

Captive breeding of the freshwater prawn Macrobrachium pilimanus (De Man, 1879): biological performances

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Abstract. *Macrobrachium pilimanus* (De Man, 1879) is one of the economically important freshwater prawns in Indonesia which experienced a population decline, rendering the importance of conducting studies related to captive breeding. The present study aimed to explore the feasibility of *Macrobrachium pilimanus* collected from Lekso River in East Java, Indonesia to be raised under captive conditions as a first step towards its captive rearing. Some biological aspects, including the morphology, growth and reproduction were observed. Morphologically, *Macrobrachium pilimanus* in the present study matched with the *Macrobrachium pilimanus* species descriptions provided by previous researchers. The newly spawned eggs were black, with length diameter of 1.45–1.65 mm and width diameter of 1.10–1.25 mm, consisted of around 30–70 eggs per female prawn. The egg incubation lasted around 36–38 days, and the newly hatched larvae were 5.10–5.55 mm long, exhibiting a completely abbreviated larval development. During captivity, it displayed a positive allometric growth pattern with the specific growth rate of 3.06–4.34% per-day and high survival rate. Sexual maturity was reached within six to seven months, and spawned repeatedly within two to three weeks after eggs hatched. These results revealed that wild *Macrobrachium pilimanus* could be spawned and raised in captivity with quite satisfying performances.

Key Words: growth, morphology, reproduction.

Introduction. Freshwater prawns of the genus Macrobrachium are important food commodities for local communities in Indonesia. Among the species of genus Macrobrachium existing in Indonesia, Macrobrachium rosenbergii (De Man, 1879), Macrobrachium sintangense (De Man, 1898) and Macrobrachium pilimanus (De Man, 1879) are considered as the most economically important species (Djajadiredja & Sachlan 1956; Holthuis 1980), for which the commercial fisheries of those species have potential to be developed. Among these species, the *M. rosenbergii* has long been highly appreciated by Indonesian communities as a source of food (Djajadiredja & Sachlan 1956; Holthuis 1980), and its aquaculture has been well established, as well as recent breeding programs (Khasani et al 2018; Khasani & Sopian 2021). The studies of M. sintangense related to aquaculture purpose in Indonesia have also been conducted (Sabar 1979; Wowor 1983, 1985; Said & Maghfiroh 2012; Said & Mayasari 2016). On the other hand, the study of *M. pilimanus* performances in aquaculture in Indonesia is still scarce. Until recently, most studies of *M. pilimanus* in Indonesia have focused on the taxonomic status (Wowor et al 2009; Trijoko et al 2015; Eprilurahman et al 2021), biological aspects in the natural habitats (Fidhiany 1985; Hernawati et al 2013; Prodhiana et al 2022), and as well as its behavioural pattern in a laboratory condition (Dwiyanto et al 2023).

Macrobrachium pilimanus is a small prawn found in freshwater riverine systems of Malayan Peninsula and larger Sunda Islands (Sumatra, Java, Kalimantan) (Holthuis 1950; Johnson 1963; Chace & Bruce 1993). This species commonly inhabits flowing to fast flowing unpolluted hill streams and rivers with sandy and rocky substrates, as well as with leaf litter, but without dense vegetation (Johnson 1963; Ou & Yeo 1995). Recently, the population of *M. pilimanus* in several Indonesian regions has declined, particularly that of Java. In Central Java regions, this species has declined, with only two individuals being found by Hernawati et al (2013) during the study in Cijalu River of Cilacap Regency and then Prodhiana et al (2022) only found 150 individuals during two months of study in Mengaji River of Banyumas Regency. In Yogyakarta regions, the *M. pilimanus* population has also declined, where Trijoko et al (2015) only found 19 individuals during their study in Opak River, and Eprilurahman et al (2021) had difficulty in obtaining an adequate number of the representative specimens (finding only 11 specimens) in Opak River, Winongo River and Sempor River. In East Java, particularly in the western region of Malang Regency, the *M. pilimanus* population has also declined. Currently this species is difficult to be found in most rivers, with limited populations existing only in certain upstream rivers and near water springs (personal observation).

The decline in *M. pilimanus* populations seems to be caused primarily due to the overfishing. In East Java, M. pilimanus is often caught using electric fishing method (personal observation), which are known to be a non-ecofriendly fishing gear. This method allows large numbers of *M. pilimanus* to be easily caught in a short period from the rivers when they are available, while a new recruitment of *M. pilimanus* takes a long time. The overfishing is driven by the high demand for *M. pilimanus* as a food source. The fresh caught *M. pilimanus* is quite appreciated by fish consumers, with a high price on traditional markets. Recently in Malang Regency, its price per-kg is around IDR 50,000-60,000, about twice more expensive than that of either tilapias (Oreochromis spp.) or African catfish (*Clarias gariepinus*), which are the most common freshwater food fishes in fish markets (personal observation). Given the importance of *M. pilimanus* as a food source and also as an effort dealing with the need of its conservation in the future, captive rearing of this species must be researched. However, there is currently no available information on its aquaculture. The present study aimed to explore the feasibility of this freshwater prawn to be raised in captive conditions as a first step towards its culture. Some biological aspects of *M. pilimanus* were observed during the course of the present study, including the morphology, growth and reproduction.

Material and Method. The present study was conducted from June 2023 through May 2024. The collection of *Macrobrachium pilimanus* was conducted in the Lekso River waters (a tributary of Brantas River), located on the border between Malang Regency and Blitar Regency of East Java Province, Indonesia. These prawns were caught in the edge sides of the river using a scoop net during June to July 2023. The 300 collected individuals were then transferred into a private fish rearing facility in Jambuwer Village of Malang Regency. Upon arriving at the rearing facility, the body weight was determined using a digital balance, while the body length (measured from posterior margin of the eye to end of the telson), total length (measured from tip of the rostrum to end of the telson) and other morphometric characters were measured using a digital calliper. The collected individuals were verified following the method of morphological characterization of *M. pilimanus* species reported in previous studies (De Man 1879; Holthuis 1950; Johnson 1963; Chace & Bruce 1993; Wowor et al 2004).

The body length and body weight were then used to analyse the length-weight relationship and condition factor. The adult male and female *M. pilimanus* (assigned by the presence of enlarged brood chamber in females and the bigger chelae and body size for the males, Figure 1a) were selected for further rearing. The females carrying eggs were separated and the eggs were carefully removed from the brood chamber using a spatula and then counted to determine the egg number (brooding fecundity). The size (diameter) of the eggs was also measured under a stereo microscope. Those females were then stocked together with the males (30 females and 30 males) in a concrete pond $(8.0 \times 0.5 \times 0.2 \text{ m})$ for further rearing. The pond was divided into four parts each using a net screen of 2 mm mesh size. The pond was filled with a clear water derived from a mountain spring, with a low rate continuous water discharge (about 0.3–0.5 litre perminute) for water circulation. Several (about equal to the number of the individuals reared) pieces of plastic pipe with 18 mm (dedicated for females) and 22 mm (dedicated for males) diameter of about 6–8 cm long were added into the pond as the nests. Several

pieces of aquatic plants (*Hydrilla* spp.) were also placed into the pond. During the rearing period, a commercial sinking artificial feed sized 0.5-1.1 mm with the crude protein content of 40% was given twice daily (in the morning and at night) at a rate of about 1% biomass per-day.

During the rearing period, a daily inspection was undertaken to observe the spawning occurrences. When the spawning took place (Figure 1b), the female was caught and then transferred into an aquarium $(30 \times 20 \times 20 \text{ cm})$ equipped with a low rate circulated water discharge (around 10-15 water drops per-second) for the egg incubation process. A piece plastic pipe was added into the aquarium as a nest. The feeding management during the egg incubation was same as previously applied in the pond rearing. When the whole incubated eggs hatched, the female brooder was removed from the aquarium and transferred back into the concrete pond for the next spawning. The newly hatched larvae were then reared within its respective incubation aquarium. An aquatic plant (Hydrilla spp.) was added into the aquarium as a shelter. A commercial sinking artificial feed sized 0.5-1.1 mm with the crude protein content of 40% was offered until apparent to satiation in the morning and night for two months of rearing. After that, the juveniles were transferred to the concrete pond for four to five months of the grow-out rearing until reaching the adult (mature) stage. The feeding management during the grow-out phase within the concrete pond was generally the same as applied in the previous rearing phase.



Figure 1. Adult male (upper) and female (lower) *Macrobrachium pilimanus* collected from Lekso River, East Java, Indonesia (a) and a spawning occurrence in captivity (b) (original images).

During the rearing period within the aquarium and pond, a monthly sampling of all test specimens for the total length, body length and body weight was conducted to determine the growth performance of *M. pilimanus* under captive conditions. When the reared *M. pilimanus* reached the mature stage, assigned by the spawning occurrences or the presences of the eggs within the female's brood chamber (ovigerous female), the body length and body weight were recorded to determine the size at first maturity. When the number of females carrying eggs within one week period reached 5–10 individuals, the eggs from each individual were carefully removed from the brood chamber and counted to determine the egg number. The egg diameter was also measured. Those females were returned to the concrete pond for further rearing and re-maturation process. When more than 50% of those females had spawned again, the re-maturation time interval was recorded. The body length and body weight data during the grow-out period and the subsequent rearing period of the adult phase were used for the length-weight relationship and the condition factor analysis.

During those rearing periods, water quality in the pond and aquarium was maintained in good condition. The water quality parameters of water in the Lekso River was assessed weekly between June and July 2023, whereas the water quality of the maintenance media was evaluated monthly from June 2023 to May 2024. Measurements were carried out in the morning (06:00-07:00 AM) and in the afternoon (02:00-03:00

PM). A thermometer was used to determine the water temperature, while a pocket pH meter was employed to measure the pH level. Dissolved oxygen, ammonia, and nitrite concentrations were analyzed using a commercial water quality tester kit (Tetra, Spectrum Brands INC, USA). Water quality parameters are presented in Table 1. Except for the current, water quality parameters in the captive condition and river waters were similar.

Table 1

Water quality parameters of the pond and aquarium for the rearing of *Macrobrachium pilimanus* and the Lekso River, East Java, Indonesia where specimens were collected

Parameters	Pond and aquarium	Lekso River
Current (m s ⁻¹)	stagnant	0.1-0.4
Turbidity (m)	clear water	0.2-0.3
Temperature (°C)	21-27	25–27
pH	6.5-6.8	6.5-6.7
Dissolved oxygen (mg L ⁻¹)	8-11	8-11
Ammonia (mg L ⁻¹)	<0.25	<0.25
Nitrite (mg L ⁻¹)	<0.25	<0.25

Data analysis. The obtained data were analyzed descriptively to describe the growth and reproduction patterns of prawn (*M. pilimanus*). The mean and standard deviation of each parameter were calculated and presented in tables and graphs. The relationship between body length and body weight was analyzed using simple regression to illustrate the population growth pattern.

Results

Morphological characteristics. The individuals of *Macrobrachium pilimanus* collected from the Lekso River in the present study were morphologically characterized by the rostrum that never extended beyond the end of the antennular peduncle, the upper margin was slightly convex with 9–13 rostral teeth, 3–4 of them were postorbital, and the ventral rostral teeth numbered 1–3, the second pereiopod had moderately inflated merus, the carpus was very short and markedly inflated (referred to as cup shaped carpus), the chela was entirely covered with long, dense, velvety hairs, the finger was more or less as long as the palm, almost not gaping, the cutting edge had 15–17 teeth, with the tips crossing one another. The chelae of the second pairs of pereiopods were unequal in size, but the shape was rather similar, either in males or females. In some individuals the right chela was larger, while in others the left one was larger. The biometric morphological characteristic of the adult *M. pilimanus* collected from the Lekso River is presented in Table 2.

Table 2

Biometric morphological characteristics of *Macrobrachium pilimanus* collected from the Lekso River, East Java, Indonesia

Characters	Values
Number of rostral teeth	(3-4)+(6-9)/1-3
Number of cutting teeth on finger of the second pereiopod	15-17
Length ratio of carpus to merus of the second pereiopod	0.50-0.64
Length ratio of carpus to palm of the second pereiopod	0.40-0.59
Length ratio of carpus to chela of the second pereiopod	0.20-0.28
Length ratio of finger to palm of the second pereiopod	0.92-1.31

Egg and larval characteristics. About two to three weeks of rearing period after its eggs were removed for the brooding fecundity determination in the present study, the female brooders of *M. pilimanus* originated from the Lekso River had spawned and were

carrying the eggs in brood chamber. The egg incubation period was around 36–38 days at a water temperature of $21-26^{\circ}$ C. During the early incubation period of approximately two weeks, the eggs had a blackish colour appearance (Figure 2a), while in the subsequent period it gradually became translucent light brown in colour (Figure 2b). The eggs were oval, with the length diameter averaging 1.55 ± 0.05 mm (1.45-1.65 mm) and the width diameter averaging 1.17 ± 0.03 mm (1.10-1.25 mm) for the black coloured eggs. The brown coloured (eyed) eggs were bigger (enlarged), with the length diameter of around 2.11 ± 0.09 mm (1.95-2.25 mm) and the width diameter of 1.46 ± 0.07 mm (1.30-1.70 mm).

Upon hatching, the larvae of *M. pilimanus* had a general morphology almost resembling the adult, except for the tail which has not differentiated yet (Figure 3). The body length of the newly hatched larvae was around 5.34 ± 0.13 mm (5.10-5.55 mm). Those larvae were not planktonic (free swimming), but exhibited a benthic activity, similar to adult *M. pilimanus*. In the following days, the tail started to differentiate, forming four uropods and a wide, large telson. At the ninth day, the tip of the telson has pointed in form, almost resembling the adult.

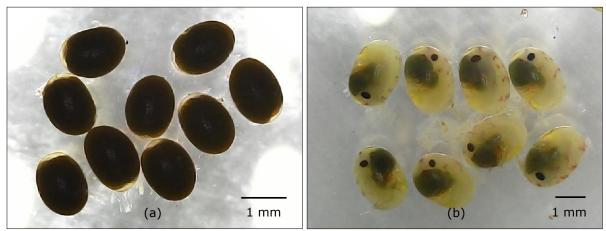


Figure 2. The eggs of *Macrobrachium pilimanus* in early (a) and late (b) incubation periods with blackish and translucent brownish appearances, respectively (original images).

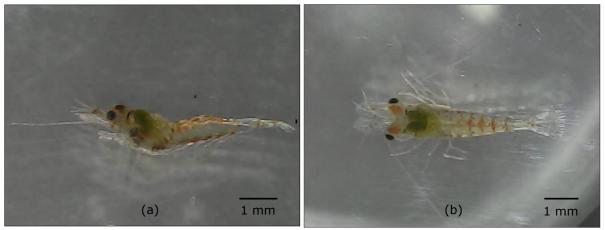


Figure 3. The newly hatched larva of *Macrobrachium pilimanus* from lateral view (a) and dorsal view (b) (original images).

Survival and growth performance. The *M. pilimanus* resulted from the spawnings in captive conditions in the present study exhibited variable growth performances. Each batch of offsprings had different specific growth rate during six or seven months of rearing, which ranged 3.06–4.34% per-day (mean final body weight ranged between 0.72–0.92 g). Offsprings with the lowest specific growth rate reached the sexual maturity (adult stage) prior to the seventh month, while those with the highest specific growth rate reached the sexual maturity within the sixth month. The monthly total length data of

the *M. pilimanus* raised in captivity in the present study with the lowest and highest specific growth rates are presented in Figure 4.

During two months of rearing in the aquarium, the larval mortalities were observed during the first week, resulting in survival rates of around 70–80%. In the subsequent rearing period, the survival rate was high (ranged 86.36–100%). Throughout the rearing in both the aquarium and concrete pond in the present study, the *M. pilimanus* did not exhibit cannibalistic behaviour, even during the moulting process. Rare cannibalism only occurred when there was a dead or a very weak dying individual due to a sickness condition. However, they exhibited agonistic behaviour related to the territory occupation and foraging the feed.

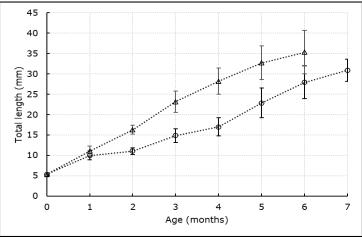


Figure 4. Monthly total length of *Macrobrachium pilimanus* raised in captivity with lowest (\bigcirc) and highest (\triangle) specific growth rates.

Length-weight relationship and condition factor. Since the body weight of *M. pilimanus* raised in captivity (within ten months) in the present study did not reach two grams, the individuals of *M. pilimanus* collected from Lekso River with body weight over two grams were firstly excluded from the analysis of length-weight relationship. The length-weight relationship of *M. pilimanus* raised in captivity exhibited an allometric pattern (Figure 5a), similar to that of *M. pilimanus* collected from Lekso River (Figure 5b), with the b values of respectively 3.23 and 3.36 (the coefficients of determination R² were 0.93 and 0.96, respectively). These allometric growth pattern revealed that the weight increase was higher than the length increase. In addition, when the larger individuals (more than two grams) collected from Lekso River were included in the length-weight relationship analysis, then the value of b was higher, at 3.45 (the graph was not shown).

During the rearing in the aquarium and concrete pond, the condition factor of *M. pilimanus* in the present study was around 2.73 ± 0.44 (ranged 1.74-3.97). The condition factor of *M. pilimanus* collected from Lekso River was somewhat similar, at around 2.58 ± 0.38 (ranged 1.71-4.17).

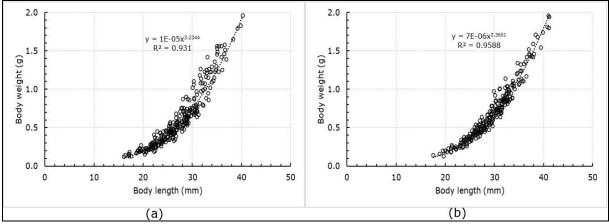


Figure 5. Length-weight relationship of *Macrobrachium pilimanus* raised in captivity (a) and that of specimens when harvested from the Lekso River, East Java, Indonesia (b).

Reproductive performance. The *M. pilimanus* obtained from spawning in captive conditions in the present study attained the first maturity (assigned by the spawning activity occurrence or the presence of eggs within the female's brood chamber) at the sixth to seventh month post hatching. The size at first maturity of female *M. pilimanus* observed was around 0.65 ± 0.12 g (approximately 28.74 ± 1.70 mm of body length), while for the male it was around 1.32 ± 0.35 g (around 34.90 ± 2.39 mm of body length).

Based on observations during the course of the present study, the spawning of the *M. pilimanus* in the captive condition might take place throughout year, with the peak spawning season occurring during the dry season (from June through October 2023). Even more, around 50% of the females were still carrying eggs in their brood chamber during the peak of rainy season (from December 2023 through March 2024). The spawning of *M. pilimanus* in the natural habitat was observed to exhibit more or less a similar pattern. However, the females carrying eggs within the brood chamber were rarely observed in the natural habitat during the peak of rainy season.

Egg number within female's brood chamber (brooding fecundity) from spawning in captive conditions in present study (in both dry and rainy seasons) ranged 32–68 eggs (48 ± 11 eggs). Compared to the brooding fecundity of female brooders of *M. pilimanus* collected from the Lekso River (31-89 eggs, 61 ± 15 eggs on average), brooding fecundity in captivity was somewhat lower. Although, on a special occasions the brooding fecundity of females raised in the captive conditions could reach 90-95 eggs, similar to that of the females collected from the Lekso River. In addition, some individuals, whether raised in captivity or collected from the Lekso River, were occasionally observed in carrying only a few incubated eggs within their brood chamber (less than 20 eggs).

Though the spawning of *M. pilimanus* was found to easily occur in the captive conditions, the hatching rate was low. Considering the number of incubated eggs within brood chamber was around 50 eggs, the hatching rate of incubated *M. pilimanus* eggs in the present study was roughly accounted for 60–75%. During the egg incubation process, some eggs in the egg chamber were shed, reducing the eggs number. Moreover, if there was a disturbance from other prawns, the number of eggs remaining in the egg chamber decreased dramatically, resulting in a very low egg hatching rate (around 20%). In addition, the study found that some prawns lost all of their eggs, leading to no successful hatching. This phenomenon was evident in both wild-caught and captive-raised prawn populations.

Discussion. Although there was no available report yet on the breeding efforts for aquaculture purpose of the *Macrobrachium pilimanus*, the larval development of *M. pilimanus* reared in laboratory conditions has been reported in detail for a long time ago from Malaysia by Chong and Khoo (1987a). In general, larval development of *M. pilimanus* in the present study agreed with that of previously reported by Chong and Khoo (1987a). As such in that of *M. pilimanus* from Malaysia, the larval development of *M. pilimanus* in the present study belonged to the completely abbreviated type, which

lacked any free swimming larval stage. As previously reviewed, three basic types of larval developmental patterns in freshwater prawns have been recognised, namely prolonged or normal type, partially abbreviated type, and completely abbreviated type (Jalihal et al 1993). The larval development of *Macrobrachium malayanum* (Roux, 1934) and *Macrobrachium ahkowi* Chong & Khoo 1987 from Malaysia were also reported to belong to the completely abbreviated type (Chong & Khoo 1987b,c,d). The freshwater prawn in Indonesia whose larval development belonged to the partially abbreviated type (with free swimming in the first larval stage) was *M. sintangense* (Sabar 1979). The larval development of *M. rosenbergii* belonged to the prolonged or normal type, with free swimming for all the larval stages (Ling & Merican 1961). The *Macrobrachium lanchesteri* (De Man, 1911), a freshwater prawn native to Thailand and the Malayan Peninsula (Johnson 1963; Holthuis 1980; Chong & Khoo 1988; Cai et al 2004) which has recently been reported as an alien invasive species in several Indonesian waters (Aprila et al 2020; Maulina et al 2020; Safira & Annawaty 2023), its larval development also belonged to the prolonged type (Chong & Khoo 1988; Wong 1994).

While the larval development of *M. pilimanus* in the present study corresponded to that of *M. pilimanus* reported from Malaysia (Chong & Khoo 1987a), their egg colours were different. The newly spawned eqg of *M. pilimanus* from Malaysia was light green and the colour unchanged prior to hatching (Chong & Khoo 1987a), whereas the newly spawned egg of *M. pilimanus* observed in the present study was black and turned into translucent light brown prior to hatching. It was yet unknown whether this egg colour difference should exactly reflect the species difference or a form of intraspecific variations. However, different species of freshwater prawns were reported in having different eqg colour. The colour of newly spawned eqg of *M. malayanum* was dark green (Chong & Khoo 1987b), whereas for *M. sintangense* it was yellowish green (Sabar 1979), and M. rosenbergii had orange eggs (Ling & Merican 1961; Ang & Law 1991). While the egg colour of M. pilimanus in the present study differed from that of M. pilimanus reported from Malaysia by Chong & Khoo (1987a), it should be noted however that morphological characteristics of *M. pilimanus* in the present study corresponded to the original description of *M. pilimanus* given by De Man (1879), as well as the species description and identification key reported in the subsequent studies (De Man 1892, 1898; Holthuis 1950; Johnson 1963; Chace & Bruce 1993; Cai et al 2004; Wowor & Choy 2001; Wowor et al 2004; Wowor 2010).

In the first descriptions of *M. pilimanus*, De Man (1879, 1892) realized that the *M.* pilimanus description might compose of several species with shared similar morphological characteristics. Then, the subsequent researchers (Holthuis 1950; Johnson 1963) treated it as a species group or species complex. Among the freshwater prawns species shared similar general morphology with *M. pilimanus*, as well as the species within *M. pilimanus* species group, M. pilimanus, M. leptodactylus (De Man, 1892), M. poeti Holthuis, 1984, and *M. empulipke* Wowor 2010 which existed in Java. Compared to the morphological data of those species that occurred in Java, the morphology of *M. pilimanus* observed in the present study was closely similar to that of *M. pilimanus* in terms of rostrum (the rostral teeth count was (3-4)+(6-9)/1-3 versus (3-5)+(6-10)/1-3 and the morphology of second pereiopods, specifically the number of cutting teeth on the finger (15-17 versus 13–17) and the length ratios of the carpus to merus (0.50–0.64 versus 0.43– 0.67), the carpus to palm (0.40-0.59 versus less than 0.5), and the finger to palm (0.92-1.31 versus 0.75-1.12) (De Man 1879; Holthuis 1950; Chace & Bruce 1993; Cai et al 2004; Wowor 2010). When comparing *M. leptodactylus* with *M. pilimanus* in the present study, M. leptodactylus exhibited greater number of postorbital rostral teeth (5-6) and different morphology of the second pereiopods, including fewer number of cutting teeth on the finger (15), longer carpus with long conical or sub-cylindrical shape (length ratio of the carpus to merus was 0.78-0.90), and a non-inflated merus (De Man 1892; Johnson 1963; Cai et al 2004; Wowor 2010). Compared to M. poeti, the M. poeti differed from *M. pilimanus* in the present study in having longer rostrum (extending beyond the antennular peduncle) with only single ventral rostral teeth, more slender second pereiopods with sub-cylindrical merus and carpus (length ratios of the palm to carpus, the chela to carpus, the finger to palm were respectively 1.50, 3, 1.33), fewer cutting

teeth on the finger of second pereiopods (only several), and the chelae of second pereiopods lacking dense velvety hairs (Holthuis 1984; Chace & Bruce 1993). From *M. empulipke*, the second pereiopods of *M. pilimanus* in the present study differed in having a greter number of cutting teeth on the finger (12–15 in *M. empulipke*), a shorter carpus (length ratios of the carpus to merus and the carpus to palm in *M. empulipke* were respectively 0.62–0.73 and 0.63–0.91, with conical shape), and a shorter finger (the length ratio of the finger to palm in *M. empulipke* was 1.40–2.01) (Wowor 2010).

Besides the colour, the eqq size of *M. pilimanus* in the present study was also slightly larger (average diameter of 1.55 mm by 1.17 mm for the newly spawned eggs and 2.11 mm by 1.46 mm for the eyed eggs) than that of *M. pilimanus* from Malaysia (1.85 mm by 1.20 mm) (Chong & Khoo 1987a) and Brunei Darussalam (1.7 mm by 1.2 mm) (Wowor & Choy 2001), while the egg diameter of *M. pilimanus* from Sumatra (2 mm by 1.5–1.75 mm) (Cai et al 2004) was similar. Previously, Holthuis (1950) observed that egg diameter of *M. pilimanus* was 1.2–1.8 mm (presumably, it was the range of the width and length egg's diameters), being roughly similar to that of *M. pilimanus* from Malaysia reported by Chong & Khoo (1987a). De Man (1892) observed the length diameter of *M. pilimanus'* eggs from Sumatra up to 2 mm, whereas those from Kalimantan measured 1.8 mm by 1.1-1.2 mm (De Man 1898). However, it should be noted that those reported *M. pilimanus* were a species group (species complex) composed from several new species described later. The variation in egg size might reflect species differences or environmental factors in their respective habitats, as seen in egg size difference of *M. malayanum* from Malaysia (1.75 mm by 1.35 mm) (Chong & Khoo 1987b) and Thailand (1.3 mm by 0.9 mm) (Cai et al 2004). The egg size of M. sintangense from several localities in Thailand was also found to be vary from 0.9 mm by 0.6 mm to as big as 1.5 mm by 1.0 mm (Cai et al 2004), whereas the egg size of M. sintangense from West Java (1.4-1.5 mm by 1.0-1.1 mm) was more or less similar to that of *M. sintangense* from West Kalimantan (1.6 mm by 1.2 mm) (De Man 1898), as well as to that of *M. sintangense* from Sabah, Malaysia (1.4 mm by 1.0 mm) (Ng 1994), and Holthuis (1950) also observed that egg diameter of *M. sintangense* from several localities in Sumatra, Java, and Kalimantan ranged between 1.0-1.5 mm. On the other hand, the egg sizes of *M. lanchesteri* from several localities were more or less similar, for which its length and width diameters was 0.9 mm by 0.7 mm for *M. lanchesteri* from Brunei Darusssalam (Wowor & Choy 2001), 0.9 mm by 0.6 mm for *M. lanchesteri* from Laos (Hanamura et al 2011), 0.8-1.1 mm by 0.6-0.8 mm for *M. lanchesteri* from Thailand (Cai et al 2004), and ranged 0.8–1.0 mm by 0.6–0.8 mm for *M. lanchesteri* from Myanmar (Cai & Ng 2002; Phone et al 2005; Saung & Htwe 2010).

Together with other freshwater prawn species belonging to the highly abbreviated type of larval development, the egg size of *M. pilimanus* was considerable bigger than that of other freshwater prawn species with either prolonged or partially abbreviated larval development, but the egg number were fewer. Unfortunately, there were no specific reports on the exact egg number of *M. pilimanus*. Previously, De Man (1892) and Holthuis (1950) only stated that the eqg number of *M. pilimanus* was small. Based on the observations in the present study, the egg number of *M. pilimanus* collected from Lekso River was around 30–90 eggs, while for the *M. pilimanus* raised in a captivity was around 30-70 eggs. The *M. sintangense* and *M. lanchesteri* have been reported in having more numerous eggs, of respectively around 90-230 eggs (Sabar 1979) and around 60-550 eggs (Phone et al 2005; Saung & Htwe 2010), whereas M. rosenbergii had much more eggs of up to 100,000 eggs (Wickins & Beard 1974; New 1990) or even up to 150,000 eggs (Ling & Merican 1961; Habashy 2013). Although its egg number was slightly fewer, the *M. pilimanus* raised in a captive condition in the present study proved to be capable of reaching the mature stage and could repeatedly spawn throughout the year. However, the hatching success was still low, as it has also been observed in the captive breeding of *M. sintangense* (Said & Mayasari 2016). In some females, the number of incubated eggs highly decreased during the incubation process, and even in several cases, all of the eggs were lost. The decreasing number of incubated eggs during the incubation process was also reported in *M. sintangense* (Said & Mayasari 2016), in *M. lanchesteri* (Saung & Htwe 2010), as well as in M. rosenbergii (Ang & Law 1991; Cavalli et al 2001; Thanh et al

2009). Hopefully, this low hatching success can be resolved in the future through a refinement of the environmental conditions required for the egg incubation.

The egg incubation process of *M. pilimanus* in the present study took place within a notably long period, ranging between 37–39 days. In comparison, the egg incubation period of *M. lanchesteri* was shorter, of around one month (Phone et al 2005), whereas the egg incubation in *M. sintangense* took place within 22–25 days (Sabar 1979; Said & Mayasari 2016), and the eggs of *M. rosenbergii* hatched in around 19 days or between 18–24 days (Ling & Merican 1961; Wickins & Beard 1974; New 1990; Habashy 2013). Regarding the egg incubation period in *M. pilimanus*, there was no report yet from other studies.

Though the egg incubation period was longer, larval development period of *M. pilimanus* and other freshwater prawns which its larval development belonged to the highly abbreviated types were remarkably shorter than other freshwater prawns with either prolonged or partially abbreviated larval development. As such that of *M. pilimanus* from Malaysia (Chong & Khoo 1987a), the larval stage of *M. pilimanus* in the present study also lasted within five to six days after hatching. Similarly, the larval stage period of *M. malayanum* lasted around five days (Chong & Khoo 1987b). In contrast, the larval stage period of *M. sintangense* was longer, around 15 days (Sabar 1979), whereas larval stages period of *M. rosenbergii* and *M. lanchesteri* were much longer, lasting around 28 (New 1990) or 30 days (Wong 1994; Saung et al 2010), respectively.

Due to its large eggs, the newly hatched larvae of *M. pilimanus* in the present study were also considerably large (the total length ranged between 5.10–5.55 mm). Since the egg size of *M. pilimanus* in Malaysia was relatively smaller than that of *M. pilimanus* in the present study, its newly hatched larvae were also slightly smaller, of around 4.25 mm of body length (Chong & Khoo 1987a). Larval size of another freshwater prawns with completely abbreviated larval development in Malaysia, *M. malayanum* and *M. ahkowi* were also large, due to their large eggs, with the body length of newly hatched larvae were respectively around 4.30 mm and 4.15 mm (Chong & Khoo 1987b,c,d). Similarly, the larval size of *M. sintangense* was large, with the total length of newly hatched larvae at around 5.0 mm (Sabar 1979). On the other hand, the larval size of *M. lanchesteri* was small, which the body length of newly hatched larvae reported by Chong & Khoo (1988) to be 3.25 mm, whereas according to Wong (1994) its total length was 2.88 mm. Furthermore, the larval size of *M. rosenbergii* was even smaller, ranging between 1.9–2.1 mm in total length (Wickins & Beard 1974).

Within six or seven months of the larval rearing, nursery and grow-out phases in captivity, *M. pilimanus* in the present study exhibited a guite high growth performance, with a positive allometric growth pattern of length-weight relationship, and a good condition factor at the end of the rearing period. Unfortunately, there was no report yet regarding the growth performance, length-weight relationship and condition factor of M. *pilimanus* from other studies. Likewise, the studies on growth, length-weight relationship and condition factor of other Indonesian freshwater prawns species were also scarce, except for *M. rosenbergii* as the most widely cultured freshwater prawn species. Regarding the growth performance, the specific growth rate of *M. pilimanus* in the present study (ranged 3.06-4.34% per-day) was comparable to that of the cultured M. rosenbergii, for which was around 1.5-2.5% per-day (Segal & Roe 1975; Davassi 2011; Khasani et al 2023). The specific growth rate of a laboratory reared *M. sintangense* juveniles previously collected from natural waters around Bogor, West Java (Wowor 1983) was approximately 1.5% per-day during ten weeks of rearing (calculated from the body weight data). Later, the *M. sintangense* resulted from the captive breeding of the brooders collected from West Java and Central Java waters were reported in having relatively high specific growth rates during 20 and 24 weeks of rearing period (Said & Maghfiroh 2012), of about 3.7–4.1% per-day (calculated from the body weight data). For the length-weight relationship, both captive and wild *M. pilimanus* in the present study exhibited a positive allometric growth pattern. The *M. rosenbergii* reared within culture conditions were also reported to exhibit a positive allometric growth pattern (Wang 1985; Sampaio & Valenti 1996; Yang et al 2016) or nearly isometric growth pattern (Kunda et al 2008), whereas those in the natural habitats were reported to exhibit either negative

or positive allometric growth patterns (Rao 1967; Rajeevan et al 2018; Indarjo et al 2021). The length-weight relationship of *M. lanchesteri* in Ulu Ngarak River, West Kalimantan exhibited a negative allometric growth pattern (Hurriyani et al 2022). In term of the condition factor, the *M. pilimanus* raised in captive conditions in the present study exhibited high value, over one (around 2.7), suggesting the good physiological condition. Similarly, the *M. pilimanus* collected from Lekso River also exhibited high values (around 2.6). These condition factor values were generally higher than that of other values reported from other freshwater prawn species. In general, the condition factor of *M. rosenbergii* was around 0.5–2 in both natural and culture conditions (Rao 1967; Kunda et al 2008; Yang et al 2016; Rajeevan et al 2018; Indarjo et al 2021). The condition factor of *M. lanchesteri* in Ulu Ngarak River, West Kalimantan was 1.01–1.02 (Hurriyani et al 2022).

During the rearing in captivity, the survival rate of *M. pilimanus* in the present study was high, comparable to that of the captive breeding of *M. sintangense* collected from several localities in Java, Sumatra, and Kalimantan (Said & Maghfiroh 2012; Said & Mayasari 2016). While the cannibalism commonly occurs for prawns and can cause substantial loss of the number prawns cultured (causing low survival rates), the rate of cannibalism depends on various factors and it is species specific (Romano & Zheng 2016). The *M. pilimanus* raised in captivity in the present study did not exhibit high degree of cannibalism occurences, even among post-moulting individuals, thus resulting in high survival. Though *M. pilimanus* in the present study exhibited an agonistic behaviour, particularly when foraging the feed, the cannibalism only occured in cases of the presence of weak, dying (due to sickness) or dead individuals. A similar cannibalistic behaviour was found in *M. acanthurus* (Wiegmann, 1836) from Jamaica (Choudhury 1971). In contrast, *M. rosenbergii* has been well known as a very cannibalstic species (Segal & Roe 1975; Nair et al 1996; Juneta-Nor et al 2020). Cannibalism during moulting was also reported for *M. sintangense* (Sabar 1979).

Prior to the sixth or seventh months post-hatching, the *M. pilimanus* raised in captivity in the present study reached sexual maturity, and was capable of spawning for the first time. The smallest size of females at the first maturity was around 26 mm of body length and for males with large claws was around 29 mm of body length, while for the small males was around 18 mm of body length. Previously, De Man (1892) found that the female *M. pilimanus* with 20 mm of body length from Singkarak Lake in West Sumatra were already carrying eggs (ovigerous). Holthuis (1950) observed that the specimen of 16 mm from Singkarak Lake has also carried eggs, although the size of ovigerous females was generally about 40 mm. For M. lanchesteri, the minimum size of ovigerous female was found to be 25 mm of body length for the specimens from Thailand (Lanchester 1901), whereas that of from Myanmar was 6.3 mm of carapace length (Phone et al 2005). The minimum size of an ovigerous female *M. sintangense* in West Java waters was 7.35 mm of carapace length (Wowor 1985). It has been widely known that non fully grown females of several *Macrobrachium* species were found to be sexually mature (Holthuis 1950). In addition, this might also be the case for males, since the small males (smaller than the mature females, resembling the females morphology, not having large claws) which were much smaller than the dominant males of *M. pilimanus* in the present study, as well as for *M. rosenbergii* and other *Macrobrachium* species (Karplus & Barki 2018) were also observed of being capable of fertilizing the females.

After the eggs hatched, the females *M. pilimanus* in the present study were commonly observed to carry eggs again within two to three weeks, resulting in a spawning frequency of every 55–60 days. Compared to *M. rosenbergii*, the rematuration period of *M. pilimanus* in the present study was longer or slightly similar, but the spawning frequency was much less frequent. In captive conditions, a female *M. rosenbergii* was reported to spawn four times within 170 days (Wickins & Beard 1974) or five to seven times within 180 days (Cavalli et al 2001; Nhan et al 2009), which equal to about one to two weeks of rematuration period.

Conclusions. The *M. pilimanus* collected from Lekso River, East Java, Indonesia could be spawned and raised in captivity with quite satisfying performances, in terms of a simple

larval development, good growth performance, high survival and the spawning might occur throughout the year, although with only a small number eggs.

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Conflict of interest. The authors declare that there is no conflict of interest.

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