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The development of gonad mantis shrimp *Harpiosquilla raphidea* Fabricius, 1798 in Banten Bay, Indonesia

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Abstract: The *Harpiosquilla raphidea* mantis shrimp in Banten Bay is very important and an abundant marine crustacean in Indonesia. It is common among the most important predators in many shallow, tropical and subtropical marine habitats. Gonad maturity level of mantis shrimp *Harpiosquilla raphidea* was identified in four phases of development. The first size mature gonad of the female mantis shrimp is 199 mm and the male is 230 mm. The gonad maturity level of the female shrimp in II, III and IV have fecundity ranging from 24.600 to 76.809,with an average of 54.082±13.456 where as the diameter of the eggs at gonad maturity level III is 510–675 µm, with an average diameter of 574±6.98 µm.

Keywords: Banten Bay, Mantis shrimp, Harpiosquilla raphidea, gonad maturity, Indonesia

Introduction

The *Harpiosquilla raphidea* mantis shrimp is an order Stomatopoda shrimp living in subtidal areas between 2-43 m with mud substrate or sandy mud (Moosa, 1975). The population of the *H. raphidea* mantis shrimp is found in several waters in the North Java Sea, such as in Banten Way Waters.

The study of the shrimp's reproduction requires a knowledge of the gonad's development on the individual shrimp. The method normally used is based on the morphological structure. The visual method may be faster but proved to be less accurate. The histological method can be used to obtain detailed analysis concerning the development of *oosit* and *spermatosit* (Wortham-Neal, 2002; Kodama *et al*., 2006).

Based on the importance of population biological data for the conservation of the mantis shrimp (*H. raphidea)* in Banten Bay waters, a reproductive aspects is needed. In general, study of the reproductive aspects of mantis shrimp (*H. raphidea)* has very rarely been reported. Indonesia, especially in Banten Bay waters, study of the reproductive biology of the mantis shrimp has not been conducted. The result of this study is expected to be benefitin formulating the first steps in the management of the mantis shrimp (*H. raphidea)* to be sustainable, both ecologically and economically, particularly in Banten Bay waters, and in Indonesia waters in general.

Material and Method

Location of Study

The study was conducted from December 2011 to February 2012 with the sample location of mantis shrimp's population in Banten Bay waters in the Province of Banten. The analysis of the reproductive biology of the structure of gonads histology and morphology was conducted in the Fish Health Laboratory, Sekolah Tinggi Perikanan Jakarta (Jakarta Fisheries University) Fisheries Health, Jakarta Fish Quarantine Agency.

Material of Study

The sample of mantis shrimps used as material for the reproductive biological study from Banten Bay waters from December 2011 to December 2012. The sample mantis shrimps were measured by length and weight, determination of sex was determined using the Wortham-Neal (2002) method. The tools used were bottom gillnet pulled by motorboat, GPS, multi checker for water quality, plastic bags, mica plastic,

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rubber bracelet, cool box, digital balance, ruler and digital camera. The tools used for making the histological network were dissecting set, tissue processor, microtome, microscope and water bath.

On the female mantis shrimp, we measured egg diameter and number of eggs. We used female mantis shrimp sample on gonad maturating level III. The egg sample taken was from the *anterior*, centre and *posterior* part. Determination of spawning type can be made by the dissemination graphic of the mantis shrimp's egg diameter with the mature gonad. The gonad histology was performed following Muladno (2006) and Wibowo (2011).

First size prediction of the mature gonad was based on Sperman-Karber method (King, 1995). The criterion for a mature gonad maturating level III, and the formula used was:

$$LogM = X_k + \frac{\bar{x}}{2} - (\bar{x}\sum Pi)$$

X_k = Medium score of logarithm at the time when gonad is 100% ripe

$\bar{x}$ =The average difference between the logarithm of the mean value

Pi = ri/ni

ri = Number of mature gonad shrimp on first class

ni = Total number of shrimps

The type of reproduction was determined based on the distribution of egg diameter according to its distribution modus.

Result

Sex Ratio

Mantis shrimp caught during the study numbered 332, consisting of 146 males and 186 females depends on difference of behaviour and the fishing factor. A Chi square test was conducted to support the result.

The Development of Gonad

The histological and morphological structure of the gonad was used to identify the level of gonad maturity of males and females. The morphological observation was based on colour, size and gonad volume, while the histological observation was based on histological structure. The development of the gonad is in four phases: gonad maturity level I (immature), gonad maturity level II (premature), gonad maturity level III (mature) and gonad maturity level IV (post-mature) following Vila *et al*., (2013) and Wortham-Neal (2002).

The morphological structure of the level 1 mantis shrimp resembles a thin thread extended along the back of the carapace to the rear of the abdomen, and is tinted translucent or transparent. The larger level II male gonads were seen clearly between the border intestinal organ, had a broken white colour (RAL 1013) and the length of gonad was not extended up to the telson (Table. 1). Level III gonads were bigger, almost filling the inner abdomen, and had creamy white colour (RAL 1015). Level IV was post-spawning, smaller than level III and were light brown in colour (RAL 1000).

Tab. 1: Assessment criteria of gonad maturity of male mantis shrimp by histology and morphology.

Gonad Maturating Level	Morphology	Histology
I (early development)	Testicle shaped like thread, around the intestinal and testicular tinted light or transparent with RAL Signal white standard, 9003 Fig. 1 (1a).	Testicle has developed and the connective tissue is already visible, tubulus sack has been filled by *Spermatosit.*
II (being developed)	Testicular strands are increasingly visible in the digestive organs and increasingly white colour with RAL *Oyster white* 1013 standard, filling 1/8 abdominal cavity or stomach. Fig.1 (1b).	Testicular is dominated by secondary *spermatosoit* and developed as spermatid.
III (mature)	Testicle fills ¼ abdominal cavity or stomach, testicle has creams colour with RAL *Light ivory* 1015 standard, shaped like branches and the dissemination extends to the telson part. Fig.2 (1c).	Developed Spermatid becomes *spermatozoa* clearly seen filling the seminifer tubulus.
IV (post-mature)	Testicle is wrinkled, pale cream colour with RAL *Green beige* 1000 standard Filling 1/5 abdominal cavity or stomach. Fig. 2 (1d).	On post-maturity, it was seen that the rest of *spermatozoa* has not been taken out.

The morphological structure of the female ovary was like a thin thread extending along the back of the carapace to the rear of the abdomen, tinted translucent or transparent. The level II ovary was bigger and clear on the border of intestinal organ, having a light yellow colour (RAL 1018) and not fully extended to the telson (Fig. 1). The level III gonad was bigger, almost filling the inner abdomen, having ark yellow colour (RAL 1003). Level IV was smaller than level III, with yellow orange colour (RAL 4002) or light orange (Table. 2).

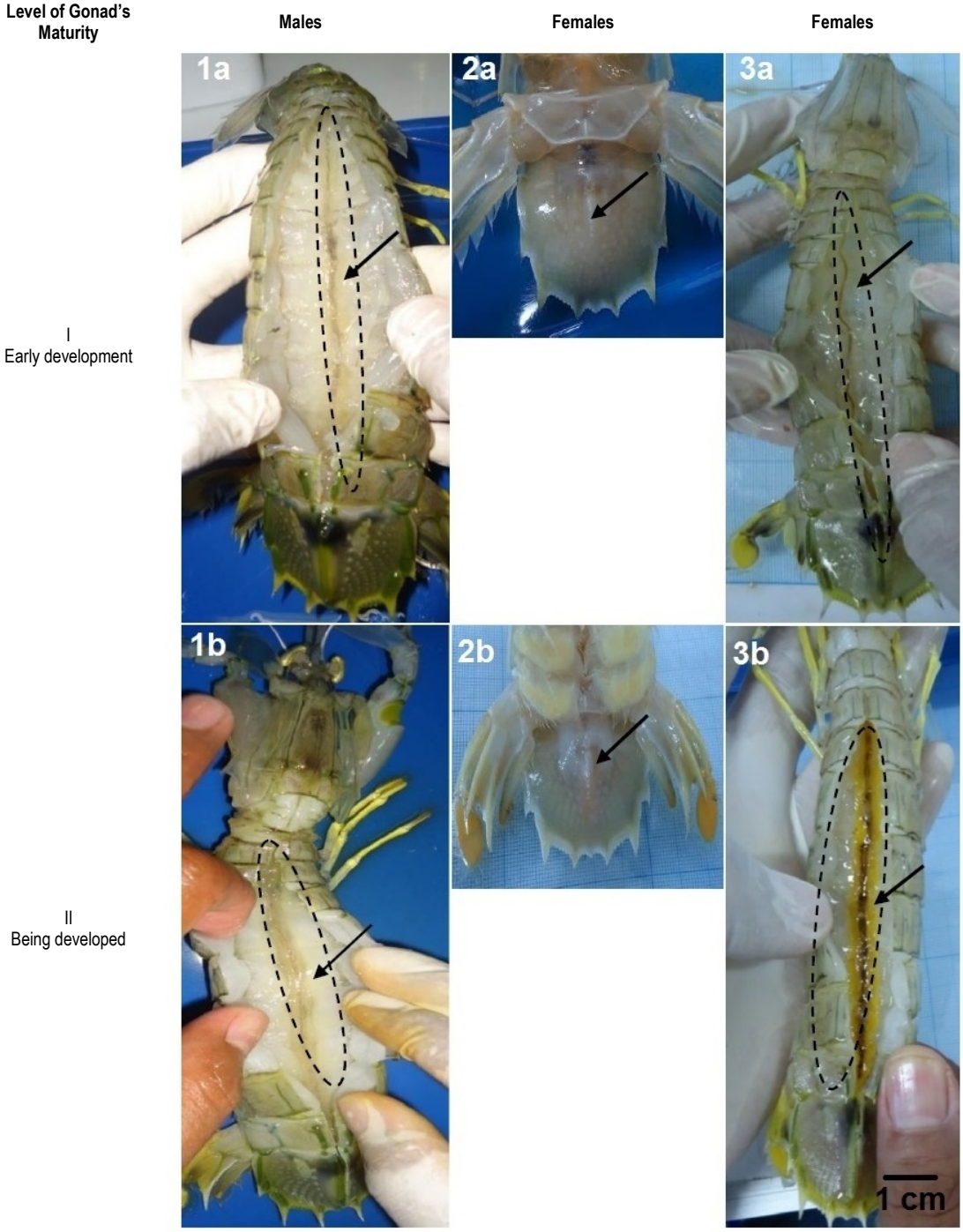

Fig. 1: Gonad's morphological structure and the male and female mantis shrimp at gonad maturating level land level II. The arrow signs show the position of the gonad.1a: gonad maturating level I male on dorsal, 1b : Male of gonad maturating level II on dorsal, 2a: Female gonad maturating level I at telson part, 2b:Female gonad maturating level II on telson, 3a: Female gonad maturating level I on dorsal part, 3b: females on gonad maturating level II on dorsalpart.

Tab. 2: Assessment criteria on gonad maturity of female mantis shrimp by histology and morphology.

Gonad Maturating Level	Morphology	Histology
I (early development)	The ovarium is transparent and light cream and between the digestive organs, but the egg is not visible yet. Figure.1. (3a). On exoskeleton lower part telson the white freckles are seen with RAL *Grey white* 9002 standard, and not the yellow colour is not seen. Fig. 1 (2a).	Cytoplasma is seen to have a clear, purple colour. The cell nucleus is clearly visible.
II (being developed)	Yellow colour with RAL *Zinc yellow* 1018 standard, has thick form and has fills 1/3 of the abdominal cavity and visible from the back end of carapace to the front end of telson. Figure.1. (3b). On the lower part of telson the yellow colour of egg starts to be visible and has been filling the telson. Fig.1 (2b).	Ovarium is dominated with secondary *oosit* that has been developing to become *ootid.*The oosit diameter is around 300–450 µm and the egg yolk is visible on s*itoplasma* with reddish colour.
III (mature)	Ovarium has orange colour with RAL *Signal yellow* 1003 standard, the form of the egg is visible and fills 2/3 of the abdominal cavity or stomach to the inner part of telson. Figure.1.(3c). The lower part of telson has a yellow colour nd clearly shows the egg has filled the telson Fig. 2 (2c).	At this phase, the egg yolk is visible, and the ovarium also shows the nucleus with the *oosit d*iameter around 478–655 µm.
IV (post-mature)	Ovarium fills ¼ of abdominal cavity, the orange colour is visible and becomes red with RAL *Sand yellow* 4002 standard, the remains of egg seen rarely. On the lower part of the telson is seen the yellow colour from the egg's remains, and the volume is smaller than gonad maturating level III Fig. 2 (2d).	Oosit is around450–600 µm.On this phase, oosit looks wrinkled with different sizes and the ovarium is also irregular.

Morphological characteristics are more pronounced in females than males. The colour and the volume of the lower part of the telson changed as the gonad developed. In level I the lower part of the telson was transparent; however, yellow colouring began to develop in level II from the upper part to the lower part. In level III the yellow colour was more intense than level II and spread out more widely inside the telson. In level IV the yellow colour started to fade compared to level III (Fig. 2).

The results of the analysis based on the histology of gonadal development indicates a change of development and size spermatosid or oosit of gonadal development Level I to IV. Histological structures were described in details in (Fig. 3).

Spawning period

The change in colour of the gonad indicated the peak of spawning period between March to April and September to October.

Discusion

The sex ratio of the mantis shrimp showed that the balance of female and male shrimp can change based on place and time. Sallam (2005) reported the result of study on mantis shrimp *Erugosquilla massavensis* with a sex ratio: 1:1. Studies on *Gonodactylids* demonstrated an increase in male mortality rate due to fighting each time they breed (Caldwell, 1986). The number of males was lower than females during shrimp breeding season in other stomatopoda species (Hamano and Matsuura 1987; Froglia and Giannini, 1989; Dittel, 1991; Wortham-Neal, 2002), *E. Massavensis* (Sallam, 2000; El-Ganainy *et al*., 2004) having a balanced sex ratio. Sex ratio determines the sustainability of species, since the number of female and male mantis shrimp can be influenced by the predator level or fighting among male shrimps (Mashar, 2010).

The level of gonad maturity can be used as the probe status of shrimp's reproduction, age and first size of mature gonad, proportion of stock that are productively mature for understanding the reproductive cycle for one population or species. The level of gonad growth in female shrimp consists of four phases based on gonad's morphological and histological characteristics. There is a close relation between gonad maturity and gonad maturity index. To determine the level of gonad maturity, it is divided into four phases of growth: beginning, starting to mature, mature and post-mature. El-Syarif *et al.,* (2012), on the other hand, classified the level of gonad maturity in six phases, classified in ten phases of gonad growth of the mantis shrimp *Oratosquilla oratoria* in Tokyo Bay. Gonad growth and maturity level reaches a maximum limit when the spawning is about to begin. In this case, the change of gonad maturity level value was close to gonad growth phase. The decrease in maturity size of first gonad of female mantis shrimp *E. massavensis* in Egyptian waters had been attributed

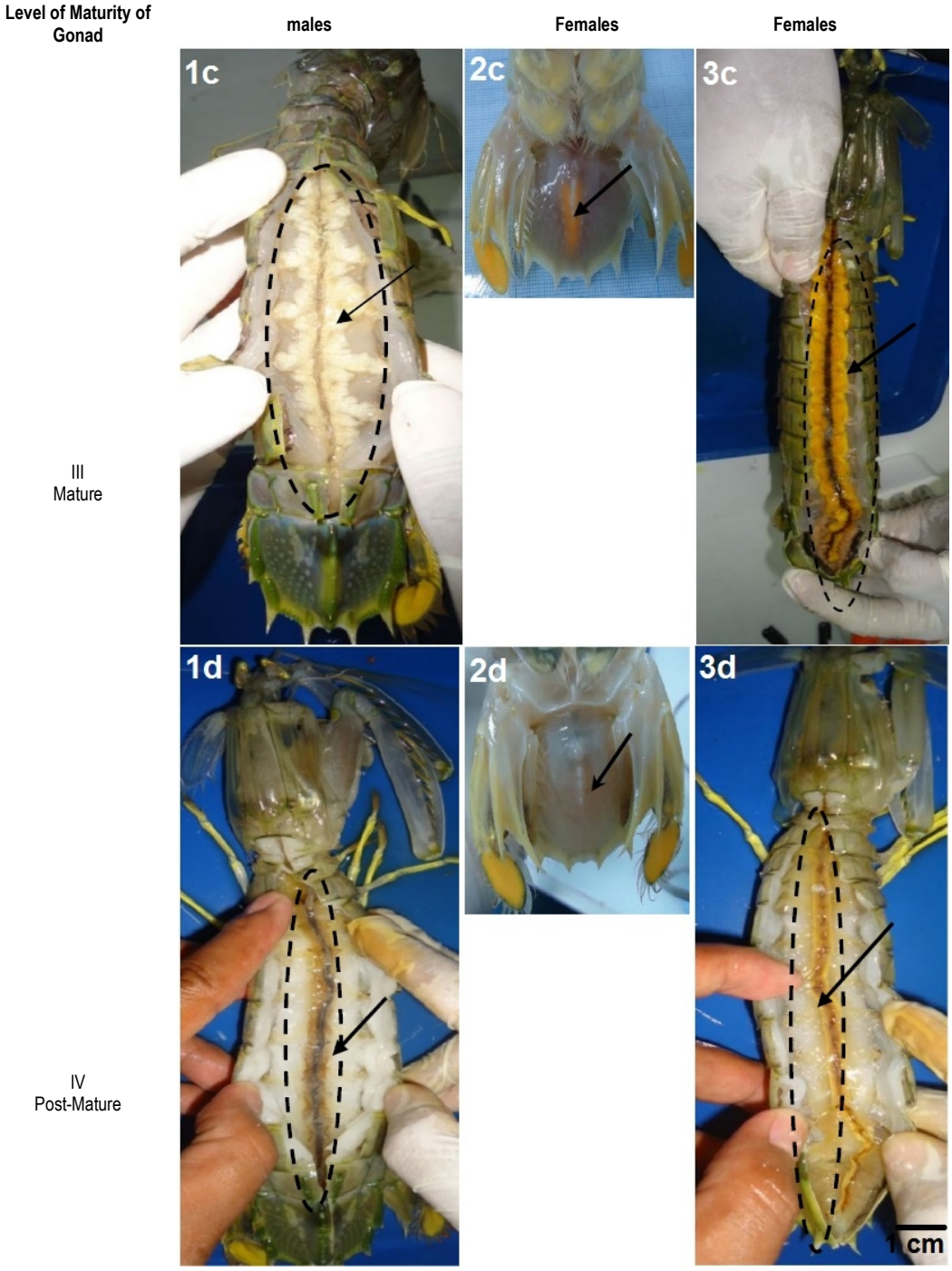

Fig. 2: Gonad's morphological structure of mantis shrimp male and female on gonad maturating level III and level IV. The arrow signs show the gonads on dorsal and the pictures show us the position of the gonad.1c: gonad maturating level III females on dorsal.1d : Male gonad maturating level IV on dorsal part, 2c: Female gonad maturating level III on telson part, 2d: Female gonad maturating level IV on telson part, 3c: Female gonad maturating levelIII dorsal part, 3d: Female gonad maturating level IV on dorsal part.

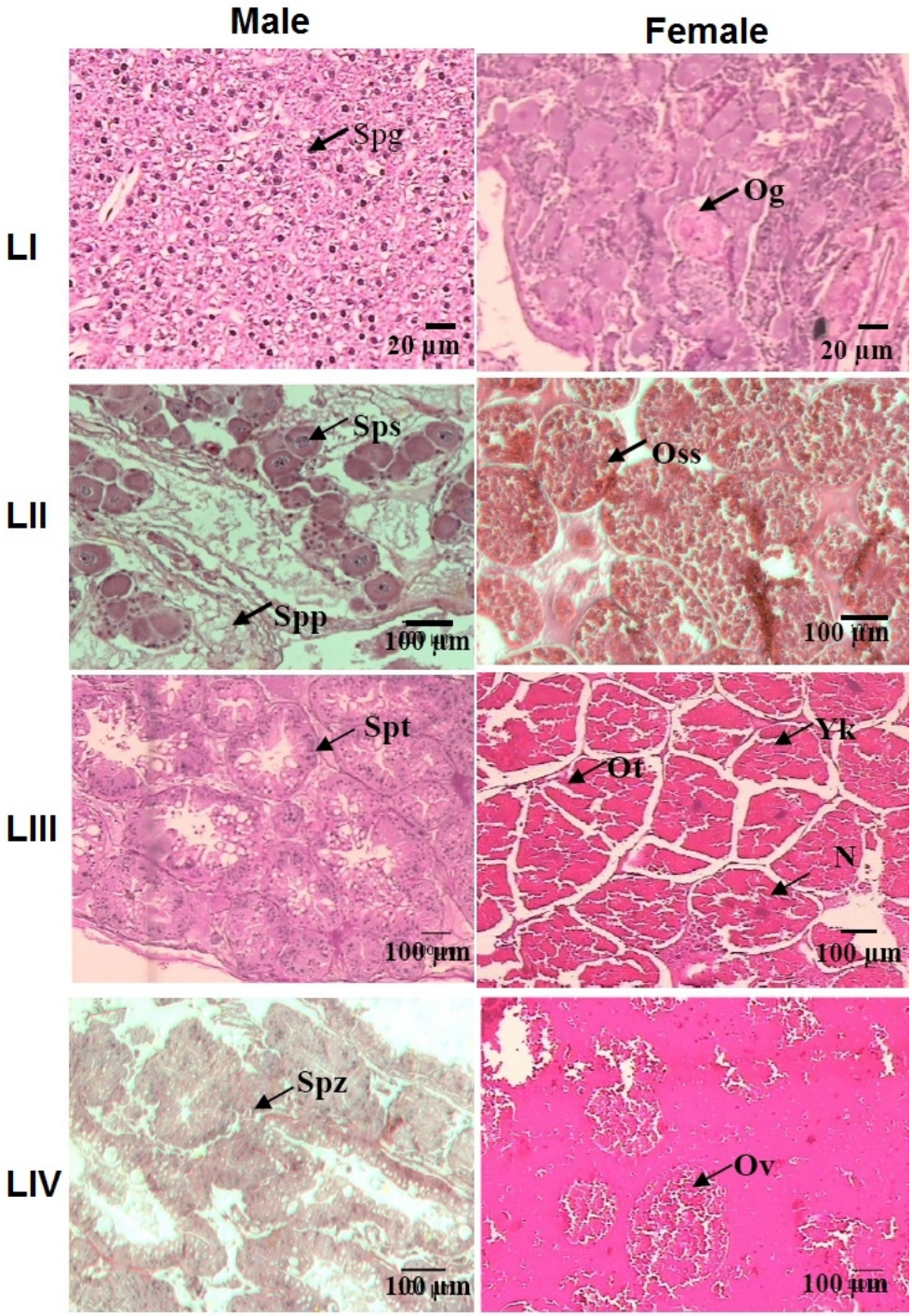

Fig. 3: The view of gonad histological structure of female and male mantis shrimp. Spg = spermagonium, Spp = spermatosit primer, Sps secondary spermatosit, Spt = spermatid, Spz = spermatozoa, Og = oogonium, Osp = oosit primer, Oss = secondary oosit, Ot = otid Ov = ovum, N = cell and Yk = yolk

by Sallam (2000) and Sallam (2005) to over-exploitation. The overfishing activity of the mantis shrimp *Oratosquillaoratoria* in Tokyo Bay (Ohtomi and Shimizu, 1991) also contributed to the change in size in gonads.

Fluctuation and the change of environment conditions had influenced the gonad maturation process. It was observed that for mantis shrimp larvae, the gonad's maturation process had a close relationship with the waters environment signals, such as the provision of food (van der Wall, 2006).

Variations in the first size of maturity was observed in the gonads for female and male mantis shrimp (*Harpiosquilla raphidea)*. The size of first male mature gonad had a total length (TL) of 230 mm or carapace length (CL) of 51.52 mm, while female mantis shrimp

had a TL of 199 mm or CL of 44.58 mm.

It was observed for the *Squilla mantis* shrimp in Caciz Bay that it had the first maturity of gonad at CL around 23.7 mm (Vila *et al*., 2013), while Abello and Martin (1993) in Delta Erbo, Spain had found the first size mature gonad of the male *Squilla mantis* shrimp around 15.2 cm and the female around 14.5 cm. On the other hand, Abello and Sarda (1989) reported the TL to be around 11.13 cm. According to Nikolsky (1963), the first size of mature gonad was probably influenced by the abundance and provision of food, temperature, lighting period and the environmental factor of water habitats or different waters. Several factors that caused the difference of the first size of mature gonad were due to population genetics, differences in rates of growth and the quality of waters (Paugy 2002); the differences of area and the fishing pressure also influenced the first size of mature gonad (Reynolds *et al*., 2001).

Type of spawning is related to the development and egg diameter could be used to identify the pattern of shrimp spawning. The fecundity ofthe mantis shrimp (*Harpiosquilla raphidea)*was found to be different from the fecundity for female *E. massavensis* (Sallam, 2000). The fecundity of *H. raphidea* was greater than other species of mantis shrimp *Harpioquilla* (Giovanardi and Piccinetti, 1983; Hamano *et al*., 1987. Fecundity was reported to be negatively correlated to water environment pressure and over-exploitation (Reaka, 1979; Sallam, 2000).

The dissemination pattern of shrimp egg diameter showed that mantis shrimp could be classified as being a total spawner.The research result of Kodama *et al*. (2006) on the mantis shrimp *Oratosquila oratoria* showed a similar dissemination pattern with the once-spawning type even though the spawning season and time were different. In controlled conditions, the mantis shrimp *Oratosquilla oratoria* showed a dissemination pattern that did not vary, but all the eggs were released during 3 days (Hamano and Matsuura, 1984). The difference of egg diameter could be determined according to the level of egg maturity, while the difference of fecundity was determined by the gonad maturity index.

Conclusion

The sex ratio between male and female mantis shrimp *Harpiosquilla raphidea* is in balance.The first size of mature gonad of the male mantis shrimp *H. raphidea* is larger than the female. The spawning peak season of mantis shrimp *H. raphidea* in Banten Bay waters is observed to be in March-April and September-October *H. raphidea* is a total spawner.

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