

Anguilla huangi Teng, Lin, and Tzeng, 2009, is a junior synonym of *Anguilla luzonensis* Watanabe, Aoyama, and Tsukamoto, 2009

Shun Watanabe · Jun Aoyama · Seishi Hagihara ·
Bunpei Ai · Rhodora V. Azanza · Katsumi Tsukamoto

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Abstract *Anguilla luzonensis* and *A. huangi* were each described in 2009 using eels obtained from northern Luzon Island. We examined the taxonomic status of these two groups of eels using morphological and molecular genetic characters. There were no significant differences in two vertebrae counts between eels of *A. luzonensis* and *A. huangi*. Mitochondrial 16S ribosomal RNA and cytochrome *b* genes sequences were obtained and compared among 28 specimens of *A. luzonensis*, the holotypes of *A. luzonensis* and *A. huangi*, and one specimen of the other 15 anguillid species. The specimens of *A. luzonensis* exhibited almost identical sequences, including the holotype, with only a few site differences, and the genetic difference between the holotypes of *A. luzonensis* and *A. huangi* was within the range of differences of specimens of *A. luzonensis*. The other anguillid species were genetically very different from *A. luzonensis* and *A. huangi*, although *A. interioris* is a closely related species. It is clear that *A. luzonensis* and *A. huangi* are the same species, and according to the principle of priority in zoological nomenclature, *A. luzonensis*

Watanabe, Aoyama, and Tsukamoto, 2009 is the valid species name, and *A. huangi* Teng, Lin, and Tzeng, 2009 is a junior synonym of *A. luzonensis*.

Keywords Synonym · Philippines · Cagayan River · Freshwater eel · Valid species name · Morphological characters · Molecular genetic characters

Introduction

The freshwater eels of the genus *Anguilla* Schrank, 1798, are widely distributed throughout the world and have catadromous life histories. The genus *Anguilla* has long been recognized as comprising 15 species [1–3], three of which are further divided into two subspecies [1]. However, two new species, *Anguilla luzonensis* Watanabe, Aoyama, and Tsukamoto, 2009 [4] and *Anguilla huangi* Teng, Lin, and Tzeng, 2009 [5] were described from the Cagayan River system on northern Luzon Island of the Philippines and from glass eels collected from the Cagayan River estuary that were reared in a culture pond in Taiwan, respectively.

Both of these newly discovered eel species had the morphological characteristics of mottled skin and broad maxillary bands of teeth. The genus *Anguilla* has been clearly divided into four different species groups based on the external morphological characters of each species. The first group has variegated skin with broad maxillary bands of teeth; the second group has variegated skin with narrow maxillary bands of teeth; the third group has nonvariegated skin with a long dorsal fin; and the fourth group has nonvariegated skin with a short dorsal fin [3, 6]. The first group consists of *A. celebesensis*, *A. interioris* and *A. megastoma*, each of which have different geographic distributions:

S. Watanabe (✉) · J. Aoyama · K. Tsukamoto
Atmosphere and Ocean Research Institute, The University of
Tokyo, 5-1-5 Kashiwanoha, Kashiwa, Chiba 277-8564, Japan
e-mail: shunwata@aori.u-tokyo.ac.jp

S. Hagihara
Division of Marine Biosciences, Graduate School of Fisheries
Science, Hokkaido University, 3-1-1 Minato-cho, Hakodate,
Hokkaido 041-8611, Japan

B. Ai
NPO Nihon Sakana no Kai, Yaizu, Shizuoka 425-0051, Japan

R. V. Azanza
Marine Science Institute, University of the Philippines, Diliman,
Quezon City 1101, Philippines

A. celebesensis being found from central Indonesia to north Luzon Island of the Philippines [1], *A. interioris* apparently being distributed longitudinally within a band of tropical latitude angled from Papua New Guinea south of the equator to Sumatra in the Indian Ocean [7–9], and *A. megastoma* being found further east in the South Pacific Ocean from New Caledonia to French Polynesia [1]. Both of the newly described anguillid eels, *A. luzonensis* [4] and *A. huangi* [5], belong to this first morphological group of freshwater eels. Teng et al. [5] showed that the morphological measurements of *A. huangi* overlap with those of *A. celebesensis* and *A. interioris*, and that these species cannot be distinguished using morphological characteristics alone. Aoyama et al. [10] had also shown that *A. celebesensis* and *A. interioris* were very similar morphologically and were different genetically. However, Watanabe et al. [4] showed statistically significant differences in 5 proportional characters and 2 meristic characters between *A. luzonensis* and *A. celebesensis* of the first group of *Anguilla*, which are possibly overlapped their geographic distribution in the Philippines.

Teng et al. [5] sequenced the complete mitochondrial DNA (mtDNA) of *A. huangi* and constructed a phylogeny of all currently recognized freshwater eel species and subspecies including *A. huangi*. The molecular phylogeny showed a strongly supported clustering of *A. huangi* and *A. interioris*, and the genetic distance between *A. huangi* and *A. interioris* was similar to that between *A. anguilla* and *A. rostrata* [5]. However, *A. celebesensis* was not a sister group with *A. huangi* and *A. interioris* in their phylogeny [5]. Furthermore, Minegishi et al. [11] also analyzed the whole mtDNA of *A. luzonensis* and suggested that the mtDNA divergence between *A. luzonensis* and *A. interioris* appeared to be roughly the same as between *A. anguilla* and *A. rostrata*.

These reports [4, 5, 11] provided the following information: (1) *A. luzonensis* were collected from the Cagayan River and the place of origin of *A. huangi* was the Cagayan River, (2) both species morphologically belong to the first group in the freshwater eels, and (3) the mtDNA of both species were different from other anguillid species, and the genetic distance between the eels with the two species names (*A. luzonensis* and *A. huangi*) and *A. interioris* were both similar to that between *A. anguilla* and *A. rostrata*. From these results, it is likely that *A. luzonensis* and *A. huangi* are synonymous names for the same species. Therefore, this confusion about having both *A. luzonensis* and *A. huangi* as names for this new anguillid species should be taxonomically solved.

The purpose of this paper is to clearly determine if *A. luzonensis* and *A. huangi* are the same species and what is the valid name of this newly discovered species. The two species are evaluated by comparing their morphological

characters with the potentially sympatric *A. celebesensis* in the first group of *Anguilla*, and their 16S ribosomal RNA (rRNA) and cytochrome *b* (*cyt b*) gene sequences of mtDNA were determined to compare the number of different sites and genetic distance between *A. luzonensis* and *A. huangi* holotypes with those within *A. luzonensis* and those between the two species and all other known anguillid species.

Materials and methods

Sample collection for *A. luzonensis*

A total of 29 specimens of *A. luzonensis* (NSMT-P 90001–90028), including the holotype (NSMT-P 90000) [4], were used in this study. These specimens (total length 244–682 mm) were collected from 29 January 2008 to 22 January 2009 in the upper reaches of the Pinacananan River (Peñablanca Province), which is a tributary of the Cagayan River of northern Luzon in the Philippines. All specimens were preserved in a 15 % formalin solution after a small piece of tissue from the right pectoral fin or liver was preserved in 99 % ethanol.

Morphological comparisons

In the first group of freshwater eels, the holotypes of *A. celebesensis* (MNHN 2150), *A. interioris* (AMS IA6075) and *A. megastoma* (MNHN A9952) were morphologically compared with the holotypes of *A. luzonensis* (NSMT-P 90000) and *A. huangi* (ASIZP0069360) (Table 1). Proportional measurements and vertebrae counts followed the methods of Ege [1] and Watanabe et al. [4, 6] and are shown in Table 1. Distances from the tip of the lower jaw to the corner of the mouth itself (length of gape), and from a perpendicular through the eye-center on the margin of the upper jaw to the angle of the gape were measured with a caliper to the nearest 0.1 mm. Other measurements shown in Table 1 were measured along the body axis, and were made with a 1-m ruler to the nearest 1 mm. The total length, head length and length of gape are abbreviated as TL, HL and LG, respectively. The total number of vertebrae (TV) as well as number of abdominal (AV) and caudal vertebrae (CV) for specimens were counted using radiographs (soft-X, Softex Co., Ltd.). We used the morphological data for the holotype (ASIZP0069360) of *A. huangi* described in Teng et al. [5].

Five of eight proportional characters and two of three vertebrae counts were already found to have statistically significant differences between *A. luzonensis* and *A. celebesensis* [4], which were predorsal fin (PD), preanal (PL)

Table 1 Comparative measurements and counts of the holotypes of *Anguilla luzonensis*, *A. huangi*, *A. celebesensis*, *A. interioris* and *A. megastoma*

Characters	<i>A. luzonensis</i> Holotype NSMT-90000 Luzon Island, Philippines	<i>A. huangi</i> Holotype ASIZP0069360 Luzon Island, Philippines	<i>A. celebesensis</i> Holotype MNHN 2150 Celebes	<i>A. interioris</i> Holotype AMS IA6075 Central Mandated Territory of New Guinea	<i>A. megastoma</i> Holotype MNHN A9952 Mangareva, Gambier Islands
Total length (mm)	528	1000	665	960	888
Measurements in % of TL					
Head length	13.6	13.0	12.2	12.0	13.2
Predorsal-fin length	33.0	30.2	30.2	28.1	28.6
Preanal length	43.4	42.5	40.3	42.7	40.5
Predorsal lengths without head length	19.3	17.2	18.0	16.1	15.4
Trunk length	29.7	29.5	28.1	30.7	27.4
Distance between the verticals through the anus and origin of the dorsal fin	10.4	12.3	10.1	14.6	11.9
Measurements in % of HL					
Length of gape	39.5	26.9	33.8	46.2	40.8
Measurements in % of LG					
Distance from perpendicular through eye-center on the margin of upper jaw to angle of gape	34.4	35.7	33.5	38.0	35.0
Counts					
Total number of vertebrae	104	106	103	— ^a	112
Number of abdominal vertebrae	41	40	39	— ^a	42
Number of caudal vertebrae	63	66	64	— ^a	70

^a The vertebrae have been removed and could not be counted in the holotype of *A. interioris*

and trunk (TR) lengths, and distance between the verticals through the anus and origin of the dorsal fin (AD) in % of TL, length of gape (LJ) in % of HL, AV and TV. The geographic distributions of *A. luzonensis*, *A. huangi* and *A. celebesensis* may possibly overlap in the Philippines but the geographic distributions for *A. interioris* and *A. megastoma* were far from the Philippines, and not enough specimens of *A. interioris* are available for statistical analysis. So, we used the specimens of *A. luzonensis*, *A. huangi* and *A. celebesensis* to statically compare morphological characters. The five proportional characters (PD/TL, PL/TL, TR/TL, AD/TL, LJ/HL) and two vertebrae counts (AV, TV) that were obtained from *A. luzonensis*, *A. huangi* and *A. celebesensis* were statistically compared with the Kruskal–Wallis test followed by pairwise comparisons with Dunn's test using Prism (version 4.0c) for Macintosh (GraphPad Software Inc.). A probability of $P < 0.05$ was used as the criteria for statistical significance. We used these morphological data for 10 specimens (ASIZP0069360–0069369) of *A. huangi* [5] and for 17 specimens of *A. celebesensis* [4] including the holotypes of both species, to compare to the 29 specimens of *A. luzonensis* including the holotypes.

Molecular genetic comparisons

The whole mtDNA sequences of one specimen of both *A. luzonensis* and *A. huangi* were already determined and published in DDBJ/EMBL/GenBank under the accession numbers AB469437 (the Paratypes of *A. luzonensis*, NSMT-P 90001) [11] and EU917054 (the holotype of *A. huangi*, ASIZP0069360) [5]. Sequences of a portion of the mitochondrial 16S rRNA and *cyt b* genes from the remaining 28 specimens of *A. luzonensis* were determined following the methods reported by Aoyama et al. [12] and Lin et al. [13]. Briefly, total genomic DNA was extracted from the ethanol preserved tissue samples according to a standard protocol, and a portion of the mitochondrial 16S rRNA (about 600 base pairs) and *cyt b* (about 1000 base pairs) genes was amplified by the polymerase chain reaction (PCR) using 4 oligonucleotide primers, L2510 and H3058 [12] and L15239 and H16468 [13]. Amplification parameters were 35 cycles of denaturation at 94 °C for 30 s, annealing at 58 °C for 30 s and extension at 72 °C for 60 s for 16S rRNA gene and 30–35 cycles of denaturation at 98 °C for 15 s, annealing at 52 °C for 15 s and extension at 72 °C for 60 s for *cyt b* gene. The PCR products were

sequenced according to the manufacturer's protocol on a Model 3130 genetic analyzer (Applied Biosystems).

For comparisons of genetic diversity of the 16S rRNA and cyt *b* mtDNA genes between *A. luzonensis* or *A. huangi* and anguillid species, we used whole mtDNA nucleotide sequences for the 15 other species of the genus *Anguilla*, which were determined by Inoue et al. [14] and Minegishi et al. [11] (DDBJ/EMBL/GenBank under the accession numbers AP007239: *A. celebesensis*, AP007241: *A. interioris*, AP007243: *A. megastoma*, AP007246: *A. bengalensis*, AP007242: *A. marmorata*, AP007248: *A. reinhardtii*, AP007238: *A. borneensis*, AB038556: *A. japonica*, AP007249: *A. rostrata*, AP007233: *A. anguilla*, AP007240: *A. dieffenbachii*, AP007244: *A. mosambica*, AP00723: *A. australis*, AP007247: *A. obscura*, AP007236: *A. bicolor*).

The individual sequences in conjunction with the homologous data for the specimens of *A. luzonensis* determined by this study and from a previous study [11], and for the specimens of *A. huangi* [5] and all species of the genus *Anguilla* [11, 13] published in the data bank, were aligned manually using DNASIS version 3.7 (Hitachi Software Engineering) and MacClade version 4 after automatic alignment with Clustal X [15] as needed. The number of variable sites and genetic distances [16] were calculated with PAUP* 4.0b10.

All of the sequences determined in the present study will appear in the DDBJ/EMBL/GenBank with the accession numbers: AB490254–AB490258, AB490260–AB490265, AB490267–AB490269 and AB490272–AB490285 ($n = 28$) for 16S rRNA: AB758625–AB758652 ($n = 28$) for cyt *b*.

Results

Morphological comparisons

The comparison of proportional and vertebral characters among the holotypes of *A. luzonensis*, *A. huangi*, *A. celebesensis*, *A. interioris* and *A. megastoma* showed that the TV and CV for *A. megastoma* were clearly higher than those of the 4 other species (Table 1). Furthermore, the type localities for *A. megastoma* and *A. interioris* were far from those of *A. luzonensis*, *A. huangi* and *A. celebesensis* (Table 1) based on information about the geographic distribution of the 5 species [1, 4, 5, 7–9]. Therefore, we compared proportional measurements and vertebrae counts among *A. luzonensis*, *A. huangi* and *A. celebesensis* (Table 2). The statistical analysis of 5 proportional characters found significant differences among the three species (Kruskal–Wallis test, $P < 0.01$, Table 3) and the 15 post hoc multiple pairwise comparisons showed that there were 9 significant differences (Dunn's test, $P < 0.05$ Table 3). However, the changes of the 5 proportional characters with increasing TL were found to have a similar tendency (positive slopes) among species except for the LJ/HL of *A. huangi* (Fig. 1).

The statistical analysis of AV and TV found significant differences among the three species (Kruskal–Wallis test, $P < 0.01$, Table 3) and the post hoc multiple pairwise comparisons showed that there were 4 significant differences (Dunn's test, $P < 0.05$, Table 3) between *A. luzonensis* or *A. huangi* and *A. celebesensis* (Fig. 2). There were no significant differences between *A. luzonensis* and *A. huangi* in AV and TV though (Table 3).

Table 2 Comparative measurements and counts of *A. luzonensis*, *A. huangi* and *A. celebesensis*

Characters	<i>A. luzonensis</i>			<i>A. huangi</i>			<i>A. celebesensis</i>		
	(244–682 mm TL, $n = 29$)			(244–1000 mm TL, $n = 10$)			(239–665 mm TL, $n = 17$)		
	Range	Mean	SD	Range	Mean	SD	Range	Mean	SD
Measurements in % of TL									
Predorsal-fin length	28.6–33.1	30.5	1.3	27.2–30.7	28.3	1.2	28.2–32.9	29.8	1.5
Preanal length	39.6–44.8	42.0	1.2	36.9–42.5	39.4	1.7	37.8–43.1	40.0	1.4
Trunk length	26.4–31.3	28.6	1.1	25.3–29.5	27.1	1.3	26.0–28.6	27.1	0.8
Distance between the verticals through the anus and origin of the dorsal fin	9.3–13.9	11.5	1.1	9.4–12.3	11.1	1.1	9.2–12.2	10.2	0.9
Measurements in % of HL									
Length of gape	35.5–46.9	40.1	2.5	24.4–39.2	29.8	4.6	28.6–42.6	36.6	3.6
Counts									
Total number of vertebrae	103–107	104.8	1.0	103–106	104.9	1.1	101–105	103.2	1.1
Number of abdominal vertebrae	40–42	41.1	0.6	40–41	40.6	0.5	38–41	39.2	1.0

Table 3 Results of Kruskal–Wallis test and Dunn’s multiple comparison test of 5 proportional and 2 meristic characters among *A. luzonensis*, *A. huangi* and *A. celebesensis*

Characters	<i>P</i> values of Kruskal–Wallis test and Dunn’s multiple comparison test		
	Species	<i>A. huangi</i>	<i>A. celebesensis</i>
Measurements in % of TL			
Predorsal-fin length		<i>P</i> = 0.0002	
	<i>A. luzonensis</i>	<i>P</i> < 0.001	ns
	<i>A. huangi</i>		ns
Preanal length		<i>P</i> < 0.0001	
	<i>A. luzonensis</i>	<i>P</i> < 0.001	<i>P</i> < 0.001
	<i>A. huangi</i>		ns
Trunk length		<i>P</i> = 0.0001	
	<i>A. luzonensis</i>	<i>P</i> < 0.01	<i>P</i> < 0.001
	<i>A. huangi</i>		ns
Distance between the verticals through the anus and origin of the dorsal fin		<i>P</i> = 0.0027	
	<i>A. luzonensis</i>	ns	<i>P</i> < 0.01
	<i>A. huangi</i>		ns
Measurements in % of HL			
Length of gape		<i>P</i> < 0.0001	
	<i>A. luzonensis</i>	<i>P</i> < 0.001	<i>P</i> < 0.05
	<i>A. huangi</i>		<i>P</i> < 0.05
Counts			
Number of abdominal vertebrae		<i>P</i> < 0.0001	
	<i>A. luzonensis</i>	ns	<i>P</i> < 0.001
	<i>A. huangi</i>		<i>P</i> < 0.05
Total number of vertebrae		<i>P</i> = 0.0002	
	<i>A. luzonensis</i>	ns	<i>P</i> < 0.001
	<i>A. huangi</i>		<i>P</i> < 0.01

Molecular genetic comparisons

A total of 580 base pairs (bp) of the mitochondrial 16S rRNA gene sequences were obtained after the alignments for 29 specimens of *A. luzonensis* including the holotype, and a total of 624 bp of the mitochondrial 16S rRNA gene sequences were obtained for the holotypes of *A. luzonensis* and *A. huangi* and one specimen from each of all other 15 species. The specimens of *A. luzonensis* exhibited almost identical sequences including the holotype with only 0–2 site differences (0–0.003 in genetic distance) and the genetic difference between the holotypes of *A. luzonensis* and *A. huangi* (2 site differences and 0.003 in genetic distance) was included in the range of these in the specimens of *A. luzonensis*, whereas the other anguillid species are considered to be genetically far distant from *A. luzonensis* or *A. huangi* with 7–31 site differences (0.011–0.052, Table 4).

A total of 1049 bp of the mitochondrial *cyt b* gene sequences were obtained after the alignments of 29 specimens of *A. luzonensis* including the holotype, the holotype

of *A. huangi* and one specimen from each of all other 15 species. The specimens of *A. luzonensis* exhibited almost identical sequences, including the holotype, with only 0–17 site differences (0–0.016 in genetic distance). The genetic difference between the holotypes of *A. luzonensis* and *A. huangi* (10 site differences and 0.010 in genetic distance) was included in the range of differences of the specimens of *A. luzonensis*, whereas the other anguillid species were genetically far distant from *A. luzonensis* or *A. huangi* with 39–103 site differences (0.038–0.107, Table 5).

Discussion

The comparisons using morphological and molecular genetic characters clearly show that *A. luzonensis* and *A. huangi* are the same species. The description paper of *A. luzonensis* [4] was published by Fisheries Science (Vol. 75, No. 2) in April 2009, while *A. huangi* was described [5] in Zoological Studies (Vol. 48, No. 6) in

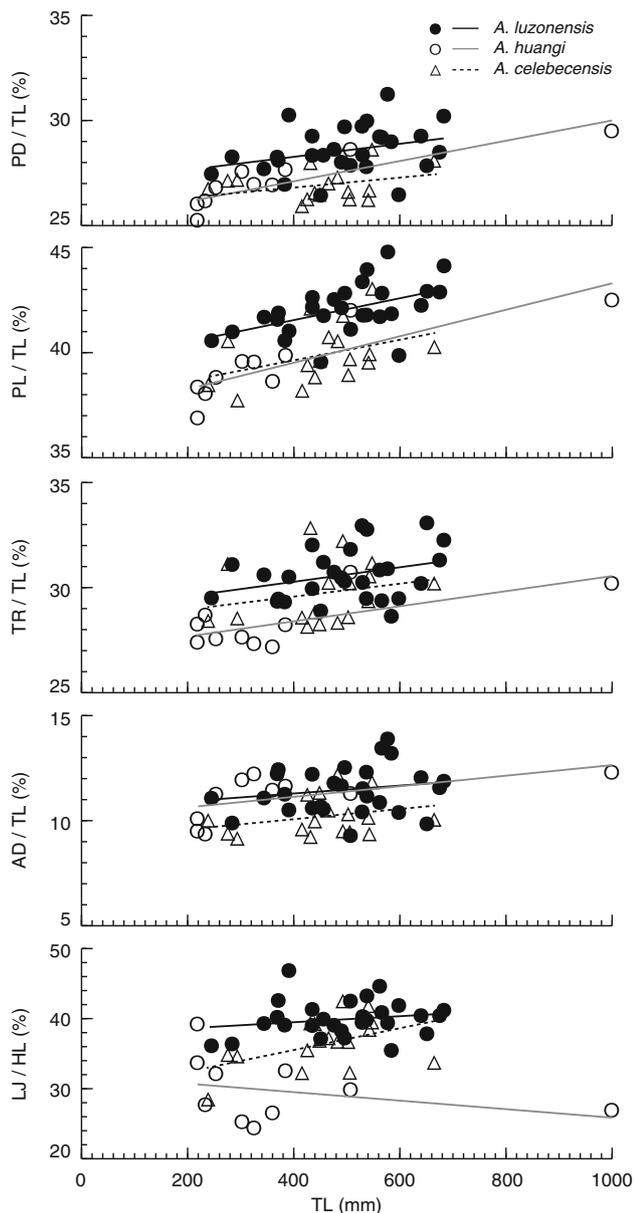


Fig. 1 Changes in the ratio of 5 proportional characters relative to total length (TL) in *A. luzonensis*, *A. huangi* and *A. celebesensis*

November 2009. When the principle of priority in zoological nomenclature [17] is applied using the publication dates of the papers describing *A. luzonensis* and *A. huangi*, *A. luzonensis* Watanabe, Aoyama, and Tsukamoto, 2009 [4] is the valid species name and *Anguilla huangi* Teng, Lin, and Tzeng, 2009 [5] is a junior synonym of *A. luzonensis*.

Although this study demonstrates that *A. luzonensis* and *A. huangi* were not different genetically or in their vertebral counts are the same species, there were 4 significant differences between *A. luzonensis* and *A. huangi* in the 5 proportional characters (Table 3). Furthermore, the change of AD/TL with TL in *A. huangi* may have been different

from those patterns in both *A. luzonensis* and *A. celebesensis* even if the much larger specimen was excluded (Fig. 1). Teng et al. [5] used cultured eels that were imported as glass eels from the Cagayan River estuary, northern Luzon Island, the Philippines and were reared in culture ponds/tanks. Because these characters are just relative length of gape to the head length, which could be affected by differences in growth rate, it is possible that the morphological differences of *A. huangi* were caused by the aquaculture process in which fish are usually fed more food and they grow faster than usual. Morphological differences between hatchery reared juvenile turbot *Scophthalmus maximus* and native individuals have also been seen [18]. The results for 2 vertebrae counts, AV and TV, which would not be affected by the aquaculture process, showed little or no differences as was the case for the molecular genetic analysis between *A. luzonensis* and *A. huangi*.

The genetic data obtained from this study, however, clearly confirmed the taxonomic identity of *A. luzonensis* and *A. huangi* by showing that they are the same species. In addition to being used in phylogenetic and population genetic analyses, mtDNA sequences have been used for evaluations of the taxonomy of animals and identification of species, such as in the expansion of use of DNA barcoding. In the case of the anguillid species that are difficult to morphologically identify to the species level [6, 10], particularly at their early life stages [7, 8, 19–22], identification using mtDNA sequences are now routinely used. Inter- and intra-species variations in the mtDNA sequence have also been investigated with a special emphasis on establishing molecular species identification methods [10, 23, 24]. The number of site differences in the whole mitochondrial 16S rRNA (1704–1712 bp) and *cyt b* (1140 bp) gene sites were reported to range from 3–10 to 0–24 between subspecies, and from 19–82 to 41–118 between species of anguillid eels [11]. The present study determined and compared the number of sequence differences in the 16S rRNA (580 or 624 bp) and *cyt b* (1049 bp) gene sequences, and the 29 specimens of *A. luzonensis* including the holotype and *A. huangi* holotype showed only 0–2 and 0–17 site differences, respectively, whereas they clearly exhibited much greater genetic divergence of 7–31 and 39–103 site differences between *A. luzonensis* or *A. huangi* holotypes and the 15 other anguillid species. These show that *A. luzonensis* is genetically distinct from all the other known anguillid species, and it is most closely related to *A. interioris* as previously reported [5, 11].

A few oceanic leptocephalus larvae of the Anguillidae, which have identical mitochondrial DNA sequences with *A. luzonensis* have been collected in the North Equatorial Current (NEC) region of the western North Pacific [25, 26]. This indicates that *A. luzonensis* likely has a spawning area somewhere in the NEC of the western North Pacific and the

Fig. 2 Frequency distribution of number of abdominal vertebrae (AV) and total number of vertebrae (TV) in *A. luzonensis*, *A. huangi* and *A. celebesensis*

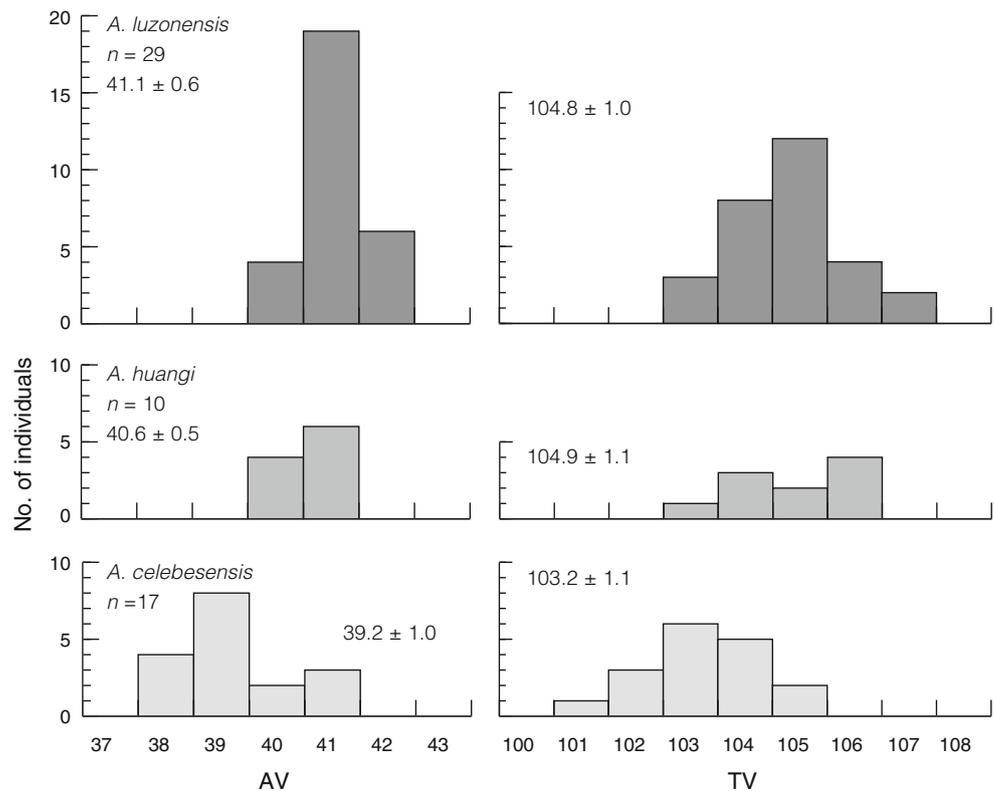


Table 4 Genetic differences in the 16S ribosomal RNA gene among 29 specimens of *A. luzonensis*, between each holotype of *A. luzonensis* and *A. huangi*, and between *A. luzonensis* or *A. huangi* and the other 15 anguillid species

Species	No. of different sites		Genetic distance	
Among 29 specimens of <i>A. luzonensis</i>	0–2		0–0.003	
<i>A. luzonensis</i> versus <i>A. huangi</i>	2		0.003	
Species	No. of different sites		Genetic distance	
	<i>A. luzonensis</i>	<i>A. huangi</i>	<i>A. luzonensis</i>	<i>A. huangi</i>
<i>A. celebesensis</i>	21	19	0.035	0.032
<i>A. interioris</i>	9	7	0.015	0.011
<i>A. megastoma</i>	21	19	0.035	0.032
<i>A. bengalensis</i>	12	10	0.020	0.016
<i>A. marmorata</i>	11	9	0.018	0.015
<i>A. reinhardtii</i>	18	16	0.030	0.027
<i>A. borneensis</i>	<u>31</u>	<u>29</u>	<u>0.052</u>	<u>0.049</u>
<i>A. japonica</i>	26	24	0.044	0.040
<i>A. rostrata</i>	22	20	0.037	0.033
<i>A. anguilla</i>	20	18	0.033	0.030
<i>A. dieffenbachii</i>	22	20	0.037	0.033
<i>A. mossambica</i>	22	20	0.037	0.033
<i>A. bicolor</i>	12	10	0.040	0.037
<i>A. obscura</i>	9	7	0.015	0.011
<i>A. australis</i>	24	22	0.020	0.016

Bolds and underlines shows minimum and maximum number of different sites or genetic distances, respectively

larvae would be transported westward to recruit to the areas around northern Luzon Island [25, 26]. Leptocephali of *A. celebesensis* or *A. interioris* have never been identified in this region though, because their spawning areas appear to

be located in Indonesian waters far south of the NEC region or in the eastern Indian Ocean [8, 19]. Therefore, *A. luzonensis* would have a spawning area and migratory route that is entirely different from those of *A. celebesensis* and

Table 5 Genetic differences in the cytochrome *b* gene among 29 specimens of *A. luzonensis*, between each holotype of *A. luzonensis* and *A. huangi*, and between *A. luzonensis* or *A. huangi* and the other 15 anguillid species

Species	No. of different sites		Genetic distance	
Among 29 specimens of <i>A. luzonensis</i>	0–17		0–0.016	
<i>A. luzonensis</i> versus <i>A. huangi</i>	10		0.010	
Species	No. of different sites		Genetic distance	
	<i>A. luzonensis</i>	<i>A. huangi</i>	<i>A. luzonensis</i>	<i>A. huangi</i>
<i>A. celebesensis</i>	78	80	0.080	0.082
<i>A. interioris</i>	39	41	0.038	0.041
<i>A. megastoma</i>	93	95	0.096	0.099
<i>A. bengalensis</i>	56	56	0.056	0.056
<i>A. marmorata</i>	56	56	0.056	0.056
<i>A. reinhardtii</i>	72	72	0.073	0.073
<i>A. borneensis</i>	88	88	0.091	0.091
<i>A. japonica</i>	81	83	0.083	0.085
<i>A. rostrata</i>	95	95	0.099	0.099
<i>A. anguilla</i>	91	95	0.094	0.099
<i>A. dieffenbachii</i>	75	77	0.076	0.078
<i>A. mossambica</i>	<u>99</u>	<u>103</u>	<u>0.103</u>	<u>0.107</u>
<i>A. bicolor</i>	97	96	0.101	0.100
<i>A. obscura</i>	59	65	0.059	0.066
<i>A. australis</i>	75	75	0.076	0.076

Bolds and underlines shows minimum and maximum number of different sites or genetic distances, respectively

A. interioris. The morphological and genetic results and this ecological evidence strongly support of the presence of *A. luzonensis* in the area of northern Luzon Island of the Philippines, but the extent of the range of this species needs to be studied further.

To clarify their geographic distribution and life history as well as their spawning area and migratory behavior, a simple and robust species identification method is urgently required. The present study however, follows the nomenclatural rule of priority and shows that the valid scientific name of this species is *A. luzonensis*, which is appropriate considering this name is linked to the place where this eel lives.

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