Gonad maturity of simping *Placuna placenta*, Linn 1758 (Bivalve: Placunidae) harvested from Kronjo Coastal, Indonesia

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ABSTRACT

Simping *Placuna placenta* is a coastal resource found in shallow water with a muddy substrate. Simping widely used as a source of food and as raw material for the decoration. Utilization of simping until now has not yet focused on recruitment, reproductive aspect. This information is important as a basis data for determining the size of the simping catch. This study aimed to find out about of reproduction aspect, determining sex ratio the size of the catch simping. This research was conduct for three months at 3 locations. The number of samples in the analysis are 36 species to determine sex ratio and gonad maturity. Sex ratio indifferent from male and female, but based on the time sex ratio of simping significant, both male and female. The length size of gonads shellfish matures form 5.50 cm and 5.85 cm at 2nd maturity stage. Length of simping mature from 6.08 cm, 6.24 cm, 6.45 cm and 7.11 cm dominant at mature stage 3, and 8.61 cm dominant at 4th maturity stage. Sex ratio of male and female are not significant between males and females (M: F=1:1). It is concluded that the level of maturity of gonads is increasing as increases of size of simping.

**Keywords:** Gonad Maturity; Kronjo Coast; Placuna; Sex Ratio

INTRODUCTION

Simping (*Placuna placenta*, Linn 1758) is a bivalves group on the coastal area that has dual sexual or called as a hermaphrodite. Early cell development (larva stage) is difficult to explain the differences between male and female. Spawning generally occurs outside the body (external fertilization) (Le Pennec *et al.*, 2003). Generally of mature simping at the adult stage of *P. placenta* at length more than 6 cm and spawning season in summer from April to May, or May to September. Gametes produced estimated that produced while spawning season between 15-21 million oocytes per spawning (Le Pennec *et al.*, 2003). There are two types of spawning patterns of simping, first type adults stage spawn partially in the spring from April or May and reach peak season at the end of August (Mason, 1983). And the second type is for shells that have not to spawn previously will be spawning in the fall (from September to January) season.

The spawning season it is also followed by a period for gonad recovery for spawning the future. Gibson (1956) found a similar spawning pattern in the Bere island Ireland coast. At Bantry Bay, simping will return to mature gonads stage after six weeks from the spawning season. Spawning occurs outside the body; fertilization occurs in the water (Mason, 1983). After spawning, gonad development is the larval phase (pelagic larvae) for a month before settle in substrate. As a resource in coastal waters that have economic potential (as a source of protein and the economy) simping exploitation tends to simping degradation. The maintenance of simping production needed a strategy for increasing stock with aquaculture program. And in this study aimed to...
determine biological information particularly gonad maturity, sex ratio, larva and stage development, and environmental information to support of simping production and sustainability.

MATERIALS AND METHODS

Site and Time
The study was conducted for three months from November 2007 until January 2008. Sample collected from the Kronjo coast at Tangerang district. The data collecting did by a sweeping method (swept area method) using garok gear (bottom gear). Area sampling is about 15 m² (1.5 x 10 m) in each station observed. The sampling location has seen in Figure 1.

![Figure 1. The map of Kronjo Coastal showed the sampling location](image)

Data collection
The sample collected was preserved on the polybag that has been labeled and preserved with formalin 10% and then analyzes in the laboratory. Laboratory activity is identifying of simping spat, pre-adult, and adult stage. Gonad maturity identifies from histology preparat from pre-adult and adult stage. Gonad preparation for histology preparat using Bouin's and alcohol 50%, 70%, 80%, 85%, 90%, 95%, and 100% alcohol, xylol, paraffin, solvents Hematoxylin and Eosin (HE).

High shell is measured by the distance dorsal to ventral maximum, using calipers with 0.01 mm accuracy. Shell height measurements carried out in Fisheries Biology Laboratory, Department of Aquatic Resource Management, Faculty of Fisheries and Marine Science, Bogor Agricultural University. Measurements of shells height needed to identify maturity level and the relationship with the development of the gonad size shells.
Data Analysis

Samples analyzed to determine and classification of sex with histology procedure takes from 7 species each sampling, with totally about 36 samples. Sample length used for gonad histology preparations from 5.5 cm, 5.85 cm, 6.08 cm, 6.24 cm, 6.45 cm, 7.11 cm and 8.61 cm. Gonad histology performed in fish health laboratory, Department of Aquaculture, Faculty of Fisheries and Marine Science, Bogor Agricultural University. Data collected are water quality, the height of shell, weight, and gonad maturity stage. Data analysis is a descriptive analysis by interpreting pictures and graphics histology sex ratio. Statistical analysis needs to evaluate of sex ratio male and female from adult simping by Chi-Square analysis.

RESULTS

The environmental parameter measured were depth, substrate, temperature, turbidity, pH, dissolved oxygen (dissolved oxygen, DO), total suspended solids (TSS), chemical oxygen demand (COD), and salinity. The results of the measurement parameters of water described in Table 1. This water analysis purpose of determining water quality conditions at these locations and its impact on the feasibility of simping life. Water temperature range from 29.00-29.67 °C (29.4±0.4) salinity from 26-28 ‰ (27±1), turbidity between 4.87-10 NTU (7.9±2.7), total suspended solids from 6.67-25 mg/l (16.4±9.2), pH 6.67-7 (6.8±2), dissolved oxygen 4.27-4.88 mg/l (4.6±0.4) and chemical oxygen demand (125.22-190.67 mg/l (153.3±33.7).

| Parameters        | Unit   | Station 1 | Station 2 | Station 3 |
|-------------------|--------|-----------|-----------|-----------|
| **Physical**      |        |           |           |           |
| Depth             | cm     | 10        | 39        | 27        |
| Temperature       | °C     | 29.00     | 29.67     | 29.67     |
| Salinity          | %      | 29.00     | 29.67     | 29.67     |
| Turbidity         | NTU    | 10.00     | 4.87      | 8.83      |
| Suspended solid   | mg/l   | 6.67      | 25.00     | 17.67     |
| **Chemistry**     |        |           |           |           |
| pH                | -      | 7.00      | 6.67      | 6.67      |
| DO                | mg/l   | 4.28      | 4.88      | 4.27      |
| COD               | mg/l   | 144.00    | 125.33    | 190.67    |

Note: 1,2,3 refer as station.

Sex ratio

Determination of sex differentiation based on gonad morphology and histology. If the color gonad orange means that female and color of the male is white. Ratio sex for male (m) higher than simping (female). Sex ratio simping shells can be seen in Table 2. Sex ratio of simping in each station are station 1 between 1, (F:M=5:1), station 2, F:M=0:1) and at station 3 is (F:M=1.09:1). Statistical analysis (Chi-square) not significant (CL 95%) between simping males and females each station. That means simping sex ratio of (P. placenta) is proportional in the habitat with value is 1:1. Sex ratio analysis based on time of sampling found that sexual proportion in November
dominant male (73%), December 67%, but in January relatively stable. Statistical analysis between sampling time of simping sex ratio is different (significant at CL 95%).

Table 2. Sex ratio of *Placuna placenta* each station

| Station | Male | Female | M: F | $\chi^2$ hit | $\chi^2$ tab | Sex ratio |
|---------|------|--------|------|-----------|-------------|-----------|
| station 1 | 6 | 4 | 1.5:1 | 0.40 | 3.84 | insignificant |
| station 2 | 0 | 3 | 0:1 | 3.00 | 3.84 | insignificant |
| station 3 | 12 | 11 | 1.09:1 | 0.04 | 3.84 | insignificant |
| Total | 18 | 18 | 1:1 | 0 | 3.84 | insignificant |

Figure 2. The sex ratio of male and female shell on the station

**Gonad maturity**

Gonad maturity is the reproductive phase of gonad to produce their new generation and sustain their population in the habitat. Simping size more than 5 cm, gonad maturity classified by morphology technique, but less size must be use histology. Gonad histology of simping population and its cell content explain in Figure 3. Gonad anatomy performance began filling the posterior gonad organs (viscera) to the interior side by side with digestion. A female gonad is yellow and older than male gonad. The gonads edge directly adjacent to the shell and the posterior part. Female gonads edges have a thin labial attached to the edge of the shell. Gonad morphology before preservation looks slightly yellowish gradually faded. However, while research did not find a hermaphrodite shell.

Morphology analysis found that the first time of simping maturity at length sizes more than 6 cm. Shells have genital simping separate (dioecious), gonad sex mature female orange, and male gonads mature sex is white. Determination of shellfish simping gonad maturity stage studied based on image analysis of gonad histology preparations — the result of gonad maturity, as shown in Table 3.
Table 3. Female gonad development each stage of *Placuna placenta*

| Height size | Stage          | Stage | Histology performance                                                                                                                                 |
|-------------|----------------|-------|-------------------------------------------------------------------------------------------------------------------------------------------------------|
| < 5.5       | Undeveloped    | 1     | Gonad performs undevelop and difficult to separate between male and female gonad.                                                                      |
| 5.50 cm     | Developing     | 2a    | In eggs cavity, oocyst cell un-mature, gonad transparency. The majority of eggs cell un-mature, partially cell cavity fill in egg cell mature and un-mature. |
| 5.85 cm     |                | 2b    | Cavity eggs occupied both mature and immature. City body dominant occupied by immature eggs.                                                            |
| 6.08 cm     |                | 3a    | In the cavity, eggs partially occupied mature or immature eggs. Cavity eggs more developed bigger than before the stage.                               |
| 6.24 cm     | Ripe stage     | 3b    | Volume eggs cavity bigger, and partially eggs develop in the cavity cell body.                                                                        |
| 6.45 cm     |                | 3c    | Eggs adhere to body cell and more develop that before the stage. Eggs color more clearly and male or female separate.                                   |
| 7.11 cm     |                | 3d    | Mature cell egg cavity, each cell fills up, and nucleus eggs visible. Cel cavity that fulfills of mature has the form eclipse and nucleus. All of the cavity cells fulfill by mature eggs. And then also found mature eggs without a nucleus cell. |
| 8.61 cm     | Spawning stage | 4a    | The whole body cavity fulfills by mature eggs. In eggs with having nucleus, cell growth up better, but also found an empty cell. In this stage, the assessing spawning process begins. |

Figure 3. Gonad morphology (female: right-above) and (male: right-under)
Maturity size close relate with length and weight of Simping. Simping sizes that observe histology are 5.50 cm and 5.85 cm for maturity stage 2 (developing). Simping shell size 6.08 cm, 6.24 cm, 6.45 cm, and 7.11 cm for maturity stage 3 (ripe), and shell size was 8.61 cm for maturity stage 4 (spawn). Gonad development each stage of simping seen in Figure 4. Based on figure 4 can be seen that the shells simping stage 2 in the developing stage. At this stage, we found the cavity follicle undeveloped in better conditions. Most of the eggs cell in cavity an immature condition (Figure 4.a). At maturity stage 3, gonad condition has been maturing (ripe). The entire cavity cell consist of mature eggs (oocyst) cell, the nucleus of eggs develop, and identified (Figure 4.b). Gonad maturity stage 4, egg cell prepare for spawning season. The oocyst has been issue by the cavity, and part of follicle cavity began empty from the end of December (Figure 4.c).

![Image](image-url)

**DISCUSSION**

Water quality status at the research site was suitable for biota growth and development. Simping larva, pre-adult and adult high abundance at clear waters with TSS levels between optimum 0-20 mg/ l (Lee et al., 1978 in Bahtiar, 2005). According to Dharmaraj et al. (2004) growth optimal in the range of salinity between 18-38 pro mill and optimum pH from 6.4-7.7.
Temperature and dissolved oxygen parameters are suitable for growth, but chemical organic content (COD) higher than the regulation limit. COD values categorize uncontaminated parameters are usually less than 20 mg / l. Organic chemical content categorizes as pollution in water body reach more than 200 mg / l, and the content of industrial waste COD value could reach 60,000 mg/l (UNESCO / WHO / UNEP, 1992 in Effendi, 2003).

Environmental parameter suitable for biota and ecosystem except chemical oxygen demand tends higher than the standard value. The accordance of Minister of Environment No 51/2004 No. 115/2003 regulations for water quality, where relatively in good condition and suitable for biota. In Virginia, estuary average monthly of temperature within 1–2°C each other also stimulate of mollusk reproduction respond (Harding et al., 2012). Ceure and Boehs show in Cachoeira (2012) river effect of salinity to gamete release in the water body. Rhine River also seems to increase stress response after stimulating by temperature and indirectly increasing the mortality rate (Petter et al., 2014). In general, the distribution of P. placenta, influenced by environmental conditions. Most of the benthic studies influenced by physicochemical, biological characteristics explored that Glycera is the dominant species in prevailing in the environment Varadharaian (2010), which has been a greater influence in April (Yonvittner et al. 2010).

Generally, the proportion male higher than females at the end of the year. This condition different from Anadara antiquates in Pakistani, where the population dominantly in mature and significant to evaluate sex ration (Jahangir et al., 2014). The sex ratio analysis is needed to ensure the sustainability of the recruitment process. Not only environment status, the success of the recruitment process depends on the sex ratio of adult stock. Others sessile species that have similarly character such as A. granosa (Narasimham, 1998); A. tuenculosa with sex ration 1:1 (Cruz et al., 1984); A. inequivalis 1.04-1 (Sahin et al., 2006) and P. placenta (1:1).

In April, sex ratio of P. placenta in Kronjo coast recorded proportional. This means that the reproductive potential support to sustaining recruit and enhancing stock for future. This data inform that sustainability reproduction and recruitment process will be more effective (Mackenzie, 1978 in Mullen and Moring, 1986) for management. Atrina serratrina (Chung et al., 2012), early maturity condition found at (F; M=10,5-15) that higher form others. The fraction of females observed within groups across. According to Ceuta and Boehs (2012) species Tagelus plebeius (Mollusca: Bivalvia) after microscopic analysis indicated that sex ratio M:F=1:06:1 and Potomida littoralis F:M=1:1,17 (P>0.05) not significant (Sereflisan et al., 2013), also Glycymeric nummariia (Crncevic, 2013), Anomalocardia brasiliiana (F:M=1:1,2) (Luz and Boehs, 2011).

In April, when the surface water temperature reached 17°C, 6.3% of the female oysters and 37.5% of male oysters were at an early development stage, exhibiting early vitellogenic oocytes and spermatogonia (Kim et al., 2010). Food supply in this period that used the amount of carbohydrate at a minimum (Shafakatullah, 2013). Different from Mytilus edulis, that substrate in the water column, more active in sorting particle while spawning period (Espinosa and Allam, 2013). Shell population which was highly abundant in the and more tourists (December-February). Santos and Boehs (2011). In case Clams from Ria de Aveiro seem to have 435 the ability to recover the reserves quickly after spawning when the SST and food 436 availability (chlorophyll) are still high (Matias et al., 2013).

Length and age-classes (Table 2) were significantly different from 1:1 only in Groups 1 and 3 (Harding et al., 2012). Maturity size close relate with length and weight (Avila-Poveda et al., 2009), where simping sizes that observe histology are 5.50 cm and 5.85 cm for maturity stage 2 (developing). According to Dharmraj et al. (2004), simping shellfish reach adulthood when length
70-100 mm. Based on Williams and Babcock (2004), simping shellfish reach maturity at the size of 6 cm.

Based on Chung et al. (2012), shell heights of sexually mature pen shells (size at 50% of group sexual maturity, GM50) that fitted to an exponential equation were 15.81 cm in females and 15.72 cm in males. Gomphina (Macridiscus veneriformis; Lamark, 1818) (Bivalvia: Veneridae) in the East Sea of Korea reach sexual maturity at size over 30.1 mm shell length (Kim et al., 2013). Annual reproduction of Lamellidens marginalis female-dominated at 70-80 mm shell length that happens from March to May (Gaikwad and Kamle, 2013). Other species like limpet Patella ferruginea change sex from females to males after spawning season at shell length were smaller than 70 mm (Guallart et al., 2013).

The gonad maturity mollusk divided into four categories based on the method proposed by Chipperfield (1953) in Setyobudiandi (2004). Observations gonad histological samples showed that the spawning A. inaequivalvis occurred during the summer period. The stages of gonad development from other species Anadara (i.e., A. granosa and A. rehombea) in various regions consist of any phase such as of rest, develop, grow, mature, and spawning stage, and its similar with Simping. Studies on A. subrenata, A. broughtoni and A. ursi in sub-tropical indicate that spawning begins from June to September (Morton, 1990; Moscose et al., 1992; Dzyuba and Maslennikova, 1982; Cruz, 1984; Yankson, 1982; Hadfield, and Anderson, 1988; Cruz, 1987; Gomez, 1985 in Sahin et al., 2006). According to Antsulevich et al. (1999) gonads of all mussel groups will mature in mid-July. This research also found of gonad mature in early or in December and January. (Aranda et al., 2003) Found that mussel spawns each semester and the peak of spawning season in July. In Kronjo, water gonad mature in December as indicating spawning after July.

Similar to Gomphina veneriformis from East Sea of Korea that early active stage star from December to March (Kim et al., 2013). Anadara antiquate gonad mature from May to August, females with developing, ripe, and spawning gonads found (Jahangir et al., 2014) and spawning season of Paphia malabarica in Dharmadon estuary star from November to December (Thomas, 2013). In the late-developing gonad, the follicles of males and females expanded and in areas between the mantle and the digestive gland. In the oogenic follicle, the oocytes are larger than in the preceding stage and are characterized by the long thin stalk that connects them to the follicular wall (Kim et al., 2010).

Freshwater mussel, resting period star from September dan October, and spawning season once a year (Callil et al., 2012). Ruditapes philippinarum (Bivalvia: Veneridae) in Western Korea early-stage maturity (shell lengths at 50% of sexual maturity (RM50) star from 15.1-20.0 mm and over 65% after 20.00 mm shell length (Chung et al., 2013), . . Indonaia caeruleus gametogenesis maturation takes place during summer and spawns during winter in September to March (Vedpathak and Pandit, 2010). And then also these species release fewer oocytes at 30°C compared to 20 °C temperature.

The essential environmental parameter effect to spawning is temperature and distribution geographic of biota. In Kronjo, this parameter has a significant effect on gonad maturity. Peters et al. (1994) researched in tropical water found the development (gonad genesis) for Crassostrea virginica 6-10 weeks after the first spawning. Pradina (1994) state the spawning occurs twice or more a year because at the anterior oocyst developed, posterior oocyst posterior at mature phase. Gonad mature phase, simping granule cell smaller than Lola (Trochus sp.) that granule content from 180-200 cell. In Korean waters. As shown in Figure 5, S. kegaki initiated oogenesis in April when
the water temperature reached 16.5°C and first partially spawned individuals were observed in July when the water temperature reached 26.7°C. (Kim et al., 2010).

CONCLUSIONS

Gonad development for simping shell, generally found in 4 stages, with nine substage maturity. Stage mature under 5 cm categories as an undeveloped stage and difficult to separate between males and females. The sex ratio for the simping population similar to another country ei 1:1 (one female: one male). Spawning stage dominant in an adult stage that found in a coastal area that influence by environmental water quality.

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