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BIODIVERSITY, DISTRIBUTION AND ABUNDANCE OF THE TROPICAL ANGUILLID EELS IN THE INDONESIAN WATERS

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ABSTRACT

In order to understand biodiversity, distribution, and abundance among the tropical anguillid eels in the Indonesian waters, inshore migration mechanism of the juvenile anguillid eel (glass eel) to the estuaries of western, central, and eastern region of Indonesian waters were examined using both morphology and genetic analyses. A total of 9 species/sub species of anguillid eels (*Anguilla bicolor bicolor*, *A. nebulosa nebulosa*, *A. bicolor pacifica*, *A. interioris*, *A. borneensis*, *A. celebesensis*, *A. marmorata*, *A. obscura* and *A. megastoma*) were recognized to recruit at the mouth of 15 rivers through Indonesian archipelago. Species/subspecies diversity and distribution of recruiting juveniles differs in the estuaries of each region. In the western region that represented by estuary of Krueng Aceh, Batang Antokan, Air Kertaun, Cibaliung, Citanduy, Progo, and Pacitan Rivers, three species and sub species (*A. bicolor bicolor, A. nebulosa nebulosa* and *A. bicolor pacifica*) were found. In the central region that represented by estuary of Mahakam, Palu, Dumoga, Poigar, Bone, and Poso Rivers, five species and sub species (*A. borneensis*, *A. celebesensis*, *A. marmorata, A. bicolor pacifica* and *A. interioris*) were found. In the eastern region that represented by estuary of Akelamo and Pami Rivers, four species (*A. marmorata, A. interioris, A. obscura* and *A. megastoma*) were found. During the 6 months investigation from May to October 2005, abundance of the juveniles was higher in the central region compared with western and eastern regions. These results were suggested that inshore migration mechanism of tropical anguillid eels recruiting in tropical estuaries of Indonesian waters differs among regions.

Keywords: Tropical anguillid eel, Biodiversity, Distribution, Abundance, Inshore migration

INTRODUCTION

Anguillid eels traditionally have been an important fish as a food resource in many eastern and western countries. In Japan, the Japanese eel (*Anguilla japonica*) has long been esteemed as an important food fish that has a unique taste. Presently, as much as 130,000 tons of eels are consumed each year in Japan (Tsukamoto, 1999). Similarly, in the European countries, the Atlantic eels (*A. anguilla* and *A. rostrata*) have been an integral part of the cuisine of several countries. However, the decrease of eel resources has been a serious problem in recent years. It is not yet clear whether this has been caused by global changes in the ocean, atmospheric system, the human impacts of over-fishing and environmental deterioration, or intra-specific or intra-population biological factors. As impact, for the last recent year import of tropical anguillid eels from the South-east Asia countries become a new trend to solve the problem. In order to counteract the decrease in eel resources it is important to understand the underlying causes and mechanisms of these changes, and to develop effective management strategies for maintaining stable eel populations. Therefore, the first goal should be to use a bio-ecological approach to gain a complete understanding of biodiversity, distribution and abundance of the freshwater eel in Indonesian waters as the greatest country in the Southeast Asian region.

Two thirds of the recognized 18 species and sub species of eel genus *Anguilla* are found in the tropical Pacific Ocean, while only 6 are found

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in temperate regions of both the Pacific and Atlantic Oceans. Seven species and sub species of tropical eels occur in the western Pacific around Indonesia (Ege, 1939; Castle and Williamson, 1974; Aoyama and Tsukamoto, 1997; Arai *et al.*, 1999a, Sugeha *et al.*, 2001; Sugeha, 2003). Both morphological and genetic studies indicate that tropical eels are more closely related than temperate eels to the ancestral eel (Ege, 1939; Castle dan Williamson, 1974; Aoyama, 1998). According to Aoyama *et al.* (2001), anguillid eels originated near present day Indonesia and dispersed to both the east and west along paleo-circum equatorial current and the authors suggested that *A. borneensis* from Borneo (Kalimantan) Island was the most basal species. Thus the long distance catadromous migrations of anguillid eel may have originated in the tropical species, and biological study of the tropical freshwater eels may provide a greater understanding of the origin of the catadromous migration of anguillid eels. The newest study on the molecular phylogeny and evolution of the catadromous eel had suggest that *A. mossambica* from Africa was the most basal species of anguillid eels (Minegishi *et al.*, 2005). However, the authors also notice that *A. borneensis* from eastern part of Kalimantan Island (Indonesia) separated from the three geographic clades (Atlantic, Oceania, Indo-Pacific) that performed by the other species of anguillid eels. In fact, the similar phenomenon also found in the molecular genetic study on the living fossil of Coelacanth from Comoro Island (Africa) and Manado Tua Island (Indonesia). Which one the most ancestral is still unclear and just based on assumption (Inoue *et al.*, 2005). This controversy had to show a complicated biodiversity in the Indonesian Waters that may possible to inhabit by ancestor of many aquatic living organisms. Therefore, it is important to study on the biodiversity, distribution, and abundance of aquatic living organisms in Indonesian Waters.

Here, we report the recent study on the biodiversity, distribution, and abundance of tropical angullid eels in the Indonesian Waters. The objective of the study was to understand the present status on the species diversity, geographic distribution, and fluctuation in abundance of the tropical anguillid eels during their inshore migration period based on morphology and genetic analyses.

METHODS

Adjustment of sampling regions: Indonesian waters has a total area of Economic Exclusive Zone (EEZ) about 2,730,000 km^2 that stretching roughly from 6°N to 10°S and from 95°E to 142°E (Tomascik *et al.*, 1997). Further, Indonesian archipelago may be roughly divided into three regions: the western region over the Sunda Shelf, the eastern region over the Sahul Shelf and central deep ocean region between the two shelves (Tomascik *et al.*, 1997). Based on these references, we divided our sampling field in the three regions: western (95°E to 110°E), central (111°E to 125°E), and eastern (126°E to 140°E) regions in order to cover biodiversity, distribution and abundance of the tropical anguillid eels in the Indonesian waters. Western region represented estuaries of Krueng Aceh, Batang Antokan, Air Kertaun, Cibaliung, Citanduy, Progo and Pacitan Rivers. Central region represented by estuary of Mahakam, Palu, Dumoga, Poigar, Bone, and Poso Rivers. Eastern region represented by estuary of Akelamo and Pami Rivers.

Specimen collection: About 5000 specimen of tropical anguillid eels collected from 15 estuaries in the Indonesian waters were used for the present study. They were caught along the beach using 2 triangular scoop nets (mouth 0.3m^2, 1mm mesh) following the sampling technique by Sugeha *et al.* (2001), except for all specimens from Mahakam (East Kalimantan) that collected using trap and net. Just after captured, the specimen were divided in two parts and fixed in 10% formalin and 99% ethanol respectively before labeled and transported to the laboratory for future analysis. A total of 327 specimens were used for genetic analysis while 325 specimens among them were used for morphology analysis.

Morphology and genetic analysis: Prior to genetic analysis, body length measurement including total length (TL) pre-dorsal length (PDL), pre-anal length (PAL), and ano-dorsal length (ADL) of 325 individuals of tropical glass eel were done to the nearest 0.1mm. Pigmentation observation was determined according to Bertin (1956) in order to adjust the developmental stage of the specimens.

Total genomic DNA (deoxyribonucleic acid) extraction from 327 specimens of glass eel was carried out following a standard protocol (Aoyama and Tsukamoto, 1997). DNA was isolated and purified using phenol-chloroform-isoamyl alcohol

(25:24:1, v/v) twice with diethyl ether, then concentrated by ethanol precipitation before finally suspended in the TE solution and stored in the freezer. A portion of the mitochondrial 16S ribosomal RNA gene was amplified via polymerase chain reaction (PCR) using the oligo-nucleotide primers that were nested in the 16SrRNA: L1854: 5'-AAA-CCT-CGT-ACC-TTT-TGC-AT- 3' (Aoyama, 1998) and H3058: 5'- TCC-GGT-CTG-AAC-TCA-GAT-CAC-GTA- 3' (Miya and Nishida, 1996). The PCR was carried out with the Gen Amp PCR system 7200 (Applied biosystem), with a 25µl reaction volume containing 13.8µl sterile distilled water, 2.5µl 10XPCR buffer (Perkin Elmer-Cetus), 2.5µl dNTP (deoxynucleotide triphospate) of 2 mM, 2.5µl each primer of 5mM, 0.4µl of Taq DNA polymerase (AmpliTaq, Perkin Elmer Cetus), and 50 to 1,000ng of template DNA. Amplification parameters were 30 cycles of denaturation at 94°C for 15sec, annealing at 55°C for 15sec, and extension at 72°C for 30sec.

To develop a method of identification of anguillid species from the Indonesian Waters, a longer double-stranded mitochondria DNA product from PCR was examined using Restriction Fragment Length Polymorphism (RFLP) analysis with the six type of restriction enzymes *Alu* I, *Hha* I, and *Bsp* 1286I (Promega); *Eco*T14I and Mva I (Takara Shuzo Co., Ltd); and BbrP I (Toyobo Co., Ltd) which made it possible to identify the species of tropical anguillid eels as described in Aoyama *et al.* (2000b), and Sugeha (2003). Restriction procedures were carried out in a 15µl final volume containing 5µl PCR product, 1µl restriction enzyme, 1.6µl restriction enzyme buffer supplied by manufacturer and 7.5µl sterile distilled waters and incubated at 37°C overnight. Restriction fragment length polymorphism (haplotype) was detected by electrophoresis on 2% agarose gel with ethidium bromide staining.

RESULTS

Morphology and genetic character of tropical anguillid eels

Prior to species adjustment using genetic analysis, there were a total of 325 specimens that were morphologically analyzed in the study based on measurement of body length. Seventy five percent (244 specimens) of these glass eel were long-finned eels species and the rest (81 specimens) were short-finned eels, based on their percentage of ano-dorsal length to total length (ADL/%TL) as reported by Sugeha *et al.* (2001). Based on the geographic distribution and the range in ADL/%TL of the tropical anguillid eel species that ever reported (Ege, 1939; Jespersen, 1942; Arai *et al.*, 1999; Sugeha *et al.*, 2001; Sugeha, 2003), it was found in this study the short-finned eels that could belongs to *A. bicolor bicolor, A. bicolor pacifica*, and *A. obscura* were about -3 to 5 in the range of ADL/%TL. The long-finned eels species has a great overlap in range of ADL/%TL (7~15) and could be identified as *A. borneensis, A. celebesensis, A. interioris, A. nebulosa nebulosa, A. megastoma,* and *A. marmorata.* Therefore, based on the external morphology analysis it was found that all specimen could be separated only in 2 categories: short-finned eels and long-finned eels. Further observation on the pigmentation development of the glass eel specimens shown that all specimens belongs to stage VA (75%) and VB (25%) and suggested that there are completed the metamorphosis stage and just enter the freshwater area or in the glass eel stage when caught.

The PCR analysis of the 16S ribosomal RNA gene from DNA mitochondria of the tropical anguillid eels showed several different restricted fragment patterns or different haplotype after RFLP-analysis. The restriction enzymes *Alu* I, *Bbr*P I, and *Mva* I showed two different haplotypes. The enzymes *Eco*T14 I and *Bsp*1286 I exhibited five different haplotypes. The restriction enzymes *Hha* I only showed one haplotypes. Thus the 16SrRNA processed by the six restriction enzymes of *Alu* I, *Bbr*P I, *Mva* I, *Eco*T14 I, *Bsp*1286 I and BbrP I, clearly exhibit unambiguous fragment pattern whose haplotypes were designated alphabetically.

All the six restriction enzymes used in the study showed a similar genetic character as ever reported except for the restriction enzymes *Bsp*1286 I that exhibited different genetic character of fragment pattern which were never described by Aoyama *et al.* (2000), Watanabe (2001), and Sugeha (2003). Based on the similarity of the restriction fragment pattern between the previous and the present study than the species-specific haplotype of the tropical anguillid eels in the present study could be precise as follows as *A. bicolor bicolor*, *A. nebulosa nebulosa*, *A. bicolor*

pacifica, *A. interioris*, *A. borneensis*, *A. celebesensis*, *A. marmorata*, *A. obscura* and *A. megastoma*.

Species/sub species diversity and distribution

Species/sub species diversity and distribution of recruiting juveniles was observed in each region of Indonesian Waters (western, center, and eastern region) (Fig. 1). Sampling area in the western region represented by estuary of Krueng Aceh (Northern Sumatera), Batang Antokan (West Sumatera), Air Kertaun (South Sumatera), Cibaliung (West Jawa), Citanduy (Central Jawa), Progo (Central Jawa), and Pacitan (East Jawa) Rivers. Three species and sub species (*A. bicolor bicolor, A. nebulosa nebulosa* and *A. bicolor pacifica*) were found to inhabit the western region of Indonesian waters. However species composition differed in each sampling area. *Anguilla bicolor bicolor* were found in estuary of Krueng Aceh and Batang Antokan Rivers. *Anguilla bicolor pacifica* were found almost in all sampling area (estuary of Air Kertaun, Cibaliung, Progo, and Pacitan Rivers). *Anguilla nebulosa nebulosa* were detected to enter estuary of Batang Antokan and Air Kertaun Rivers.

In the central region that represented by Muara Muntai of Mahakam River and estuary of Palu, Dumoga, Poigar, Bone, and Poso Rivers, five species and sub species (*A. borneensis*, *A. celebesensis*, *A. marmorata, A. bicolor pacifica* and *A. interioris*) were found. Species composition differs in each sampling area. *Anguilla borneensis* was the only one species found in Muara Muntai of Mahakam River (East Kalimantan). *Anguilla celebesensis* was detected to enter estuary of Palu River (West Sulawesi) together with *A. marmorata and A. bicolor pacifica*. Estuary of Dumoga River (North Sulawesi) was entered by *A. celebesensis, A. marmorata, A. bicolor pacifica,* and *A. interioris.* Estuary of Poigar River (North Sulawesi) were inhabited by *A. celebesensis, A. marmorata,* and *A. bicolor pacifica* while estuary of Poso River (Central Sulawesi) were inhabited by *A. marmorata, A. celebesensis, A. bicolor pacifica, and A. interioris*. Estuary of Bone River (Gorontalo) was entered by *A. marmorata, A. bicolor pacifica,* and *A. celebesensis*.

In the eastern region that is represented by estuary of Akelamo and Pami Rivers, four species (*A. marmorata, A. interioris, A. obscura* and *A. megastoma*) were found. *Anguilla marmorata* were found to enter both estuaries, but *A. obscura* only found in the specimen from estuary of Akelamo River (Halmahera) while *A. interioris* only detected in the specimen from Pami River estuary (West Papua). *Anguilla megastoma* was

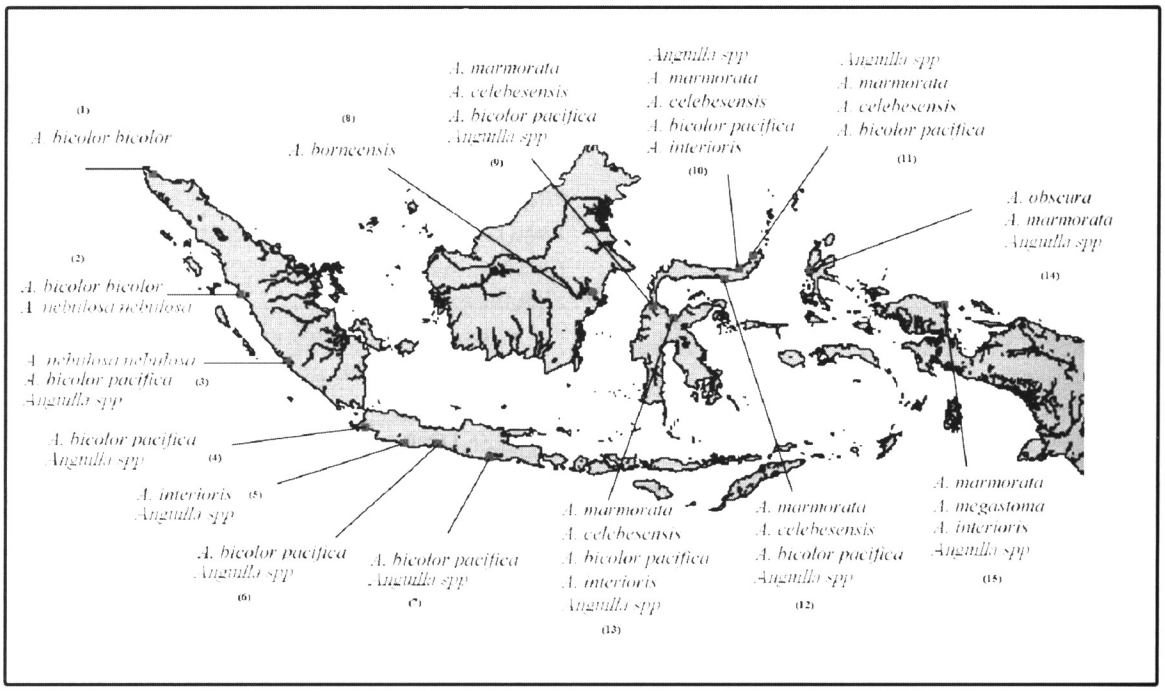

Figure 1. Present status on the geographic distribution of tropical anguillid eels in the Indonesian Waters.

Table 1. Species composition of the tropical anguillid eels recruited in the 15 estuaries around Indonesian Waters

No.	Sampling location	Region	Species/subspecies
1	Estuary of Krueng Aceh River	West	*A. bicolor bicolor*
2	Estuary of Batang Antokan River	West	*A. bicolor bicolor, A. nebulosa nebulosa*
3	Estuary of Air Kertaun River	West	*A. bicolor pacifica, A. nebulosa nebulosa, Anguilla* spp
4	Estuary of Cibaliung River	West	*A. bicolor pacifica, Anguilla* spp
5	Estuary of Citanduy River	West	*A. interioris, Anguilla* spp
6	Estuary of Progo River	West	*A. bicolor pacifica, Anguilla* spp
7	Estuary of Pacitan River	West	*A. bicolor pacifica, Anguilla* spp
8	Estuary of Mahakam River	Central	*A. borneensis*
9	Estuary of Palu River	Central	*A. celebesensis, A. marmorata, A. bicolor pacifica, Anguilla* spp
10	Estuary of Dumoga River	Central	*A. celebesensis, A. marmorata, A. bicolor pacifica, A. interioris, Anguilla* spp
11	Estuary of Poigar River	Central	*A. celebesensis, A. marmorata, A. bicolor pacifica, Anguilla* spp
12	Estuary of Bone River	Central	*A. celebesensis, A. marmorata, A. bicolor pacifica, Anguilla* spp
13	Estuary of Poso River	Central	*A. celebesensis, A. marmorata, A. bicolor pacifica, A. interioris, Anguilla* spp
14	Estuary of Akelamo River	East	*A. marmorata, A. obscura, Anguilla* spp
15	Estuary of Pami River	East	*A. marmorata, A. interioris, A. megastoma, Anguilla* spp

found to enter estuary of Pami River. All species and sub species diversity and distribution was shown in Table 1.

Abundance

During the 6 months investigation from May to October 2005, abundance of the juveniles of tropical angullid eels was higher in the central region compared with western and eastern regions (Fig. 2a). As the most abundant area, center region that represented by Dumoga River and Palu River estuaries has a total number of catch up to 50000 individuals during the sixth month of investigation. Eastern region was in the second position with a total number of catch more than 100 individuals during the six month investigation. Western region that represented by Batang Antokan and Citanduy River estuaries has only less than 50 individuals during the six month of investigation.

Fluctuation in abundance was recorded during the six month of investigation and has to show differences in pattern of abundance between the three regions (Fig. 2b). In the western region, glass eel appeared in May but drastically decreased from June to September, and no specimen collected in October. In the central region, glass eel appeared in June and drastically increased from July to August before decreased from September to October. In the eastern region, no glass eels collected in May and June, but glass eels appeared from July and gradually increased from September to October.

Time in peak of abundance was clearly detected in the central region. August was the time of peak in abundance for glass eels that entering Dumoga River estuary while July was the time of peak in abundance for glass eels that entering Palu River estuary. Time in peak of abundance in the eastern region was in September and October but was not clearly seen in the western region since the number of catch was very few. However, a relatively larger number of catch in May in Cibaliung River estuary could be indicating as the time in peak of abundance in western region of Indonesian Waters compared with other month.

It could be seen that time in peak of abundance in central region falls in the middle of the year, western region was in the early of the year, and eastern region was in the late of the year. Further, time in peak of abundance in western and eastern regions occurs during rainy season while in central region occurs in the dry season.

DISCUSSION

Biodiversity and distribution of tropical anguillid eels

Using external morphological analysis of the quantitative catch sample of glass eels at some estuaries around Indonesian Waters, the same several species of *Anguilla* were identify by morphology and genetic analysis based on Aoyama *et al.* (2000), Arai *et al.* (1999), Watanabe (2001) and Sugeha (2003) studies. These were short-finned eels with a range of ADL/%TL about -3 to 5 and long-finned eels with the range of ADL/%TL 7 to 20. The three sub-species of short-finned eels (*A. bicolor bicolor*, *A. bicolor pacifica*, and *A. obscura*) could be distinguished genetically by

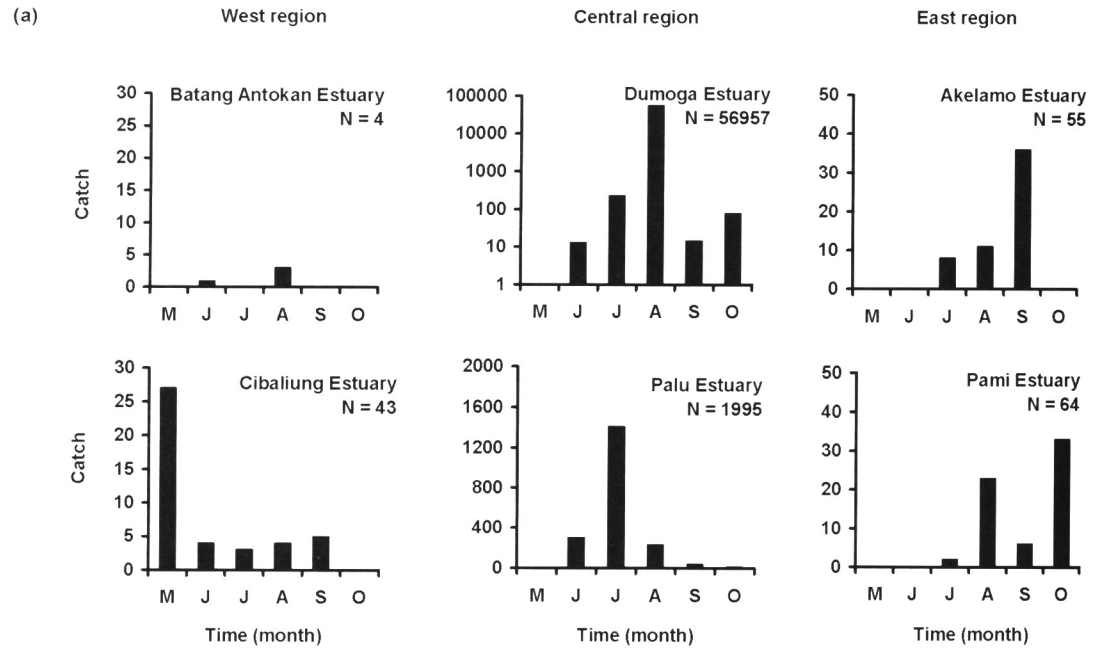

Figure 2(a). Fluctuation in abundance of tropical anguillid eels recruiting in the estuaries of Indonesian Waters from May to October 2005.

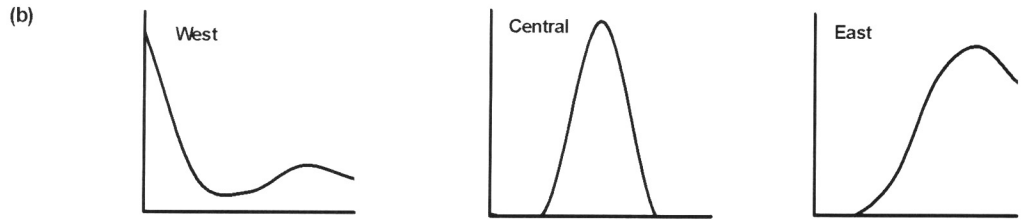

Figure 2(b). Three different patterns in abundance detected in the three different regions of Indonesian Waters.

different haplotype but their range of ADL/%TL overlapped from -1 to 3 and could not be used for species/sub-species identification. Similar condition was observed in the long-finned eels. Genetically the long-finned eel species of *A. nebulosa nebulosa, A. borneensis, A. interioris, A. celebesensis*, *A. marmorata* and *A. megastoma* could be identify based on their species-specific haplotype but morphologically they could not be distinguished since their range of ADL/%TL showed a great overlap from 7 to 20. It was found also that all characters of body measurement outside ADL/%TL showed overlapping in ranges including total length (TL), pre-dorsal length (PDL), and pre-anal length (PAL). These finding reconfirmed that the external morphology (body measurements) alone could not be used for species identification of tropical anguillid eels around Indonesian waters. This study also has to complete similar study on the genetic diversity that ever reported. The present study has been rising up a new perspective on the critical condition of species identification of anguillid eels in the tropic. It has become important to understand that the morphological criteria of short-finned and long-finned anguillid eels could not be applied for the tropical eel species and new criteria of taxonomy works in the tropical angullid eels are strongly required based on confirmation and crossing check between morphology and genetic study.

The PCR-RFLP analyses clearly identified the species of the 327 specimens of anguillid eels from some estuaries around Indonesian Waters based on the species specific haplotype found in the study. There are nine species and sub species that are

genetically identifiable in the study as the following: *A. nebulosa nebulosa*, *A. interioris*, *A. marmorata*, *A. borneensis*, *A. celebesensis*, *A. bicolor pacifica*, *A. bicolor bicolor*, *A. obscura*, and *A. megastoma*. The nine species and sub species recognized in the present study completely supported by previous study on the diversity of anguillid eel species in the Indonesian Waters (Aoyama *et al.*, 2001). The author reported about 7 species and sub species which inhabit the region. Interestingly, *A. obscura* has been found in the present study even the range of distribution of the species was located in the eastern part of Indonesian Waters (Ege, 1939; Aoyama, 1998; Watanabe, 2001). Also, the long-finned eel species of *A. megastoma* that usually reported to be found and distributed in the eastern part of Papua New Guinea to the southern part of Pacific Ocean (Ege, 1939; Watanabe, 2001) were found in the Western Papua. Finding on the changes in the geographic distribution of the tropical eels around Indonesian waters might be the cause of change in distribution pattern and migration route of the organism regulated by global climate changes. The other possibility that the sampling area conducted in the previous studies still not yet covering the range of distribution of *A. megastoma*. A new range of distribution of *A. megastoma* carried out from the present study would expand the range of distribution of the species in the world.

Especially for *A. bicolor*, both genetic sequence and PCR-RFLP analysis had been applied in previous study by Aoyama (2000) and Sugeha (2003). The authors found two different haplotypes derivate from *A. bicolor* that could be used to recognize and distinguish genetic characters of sub species inform different restriction fragment pattern between *A. bicolor bicolor* and *A. bicolor pacifica*. In the present study, the similar genetic characters also found. This finding supported the previous studies and had proven that the two sub species of the tropical short-finned eels of *A. bicolor* could be separated using PCR-RFLP analysis and they were inhabiting the Indonesian Waters. Some new genetic characters have also been found but not have been reported in the present study namely *Anguilla* spp. All the genetic characters showed different haplotypes that never reported in previous studies. The result suggested a possibility of new finding on the new species in the genus. The possibility of new finding on the intra-specific variation of tropical anguillid eels also promised a study on their population structure around Indonesian Waters. In general, study on the molecular genetic of tropical anguillid eels in the Indonesian Waters using an advance method of DNA sequencing analyses will be an important research works in the future.

Inshore migration mechanism of tropical anguillid eels

Even the study was not covering a complete one year study in fluctuation in abundance but from the pattern of abundance it could be seen a tendency that all regions has to perform a complete year round inshore migration pattern. These results suggested that inshore migration mechanism of tropical anguillid eels recruiting in tropical estuaries of Indonesian Waters differ in each region. Differences in the inshore migration mechanism in the Indonesian Waters were understandable since this region was well known as a remote area with a complex oceanographic condition that might be affecting and playing an important role on the successful recruitment of juveniles anguillid eels to reach their growth area. Oceanographic condition may affecting migration activity of tropical anguillid eels during oceanic migration period from their spawning ground in the open ocean, inshore migration period through the estuary in the coastal area before finally migrate upstream to growing up for several years in the freshwater area.

Marine scientists (Schmidt, 1922; McCleave, 1993; Tsukamoto, 1999; Miller *et al.*, 2002) suggested that oceanographic factors such as water current, salinity, and temperature in the ocean had been playing an important rule on successful migration of planctonic organisms such as leptocephalus larva of anguillid eels as a kind of ichthyoplancton. Indonesian throughflow and Eddy Current were typical currents in the Indonesian Waters and are proposed to be important environmental factors that affect oceanic migration activity of tropical anguillid eels during their early life history stage. Indonesian archipelago is located between the two great oceans (Indian and Pacific Ocean). Because of the strategic position, western region of Indonesian Waters mostly influenced by oceanographic character of Indian Ocean while eastern region mostly influenced by oceanographic character of Pacific Ocean. Beside that specific

oceanic tides propagation into Indonesian archipelago from the Pacific and Indian Ocean has been playing important roles in the transfer and mixing properties in Indonesian Seas (Tomascik *et al.*, 1997). The typical oceanographic condition might be resulting on the typical migratory behavior of the tropical anguillid in the Indonesian Waters. If this idea is true than it could be understand why biodiversity, distribution, and abundance of tropical angullid eels in the Indonesian Waters varied between western, center, and eastern region. Central region were famous as "Wallacea Line" region and reported by many marine biologists as the most rich region that were inhabited by specific character of marine living organisms. It would be one reason why higher diversity of tropical anguillid eels were also found in the central region of Indonesian Waters.

One important thing carried out from the present study is that the species diversity and geographic distribution of tropical angullid eels in the Indonesian Waters changes compared with previous study reported by Ege (1939) and Jespersen (1940). Further study is needed to carry out in the future in order to clarify weather the reformation of biodiversity and distribution in the tropic was triggered by global climate changes or other factors that proposed in the present study.

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