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REPRODUCTIVE BIOLOGY OF LONGTAIL TUNA (*Thunnus tonggol*) IN THE JAVA SEA

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ABSTRACT

Longtail tuna (*Thunnus tonggol*) is one of common economically important pelagic fish species in Indonesia. The objective of this study is examining the biology of reproduction, consisting of length of weight relationship, sex ratio, maturity stage, gonado somatic index (GSI), length at first capture, and length at first maturity and spawning pattern. A total of 633 longtail tuna, ranging 29-58 cmFL and consisting of 293 males and 340 females, were collected from the Java Sea between April 2018 and March 2019. The results showed that the longtail tuna growth pattern was isometric. The sex ratio was not significantly different between male and female. The length of first capture longtail tuna of drift gillnet (43.2 cmFL) was bigger than the length at first maturity (42,3 cmFL). This indicates that the most of longtail tuna caught by drift gillnet have already spawned. The peak's spawning season occurred in May and November, with fecundity ranging from 783,597 - 1,579,160 eggs. Longtail tuna has multiple spawning pattern.

Keywords: Biology; *Thunnus tonggol*; spawning season; the Java Sea

INTRODUCTION

Longtail tuna (*Thunnus tonggol*) is an epipelagic species in tropical and subtropical waters in the Indo Pacific region between 47°N to 33°S. This species can grow to a maximum size of total lengths of 142 cm (TL) and weight 35.9 kg (Froese & Pauly, 2020). Longtail tuna is quite unique compared to other *Thunnus* species that are commonly found in the deep sea. This species is almost exclusively confined to neritic areas less than 200 m depth, but generally less than 50 m, and is rarely found offshore (Yesaki, 1994). According to Cheunpan (1984), longtail tuna is usually caught in the waters around 15-30 nautical miles from shore land at depths of 20-45 m. The fish commonly live in the waters with temperatures of 16-31!, avoiding low salinity and high turbidity. Longtail tuna is found in Indo Pacific waters from southern Japan, the Philippines, Papua New Guinea to Australia and westward from Indian waters, the Arabian Peninsula, the Red Sea to the Somalian coast. (Collete & Nauen, 1983). In Indonesian waters, longtail tuna is commonly found along the Java Sea, the Natuna Sea/southern part of the South China Seas to the Malacca Strait.

Longtail tuna (*Thunnus tonggol*) in the Java Sea is commonly performed as fish schools with other neritic tuna kawa-kawa (*Euthynnus affinis*) and frigate tuna (*Auxis thazard*). Landing data in Pekalongan Fishing Port, one of the largest fishing ports in the north coast of Java, indicates that longtail tuna has the largest proportion among the other neritic tuna at about 48% (Pekalongan Fishing Port Nusantara 2016). This annual report showed that the landed fish was mainly caught by two types of fishing gears, comprising 33% from drift gillnet and 67% from purse seine (Wujdi & Suwarso, 2014). The annual national catch statistics of longtail tuna increased from 95,325 tons in 2006 and reached its peak production with 117,783 tons in 2011 then declined to 84,022 tons in 2012 to its lowest at 65,651 tons in 2016 (DGCF, 2017). Longtail tuna catch in the Java Sea reached 12,553 tons or 19.2% from nasional catch andlongtail tuna catch in Pekalongan was 19.68% from the Java Sea catch, or equal to 1.80% of national catch (DGCF, 2017).

Worldwide study on biological parameters of this species has been published by several authors, such as in Pacific Oceania (Wilson, 1981), west Thailand waters (Yesaki, 1982), Gulf of Thailand (Cheunpan, 1984), Manglore East Indian waters (Muthiah, 1985), north and eastern Australia (Griffiths *et al*., 2010), Taiwan (Wei *et al*., 2011), Persian Gulf (Hedayatifard, 2007; Kaymaran *et al*., 2013), along Indian coastal waters (Abdussamad *et al.*, 2012; Koya *et al*., 2018),

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and the South China Sea (Hidayat & Noegroho, 2018). Nevertheless, studies of this species on the Java Sea are still limited and need to be updated.

The focus of this study is to provide information on the reproductive biology of longtail tuna that could provide an important reference to describe the stock status as the baseline to build management guidance in the Java Sea and its adjacent waters.

MATERIALS AND METHODS

Data Collections

Samples of longtail tuna were collected from drift gillnet and mini purse seine fishing that were operated in the Java Sea (Figure 1). Random samples were collected monthly from April 2018 to March 2019 at Pekalongan Fishing Port, Central Java, Indonesia, with geographical position 6º50’42"S- 6º55’44"S and 109º37’55"E- 109º42’19"E. Whole weight (to the nearest 0.1 g) and fork length (to the nearest 0.1 cm) were measured and collected as the baseline of this study. Gonads were removed and weighed to the nearest 0.1 g, then preserved in Gilson’s fluid for further examination on fecundity and oocyte diameter. Furthermore, for histological analysis, gonad samples were preserved in formalin 4 %. Sample analyses were conducted at the Laboratory of Research Institute Marine Fisheries, Cibinong Bogor. Measuring biological aspects (length, weight, and gonad maturity) were conducted in the Fishery Laboratory at the Pekalongan Fishing Port, Central Java. Analysis of fecundity and histological examination of ovaries were conducted at the laboratory of Research Institute Marine Fisheries, Cibinong Bogor. The determination of gonad maturity stages followed the criteria of I to V (Holden & Raitt, 1974).

Figure 1. Sampling site of longtail tuna in the Java Sea.

Data Analyses

The Length–Weight relationship was determined by Effendie (2002):

$$W = aL^b \quad \text{.........} \quad (1)$$

Where W is the total weight of fish (gram), L is the fork length (cm), a and b = constants. In order to verify if calculated b was significantly different from 3 (allometric), the Student t-test (Sokal & Rohlf, 1987) was conducted.

The sex ratio was expressed as:

$$X = M:F \quad \text{.........} \quad (2)$$

Where X is the sex ratio, M is the number of male fish observed, and F is the number of female fish observed. Chi-square (X^2) test was used to examine the homogeneity of the sex ratio. The sex ratio was tested by chi-square according to Sugiyono (2004) as follows:

$$X^2 = \sum_{i=l}^{k} \frac{(fo - fn)^2}{fn} \quad \text{.........} \quad (3)$$

Where X^2 = Chi–square, $f0$ = observed frequency, and fn = expected frequency, with the null hypothesis (H_0): there is no significant difference between the number of male and female fish.

The length at first capture (Lc) is determined based on the relationship between the length of the fish (Xaxis) and the number of fish (Y-axis) to form an S curve. The estimate length at first capture (*Lc*) was estimated using the following formula by Sparre & Venema (1998):

$$S_L est = \frac{1}{1 + \exp(S_1 - S_2 * L)} \quad ....(4)$$

$$L_{50\%} = \frac{S_1}{S_2} \quad ....(5)$$

Where $S_L est$ is a logistic curve, S_1 and S_2 are constants.

Estimation of the length at first maturity (*Lm*) used the logistic function approach (King, 1995) with the equation:

$$P_{Lm} = \frac{1}{1 + \exp(aL + b)} \quad ....(6)$$

Where P_{Lm} is the proportion of the number of mature fish with length *L* to the number of immature fish with length of *L* *a* and *b* are the curve parameters (a<0 and b>0), so that the length at 50% mature gonad (*Lm*) is the same as –a/b.

The gonadosomatic index (GSI) is a comparison of gonad weight to fish weight.This index value over time will get greater until reaching the peak limit, then there will be a decrease (King, 1995). By knowing this GSI, the fish spawning period can be estimated. The GSI was analyzed according the Effendie formula (2002):

$$GSI = W_g / W_t \times 100\% \quad .... (7)$$

Where *GSI* is gonadosomatic index (%), W_g = weight of gonad (gr), W_t = total weight of fish (gr).

The fecundity analysis was conducted on adult female fish with gonad maturity stage of III and IV. The measurements of diameter size and number of oocytes were done by using a microscope 4 x 10. The observation of the number and distribution of oocyte size used egg samples as much as 0.5 grams. The fecundity was calculated gravimetrically by the formula Holden & Raitt (1974):

$$F = n \cdot G / g \quad ....(8)$$

Where *F* is fecundity, *n* = number of eggs in subsample, *G* = gonad weight, *g* = weight of gonad subsample (0.5 gram).

The relationships between fecundity (*F*) and length (L) or weight were expressed by the following equation (Sujatha *et al.*, 2014):

$$F = a L^b \quad .... (9)$$

Where *F* is the fecundity of eggs, *L* is fork length (cm) or fish weight (g), *a* and *b* = constants.

RESULTS AND DISCUSSION

Results

Length-Weight Relationship

A total 633 specimens of longtail tuna were investigated. The length ranged 29-58 cmFL, and weight 433-3190 g, the unisex the length-weight relationship of *T. tonggol* was estimated as $W=0.019L^{2.9838}$ (R^2=0.9677), where the value of *b* was 2.9838 (Figure 2).

The next analysis used Student's *t*-test on the value of *b* of the whole samples of male and female longtail tuna, with 95% confidence level (p=0.05). The value of *tcount* = 0.0000170738 was smaller than t_{table} =1.984. This result showed that *b* was not significantly different from 3, which indicated isometric growth pattern. The isometric length-weight relationship means the weight gain is proportional to the increase in length (Effendi, 2002).

The observations of monthly length-weight parameters of longtail tuna obtained are as in Table 1 below. It shows that all monthly length-weight relationships of longtail tuna were isometric (after conducting *t*-test).

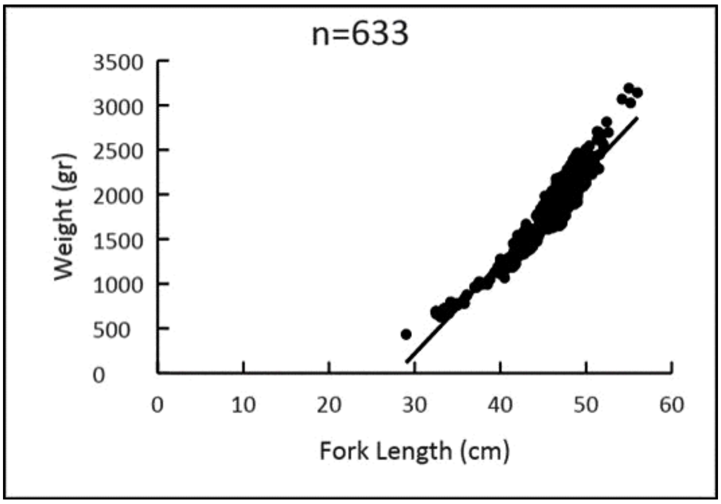

Figure 2. Length–weight relationships of longtail tuna in the Java Sea.

Table 1. Monthly length-weight parameters of longtail tuna in the Java Sea

Month	*n*	*a*	*b*	R^2	*t*-test		Growth
					tcount	*ttable*	
Apr	47	0.0563	2.7310	0.820	0.000245	2.021	isometric
May	48	0.0454	2.7702	0.894	0.000218	2.021	isometric
Jun	36	0.0212	2.9682	0.969	0.000045	2.042	isometric
Jul	58	0.0134	3.0819	0.822	0.000082	2.021	isometric
Aug	37	0.0717	2.6571	0.826	0.000360	2.042	isometric
Sep	54	0.0279	2.8945	0.923	0.000100	2.021	isometric
Oct	51	0.0989	2.5684	0.680	0.000410	2.021	isometric
Nov	52	0.1335	2.4848	0.709	0.000520	2.021	isometric
Dec	54	0.0371	2.8047	0.990	0.000235	2.021	isometric
Jan	50	0.0189	2.9938	0.980	0.000008	2.021	isometric
Feb	97	0.0220	2.9486	0.978	0.000065	1.984	isometric
Mar	49	0.0208	2.9626	0.975	0.000043	2.021	isometric

Sex Ratio

The sex ratio of all male and female specimens was 0.9:1. A chi-square test accepted the null hypothetical ratio resulted, which was no significantly difference between male and female with 95% confidence level (p=0.05). It means the sexes were equally distributed in the population.

Gonadosomatic Index

The average of monthly gonadosomatic index (GSI) for female ranged from 0.08-4.4 and for male ranged 1.0-5.5. The GSI for male and female had two peaks with the highest indeces were in April and October then decreased in the following months that indicated the spawning happened in May and November (Figure 3.)

Gonad Maturity Stage

The results showed that the monthly distribution of gonad maturity stage I to IV are fluctuated. Gonad maturity stage IV of male and female longtail tuna fish was found every month, with the two highest percentage occuring in September and October (Figure. 4 and 5).

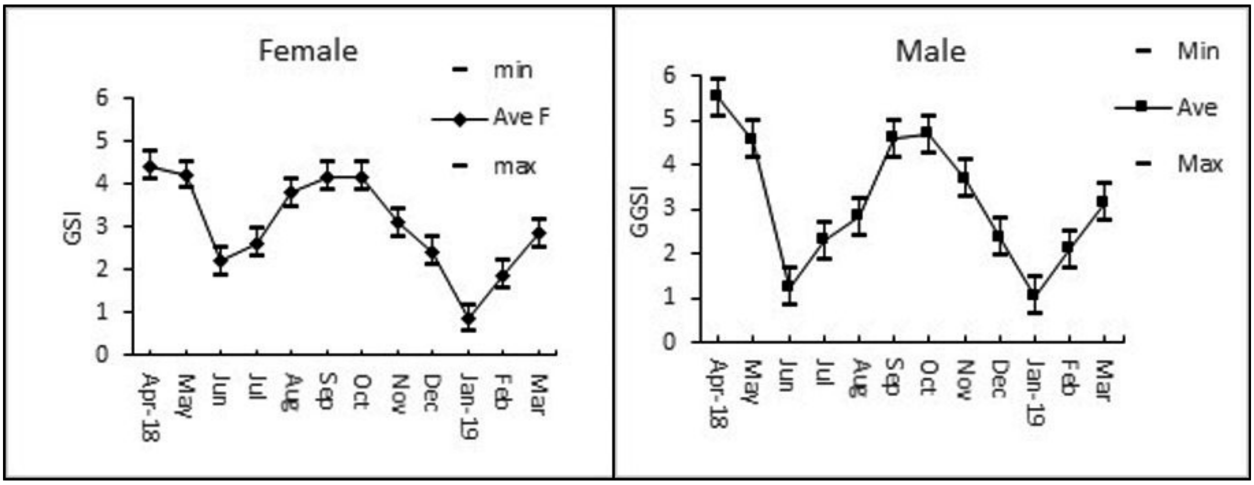

Figure 3. Monthly gonadosomatic index of female and male longtail tuna in the Java Sea.

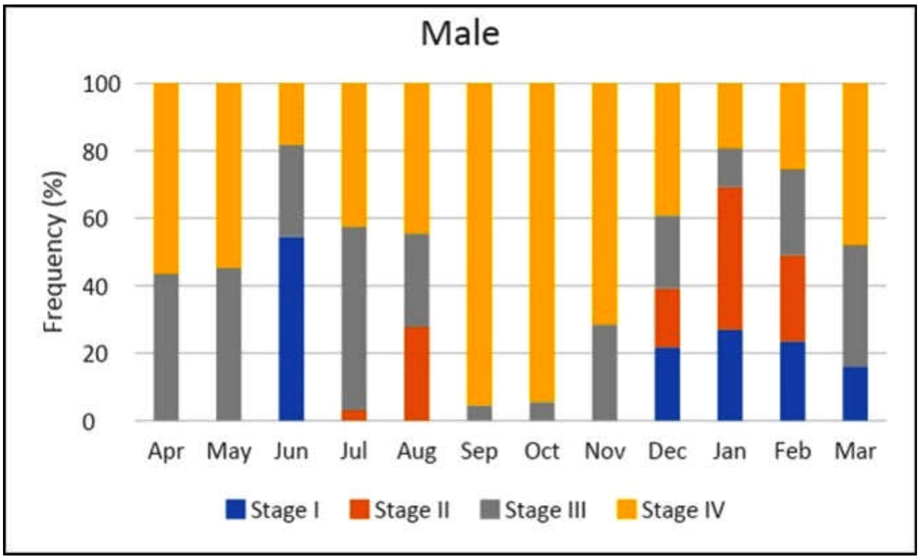

Figure 4. Monthly gonad maturity stages of male longtail tuna in the Java Sea.

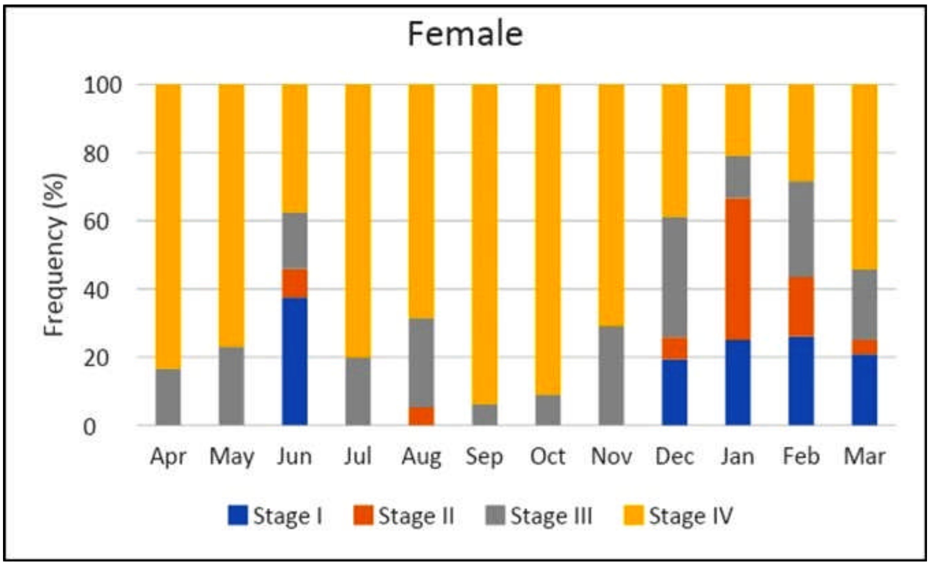

Figure 5. Monthly gonad maturity stages of female longtail tuna in the Java Sea.

*Size at First Capture (*Lc*) and Size at First Maturity (*Lm*)*

Estimation of the size at first capture (*Lc*) of longtail tuna from drift gillnet and small purse seine was 43.2 cm and 26.4 cm, respectively (Figure 6). The estimated size at first maturity using the logistic model was estimated 42.3 cm.

Fecundity and Size of Oocytes

The fecundity of longtail tuna (*Thunnus tonggol*) in the Java Sea was 783,597 - 1,579,160 eggs, from mature females ranging 38-52 cmFL. The oocyte diameter was 0.21-0.99 mm, with average 0.56 mm. The oocyte frequency distribution has two modes (heterogen) (Figure 7).

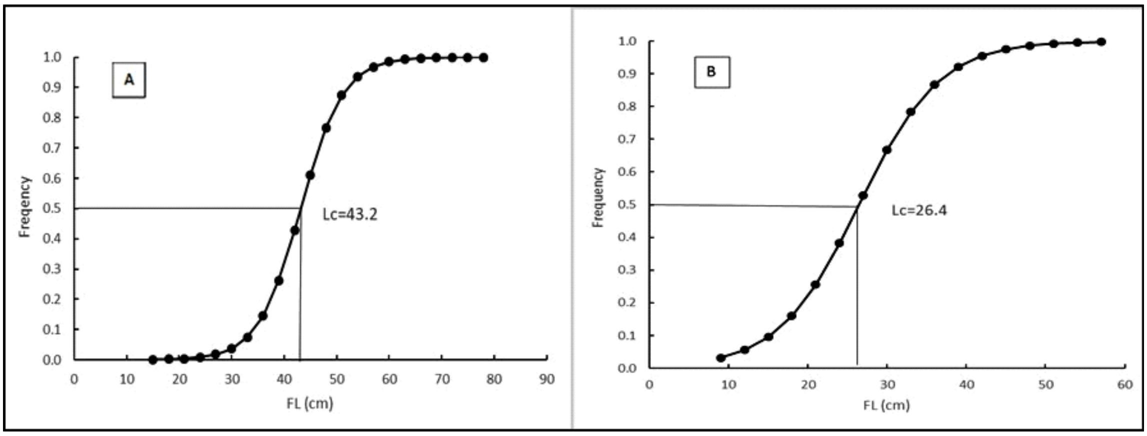

Figure 6. The length at first capture (*Lc*) of longtail tuna caught by drift gillnet (A), and mini purse seine (B).

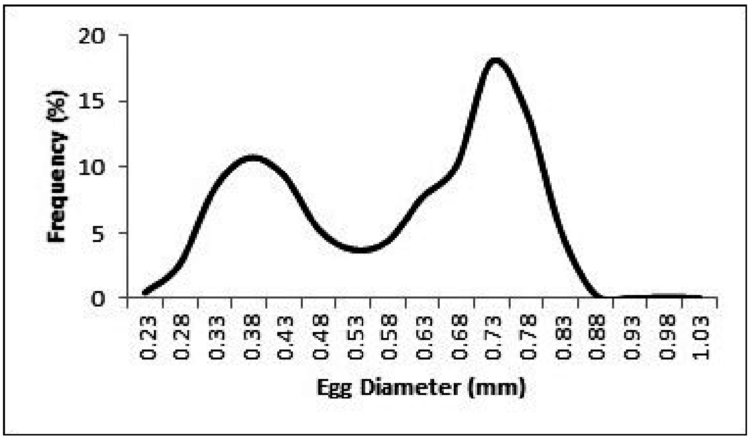

Figure 7. Oocyte diameter of longtail tuna in the Java Sea.

The relationships between fecundity (*F*) and variables length (*FL*) as well as body weight (*W*) are described in Figure 8. There are significant positive relationships between the fish length or weight and fecundity.

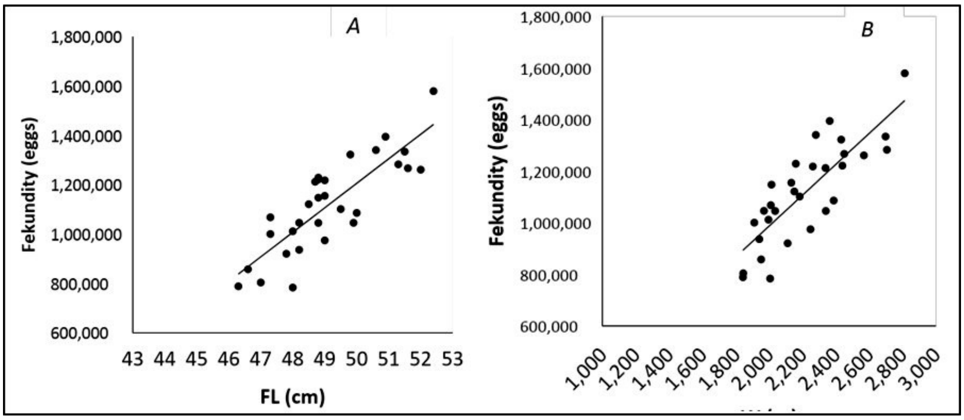

Figure 8. Relationships of fecundity-length (A) and fecundity-weight (B) of longtail tuna.

Oocyte Development

The development of oocytes determined based on histological examination showed at least four stages of sexual maturity, while stage V was not found during this study (Figure 9).

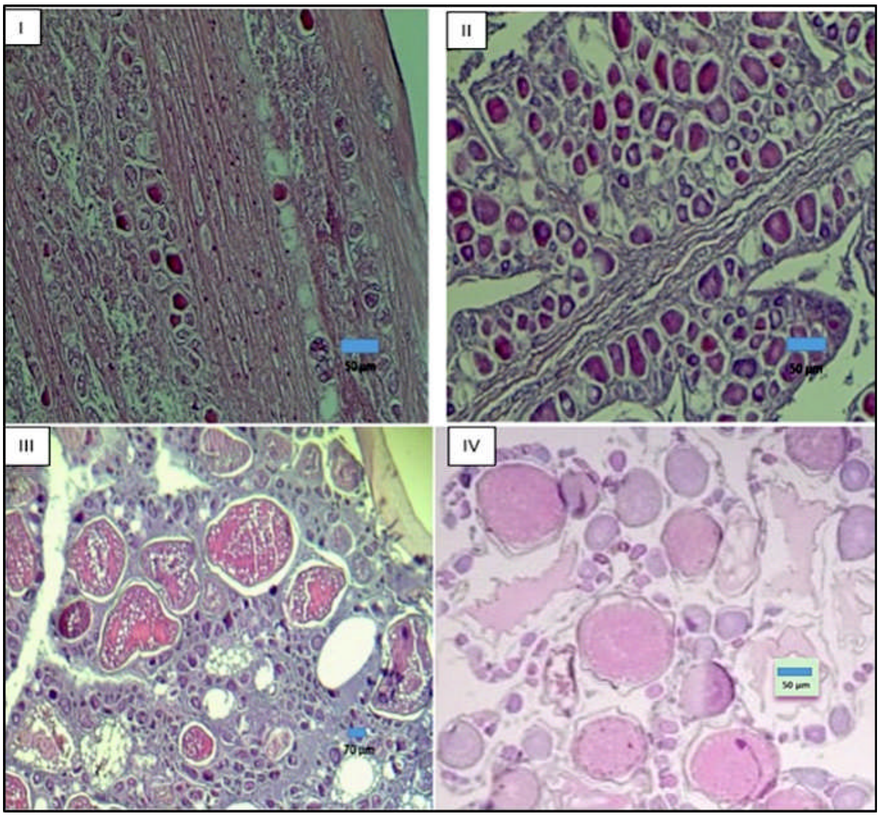

Figure 9. Histological section of the ovaries of longtail tuna in the Java Sea. (I) stage I and (II) stage II: Primary growth oocytes, (III) stage III: Vitellogenesis, (IV) stage IV: Fully yolked oocytes.

Stage I, is still in the oogenesis stage, the gonadal development process of forming an oocyte. The oocyte is still small and the nucleus is round or oval with a thicker cytoplasm. Stage II, oocytes begin to grow and the cell nucleus is appeared to increase in size and the yolk spread. Stage (III) is maturing or vitellogenesis. The amount and size of the yolk is increasing and clearly visible in all areas of the oocyte. Oil droplets appear in the cytoplasm, the cell nucleus concentrated in the central oocyte. Stage IV, is maturation. Plenty of egg yolks reach fully yolked oocytes, oil droplets increasingly spread from around the cell nucleus to the periphery oocytes. The core cells migrate around the oocytes and are usually replaced by larger oil droplets.

Discussion

The samples used were both male and female fish, which were not separated because longtail tuna is not a dimorphism species, meaning there are no significant differences among external morphological characteristics between male and female (Schaefer, 2001). Furthermore, Griffiths *et al*. (2017) stated that there were no statistical differences found in the morphometric characteristics between sexes in longtail tuna. The length weight relationship in our study showed that the *b* value had no significant difference with 3, which indicates an isometric growth pattern. Several studies on length-weight relationship in this species have isometric growth pattern, such as Abdussamad *et al*. (2012) in Indian waters, Kaymaram *et al*. (2013) and Yasemi *et al*. (2017) in Persian Gulf waters, and Hidayat & Noegroho (2018) in the South China Sea. However, the studies of Ahmed *et al*. (2016) in Pakistan waters, Griffiths *et al*., (2017) in Australian waters, Darvishi *et al*. (2003) and Khorshidian & Carrara (1993) in the Persian Gulf showed negative allometric growth patterns. These differences in the exponent *b* value of length-weight relationship are probably related to ecosystem and biological conditions, like spawning period, sexual maturity, feeding behavior, and food competition (Ghosh *et al*., 2010). Furthermore, Froese (2006) states the variation in the *b* value could be affected by geographic locations, sampling area, seasons, size range, and ecological factors such as temperature. The negative allometric growth of fish might be attributed to the environmental conditions and insufficient food availability (Arslan *et al*., 2004; Jisr *et al*., 2018).

The knowledge of sex ratio is important to understand the relationship among individuals, the environment and the state of the population (Vicentini & Araújo, 2003). The sex ratio of longtail tuna in this study was not significantly different between male and female, showing the similar number of male and female fish in the population. The sex ratio of longtail tuna in other regions of the world was also found not significantly different between male and female (Wilson, 1981; Yesaki, 1982; Griffiths *et al*., 2010; Hassadee *et al*., 2014). The insignificant difference between the sexes implies the non-existence of seasonality in sex distribution within the population (Jega *et al*., 2017). However, the sex ratio may vary from the expected 1:1, influenced by several factors, such as adaptation of the population, reproductive behavior, food availability, and environmental conditions (Vandeputte *et al.,* 2012).

The length at first mature (*Lm*) of longtail tuna caught in the Java Sea on this study was 42.3 cm FL. The size at first mature (Lm) of this fish in Thailand waters was 39.6 cm (Chuenpan, 1985). *Thunnus tonggol wa*s reported to attain sexual maturity at 39.6 cm in the Persian Gulf and the Oman Sea (Hedayatifard, 2007) and at 37 cm in Taiwan waters (Chiang *et al.,* 2011). Yesaki (1982) and Cheunpan (1984) from Thailand recorded 43 and 40 cm, respectively, as the length at maturity. Pillai and Ganga (1985) and Abdussamad *et al.,* (2012) in Indian waters reported the length at first maturity (*Lm50*) as 51.1 and 50 cm, respectively. Hidayat & Tegoeh (2018) stated that the *Lm* value of this fish in the South China Sea was 41.1 cm FL. It appears that the size at maturity of this species varies in several locations. The differences in size at maturity for the same species may be due to variations in geography and fishing pressure (Morgan & Colbourne 1999; Claereboudt *et al*. (2005); Moresco & de Bemvenuti (2006); (Haig *et al*., 2015). This variation might be different in environmental variables rather than genetic differences (Duponchelle & Panfilli, 1998). Tormosova (1983) suggested that stock density, food, and water temperatures may influence the growth of fish and affect the age and size at maturity. Yoneda & Wright, (2004) stated that variation in size maturity may reflect population differences.

The estimation length at first capture (*Lc*) of longtail tuna from drift gill net and small purse seine was 43.1 cm and 25.9 cm, respectively, while the length at first maturity (*Lm*) using a logistic model was estimated 42.3 cm. The values of drift gillnet Lc was bigger than its Lm value, indicating that most of the longtail tuna caught by drift gillnet were the mature fish that had spawned at least once before being caught. This type of fishing gear is recommended. In contrast, the sizes of longtail tuna caught by small purse seine were mostly under the length at first maturity, meaning they were immature fish.

The development of the monthly GSI simultaneously with macroscopic stages showed that both male and female gonad development reached the highest in April and October then decreased in the following month. According to Widodo (1986), King (1995), and Claereboudt *et al*. (2005), spawning season occured about one month after the highest percentage of mature fish. Therefore, the estimated spawning seasons of longtail tuna in the Java Sea were around May and November. The spawning season of longtail tuna in Thailand waters occurred in May and August (Hassadee *et al.,* 2014), in Indian waters occured in October-November (Abdussamad *et al.,* 2012), and in the waters of Taiwan around December (Wei *et al*., 2011). The diversity of the spawning season and periodicity exists due to the varied ecological environments (Agarwal, 2008; Jega *et al*., 2018; Papoulias *et al*., 2006). The water temperature, photoperiod, and prey items may influence the spawning season of fish (Tormosova, 1983; Papoulias *et al*., 2006; Ma *et al*., 2012).

The fecundity of longtail tuna in the present study was estimated between 783,597 - 1,579,160 eggs. The information on the fecundity of longtail tuna is limited. The previous estimates varied between 0.2 and 2.0 million oocytes (Klinmuang, 1978; Wilson, 1981; Hedayatifard, 2007; Abdussamad *et al.,* 2012). Lambert (2008) said that fecundity varies in relation to parental quality (size, condition), food availability, as well as environmental and evolutionary factors (stock biomass, fishing pressure). Jega *et al*. (2018) stated that the differences in fecundity and reproductive potential were affected by the size of fish, food availability, temperature, salinity and genetics.

The oocytes distribution has more than one modes, especially those in mature fish, indicating that this species was categorized as batch or multiple spawners (Murua & Saborido-Rey, 2003; Lowerre-Barbieri *et al.*, 2011; Schaefer, 2001). The histological oocyte development in mature fist (stage III and IV) showed that the oocyte is heterogen, showing that the ovarian development of the longtail tuna belongs to the asynchronous oocyte development group (Murua & Saborido-Rey, 2003).

CONCLUSIONS

The longtail tuna (*Thunnus tonggol*) in the Java Sea has an isometric growth pattern. The sex ratio was not significantly different between male and female. The longtail tuna tuna caught by drift gill net was mature fish, while from purse seine was immature. The spawning season occurred in May and November, with fecundity ranging 783,597 - 1,579,160 eggs. The longtail tuna was multiple spawner, with double spawning pattern.

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Appendix 1. A five-point maturity scale for partial spawners

Stage	**State**	**Description**
I	Immature	Ovary and testis about 1/3rd length of body cavity. Ovaries pinkish, translucent; testis whitish. Ova not visible to naked eye.
II	Maturing virgin and recovering spent	Ovary and testis about 1/2 length of body cavity. Ovary pinkish, translucent; testis whitish, more or less symmetrical. Ova not visible to naked eye.
III	Ripening	Ovary and testis is about 2/3rds length of body cavity. Ovary pinkish-yellow colour with granular appearance, testis whitish to creamy. No trans- parent or translucent ova visible.
IV	Ripe	Ovary and testis from 2/3rds to full length of body cavity. Ovary orange-pink in colour with conspicuous superficial blood vessels. Large transparent, ripe ova visible. Testis whitish- creamy, soft.
V	Spent	Ovary and testis shrunken to about 1/2 length of body cavity. Walls loose. Ovary may contain remnants of disintegrating opaque and ripe ova, darkened or translucent. Testis bloodshot and flabby.

Source: Holden & Raitt (1974)