

## ASPECTS OF LEOPARD CORAL GROUPEL (*Plectropomus leopardus*) REPRODUCTION IN INDONESIA

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### ABSTRACT

Leopard coral grouper, *Plectropomus leopardus* is one of the most economically important finfish fish in Indonesia and the demand for the grouper is rapidly increasing in Asia and the Pacific. Grouper exports from Bali were 1,613 mt in 2001, 2,082 mt in 2002 and 2,861 mt in 2003. Understanding the reproductive biology of fishes is an important component in developing mariculture and in the management of capture fisheries. This study reports on the reproductive biology of 86 coral groupers collected from various locations in Indonesia. The length and weight of these fish were recorded and related to gonad development. There was a strong relationship between length and weight; weight being proportional to the length raised to the power (b value) 3.2. As the value of b was greater than 3, this suggests that growth is allometric. Histological analysis 73% of the fish were immature, 19% were in transition from females to males, only 4% were male, and only 2 fish (2%) had mature gonads: these were female. The sex of 2 fish could not be determined. From these data it can be seen that the leopard coral grouper has asynchronous gonad development. The two fish that were mature contained 343,980 and 429,259 oocytes and three distinct sizes of oocytes could be found. This suggests that the grouper is a multiple spawner. If fish are required for brood stock, this study has shown that only those with a length greater than 35 cm in standard length should be taken from the wild.

**KEYWORDS:** reproductive biology, fecundity, grouper

### INTRODUCTION

Leopard coral grouper (*Plectropomus leopardus*) is one of the most economically important finfish fisheries in Indonesia. Understanding the reproductive aspects of fishes is an important component in developing mariculture and in the management of capture fisheries. The demand for grouper is rapidly increasing in Asia and the Pacific. Grouper are marine species that have a wide market and achieve a relatively high price particularly when sold internationally in countries such as Singapore, Hongkong, Taiwan, and Southern China (Chou & Lee, 1998 in Rimmer *et al.*, 2004). The culture of grouper has not yet been developed successfully and the majority of grouper trade is currently based on wild-caught

fish. According to the Laboratory of Quality Control of Fisheries Product, the export of grouper from Bali Province was 1,613,639 kg in 2001; 2,082,276 kg in 2002; and 2,861,121 kg in 2003, respectively.

Culture of coral grouper still relies on a supply of juveniles from the wild, whereas their existence is dependent on season. There is an anticipated continuing need for juvenile with the increasing demand of grouper for breeding in aquaculture. To support these mariculture activities we identified a need for investigation of the reproductive biology of the grouper and this study was directed at examining certain reproductive aspects: the relationship of reproductive parameters to length and weight, the stages of gonad

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maturation, and the gonad somatic index (GSI) of wild-caught leopard coral grouper (*P. leopardus*).

**MATERIALS AND METHODS**

**Field sampling.** *Plectropomus leopardus* (Figure 1) were collected from Bawean Island, Karimun Jawa Archipelago, Sirai Island (Riau Archipelago) and Ujung Kulon (Sunda Strait) (Figure 2) from April to October 2003. All fish were measured (standard length in mm) and weighed (g). Gonads were removed and preserved in 4% formaldehyde for routine histology (Luna, 1968).

**Laboratory study.** Ovaries were thawed, blotted dry and weighed ( $\pm 0.001$  g). Sub-samples were taken and preserved in 70% ethanol for subsequent histological preparation. For histological processing each ovary was dehydrated in ethanol, cleared in xylene, embedded in paraffin, sectioned to 6  $\mu$ m and stained with Harris's haematoxylen and eosin (Luna, 1968). Histological examination was defined following the stages of Hunter &

Goldberg (1980); Cyrus & Blaber (1984); Andamari *et al.* (1998). Ovaries were classified into one of the following six developmental stages, and the presence of post ovulatory follicles was noted.

- Stage I Immature ovary with unyolked oocytes
- Stage II Developing/resting (pre-vitelogenic oocytes)
- Stage III Maturing (yolk precursor stage; some non staining yolk)
- Stage IV Ripe (non staining yolk; developed chorion)
- Stage V Running ripe (homogeneous red-staining yolk; oocytes hydrated; development complete)
- Stage VI Spent (atresion of ripe oocytes plus pre-vitelogenic oocytes; presence of post-vitelogenic follicles)

Fish that had ripe egg were used to estimate batch fecundity following the approach of Bagenal (1978) and Effendie (1997).

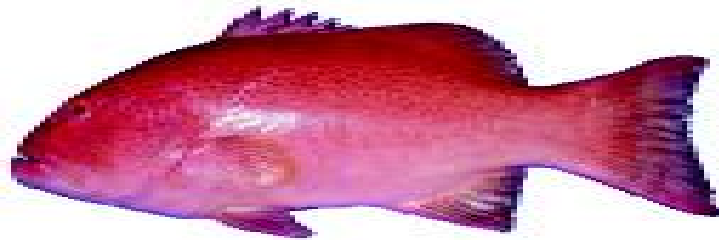


Figure 1. Morphology of leopard coral grouper (*P. leopardus*) from Karimun Jawa

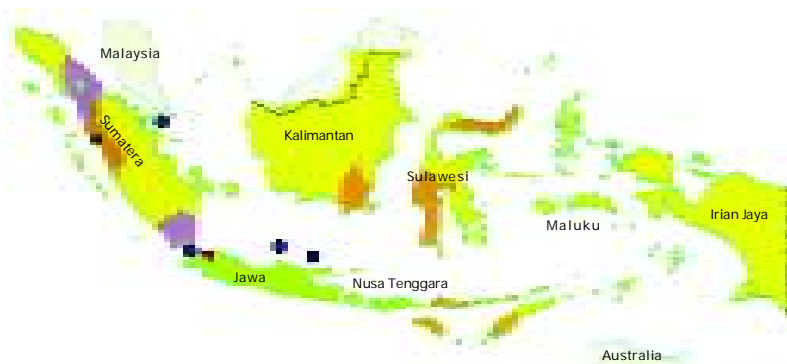


Figure 2. Map of the sampling collection of *P. leopardus* in Bawean Island, Karimun Jawa Archipelago, Sirai Is. (Riau archipelago) and Ujung Kulon (Sunda Strait) sampling location in 2003

$$F = (Wg/Ws)n$$

Where: F = batch fecundity  
Wg = gonad weight (g)  
Ws = gonad weight in subsample (g)  
N = number of eggs in subsample

Gonadosomatic index of the ovaries was determined by the relationship (Nikolsky, 1969):

$$GSI = Wg/W \times 100\%$$

Where: GSI = gonadosomatic index (%)  
Wg = gonad weight (g)  
W = weight (g)

A length-weight relationship was determined for the fish based on the relationship (Royce, 1984):

$$W = aL^b$$

Where: W = weight (g)  
L = length (mm)  
a,b = constant

## RESULTS AND DISCUSSIONS

**Length-weight relationship.** The results are based on the collection of 86 *P. leopardus*.

A detail description of the length and weight of the fish by collection region can be seen in Tables 1 and 2. The length-weight relationship of *P. leopardus* is shown in Figure 3. The value b appears to not be equal to 3 (based on t-test), so it can be concluded that coral grouper exhibit a positive allometric growth pattern (b = 3.2) i.e. the growth in length is not directly proportional to growth in weight.

**Gonadosomatic index.** This index was used to determine the gonad maturation stage where the gonads could not be investigated by histology (Nikolsky, 1969). The results of four monitoring locations can be seen in Figures 4 and 5. It appears that *P. leopardus* reach gonad maturity when the GSI > 1.5%. However, there is not a direct relationship between GSI and fish length or between GSI and fish weight. This means that the longer fish or heavier fish do not necessarily have a higher GSI. The stage of gonad maturity is possibly more directly linked to timing of spawning season.

**Gonad Maturity Stage and Fecundity.** Sex and stage of gonad maturity was determined for all the sampled leopard coral

Table 1. Mean and range of *P. leopardus* standard length (mm) sampled from four locations

Locations	Month (2003)	n	Standard length (mm)		
			Min	Max	Mean ± SE
Bawean Is.	April	4	285	473	381.8 ± 39.31
Sirai Is. (Riau)	May	30	194	393	261.5 ± 8.70
Karimun Jawa Arc.	July	31	271	432	345.35 ± 9.50
Ujung Kulon	October	21	163	355	252 ± 8.68
<b>Total</b>		<b>86</b>	<b>163</b>	<b>473</b>	<b>295 ± 7.22</b>

Table 2. Weight (g) mean and range of *P. leopardus* sampled from four locations

Locations	Month (2003)	n	Weight (g)		
			Min	Max	Mean ± SE
Bawean Is.	April	4	626	2,900	1,912.8 ± 486.53
Sirai Is. (Riau)	May	30	166	1,743	487.6 ± 60.34
Karimun Jawa Arc.	July	31	540	2,606	1,312.31 ± 114.62
Ujung Kulon	October	21	214	1,405	542.20 ± 66.44
<b>Total</b>		<b>86</b>	<b>166</b>	<b>2,900</b>	<b>864.51 ± 71.27</b>

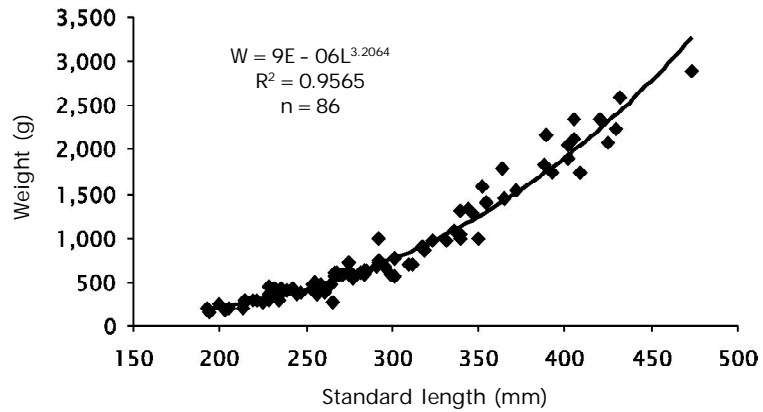


Figure 3. Length-weight relationship of *P. leopardus* collected from 4 locations

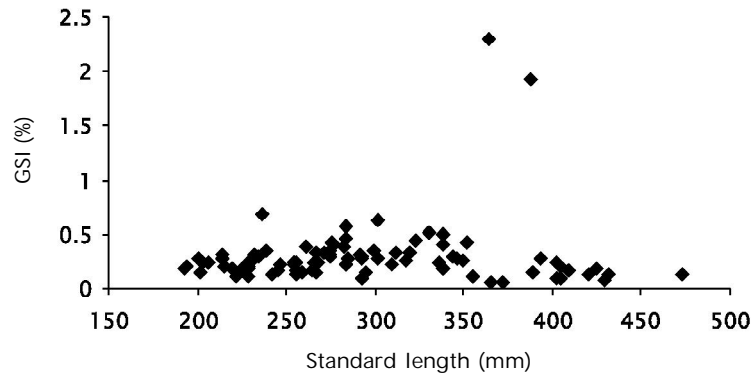


Figure 4. Gonadosomatic Index (GSI) and body length relationship of *P. leopardus* from four locations (n=86)

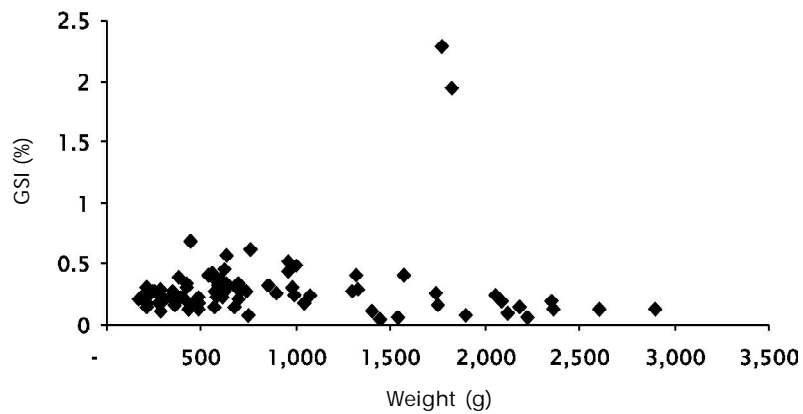


Figure 5. Gonadosomatic Index (GSI) and body weight relationship of *P. leopardus* from four locations (n=86)

grouper and results are detailed in Table 3. Of the 86 fish, sex could not be determined for 2 fish (2%), 63 (73%) were immature (Figure 6A), 16 (19%) were in transition from female to male (Figure 6B), only 3 fish (4%) were males, and only 2 fish (2%) with mature gonads at stage V or higher (Figure 6C).The smallest

female with immature gonads had standard length of 20 cm and body weight of 250 g, while the smallest in transition phase from female to male was 32.3 cm SL and weighted 962.5 g. This study was unable to determine length at first maturity because of the influence of seasonality and the number of

Table 3. Gonad maturity stage of *P. leopardus* sampled at several locations

Location	Month (2003)	Sex	N	Gonad maturity stage					
				I	II	III	IV	V	VI
Bawean Is.	April	Male	2	0	0	0	2	0	0
		Female	2	1	0	0	0	1	0
Sirai Is.	May	Female	30	30	0	0	0	0	0
Karimun Jawa Arc.	July	Male	1	0	0	0	1	0	0
		Transition	15	0	0	0	0	0	0
		Female	15	14	0	0	0	1	0
Sunda Strait	October	Unidentified	2	0	0	0	0	0	0
		Transition	1	0	0	0	0	0	0
		Female	18	18	0	0	0	0	0

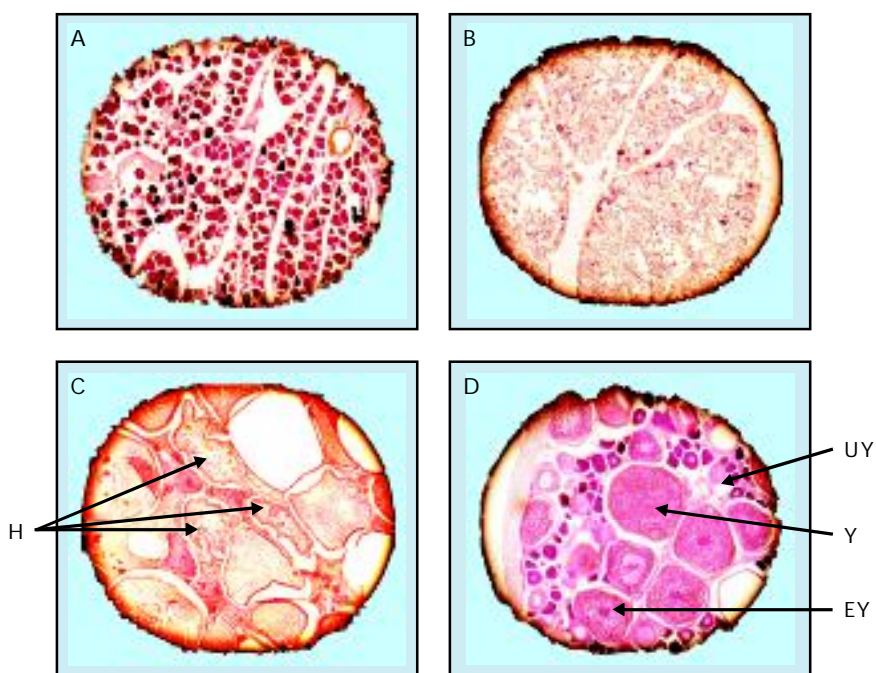


Figure 6. Developmental stage of oocytes in *P. leopardus* ovaries. (A) immature ovary, (B) transition, (C) prespawning ovary with hydrated oocyte, (D) asynchronous oocyte, UY (unyolked), EY (early yolked), and Y (yolked). H&E 100X

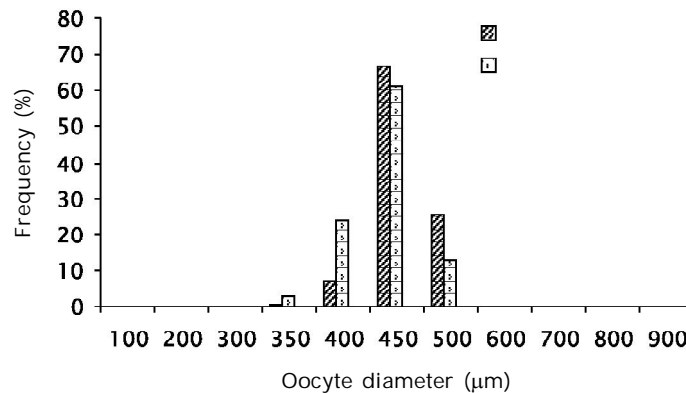


Figure 7. Size-frequency distribution of *P. leopardus* oocyte diameter (by 50 µm intervals) in mature gonad stages

sample was very small. According to Goeden (1978) in Heemstra & Randall (1993), the spawning season of leopard coral grouper occurs at the end of November – beginning of December.

*P. leopardus* are protogynous hermaphrodites i.e. they have the ability to change sex from female to male. The length of the smallest mature male was 30 cm SL (age 3 years) while the smallest mature female was 21 cm SL (age 2 years) and the largest 47 cm SL (4 years). Two of the mature females in this study, of 38.8 cm SL and 36.4 cm SL had batch fecundity of 343,980 and 429, 259 oocytes respectively, and a relative fecundity of 241.8 and 189.0 oocytes/grams body weight respectively. By comparison Goeden (1978) in Heemstra & Randall (1993) estimated the fecundity of 4 year old *P. leopardus* to be 457,900 oocytes. Sampled *P. leopardus* showed asynchronous gonad development i.e. in a single gonad several stages of development were observed, meaning that in one spawning season *P. leopardus* are able to spawn several times (as shown in Figure 6D). The diameter of oocytes ranged from 425–475 µm for mature gonad (stage V) but with oocytes not fully hydrated. By comparison, in Gondol Research Institute reported the diameter of *P. leopardus* eggs in plankton to be 700 to 800 µm.

#### CONCLUSIONS

Of 86 *P. leopardus* sampled from 4 locations in the wild only 2 fish had mature gonads. Examination of batch fecundity in these gonads revealed totals of 429,259 and 343,980

oocytes. Leopard coral grouper are protogynous hermaphrodites and exhibit asynchronous gonad development (i.e. multiple spawners).

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