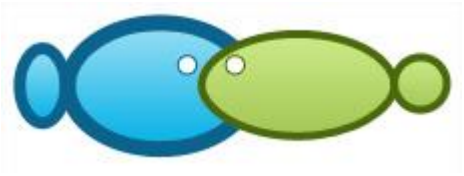

Analysis of genetic diversity and reproductive performance of the Blue Swimming Crab (*Portunus pelagicus*) from several waters in Indonesia

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Abstract. The aim of this research was to analyse the genetic diversity and reproductive performance of *Portunus pelagicus* from several Indonesian waters. The information obtained can be used as a basis for the development of breeding programs to support cultivation. Sample collection was conducted in March–November 2017 at Raja Ampat (Papua Sea), Maumere (Flores Sea), Jepara (Java Sea) and Barru (Makassar Strait). Genetic data was collected based on the CO1 (Cytochrome Oxidase sub-unit 1) gene sequences. The reproductive performances determined were the size of crabs, fecundity and egg diameter. The DNA genome extraction was conducted using the phenol-chloroform method and COI gene was amplified using Polymerase Chain Reaction with universal primers, COIa (5'-AGTATAAGCGTCTGGGTAGTC–3') and COIf (5'-CCTGCAGGAGGAGGAGATCC–3'). The results showed that there were differences in genetic distances between samples from different locations. The furthest *P. pelagicus* genetic distance (14.300) was found between the Makassar Strait and the Java Sea, while the nearest genetic distance (8.540) was between the crabs from Java Sea and Papua Sea. Variations of the egg diameter, fecundity and egg weight were also found between crabs from different sampling locations. The *P. pelagicus* from the Makassar Strait had the highest fecundity (137.157±25.059), while the lowest fecundity was found in sample from Raja Ampat (99.711±25.491). However, *P. pelagicus* from Makassar Strait had the smallest egg diameter (10.11±0.41 µg). The largest egg diameter (µm) was found in the *P. pelagicus* sample from Jepara (11.15±0.66 µg). Multivariate analysis results showed that there was a significant difference in reproduction performance of *P. pelagicus* from the 4 sampling locations. The different geography could have affected the genetic variation of the *P. pelagicus*, consequently affecting its size and reproductive performance.
Key Words: *Portunus pelagicus*, reproduction, fecundity, egg, genetic diversity.

Introduction. Blue Swimming Crab (*Portunus pelagicus*) is a valuable fishery commodity with a high economic value. *Portunus pelagicus* is distributed along the coast of Indonesia and most of the *P. pelagicus* production comes from fishing activities. However, a decrease in the production of *P. pelagicus* can be observed, not only in quantity but also in size. Based on previous studies, the *P. pelagicus* from all over the world have been over-exploited, resulting in a population decline (Hidayani et al 2015; Fujaya et al 2016a; Fujaya et al 2016b; La Sara et al 2017; Johnston et al 2011; Kunsook et al 2014). This is studied not only from a biodiversity point of view, but also from a coastal community depending on crab point of view.

There are many ways to conserve the *P. pelagicus* population: catch limitation, restocking juveniles from hatcheries and the pond aquaculture. However, these methods require detailed information regarding the species, particularly the morphological, genetical and reproductive aspects.

Initially, *P. pelagicus* was regarded as a monophyletic species (Mantelatto et al 2007), so in several countries, it was managed as a single species (Klinbunga et al 2010;

Sienes et al 2014). However, a previous study by Lai et al (2010) found that *P. pelagicus* was a complex of species consisting of 4 species members, *P. pelagicus*, *P. segnis*, *P. reticulatus* and *P. armatus*. Asphama (2014) and Fujaya et al (2016a) found that the *P. pelagicus* from the Makassar Strait present high variety and are different from *P. pelagicus* studied by Lai et al (2010). These results are based on a morphometric analysis and mitochondria DNA Cytochrome Oxidase 1 (CO1) analysis. Based on the morphometric analysis by Hidayani et al (2015), RAPD and 16SrRNA (Fujaya et al 2016a), high variation was observed among *P. pelagicus* populations from the Makassar Strait. There was also found a unique-specific tape at 450 and 600 bp that used the OPA-11 primer. The result indicated that there was a possibility for hybridization among populations.

Developing a *P. pelagicus* culture requires a detailed study regarding the genetic relationships and reproductive performances of the blue swimming crab. This information is very important for designing breeding programs to produce high-quality juveniles for *P. pelagicus* aquaculture. The aim of the study was to examine the genetic distance, size, fecundity and egg diameter of the *P. pelagicus* broodstock from several Indonesian waters.

Material and Method

Study sites and sample collection. The study was conducted from March to November 2017. *Portunus pelagicus* were collected from Raja Ampat (Papua Sea), Maumere (Flores Sea), Jepara (Java Sea) and Barru (Makassar Strait). The DNA extraction and analysis were conducted at the Faculty of Science, Hasanuddin University. The fecundity and egg diameter were measured at the Hatchery Technology Laboratory, Faculty of Marine Science and Fisheries, Hasanuddin University.

Figure 1. Map of study sites in Indonesian archipelago. 1 - Raja Ampat (0°55'50"S, 131°07'50"E); 2 - Maumere (8°37'12.00"S; 122°13'12.00"E); 3 - Jepara (6°35'0"S, 110°39'0"E); 4 - Barru (4°7'36"S, 119°37'25"E).

The *P. pelagicus* samples were collected from crab fishermen. The *P. pelagicus* samples from Maumere and Barru were caught using a gill net. The *P. pelagicus* samples from Raja Ampat were collected from the fishermen at Jeffman Island, caught by a gill net. The *P. pelagicus* samples from Jepara were caught by bubu traps. About 100 crabs were collected from each site. The female and the male crabs were separated. The measurement of weight (whole body and eggs), length (measured from the anterior limit to the posterior limit of the carapace) and width (measured from the tip of the longest spine on the side of the crab's body) of the carapace was done *in situ* (Figure 2). DNA samples were collcted from the muscle located at the foot swim edge, accounting for 50

mg. The samples were stored and fixed by 70% methanol, with a ratio of 1:10 muscle to methanol).

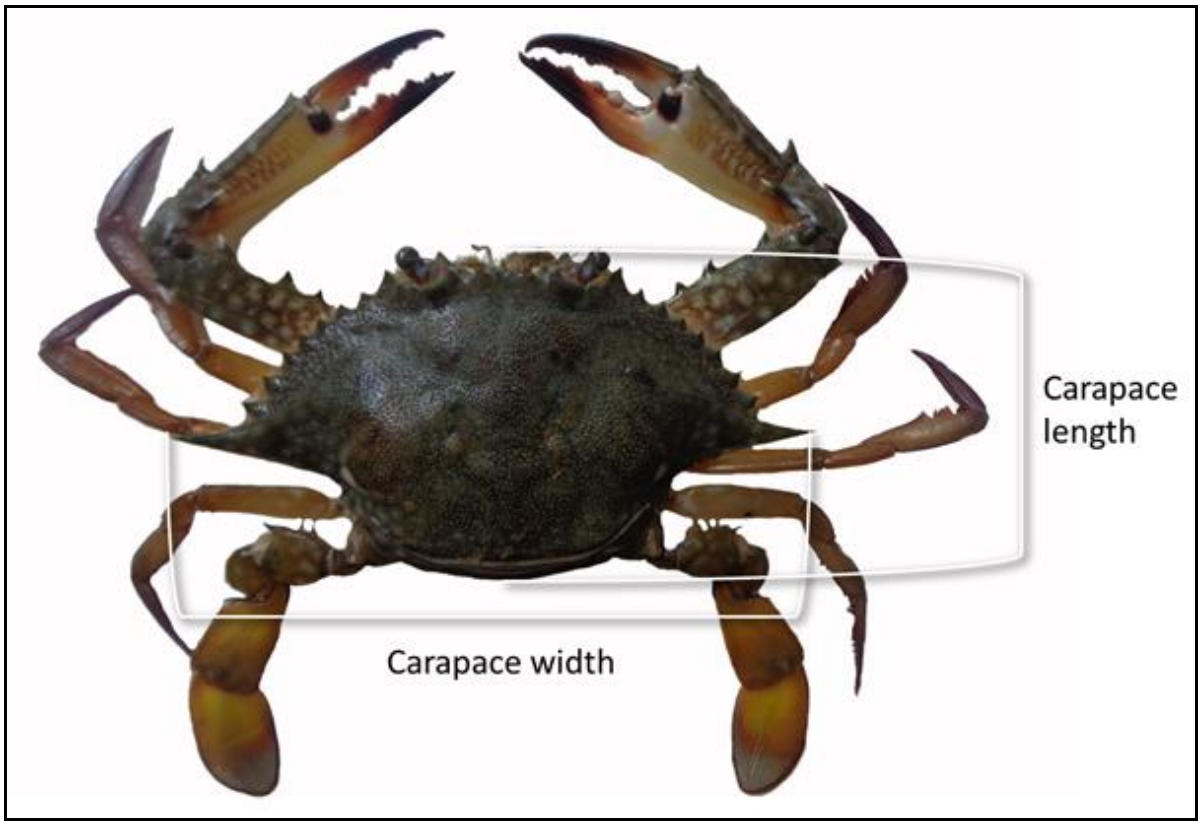

Figure 2. Morphology measurements of *Portunus pelagicus* (female).

Egg samples were collected from the berried females. All eggs from the female crabs were weighed (digital scale with the accuracy of 2 decimal places). The egg diameter was measured for 10% of the eggs. The egg samples were fixed by Gilson solution.

DNA extraction. The DNA genome extraction was performed using the phenol-chloroform method described by Parenrengi (2001), as follows:

1. Digestion buffer (0.5 M NaCl, 0.001 M EDTA, 1% SDS, 0.8% Triton-X, and 0.1 Tris-HCl at pH 9.0) was added into a 1.5 mL microcentrifuge tube containing 50 mg of crab muscle and then 40 µL of SDS 10% and 40 µL of Proteinase K (20 mg/mL) were added. The tube was shaken gently and incubated at 55°C for 1 to 3 h
2. The sample was treated with 25 µL of RNase (20 mg/mL) and was left at room temperature for 15 to 30 minutes.
3. The samples were treated with 500 to 600 µL of phenol:chloroform:isoamyl alcohol (25:24:1) and gently homogenized. The samples were left at room temperature for 10 minutes before centrifugation at 13000 rpm for 4 minutes.
4. The top layer of the aqueous solution was removed and dispersed into a new microcentrifuge tube. The step of adding phenol:chloroform:isoamyl alcohol was repeated three times.
5. The samples were treated with 500 µL of chloroform:isoamyl alcohol (24:1) and were centrifuged at 13000 rpm for 2 minutes.
6. The upper aqueous layer was mixed with 1 mL of ice-cold absolute ethanol by rapid inversion of the tubes several times. Then, it was centrifuged at 6000 rpm for 30 minutes.
7. The DNA precipitated at the bottom of the tubes as a white pellet. It was collected and washed with 500 µL of 70% of ethanol and then centrifuged at 6000 rpm for 15 minutes.

8. The DNA was allowed to dry at room temperature for 20 minutes and resuspended with 50 µL of sterile distilled water (SDW) for at least 24 h at room temperature to fully dissolve before proceeding to the next step. To determine a successful extraction, 7.5 µL of DNA genome and 2.5 µL of Loading Dye was electrophoresed in 0.8% agarose gel in 1 X TBE (Tris-Boric acid-EDTA) at 50 volts. The gel was stained with 0.5 µg/mL ethidium bromide solution. A Hind III marker was used as a DNA genome extraction marker. The DNA genome samples were kept in -20°C to avoid DNA degradation.

COI gene isolation. COI gene was amplified based on Lai et al (2010), using the Polymerase Chain Reaction with the universal primers COIa (5'-AGTATAAGCGTCTGGGTAGTC-3') and COIf (5'- CCTGCAGGAGGAGGAGATCC-3'). This pair of primers was used to strengthen a COI fragment that matched with the position of 5' to 3' from 681-1294 mitochondria genome. A PCR was run in a 50 mL volume that consisted of 5 mL of 10 × Taq Polymerase Buffer, 5 mL of 25 mM $MgCl_2$, 5 mL of 0.5 mM dNTP, 1 unit of Taq polymerase (Perkin Elmer) and 0.5 mL of each 25 pmol/mL COIa and COIf.

Nucleotide sequence alignment. The PCR result of the COI gene was purified with the Gel/PCR DNA Fragment Extraction (Geneaid) kit. The samples were then sequenced by using the automatic sequencer AB-3130 (Applied Biosystem). The sequencing result was read manually using the Sequence navigator program.

Egg sample measurement. The reproductive characteristics were measured, namely the fecundity and egg diameter. The fecundity (the number of egg stored under the abdomen) was measured using a combination of the gravimetric and volumetric methods.

$$F = TGW/SGW \times ES$$

Where: F – fecundity; TGW - total gonad weight; SGW - sample gonad weight; ES - number of eggs.

The number of eggs was counted using the volumetric method. The weighted eggs were diluted with 100 mL distillate water and homogenized. 1 mL of the homogenized sample was placed into the Sedgwick-Rafter for counting the number of eggs under the microscope. The measurement of eggs was done in 3 replicates. The number of eggs was counted using a formula as below:

$$ES = V \times X$$

Where: ES - number of eggs; V - dilution volume (mL); X - number of egg per mL.

The egg diameter was measured under a microscope, using the binocular microscope and an ocular micrometer with 0.1 mm accuracy.

Data analysis. The sequence results were analyzed by the Genetyx Versi 7 program (Genetyx Coorporation) to obtain the sequence consensus from the forward and reverse sequence. To determine the sequences similarity, they were compared with the existing sequences from the GenBank, using the BLAST-N program (basic local alignment search tool-nucleotide). A phylogeny tree was built using Megablast ver. 6 program. The egg data was analyzed using the Kruskal-Wallis test with a significance level of 5%. Multiple pairwise comparisons were made using Dunn's procedure/Two-tailed test to determine the area of difference.

Results and Discussion. The size of the females *P. pelagicus* caught at each landing site was different. Samples from each study site have a size variation not only in the carapace length and carapace width, but also in the body weight (Table 1).

Table 1

Female crab size (mean ± standard deviation) from the study sites

Landing Site	*Carapace Length (cm)*	*Carapace Width (cm)*	*Body Weight (g)*
Raja Ampat	6.21 ±0.79	13.47±1.47	170.08±56.3
Maumere	6.16±0.83	12.79±1.43	145.26±49.84
Jepara	5.59±0.79	11.96±1.43	106.72±47.87
Barru	6.06±3.01	11.02±0.80	94.51±26.59

Genetic distance. The result of the CO1 (Cytochrome oxidase subunit 1) gene sequence analysis showed that the *P. pelagicus* from Indonesian waters had a high variation, not only between different populations but also within the same population. The genetic intrapopulation distance of *P. pelagicus* from Maumere, Raja Ampat, Jepara and Barru varied from 0.000 to 4.847, while the genetic distance between populations varied between 8.540 and 14.300 (Table 2; Figure 3). The shortest genetic distance of the *P. pelagicus* was found between Jepara and Raja Ampat populations, and also between Raja Ampat and Maumere. The furthest genetic distance of *P. pelagicus* was found between the samples from Jepara and Barru.

Table 2

Genetic distance of female crab between study sites

	Jepara	*Raja Ampat*	*Maumere*
Raja Ampat	8.540		
Maumere	12.326	9.134	
Barru	14.300	9.909	11.670

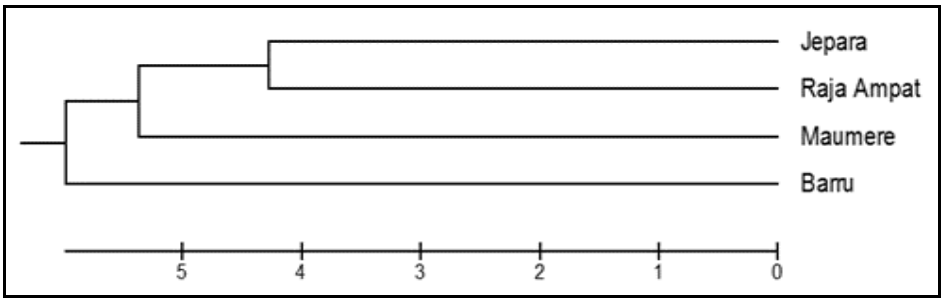

Figure 3. Phylogeny tree of female crab (*Portunus pelagicus*) from the study sites.

Reproductive performance. Fecundity was the number of fertilized eggs that were carried by the female (the number of eggs stored under the abdomen). Our findings showed that the fecundity varied between the catching areas of *P. pelagicus* (Figure 4). The highest fecundity was found in *P. pelagicus* from Barru, 137.157±25059 (mean ± standard deviation), while the lowest fecundity was observed in the samples from Raja Ampat (99711±25491). Interestingly, the *P. pelagicus* female brood stock from Barru had the smallest body size, but produced the highest number of eggs (Figure 5). The largest egg diameter (µm) was found in the *P. pelagicus* from Jepara (11.15±0.66) and the smallest was found in the samples from Barru (10.11±0.41) (Figure 6).

The results showed that there were variations in size, reproductive performance and genetic distance in the 4 study areas. Raja Ampat, located at the eastern tip of Indonesia, has *P. Pelagicus* of larger size compared to other regions. Barru, located in the middle of Indonesia, in the Wallacea region, had blue swimming crabs with the smallest body size and egg diameter (Table 1; Figure 6), but with the highest number of eggs

(Figure 4; Figure 5). This difference is supported by the results of genetic distance and phylogeny tree analysis. These observations provide confirmation that differences in swimming crab reproduction performance from various regions in Indonesia are influenced by genetic differences.

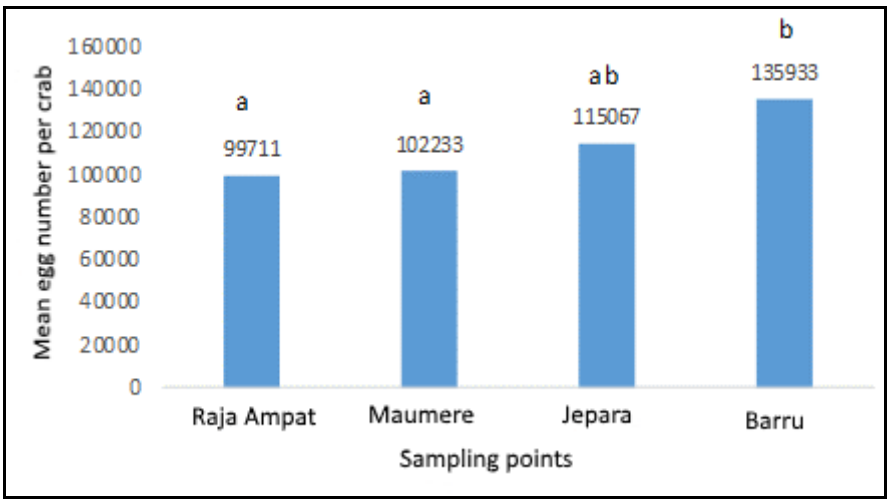

Figure 4. Mean fecundity (egg number) of *Portunus pelagicus*; different letters indicate significant difference (P<0.05).

Our finding confirmed the previous study, although they have not clearly confirmed whether the *P. pelagicus* in Indonesia were a different species or sub-species from the species that was found by Lai et al (2010). However, the differences of genetic distance and reproductive performance have supported strongly the assumption of our findings. Elrod & Stansfield (2007) have mentioned that the genetic variation of organisms in populations occur mainly due to three mechanisms: mutation, free of pair allele or recombination and gene migration.

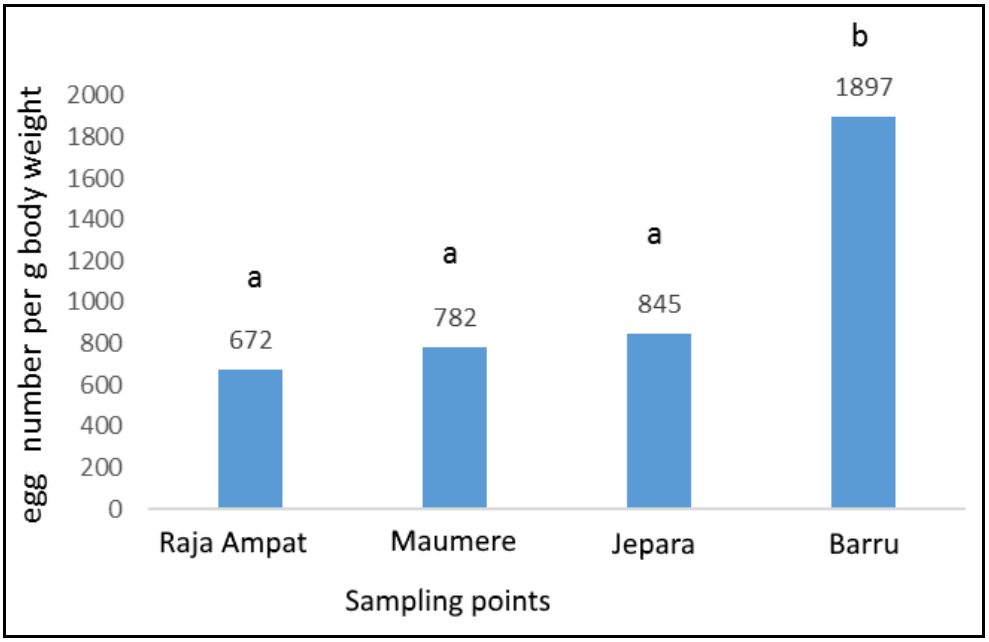

Figure 5. Mean of egg number per gram of body weight of *Portunus pelagicus*; different letters indicate significan difference (P<0.05).

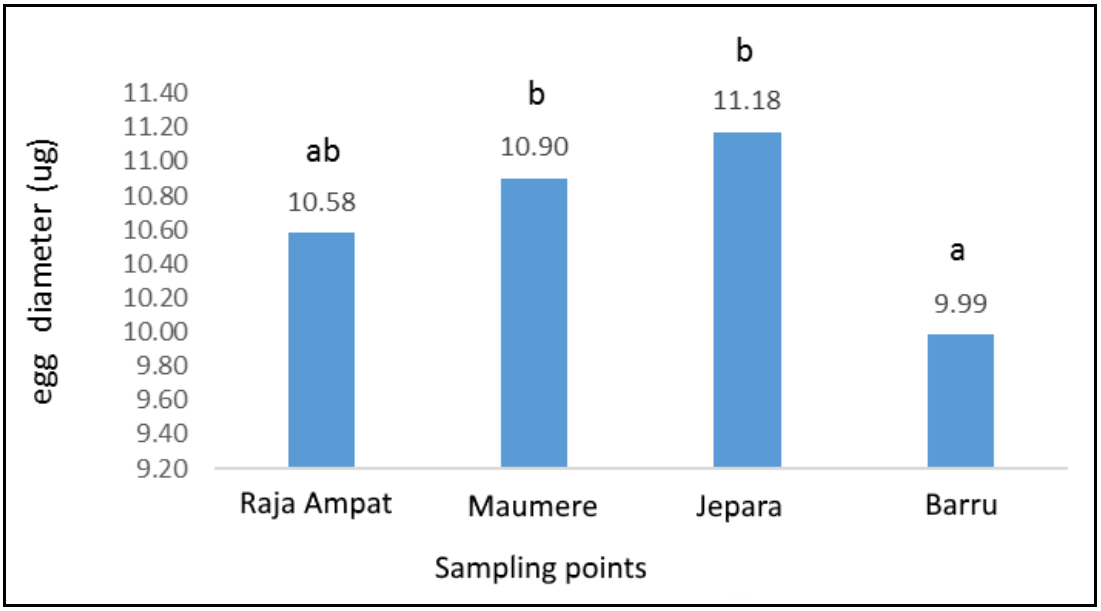

Figure 6. Mean of egg diameter (µm) of *Portunus pelagicus*; different letters indicate significant difference (P<0.05).

The level of individual variations inter and intra populations have described an existing status of species in nature. Genetic variations of the *P. pelagicus* from Indonesian waters could possibly be due to different environmental conditions. The Indonesian waters could be divided into three sub-regions that have a geological history correlation: Sunda Shelf, Wallacea region and Sahul Shelf. Sahul Shelf is a region including Australia, Tasmania and New Guinea, and the shallow sea between Australia and East Timor. Sunda Shelf was above water for a substantial part of the Miocene epoch, arguing that there was an exchange in biodiversity between different, now separated regions, of the Asian continents. The connection between Wallacean Island, Sunda Shelf in the west and Sahul Shelf in the east was covered by water the Eocene and early Pleistocene (Boer et al 2015).

Genetic variation of the population is an important factor that could affect the response of a population to natural or artificial selection. Individuals that have a high genetic variation have a high fitness component (growth rate, fecundity, viability, immunity to environmental changing and stress level). On the other hand, when individuals have low genetic variation, it can affect the ability of the species to respond to environmental changes (Imron et al 1999).

Previous studies by Overturf et al (2003) and Wachirachaikarn et al (2009) showed that the genetic variations on a molecular level could not guarantee the variations in the quantitative phenotypic characteristic. However, a high genetic variation could be a basic factor for the breeding program in a long term. Our findings show that genetic variations in the CO1 level were strengthened by a phenotypic character, such as body size, fecundity or egg diameter. This information is important for supporting a breeding program and for developing *P. pelagicus* aquaculture in the future.

Fecundity is an output of reproduction that is characterized by the number of eggs that can be produced by the female individuals during spawning (Kumar et al 2000; Ikhwanuddin et al 2012). Fecundity is also one of the parameters of reproductive performance. Several previous studies showed that this parameter varies. Arshad et al (2006) found that crab fecundity varied between species and also within species due to different factors, such as age, size, food, ecological conditions. The number of eggs that could be produced by the female varied depending on the size of the individual crab. The main intrinsic factors that contribute to the fecundity differences among females in the same population are the variation in size, the dietary history that correlated with the availability of food in their habitat, the age at sexual maturity and the first reproductive

age that could affect the next reproduction process. The main extrinsic factor is the intra and inter-specific competition (Ramirez-Llodra 2002; Zairion et al 2015).

The female brood stock of a bigger size can produce more eggs than those of a smaller size (Hamasaki et al 2006; Ikhwanuddin et al 2012; De Lestang et al 2003; Arshad et al 2006). Kumar et al (2003) explained that fecundity can increase by 83.9% with the increase of the carapace width from 10.5 to 12.5 cm. Ikhwanuddin et al. (2012) also mentioned that the number of egg production had increased linearly with the increasing carapace width and body weight. Similar results were also found by Kumar et al (2000) for *P. pelagicus*, but it is also true for other Portunid species (De Lestang et al 2003). However, our findings showed that the brood stock with a small size from Barru Regency has a higher fecundity due to a smaller egg diameter. This finding indicates that a reproductive strategy in coping with pressure in population due to high mortality because of fishing and habitat degradation can be applied.

Our finding is very important in improving the genetic quality of brood stock for crab aquaculture. For the purpose of aquaculture, hybridization is one method to improve the genetic quality and to obtain a high quality brood stock in a shorter period through increasing the heterogeneity and the dominant genetic variation (Guy et al 2009). The hybrid could have several characteristics from both the parents.

Cross breeding in fish and prawn with a high genetic distance improved the performance of the reproductive and immune systems (fitness) in the Guppy fish (*Poecilia reticulata*) (Shikano & Taniguchi 2002) and an increasing growth rate in the Pacific blue prawn (*Litopenaeus stylirostris*) (Goyard et al 2008). Falconer et al (1996) found that the heterocyst expression of two populations that were cross bred depended on the differences in the gene frequency and interaction between the dominant and epistasis gene. However, the appearance of a heterocyst on cross breeding was difficult to predict and may depend on the fish age (Granier et al 2011).

Conclusions. DNA Cytochrome Oxidase 1 (CO1) in mitochondria could be used for predicting the variation of *P. pelagicus* reproductive performance. The different geography might affect the genetic variation of *P. pelagicus*, consequently affecting its size and reproductive performance. We suggest conducting a further study related to the cross breeding between the *P. pelagicus* brood stock from Barru with the populations from Jepara, Raja Ampat and Maumere to produce a high quality population.

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References

Arshad A., Efrizal K. M., Saad C., 2006 Study on fecundity, embryology and larval development of blue swimming crab *Portunus pelagicus* (Linnaeus, 1758) under laboratory conditions. Research Journal of Fisheries and Hydrobiology 1(1):35-44.

Asphama A. I., 2014 [Genetic variety analysis of *Portunus pelagicus* (Linnaeus, 1758) species complex from Barru waters based on morphology and DNA characters]. MSc Thesis, Hasanuddin University, Makassar, 113 p. [in Indonesian].

Boer H. J., Steffen K., Cooper W. E., 2015 Sunda to Sahul dispersals in Trichosanthes (Cucurbitaceae): a dated phylogeny reveals five independent dispersal events to Australasia. Journal of Biogeography 42(3):519-531.

De Lestang S., Hall N., Potter I. C., 2003 Changes in density, age composition, and growth rate of *Portunus pelagicus* in a large embayment in which fishing pressures and environmental conditions have been altered. Journal of Crustacean Biology 23(4):908-919.

Elrod S., Stansfield W., 2007 [Genetika]. Jakarta, Erlangga, pp. 78-191. [in Indonesian].

Falconer D. S., Mackay T. F., Frankham R., 1996 Introduction to quantitative genetics (4th edition). Trends in Genetics 12(7):280.

Fujaya Y., Asphama A. I., Hidayani A. A., Parenrengi A., Tenriulo A., 2016a High genetic variation of *Portunus pelagicus* from Makassar Straits revealed by RAPD markers and mitochondrial 16S rRNA sequences. African Journal of Biotechnology 15(7):180-190.

Fujaya Y., Trijuno D. D., Aslamyah S., Alam N., 2016b Domestication and selective breeding for producing fast growing and high meat quality of blue swimming crab (Portunus pelagicus). AACL Bioflux 9(3):670-679.

Goyard E., Goarant C., Ansquer D., Brun P., De Decker S., Dufour R., Galinié C., Peignon J. M., Pham D., Vourey E., 2008 Cross breeding of different domesticated lines as a simple way for genetic improvement in small aquaculture industries: Heterosis and inbreeding effects on growth and survival rates of the Pacific blue shrimp *Penaeus (Litopenaeus) stylirostris*. Aquaculture 278(1):43-50.

Granier S., Audet C., Bernatchez L., 2011 Heterosis and outbreeding depression between strains of young-of-the-year brook trout (*Salvelinus fontinalis*). Canadian Journal of Zoology 89(3):190-198.

Guy J. A., Jerry D. R., Rowland S. J., 2009 Heterosis in fingerlings from a diallel cross between two wild strains of silver perch (*Bidyanus bidyanus*). Aquaculture Research 40(11):1291-1300.

Hamasaki K., Fukunaga K., Kitada S., 2006 Batch fecundity of the swimming crab *Portunus trituberculatus* (Brachyura: Portunidae). Aquaculture 253(1):359-365.

Hidayani A. Asphama I., Trijuno D. D., Tenriulo A., Parenrengi A., Fujaya Y., 2015 The Morphometric Character and Mitochondrial 16S rRNA sequence of *Portunus pelagicus.* Aquacultura Indonesiana 16(1):1-9.

Ikhwanuddin M., Azra M., Siti-Aimuni H., Abol-Munafi A., 2012 Fecundity, embryonic and ovarian development of blue swimming crab, *Portunus pelagicus* (Linnaeus, 1758) in coastal water of Johor, Malaysia. Pakistan Journal of Biological Sciences 15(15):720-728.

Imron K. S., Sumantadinata K., Soewardi K., 1999 [Genetic variation in cultured stocks of Tiger Shrimp (*Penaeus monodon*) in Indonesia]. Indonesian Fisheries Research Journal 1:10-18. [in Indonesian].

Johnston D., Harris D., Cauti N., Thomson A., 2011 Decline of a blue swimmer crab (*Portunus pelagicus*) fishery in Western Australia-History, contributing factors and future management strategy. Fisheries Research 109:119-130.

Klinbunga S., Yuvanatemiya V., Wongphayak S., Khetpu K., Menasveta P., Khamnantong B., 2010 Genetic population differentiation of the blue swimming crab *Portunus pelagicus* (Portunidae) in Thai waters revealed by RAPD analysis. Genetics and Molecular Research 9(3):1615-1624.

Kumar M. S., Ferguson G., Xiao Y., Hooper G., Venema S., 2000 Studies on reproductive biology and distribution of blue swimmer crab (*Portunus pelagicus*) in South Australian Waters. SARDI Research Report Series 47:1324-2083.

Kumar M. S., Xiao Y., Venema S., Hooper G., 2003 Reproductive cycle of the blue swimmer crab, *Portunus pelagicus*, off southern Australia. Journal of the Marine Biological Association of the UK 83(05):983-994.

Kunsook C., Gajaseni N., Paphavasit N., 2014 A Stock Assessment of the Blue Swimming Crab Portunus pelagicus (Linnaeus, 1758) for Sustainable Management in Kung Krabaen Bay, Gulf of Thailand. Tropical Life Sciences Research 25(1):41-59.

La Sara, Muskita W. H., Astuti O., Safilu, 2017 Some population parameters of blue swimming crab (*Portunus pelagicus*) in Southeast Sulawesi waters, Indonesia. AACL Bioflux 10(3):587-601.

Lai J. C., Ng P. K., Davie P. J., 2010 A revision of the *Portunus pelagicus* (Linnaeus, 1758) species complex (Crustacea: Brachyura: Portunidae), with the recognition of four species. The Raffles Bulletin of Zoology 58(2):199-237.

Mantelatto F. L., Robles R., Felder D. L., 2007 Molecular phylogeny of the western Atlantic species of the genus *Portunus* (Crustacea, Brachyura, Portunidae). Zoological Journal of the Linnean Society 150:211-220.

Overturf K., Casten M., LaPatra S., Rexroad C., Hardy R., 2003 Comparison of growth performance, immunological response and genetic diversity of five strains of rainbow trout (*Oncorhynchus mykiss*). Aquaculture 217(1):93-106.
Parenrengi A., 2001 Studies on genetic variability of groupers (Genus: Epinephelus) from Indo-Malaysian waters using PCR-RAPD Analysis. MSc Thesis, Kolej University Terengganu. Universiti Putra Malaysia, 177 p.
Ramirez-Llodra E., 2002 Fecundity and Life-History Strategies in Marine Invertebrates. Advances in Marine Biology 43:87-170.
Shikano T., Taniguchi N., 2002 Relationships between genetic variation measured by microsatellite DNA markers and a fitness-related trait in the guppy (*Poecilia reticulata*). Aquaculture 209(1):77-90.
Sienes P. M. Q., Willette D. A., Romena L. R., 2014 Genetic diversity and the discovery of a putative cryptic species within a valued crab fishery, *Portunus pelagicus* (Linnaeus 1758), in the Philippines. Philippine Science Letters 7(2):317-323.
Wachirachaikarn A., Rungsin W., Srisapoome P., Na-Nakorn U., 2009 Crossing of African catfish, *Clarias gariepinus* (Burchell, 1822), strains based on strain selection using genetic diversity data. Aquaculture 290(1):53-60.
Zairion Y. W., Boer M., Fahrudin A., 2015 Reproductive biology of the blue swimming crab *Portunus pelagicus* (Brachyura: Portunidae) in east Lampung waters, Indonesia: fecundity and reproductive potential. Tropical life sciences research 26(1):67-85.

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