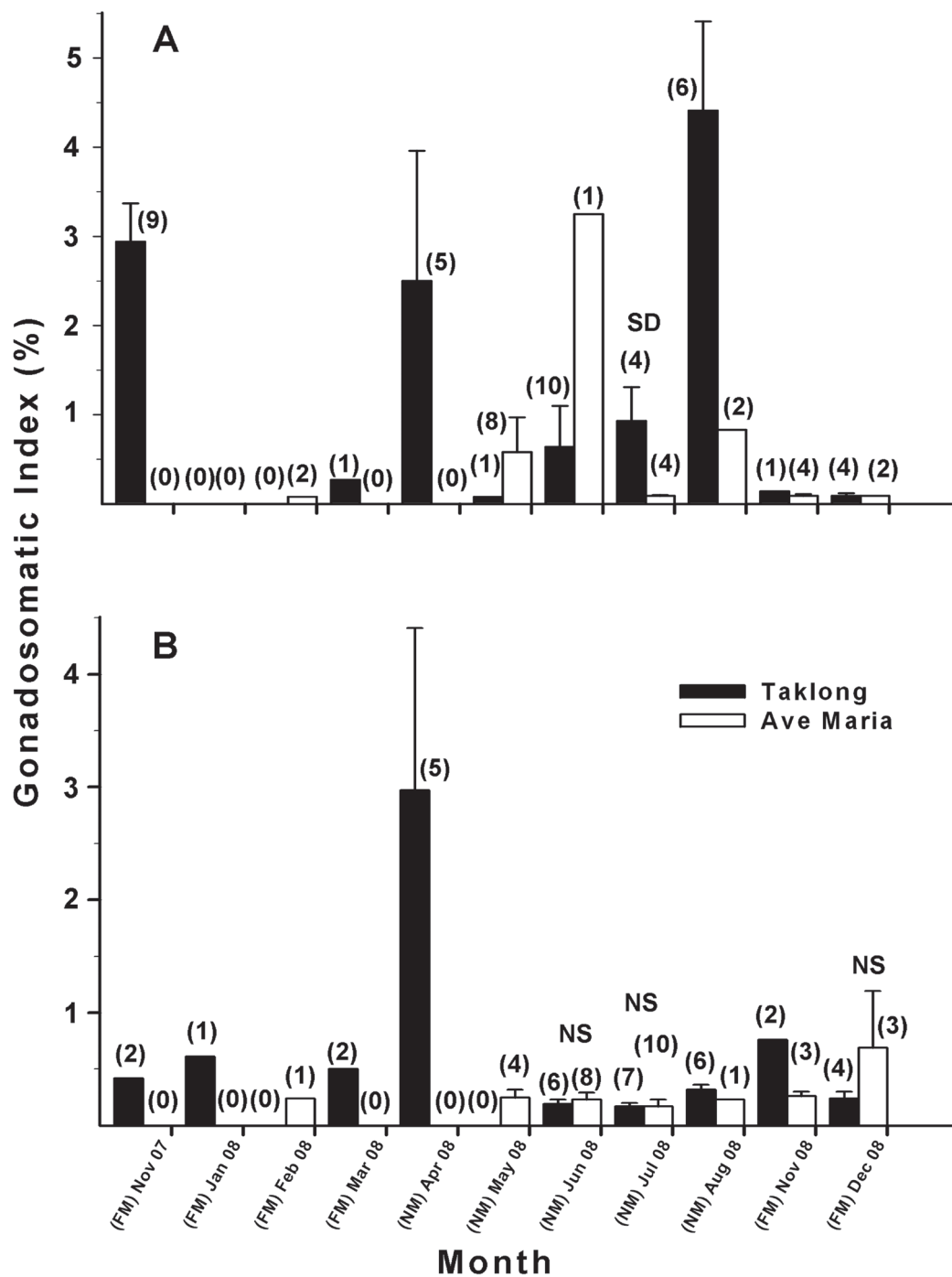


Fig. 2. Gonadosomatic index of male (A) and female (B) *Siganus guttatus* sampled from the oil spill affected and unaffected areas in Guimaras Province. Line above a bar indicates standard error of the mean. Numbers in parenthesis indicate number of samples collected. SD above the paired bars indicate significantly different. NS, above the paired bars indicate no significant difference. FM, full moon phase. NM, new moon phase.

as also showed no distinct difference between the oil spill affected and unaffected area. Gonad samples from the males collected from the period November to March which was towards the full moon phase showed that rabbitfishes were immature and early maturing. The testes contained few residual spermatozoa and germ cells at different stages of spermatogenesis.

The ovary of the females contained numerous oogenesis. From period April to August sampling was during the new moon phase, late maturing gonads were observed. The lumen of the testes were filled with spermatozoa, but some still contained developing germ cells in the lobules. Majority of the female gonads contained oocytes at the tertiary yolk stage

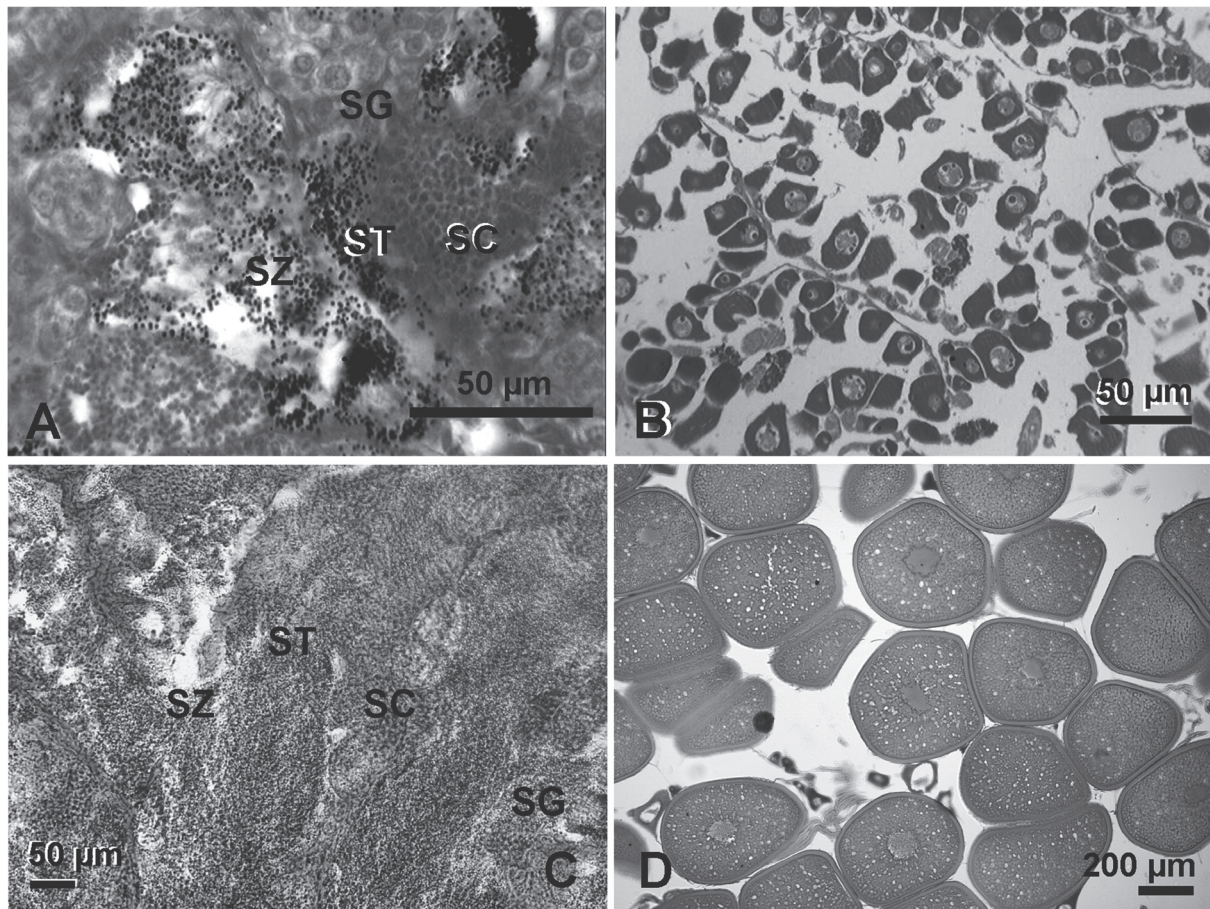


Fig. 3. Cross sections of male and female *Siganus guttatus* sampled from Guimaras Province. A. Maturing male gonad sampled 20 February 2008 (full moon phase) from the oil spill unaffected area (Ave Maria Sanctuary) showing different germ cells with few spermatozoa in the lumen. B. Immature female gonad sampled 25 November 2007 (full moon phase) from the oil spill affected area (Taklong Island) showing abundance of oogonia. C. Late maturing male gonad sampled 4 July 2008 (new moon phase) from the oil spill unaffected area (Ave Maria Sanctuary) showing abundance of different germ cells but few spermatogonia. D. Late maturing female gonad sampled 8 April 2008 (new moon phase) from the oil spill unaffected area (Ave Maria Sanctuary) showing mostly advance stages of oocytes. SC, spermatocyte. SG, spermatogonia. ST, spermatid. SZ, spermatozoa.

and final oocyte maturation. Moreover, there were no deformities or abnormalities observed in the gonads of rabbitfish from the oil spill affected area.

Discussion

Results our study showed that there were no difference in the GSI and histological observations of the gonads of male and female rabbitfish collected from oil spill affected and unaffected area from November 2007 to December 2008. The gonads in both areas were immature and early maturing stage during the full moon phase indicating that the germ cells are developing for the next spawning period. During the new moon phase, the gonads are in the late maturing stage and are ready to spawn sometime around the first quarter phase.⁵⁾ Other species of rabbitfishes also show lunar periodicity in their gonadal maturation similar to our observations.⁶⁻⁹⁾ These

indicates that gonad maturity of the rabbitfishes were already not affected by the oil or dispersed oil more than a year after the oil spill. The rabbitfishes in the oil spill affected area already showed normal gonad maturation activity probably due to fact that the seagrass beds which their feeding ground may have fully recovered from the effects of the oil spill and the dispersed oil. Green and chara algae have been shown to be able to detoxify carcinogen from polycyclic aromatic hydrocarbons (PAHs) while brown algae are able to bioaccumulate large quantities of carcinogen which may have a serious effect on the environment.⁶⁾

It seems that further studies on the effect of exposure to dispersed oil on gonad maturation and spawning of rabbitfish has to be undertaken. At present, we are conducting an experiment on the effect of exposure to dispersed bunker oil on the spawning of rabbitfish.

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