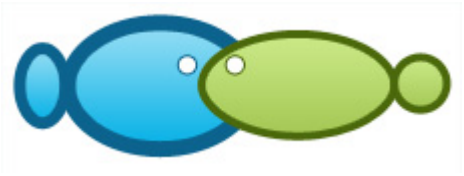

Domestication and selective breeding for producing fast growing and high meat quality of blue swimming crab (*Portunus pelagicus*)

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Abstract. Aquaculture has not yet made a significant contribution in meeting the market demand. This research aims to produce a superior *Portunus pelagicus* brood stock candidate through domestication with selective breeding for faster growth. The study was conducted over two years (2013-2014) at educational fish ponds of Hasanuddin University, located in Barru Regency, Province of South Sulawesi Indonesia. There are several steps of domestication: selecting wild crab stock (G0) generation, improving method for spawning, hatching, and larval rearing in hatchery, growing-out in brackish water pond for 90 days to select fast growing and high meat quality of brood stock (G1), repeating all steps until third generation of domesticated stock (G3) produced. The result showed that there was significantly increasing in carapace width from G1 to G3-stock ($p < 0.01$). The G3 carapace width growth reached 1.17 and 1.12 mm day^{-1}, while the G1 was 1.02 and 1.03 mm day^{-1} for female and male crab, respectively. There was no significant difference in protein, fat, and nitrogen-free between G0 and G3-crab. This domestication procedure producing high growth rate and good quality of crab meat can be applied in crab aquaculture.

Key Words: *Portunus pelagicus*, domestication, selective breeding, crab, growth.

Introduction. Blue swimming crab (*Portunus pelagicus*) is one of important fisheries commodities in the world including in Indonesia. This marine crab is demanded highly for domestic and overseas market and it is exploited by many countries (Kangas 2000; Svane & Hooper 2004; Sawusdee & Songrak 2009; Ehsan et al 2010; Mehanna et al 2013; Nieves et al 2013; Gadhavi et al 2013). Blue swimming crab is very popular as a seafood, delicious and exclusive in China and the United States, the two largest consumers of crab in the world. However, in recent years, the production of crabs has decreased both in number and size. Some research from a number of countries reported that there has been overexploitation of the crab resulting population declined (Johnston et al 2011; Mehanna et al 2013; Harris et al 2014; Kunsook et al 2014).

Aquaculture is one way to address the threat of crab population sustainability due to overexploitation. For example, in 2010 capture production of blue swimming crab was 200,000 tons, while aquaculture production was around 20 to 30 tons (FAO 2014). Data showed that there is still low contribution of crab aquaculture to the market demand. In Indonesia and many countries in the world, the crabs farming is still in early phase. There are still many problems in blue swimming crab aquaculture, such as cannibalism, low growth rate, low survival rate and low quality of meat especially when wild seeds of crab are used for growing-out. Some researchers reported that lower survival rate of larvae is still the main problem of blue swimming crab hatchery (Soundarapandian et al 2007), which is caused by various factors like disease (Govindasamy & Srinivasan 2012; Talpur et al 2011a, b), molting syndrome (Hamasaki et al 2002) and cannibalism (Soundarapandian et al 2007). Although, many efforts have been done to address these issues such as probiotics application for vibrio-pathogenic control and arrangement of natural diet amount and feeding time (Juwana et al 2010), but the mortality is still high and survival rate is unstable.

Growing blue swimming crab from hatchery rise seed which was from domesticated brood stock is one possible solution to overcome the problem of blue swimming crab aquaculture. Domestication is the acclimatization of an organism to a culture condition (Hoa 2009). The main process in domestication is repeated selection for producing the best quality generation in culture condition. Through the process of gradual adaptation and selection, the crab is able to adapt to the culture environment. Hence, the adapted crab easily produces high number and high quality offspring and the offspring is more able to survive than the undomesticated crab. In the cultivation condition, direct selection of animals that have the best behavior and rapid growth has been conducting on the many fish species such as Atlantic salmon (*Salmo salar*), rainbow trout (*Oncorhynchus mykiss*), Coho salmon (*Oncorhynchus kisutch*), tilapia (*Oreochromis mossambicus*), carp (*Cyprinus carpio*), channel cat fish (*Ictalurus punctatus*), and sea bream (*Sparus aurata*). Domestication through selective breeding has also been conducted on shrimp, oyster, scallops, and others. Selected species will grow better and become more resistant to disease and ultimately improve productivity (Gjedrem & Baranski 2009).

The aim of the study is to produce high quality broodstock through domestication that generating superior offspring of blue swimming crab for aquaculture. Characteristics of superior offspring for aquaculture include fast growth, resistant to diseases, adapt well to cultivation condition, and it has a high quality of meat as required by market. This paper describes the process of domestication and selective breeding of blue swimming crab until three generations, and the results of domestication that produces high quality of crab seed which suitable for aquaulture.

Material and Method. The study was conducted from April 2013 to November 2014 at educational hatchery and fish ponds of Hasanuddin University, located in Barru Regency, Province of South Sulawesi, Indonesia (Figure 1).

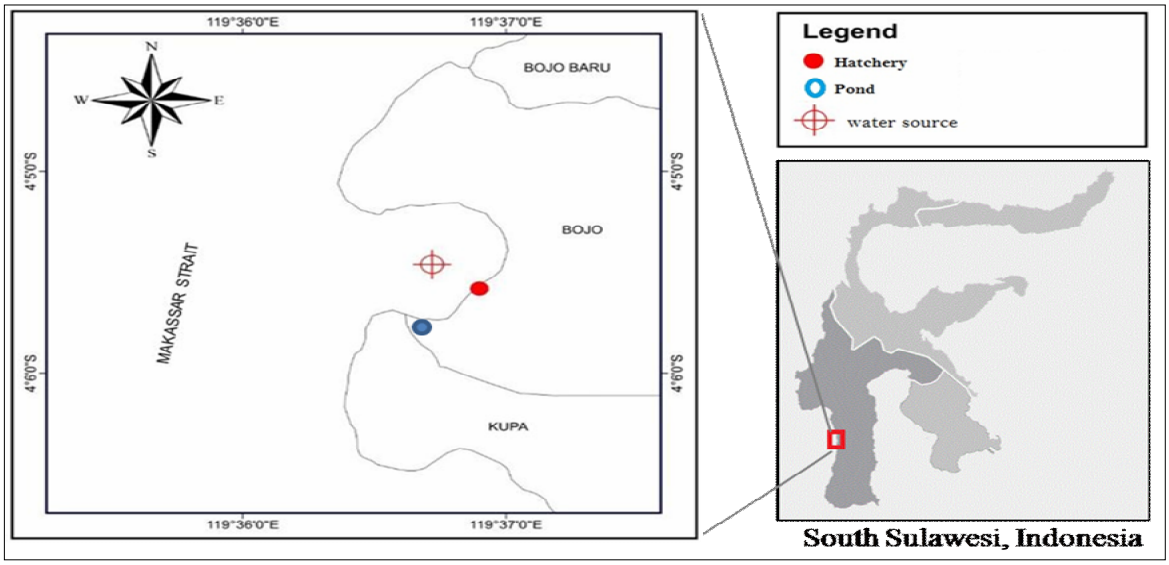

Figure 1. Location of the wild broodstock collection, larval rearing and crab rearing for domestication.

Broodstock collection. Wild broodstocks crab were collected from crab fisherman around the Makassar Strait, while the domesticated broodstock was collected from the educational fish ponds of Hasanuddin University at Bojo village, Barru Regency, South Sulawesi. The broodstock was transferred into backyard hatchery and it was kept in the 2 tonnes of sandy substrate concrete tank about one to two weeks until spawning. After spawning, the brooder maintained in the same tank until the eggs turn to black in colors which takes about 8 to 10 days. The broodstocks were fed twice daily at 07.00 AM and 19.00 PM with marine worm ad libitum.

Eggs hatching and larval rearing. The broodstock with black eggs indicated ready to hatch. That broodstock was then removed into another 2 tones concrete tank. Soon after the egg hatched, the broodstock was removed back into the broodstock tank, then the zoea was transferred into larva rearing tank. The larva was fed rotifer, artemia nauplii, and shrimp feeds (Japonicus No.0-2). Water quality of larva rearing was maintained at 30-32^{o}C, 31-33 ppt, and 4-5 mg L^{-1}, for temperature, salinity, and dissolved oxygen, respectively. Shelter (plastic band) was installed in the rearing tank when the larva reached to megalopa phase. Harvest was conducted when the larva reached day 5 of crab juvenile.

Grow-out in the brackish water pond. The grow-out culture was conducted in a 0.1 ha earthen pond by stocking 1 ind m^{-2} of day 5 crab juvenile. Ten days before stocking, to boost production of phytoplankton and zooplankton in the pond, inorganic manures such as urea and super phosphate were applied in the ratio of 100 kg ha^{-1} and 50 kg ha^{-1} respectively. Seaweed (*Gracilaria* sp.) as crab shelter was also stocked in the pond (1 kg m^{-2}). The crabs was fed twice daily with dried trash fish at a dose of 2% of body weight day^{-1}.

Selective breeding. Individual selection refer to Tave (1995) was used in this study. Wild crab broodstock from different location were collected and they were breeded to get crab instar (Figure 2). Crab seeds from different wild brooders were pooled and grown-out in the pond for 3 months. On day 90, water in the pond was drained out to collect all the crabs. Harvested crabs were grouped based on sex and length (carapace width). Crab with carapace width over 100 mm were selected and further reared until spawning while the smaller size was rejected.

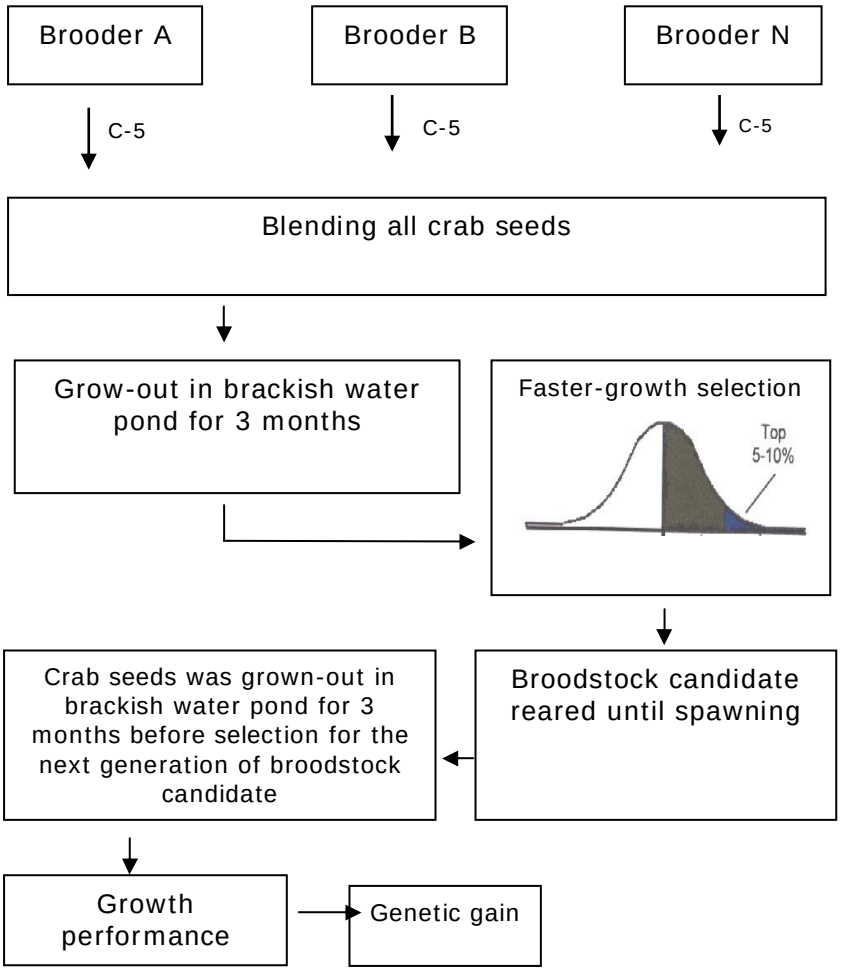

Figure 2. Procedure of blue swimming crab selection based on the growth (C-5: crab-5 stage seed).

Parameters measured. Growth performance measurement was conducted as indicator of genetically potential improvement of the crab. Parameters measured were growth rate, and meat quality. Growth rate was analyzed based on the carapace width (CW). The CW was measured from both the tip of antero-lateral carapace spine by digital caliper. Growth rate (mm day^{-1}) was calculated from differences of CW between final and initial rearing devided the rearing time ($CW_t - CW_o t^{-1}$). Meat quality of the crab was examined based on chemical composition, organoleptic test and meat yield. Parameters of chemical composition were crude protein, lipids, crude fiber, moisture, ash, and nitrogen free extract according to AOAC (2000). Organoleptic test and meat yield were analyzed by panelis from exporter company named PT. Sumber Mina Bahari in East Java.

*Data analysis***.** Growth rate data was analyzed by T Test and meat quality observation was explained descriptively.

Results and Discussion. There were fifteen from twenty wild broodstock collected from the sea successfully spawned and their eggs hatched into zoea and reaching crab phase in the hatchery (Table 1). Performance of the broodstock may vary due to catching or handling by fishermen in the field or different biological status of the broodstock and this could resulted in the differences in reproduction capacity.

Observation on the larva rearing showed high mortality was still a limiting factor. The highest mortality occurred mostly at early zoea phase until day-5 or before zoea-3 stage. Critical time at the initial zoea phase was assumed due to parasite contamination and fluctuation of water quality parameters especially temperature. Fujaya et al (2014) found that larva crab mortality was caused by several possible factors such as molting syndrome, cannibalism, fungi, abnormality, diseases, feed, water quality and genetic. Survival rate of crab larva in this research was higher than was found by Ikhwanuddin et al (2013). They found the surival rate from zoea – crab phases fed by combination of rotifer, artemia and artificial diets was 0.001–0.47%. Ikhwanuddin et al (2012) found that the swimming crab larva cultured at salinity of 30 ppt and temperature of 30°C attained survival rate was 0.02–0.45%.

Table 1

Wild broodstock performance of blue swimming crab to be used for domestication

No.	*Carapace width (CW: mm)*	*Body weight with eggs (g)*	*Egg batch weight (g)*	*Number of zoea hatched*	*Number of fifth crab instar (C-5) recovered*	*Survival of C-5 (%)*
1	8.5	100				
2	8	110	30	120000	905	0.75
3	8	90	30	150000	973	0.65
4	8	90	30	100000	610	0.61
5	9	120	30	120000	784	0.65
6	11	130	30	150000	1034	0.69
7	12	230	40	200000	610	0.31
8	9	140	50	250000	398	0.16
9	10	190	80	350000	257	0.07
10	9.5	160	60	260000	784	0.30
11	7	90	40	150000	274	0.18
12	10	150	50	210000	1034	0.49
13	8	90	30	150000	2015	1.34
14	8	90	30	150000	3015	2.01
15	9	110	20	100000	464	0:46

Crab seed (first generation; G1) from wild broodstock could adapt with pond conditon. Survival rate (SR) after three months rearing was 58.8–67.4% (Table 2). This SR was relatively higher than the results of crab culture in India (Maheswarudu et al 2008). They found that SR of blue swimming crab after 135 days of rearing in pond was 32% at the

stocking density of 2.6 first crab juvenile m^{-2}. Maheswarudu et al (2008) also reported that the crab reach to maturity after 5 months (20 days larval rearing + 134 days grow out). In this study, berried female found after less than 4 months (20 days larval rearing + 90 days grow out). Some crab attained maturity in grow-out pond indicated that the crab well adapted in the rearing pond condition and it succeeded in completing nearly all their life cycle. This was a vital requisite to any potential candidate for aquaculture. This finding supported for the purpose of domestication as proposed by Liao & Huang (2000) that domestication in aquaculture is a control of life cycle of an organism including manipulation of breeding in the captivity.

Table 2

Performance of first generation (G1) of domesticated crab after 3 months rearing in the ponds

No. pond	*Number*				*Survival (%)*	*Berried female (%)*	*Sex ratio (M:F)*
	Stocking	*Harvest*	*Male*	*Female*			
1	1000	674	331	343	67.4	0	49:51
2	1000	664	318	346	66.4	0	48:52
3	1000	588	291	297	58.8	12	49:51
4	1000	601	287	314	60.1	31	48:52

Size distribution of harvested crab was relatively different among the ponds (Figure 3). This may caused by differences in pond fertility. Bottom texture of No. 1 and 2 ponds was sandy bottom while No. 3 and 4 ponds were loam sandy bottom. Bottom texture of pond has influenced the fertility of waters body and directly affected plankton abundance. This condition has an effect on the better water quality especially during early period of rearing.

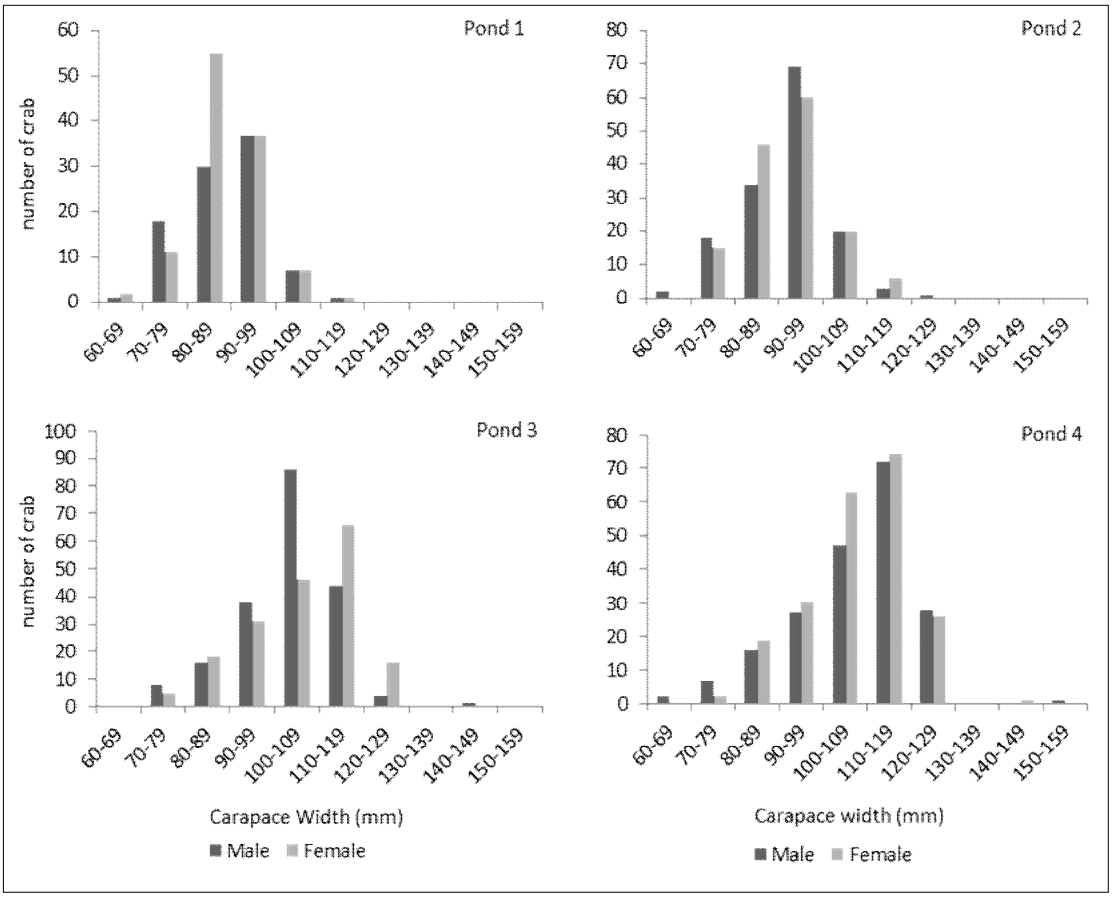

Figure 3. Size distribution of first domesticated crab generation (G1) after 3 months rearing in the ponds.

Size improvement and uniformity of size has began to appear after three generation of selection. Number of crab from domesticated crab with CW more than 100 mm increase over generation, that was 50% of the population in the first generation (G1), 73% in the second generation (G2) and 78% in the third generation (G3) (Figure 4). Growth rate increased from 1.0 mm day^{-1} in G1 to 1.1–1.2 mm day^{-1} in G3 (Table 3). Growth rate of domesticated crab was considerably high than the growth rate of wild male and female of blue swimming crab along the Southeast Coast of India were 11.0 and 9.6 mm month^{-1} and it can reached 145.2 and 132.5 mm, respectively (Sukumaran & Neelakantan 1997).

In the present study, differences in growth rate between G1 and G3 have shown a significant improvement ($p < 0.01$) (Table 3). Even though, Gjedrem & Baranski (2009) said, domestication is a slow process and requires a long time to produce a better adapted animal to its environment, but these results shows that the progress of growth improvement was relatively fast if followed by a faster-growing selection.

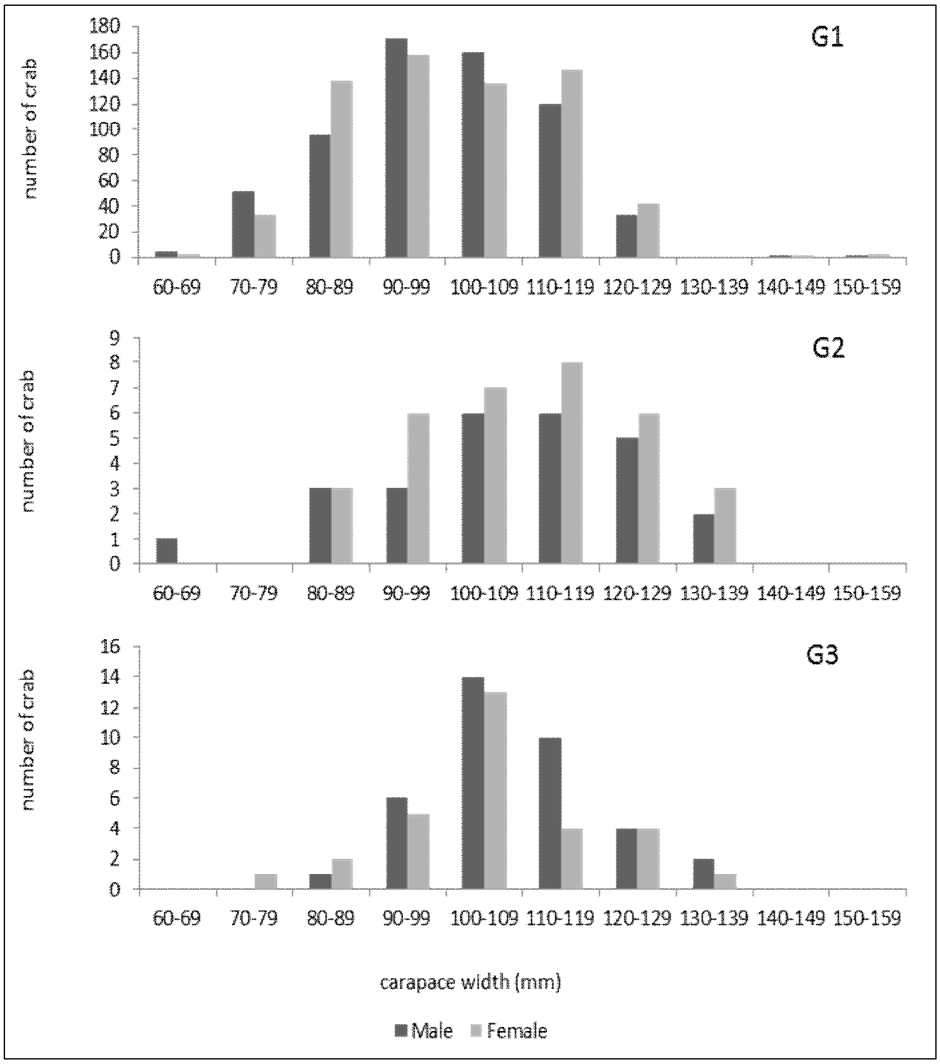

Figure 4. Size distribution of domesticated crab after 3 months rearing in the pond G1, G2, G3 were first, second, and third generation of domesticated crab, respectively.

The first domesticated aquatic species was carp which was conducted in 3000-4000 years ago in china. Atlantic salmon is the highest production species in the world today, was originally resulted from domestication which was started in Norway and Scotland in the late 1960s. Farming of Pacific white shrimp (*Litopenaeus vannamei*) began in the 1980s using wild captured larvae, then shift to hatchery raised larva era in the period of 1988 to 1996, finally the shrimp industry is growing very rapidly after entering the breeding era in 1997. This rapid industry growth was primarily driven by the domestication, breeding,

and world wide spread of *L. vannamei* from the Western Hemisphere into Asia. As a result, world aquaculture of *P. vannamei* increased from only 10% in 1998 to 75% of total shrimp production in 2006. Thailand shrimp revolution was characterized by the use of domesticated *P. vannamei* based on the fast growth and resistant disease bred (Wyban 2007).

Table 3

Growth rate of domesticated crab after 3 months rearing in the ponds

Generation	*Sex*	*Carapace width (CW) (mm)*	*Growth rate (mm day^{-1})*
G1	Male	97.0	1.02
	Female	97.7	1.03
G2	Male	105.4	0.99
	Female	109.9	1.04
G3	Male	110	1.17
	Female	106	1.12

Significant value ($p < 0.01$) between G1 and G3.

Prospects for increasing production of blue swimming crab through domestication is promising. Beside increased in growth rate and ability to reproduce in the cultivation condition, the domesticated crab also has good quality of meat. Crab meat proximate analysis indicated that the chemical composition of 3rd generation (G3) of domesticated crab was relatively similar to the wild crab (Table 4). Protein content of domesticated crab in this study was similar to results obtained by Chaiyawat et al (2009) which ranged between 10-15% depending on the type of feed given. Crabs, after 8 weeks of being fed with trash fish have a protein content of 13.99±0.91 while those who were fed red seaweed has a protein content of 15.12±0.38. However, lipid content in this study was lower than the results found by Chaiyawat et al (2009), which was only around 0.08% while the lipid content of the crab given a feed of combination of trash fish and seaweed was 0.40±0.47%. In this study, the crab was fed with trash fish, while the seaweed was grown in the pond as a shelter for the crab. Referring to the research of Chaiyawat et al (2009), it was assumed that the crab in this pond use the seaweed as feed.

Table 4

Chemical composition of domesticated and wild blue swimming crab meat

Sample fraction	*Crab sample*	*Composition (%)*					
		Crude protein	*Fat*	*Crude fiber*	*Moisture*	*Ash*	*Nitrogen free extracts*
Fresh	Domesticated	15.89	0.08	0.04	79.85	1.31	2.84
	Wild	14.18	0.08	0.03	81.85	2.55	1.32
Dry weight	Domesticated	78.85	0.41	0.18	-	2.04	18.53
	Wild	78.25	0.43	0.17	-	1.74	19.41

Organoleptic and meat yield analysis showed that the G3 crab has been qualified as export standard crab meat in term of colour and typical crab meat flavor. The meat yield was relatively high (37%). This value was higher than the wild crab with only 25-30% (PT. Philips Seafood Indonesia 1997). Based on these descriptions, domestication of blue swimming crab has very good prospects in the development as an aquaculture species. Although the rate of genetic improvement through domestication takes slowly, but the advantage of domestication is to allow the control or manipulation of the production and reproduction. Artificial selection and hybridization can be done and diseases or predators can be removed to improve production efficiency (Liao & Huang 2000).

However, domestication also has some negative effects. During culture period, the number of crab broodstock in the captivity is limited hence it may loss the genetic variation (Falconer 1989) and increase of homozygotes (inbreeding) (Agnese et al 1995) become a threat. Therefore, it needs special attention in designing and implementing selective breeding for aquatic species including the blue swimming crab. Selective

breeding is one method to speed up the desired properties of genetic improvement during domestication. Cross breeding can be done to make useful of non-additive gene effect, polyploidy, or sex manipulation also may be applied for further improvement (Gjerde & Rye 1998). For aquaculture purpose, generally genetic improvement focuses on the characters of increase of growth rate and disease resistant. Benefit of higher growth rate is shortening of culture period, hence increase food conversion ratio. Use of domesticated stock enables farmers to improve the production system continuously based on market demand and prices (Wyban 2007).

Conclusions. The individual selection of blue swimming crab up to three generations produced a higher growth rate than wild broodstock, meanwhile the quality of domesticated crab was similar with wild crab. Furthermore, domesticated broodstock followed by a fast-growing selection of crab have a high potential prospects for blue swimming crab aquaculture.

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