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MITOCHONDRIAL DNA DIVERSITY IN FOUR POPULATIONS OF INDONESIAN FRESHWATER GIANT PRAWN (*Macrobrachium rosenbergii*)

Keragaman DNA Mitokondria pada Empat Populasi Udang Galah (*Macrobrachium rosenbergii*) asal Indonesia

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ABSTRACT
*Mitochondrial DNA has been widely applied to analyze fish genetic diversity, especially the cytochrome oxidase subunit I (COI) gene, which can be used as a marker of typical variation patterns, both intraspecific and interspecific crossbreeding. Haplotype diversity was observed in four populations of giant freshwater prawns (*Macrobrachium rosenbergii*) originating from Peureulak River in Aceh, Tabuk River in South Kalimantan, Bengawan Solo River in East Java, and the Siratu strain. Haplotype diversity was observed in the mitochondrial DNA COI gene of 1516 bp and restricted using 5 enzymes, namely AvaII, MspI, RsaI, HaeIII, and TaqI. The diversity of mtDNA COI haplotypes in the population of Peureulak River, Tabuk River, Bengawan Solo River, and Siratu were 0.8183, 0.6222, 0.7461, and 0.8044, respectively. The dendrogram of genetic distance showed that the Bengawan Solo River populations and the Siratu strain were in the same cluster, while Peureulak River and Tabuk River populations were in another group. The four populations can be used as genetic sources in crossbreeding activities based on haplotype data for each population, haplotype diversity, and genetic distance.*

Keywords: *COI, genetic distance, haplotype diversity,* Macrobrachium rosenbergii*, mtDNA*

ABSTRAK
DNA Mitokondria telah banyak diaplikasikan untuk analisa keragaman genetik ikan, terutama gen cytochrome oxidase subunit I (COI) yang dapat digunakan sebagai penanda pola variasi yang khas baik pada persilangan intraspesifik maupun interspesifik. Keragaman haplotipe diamati pada empat populasi udang galah (*Macrobrachium rosenbergii*) yang berasal dari Sungai Peureulak di Aceh, Sungai Tabuk di Kalimantan Selatan, Sungai Bengawan Solo di Jawa Timur dan strain Siratu. Keragaman haplotipe diamati pada gen COI DNA mitokondria yang berukuran 1516 bp dan direstriksi menggunakan 5 enzim, yaitu AvaII, MspI, RsaI, HaeIII, dan TaqI. Keragaman haplotipe mtDNA COI pada populasi Sungai Peureulak, Sungai Tabuk, Sungai Bengawan Solo dan Siratu masing-masing adalah 0,8183; 0,6222; 0,7461 dan 0,8044. Dendrogram jarak genetik menunjukkan bahwa populasi Sungai Bengawan Solo dan galur Siratu berada dalam 1 kluster yang sama, sedangkan populasi Sungai Peureulak dan Sungai Tabuk berada pada kluster lainnya. Berdasarkan data haplotipe untuk setiap populasi, keragaman haplotipe dan jarak genetik keempat populasi tersebut dapat dijadikan sebagai sumber genetik dalam kegiatan persilangan.

Kata Kunci: COI, jarak genetik, keragaman haplotipe, *Macrobrachium rosenbergii*, mtDNA

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INTRODUCTION

Giant freshwater prawn (*Macrobrachium rosenbergii*) is a species of freshwater shrimp that has the potential to be developed to support animal protein-based of national food self-reliance and security programs. Giant freshwater prawns are Indonesian freshwater fishery commodities with high economic value for domestic consumption and export. The market demand that can be met is still limited to domestic consumption, especially in seafood restaurants, high-class restaurants, and high-star hotels. Meanwhile, export demand from Japan, the United States and European countries, and Singapore has not been fulfilled.

The development of giant freshwater prawn cultivation in Indonesia is relatively lower compared to other countries such as China, Vietnam, India, Thailand, and Bangladesh. Until this time, China is the number one producer of giant freshwater prawns, with a total production of 132,678 tons in 2016, while Indonesia is in 4th position after Bangladesh and Thailand, with a total output of 11,708 tons (Farook et al. 2019). The productivity of Indonesian giant freshwater prawns is still low, from 2010 to 2016, only increasing by 2.86% per year. Currently, the cultivation of giant freshwater prawns has developed in other countries such as the United States of America and China (Zafar et al. 2015). FAO (2020) reports that the global production of giant freshwater prawns has not shown a significant increase. Its production in 2018 reached 234,400 tons, only a rise of 7.67% compared to production in 2010. Furthermore, the production of giant freshwater prawns was only 2.5% of the total crustacean production.

In Indonesia, giant freshwater prawn seeds are currently being produced in hatcheries using wild catches for cultivation purposes. Domestication efforts and genetic improvement of giant prawns have been carried out. Giant freshwater prawns GMacro II and Siratu are two giant freshwater prawn strains produced by genetic improvement efforts released by the government in 2014 (KKP 2014) and 2015 (KKP 2015). Due to the limited availability of genetically improved giant freshwater prawns broodstock, the breeders make efforts to bring in natural broodstock from various locations to produce quality seeds. Crossbreeding between populations that are carried out usually comes from populations from different geographies. Crossbreeding between populations/ strains is common in fish farming to avoid inbreeding. Crossbreeding between controlled populations/ strains is expected to produce offspring that have better growth characteristics, are more resistant to disease, and have low levels of abnormalities (Tave 1993).

The crossbreeding process will be more effective if the genetic quality can be identified first. Genetic quality can be analyzed through the level of gene diversity because high genetic diversity in a population means that there are many individuals with a variety of different traits. Genetic diversity is significant for a population to adapt to a dynamic environment (Tave 1993). Aliah (2014) has carried out an analysis of genetic diversity on several tilapia strains (*Oreochromis niloticus*), which will be used to develop hybrid Salina tilapia resistant to high salinity. Then the same was done by Mastrochirico-Filho et al. (2019) on pacu fish (*Piaractus mesopotamicus*) to obtain a parent base population used in breeding activities for this species. Several studies on crosses between populations or strains that produce offspring that have better growth and survival have been reported by Aliah (2014) on Salina tilapia (*Oreochromis sp.*), Sunarma et al. (2016) on African catfish (*Clarias gariepinus*), Zeng at al. (2017) on mandarin fish (*Siniperca chuatsi*) and several strains of tilapia, *O. niloticus* (Novelo et al. 2021).

Several methods that can be used to analyze genetic diversity in fish include DNA microsatellites (Cordova-Alarcon et al. 2019), PASA (PCR amplification of specific alleles), and SNP (single nucleotide polymorphism) (Dudu et al. 2015). Genetic diversity can be estimated molecularly. One of them is by using genetic markers of mitochondrial DNA. The mitochondrial DNA control region (mtDNA CR) is the most variable part of the mtDNA, composed of a conservative central area with relatively divergent left and proper domains. It involves 3-5 times more rapidly than the other segments of the mitochondrial genome (Cheng et al. 2012, Li et al. 2022). In addition, the type of mutation in mtDNA is simple, namely base substitution or base

length mutation, and occurs mainly in small non-coding regions; thus, the mtDNA polymorphism is a neutral genetic marker. The COI (Cytochrome Oxidase I) region of mtDNA is generally applied in the genetic structure of the fish population (Ceruso et al. 2020) and reliable DNA barcodes for the identification and discrimination of fish species (Gao et al. 2020).

Mitochondrial DNA sequences or sequenced *mitogenomes* of giant freshwater prawns have been studied, including giant freshwater prawns from China with the size of 15,767 bp (Li et al. 2021), giant freshwater prawns from Indonesia with the size of 15,772 bp with the GenBank accession number of AY659990 (Ma et al. 2011) and giant freshwater prawns from China 15,766 bp (Li et al. 2019). Mitochondrial DNA sequences from several *Macrobrachium* genera, including *M. nipponense*, have a mitochondrial genome sequence of 15,806 bp (GeneBank ID NC_015073.1), and *M. lanchesteri* has a shorter mitochondrial sequence of 15,694 bp with GeneBank ID NC_0122217.1 (Ma et al. 2011). The mitochondrial DNA genetic markers have been successfully used to analyze differences in genetic diversity between natural and aquaculture populations (Nguyen Thanh et al. 2015), identify various types of tilapia species (Wu and Yang 2012), and evaluate the restocking program in fish species (Li et al. 2016, Maidin et al. 2017).

This study was conducted to identify the genetic diversity of four populations of giant freshwater prawns (*M. rosenbergii*) originating from the Peureulak River (Aceh), Tabuk River (South Kalimantan), Bengawan Solo River (East Java), and the Siratu (genetically improved strain), which will be used as parent base in the process of crossbreeding. The population was selected based on differences in the area's geographic location to obtain high potential for diversity, namely natural giant freshwater prawns from Sumatera Island, Kalimantan Island, Java Island, and genetically improved giant freshwater prawns. Genetic diversity was carried out using mitochondrial DNA COI gene markers. Hopefully, the genetic diversity data obtained can be used as primary data to determine parent combinations in the crossbreeding process.

Figure 1. Giant freshwater prawn, *M. rosenbergii*

MATERIALS AND METHODS

Location and time

Research on mitochondrial DNA diversity in four populations of Indonesian giant freshwater prawns (*M. rosenbergii*) was conducted at the Bioaquaculture Laboratory, Center for Agricultural Production Technology located at LAPTIAB BRIN, PUSPIPTEK, South Tangerang, Banten. This research was conducted from 2017-to 2018.

Materials

The 194 samples of caudal fin pieces were collected from 4 genetic sources of giant freshwater prawns (Figure 1), namely, 62 samples from the Peureulak River in Aceh (4°55'58" N 97°55'45" E, alt. 93 mamsl), 44 samples from the Bengawan Solo River in East Java (7°20'28" S 111°41'23" E, alt. 295 mamsl), 18 samples from the Tabuk River in South Kalimantan (3°17'49" S 114°42'14" E, alt. 37 mamsl) and 42 samples of giant freshwater prawns of Siratu strain from the Pelabuhan Ratu Prawn Hatchery Center (6°57'33" S 106°28'14" E, alt. 1.055 m mamsl). Giant freshwater prawns of the Siratu strain are the result of genetic improvement carried out by BBPBAT-KKP Sukabumi and have been released by the Minister of Fisheries and Marine Affairs in the Decree of the Minister of Fisheries and Marine Affairs Number 25/KEPMEN-KP/2015 on April 4, 2015 (KKP 2015).

The DNA extraction was done using the modified standard SDS-phenol/chloroform method (Green and Sambrook 2012). The DNA was extracted from 50 mg pleopod of giant freshwater prawn samples. The 700 µL of lysis solution (10 mM Tris-HCl pH 7.5, 1.5 M NaCl, 10 nM EDTA pH 7.5, 0.5% SDS, and 4 M Urea) and 5 µL

Proteinase K (1 mg μL^{-1}) were mixed and vortexed, before the incubation for 24 hours at 37ºC. After the incubation, 700 µL of phenol/chloroform/isoamyl alcohol (25:24:1) and chloroform/isoamyl alcohol (24:1) were added. The supernatant was taken and added to 10% 3M sodium acetate and 2 times the volume of absolute ethanol, followed by incubation on ice for 30 minutes and centrifugation at 5000 rpm for 10 minutes. The pellet was added with 1 mL of 70% ethanol and centrifuged again at the same speed. The supernatant was discarded, and the pellet was dried. The pellet was dissolved in TE buffer (10 mM Tris-HCl pH 7.2; 1 mM EDTA pH 8) with a volume of 20-100 µL and stored at 4ºC.

Cytochrome Oxidase I (COI) regions in mtDNA were selected to detect haplotype diversity. The D-loop regions were amplified using COI UD forward 5'-ACG CAA CGG TGG CTT TTC-3' primer (GenBank No. Acc. AY659990.1) and reverse 5'-TAG TTA GCT GTT AGG GGG AT-3' (Liu et al. 2007) with a target of 1516 bp. The PCR reaction proceeded as follows: pre-denaturation at 95ºC for 5 minutes followed by 35 cycles (95ºC for 1 minute, 56ºC for 30 seconds, and 72ºC for 1 minute). Furthermore, the diversity of COI mtDNA genes was analyzed using 5 restriction enzymes, namely *Ava*II (G/GCC), *Msp*I (CC/GG), *Rsa*I (GT/AC), *Hae*III (GG/CC), and *Taq*I (T/CGA). The enzyme restriction reaction used a total volume of 10 µL with the following composition: 1 µL of sample DNA, 1 U of *Ava*II enzyme, 2 µL of buffer O, and 6 µL of nuclease-free water.

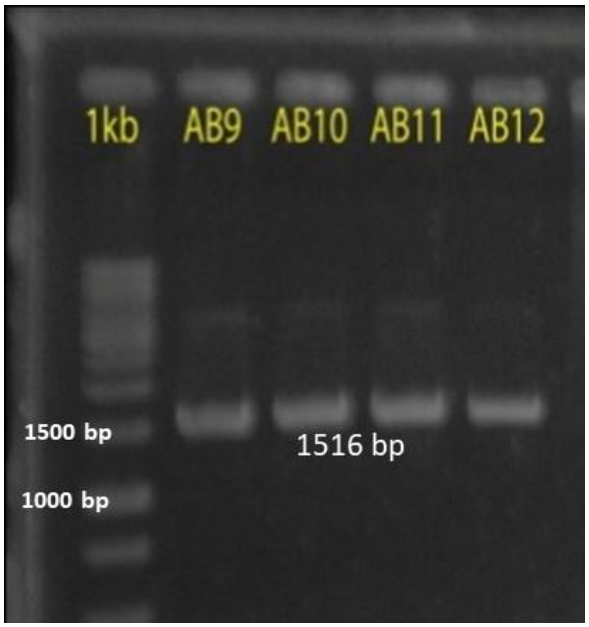

Figure 2. COI gene of mtDNA region in four female giant freshwater prawns from Peureulak River, Aceh (AB9-AB12) (1516 bp)

Analysis of mtDNA variation was carried out to detect genetic variation of giant freshwater prawns based on the presence or absence of restriction sites on mtDNA COI. Each restriction enzyme has a different restriction pattern, and each individual has a restriction pattern that may be different for each enzyme used. Based on the restriction pattern, a haplotype was made. Haplotype diversity (h) in a population is calculated according to Nei's (1978) equation:

$$h = 2n\,(\textstyle\sum_{i=1} xi^2)/(2n-1)$$

where,
h : haplotype diversity
n : sample size
xi: haplotype frequency of sample-i

The genetic distance between populations is presented in a UPGMA matrix and dendrogram, which was analyzed from haplotype data using the TFPGA (tools for population genetic analyses) program ver 1.3.

RESULTS AND DISCUSSION

The COI mtDNA gene from 166 giant freshwater prawn samples was amplified using primers COI UD forward (5'-ACG CAA CGG TGG CTT TTC-3') and COI UD reverse (5'-TAG TTA GCT GTT AGG GGG AT-3') with the size of 1,516 bp (Figure 2). Cytochrome Oxidase I (COI) region is one of the protein-coding for mtDNA. The COI gene is commonly used as a genetic marker in population studies. Species with very closely genetic related, can be categorically distinguished using mtDNA COI sequence differences at particular nucleotide positions (In et al. 2017).

The restriction type of *Ava*II and *Taq*I enzymes produced two patterns (Figure 3a), *Hae*III enzymes had three patterns (Figure 3b), *Msp*I enzymes had four patterns, and *Rsa*I enzymes produced three patterns (Figure 3c). The results of the restriction of the COI mtDNA area using five enzymes *Ava*II, *Msp*I, *Rsa*I, *Hae*III, and *Taq*I in four populations of giant freshwater prawns

produced 30 haplotypes. The giant freshwater prawn population from Peureulak River, Aceh has 15 haplotypes, the population from Tabuk River, South Kalimantan, has five haplotypes, the population from Bengawan Solo River has eight haplotypes, and the population strain Siratu has 10 haplotypes (Table 1). The AABAA haplotype was found in three observed populations, namely the population of Tabuk River, Peureulak River, and Bengawan Solo River, with the frequencies of 0.5556, 0.2742, and 0.0227, respectively. Two haplotypes, namely ABCBA and ABCCA, were found in Bengawan Solo River and Siratu populations. In comparison, two other haplotypes, AABBA and AABCA, were found in the populations of Peureulak River and Bengawan Solo River. Several haplotypes were found dominant in each population, namely AABAA in the Tabuk River population with the frequency of 0.5556, ABCAA in the Siratu population with the frequency of 0.3571, and ABCBA in the Bengawan Solo River population with the frequency of 0.4545. Furthermore, two haplotypes were found in the Peureulak River population, namely BABAB and AABAA, with the frequencies of 0.3065 and 0.2742, which were relatively high compared to other haplotypes in the population.

The diversity of haplotypes in a population is highly dependent on the number of detected haplotypes and the frequency of each haplotype. The haplotype diversity observed in the four giant freshwater prawn populations was 0.8183 in the Peureulak River population, 0.8044 in the Siratu

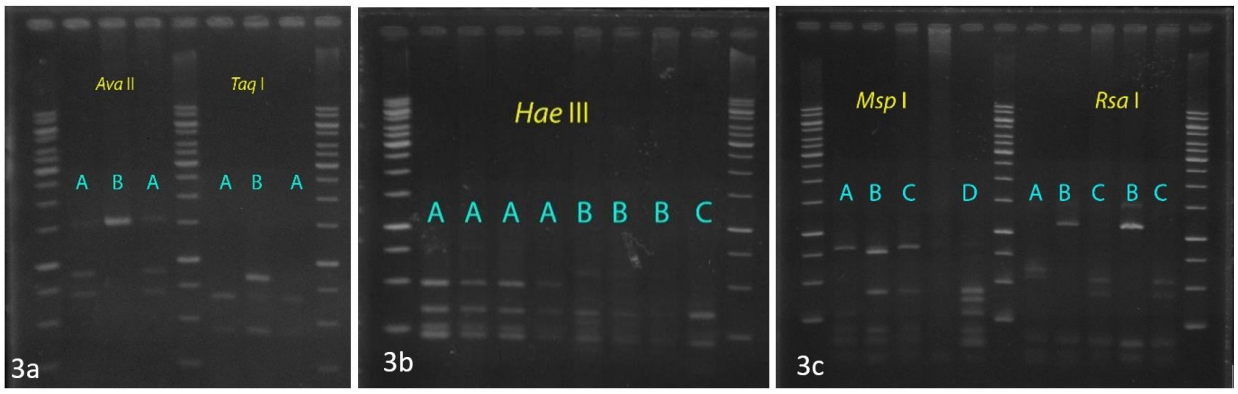

Figure 3. Restriction patterns of COI gene of mtDNA in 4 populations of giant freshwater prawn (*M. rosenbergii*) by using AvaII and TaqI (a), *Hae*III (b), *Msp*I and *Rsa*I (c) enzymes

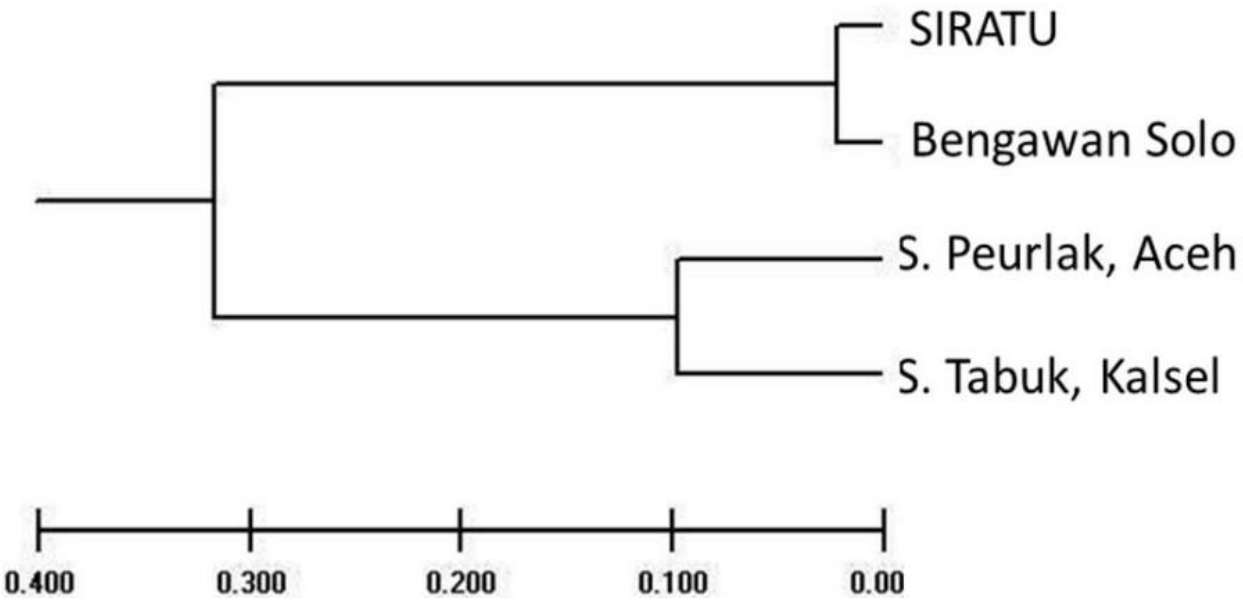

Figure 4. UPGMA dendrogarn based on Nei's genetic distance among 4 populations giant freshwater prawn (*M. rosenbergii*) on COI mtDNA sequences

Table 1. Haplotype diversity in 4 populations of giant freshwater prawn (*M. rosenbergii*) based on haplotype frequency of mtDNA COI gen with 5 restriction enzymes (*Ava*II, *Msp*I, *Rsa*I, *Hae*III, and *Taq*I)

No.	Haplotype	S. Peurlak		B. Solo		S. Tabuk		SIRATU	
		n	frequency	n	frequency	n	frequency	n	frequency
1	AAAAA	2	0.0323			1	0.0556		
2	AAABA							1	0.0238
3	AABAA	17	0.2742	1	0.0227	10	0.5556		
4	AABAB	5	0.0806						
5	AABAC					1	0.0556		
6	AABBA	3	0.0484	1	0.0227				
7	AABCA	1	0.0161	5	0.1136				
8	AABCB	2	0.0323						
9	AACAA	1	0.0161						
10	AACAB	1	0.0161						
11	AACBA			6	0.1364				
12	AACCA			5	0.1136				
13	AADCB	1	0.0161						
14	ABBAA					5	0.2778		
15	ABBBA			2	0.0455				
16	ABCAA							15	0.3571
17	ABCBA			20	0.4545			6	0.1429
18	ABCCA			4	0.0909			8	0.1905
19	ACCAA							1	0.0238
20	ACCBA							4	0.0952
21	ACCCA							1	0.0238
22	ADBAA	1	0.0161						
23	ADCBA							1	0.0238
24	BAAAA	1	0.0161						
25	BABAA	4	0.0645			1	0.0556		
26	BABAB	19	0.3065						
27	BABBA							4	0.0952
28	BABBB	3	0.0484						
29	BABCB	1	0.0161						
30	BBBBA							1	0.0238
No. of Sample			62		44		18		42
No. of Haplotype			15		8		5		10
Haplotype Diversity			0.8183		0.7461		0.6222		0.8044

population, 0.7461 in the Bengawan Solo River population, and 0.6222 in the Tabuk River giant freshwater prawn population (Table 2). Referring to the genotypic diversity index according to Nei (1978), namely 0.1-0.4 for low diversity, 0.5-0.7 for moderate diversity, and 0.8-1.0 for high diversity. The haplotype diversity of giant freshwater prawns from the Tabuk River was classified as moderate, while giant freshwater prawns from Peureulak River, Bengawan Solo River, and Siratu strains were classified as high. The lower haplotype diversity in the shrimp population from Tabuk River was probably caused by the smaller number of samples observed compared to the sample size of three other giant freshwater prawn populations.

The more similar haplotypes the giant freshwater prawn populations observed, the closer their kinship or the smaller genetic distance value. The genetic distance value in the form of a matrix among four giant freshwater prawn populations is presented in Table 2. The genetic distance between the giant freshwater prawn population of Bengawan Solo River and the Siratu strain is the closest at 0.0202, while the genetic

Table 2. Genetic distance between populations of giant freshwater prawns (M. rosenbergii) from Peureulak River (Aceh), Bengawan Solo River (East Java), Tabuk River (South Kalimantan) and Siratu

	Peureulak River	Bengawan Solo River	Tabuk River	Siratu
Peureulak River	*******			
Bengawan Solo River	0,3599	*******		
Tabuk River	0,0959	0,2766	*******	
Siratu	0,3603	0,0202	0,2641	*******

distance between the Peureulak River population and Siratu is the farthest with a value of 0.3603.

The genetic distance dendrogram presented in Figure 4 shows that the giant freshwater prawns from Bengawan Solo River and Siratu are closely related to form a cluster far apart from giant freshwater prawns derived from Peureulak River and Tabuk River. Furthermore, the genetic distance of the giant freshwater prawn Siratu – Bengawan Solo River was much smaller (0.0202) than the genetic distance between the giant freshwater prawn populations from Peureulak River and Tabuk River (0.0959).

In this study, mitochondrial DNA genetic markers have been carried out to identify and analyze the diversity of the giant freshwater prawn population, which will be used as the primary parent population in crossbreeding activities. Mitochondrial DNA genetic markers successfully examined the differences in genetic diversity of natural and aquaculture populations of giant freshwater prawns in China and Vietnam using partial sequences of the 16S rDNA gene and mitochondrial DNA COI gene (Nguyen Thanh et al. 2015). This method was also applied to identify various types of tilapia: the *Oreochromis* genus, *Sarotherodon* genus, *Tilapia,* and the hybrid *O. massambicus* and *O. niloticus,* which have been introduced to Hawaii (Wu and Yang 2012). Moreover, Zhang et al. (2013) found a decrease in the genetic diversity of sturgeon fish (*Acipenser sp.*) in nature due to the presence of hybrid sturgeon fish second-generation mass production in hatcheries released into the wild. Zhang et al. (2016) proved that mitochondrial DNA markers were inherited "maternally" in the hybridization process between 4 fish species, *Megalobrama sp* and *Parabramis pekinensis* first and second generations. Furthermore, mitochondrial DNA genetic markers were used to evaluate the restocking program on the endangered species *Percocypris pingi* in the Yalong River, China (Li et al. 2016). They assessed the success of restocking activities of giant freshwater prawns (*M. rosenbergii*) to the Petagas River in the Sabah region, Malaysia, between 2012 – 2015 (Maidin et al. 2017).

According to Nei (1978), the genetic diversity of giant freshwater prawn populations from the Peureulak River and Siratu strains is relatively high. This indicates that genetic improvement activities in giant freshwater prawns have been successful and can be used to produce good quality seeds of giant freshwater prawns as long as the parent management is carried out properly. Several comparisons of genetic diversity between natural and cultivated populations have been completed to anticipate the effect of inbreeding in cultured populations whose seeds are produced in hatcheries. Nguyen Thanh et al. (2015) analyzed the mtDNA diversity in cultured prawn populations from Zhejiang, Guangdong, and Guangxi provinces in China and two natural populations from the Mekong River and Dong Nai River in Vietnam. The results showed that wild populations had higher genetic diversity than cultured populations. Bala et al. (2017) also reported that the genetic diversity of the giant freshwater prawn population in Bangladesh of natural origin was higher than the genetic diversity of the giant freshwater prawn population in the hatchery. Furthermore, Li et al. (2016) reported that the restocking activity of *P. pingi* fish species hatchery products in Moulo River, China, did not affect the level of genetic diversity of natural populations, which remained high and higher than the genetic diversity of hatchery populations, even though the number of fish species in the nature is much decreased due to anthropogenic factors.

On the other hand, the genetic diversity of giant freshwater prawn populations in four rivers in the Malaysia Peninsula has decreased its heterozygosity, and inbreeding occurs due to overfishing of giant freshwater prawns to produce seeds in hatcheries (Atin et al. 2017). Furthermore, Binur and Pancoro (2017) observed the diversity of four microsatellite DNA loci of Indonesia's giant freshwater prawn hatchery population. The results indicated inbreeding in the moderate category at the post-larval stage at 4 giant freshwater prawn hatchery locations on Java Island. Therefore, using post larvae from these four hatcheries is not recommended for potential broodstock. Different results were reported by Li et al. (2017); in spotted barbell fish (*Hemibarbus maculates*), cultured populations produced from controlled hatcheries have better genetic diversity than natural populations that are naturally selected due to the decrease in environmental quality.

The kinship pattern of a population occurs because of the spread and process of migration (gene flow), which moves genetic material from one population to another. If there are two populations with different genetic structures and then there is a transfer of genes between populations within a specific time, it will be genetically similar. The transfer of genes between populations can be caused by the movement of individuals or populations naturally due to the location of the habitats of these two populations being close together or due to human intervention.

In this study, the kinship pattern among four observed giant freshwater prawn populations could be analyzed from the resulting genetic distance. The Java Island cluster consisting of the Bengawan Solo River population and the Siratu strain had a lower genetic distance value than the outside of the Java Island. The Java Island cluster consists of Peureulak River and Tabuk River populations, with high genetic distance values between them, reflecting the absence of gene flow from these two natural populations. Furthermore, the results showed that not even one haplotype was detected simultaneously in the 4 populations.

This genetic diversity and distance observed in this study are valuable basic information for determining the combination of parental populations in cross-breeding to obtain offspring with the desired traits. The results of the analysis showed that the genetic diversity of the four giant freshwater prawn populations is relatively high (0.6222 – 0.8183) and has a relatively large genetic distance so that it can become the primary population for cross-breeding. The hybrid/crossbreed giant freshwater prawns are expected to be positive heterosis, which means they have better characteristics than their parents.

The National Research and Innovation Agency (known as BRIN) has initiated programs to improve the genetic quality of giant freshwater prawns, one of which is cross-breeding activities to obtain hybrids with superior characteristics. For this crossing activity, giant freshwater prawns have been obtained from three different geographic locations, plus the genetically improved giant freshwater prawn strains released by the government. Based on the results of the analysis of genetic diversity and genetic distance of the four observed populations of giant freshwater prawns, namely Peureulak River, Tabuk River, Bengawan Solo River, and Siratu, a preliminary trial of reciprocal crossbreeding between populations of (1) Peureulak River and Bengawan Solo River, (2) Bengawan Solo River and Siratu and (3) Peureulak River and Siratu. The results of rearing for 120 days showed that reciprocal crossbreeding of (1) Peureulak River and Bengawan Solo River at the juvenile stage had daily growth rates and better survival rates than reciprocal crossbreeding of (2) Bengawan Solo River and Siratu and (3) Peureulak River and Siratu.

CONCLUSION

The diversity of mitochondrial DNA COI gene haplotypes observed in giant freshwater prawn populations from Peureulak River, Tabuk River, Bengawan Solo River, and Siratu strains showed relatively high diversity ranging from 0.6222 to 0.8183. The genetic distance between these four populations ranged from 0.0202 to 0.3603 and formed two clusters: the Bengawan Solo River population – the Siratu strain cluster, and the Peureulak River population – Tabuk River population cluster. Based on the haplotype diversity data and genetic distance, these four giant freshwater prawn populations can be used as genetic sources for crossbreeding activities.

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