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GENETIC DIFFERENTIATION OF *OSTEOCHILUS HASSELTII* (TELEOSTEI: CYPRINIDAE) IN THAILAND

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ABSTRACT. - *Osteochilus hasseltii* in Thailand is divided into two population groups, viz. the Mekong-Chao Phraya group and Mae Khlong-Malay Peninsula group, by genetical differentiations (D=0.3706-0.5709). In the morphological characters, these two groups are distinguished solely by the number of total vertebrae and total lateral line scales. However, because of the wide overlaps in ranges of these characters, the differences are not recognized at the individual level.

KEY WORDS. - *Osteochilus hasseltii*, genetic differentiation, Thailand, Cyprinidae.

INTRODUCTION

Fishes of the genus *Osteochilus* are small to medium size cyprinids consisting of 25 species distributed in Southeast Asia (Karnasuta, 1993; Kottelat, 1995). Although most species of the genus show restricted distribution, *O. hasseltii* is found northwest from the middle Irrawaddy, east to the Indochinese Peninsula and south to Java, nearly overlapping with the distribution of the entire genus. Southeast Asia is divided into subareas based on the distribution patterns of fish genera or species (e.g. Kottelat, 1989; Rainboth, 1991). These subareas coincide with geographic features and contain many endemic species. In recent years, by the use of morphological characters, *Botia hymenophysa,* a widely distributed cobotid in Southeast Asia was divided into the two species, i.e., *B. hymenophysa* (Malaya, Sumatra, Borneo) and *B. helodes* (Mekong-Chao Phraya basin) (Kottelat, 1984). Based on genetic characters, Kornfield & Carpenter (1984) concluded that the cyprinid *Puntius binotatus* from Mindanao should be considered distinct from the Bornean forms. Furthermore, Dodson et al. (1995) suggested that *Hemibagrus nemurus* was almost certainly a mixture of species based on the mt DNA. These results indicate a necessity to reexamine the widespread species in Southeast Asia. In this study, the degree of genetic features of local populations of *O. hasseltii* in Thailand was examined using the allelic variations of enzymes, and some morphological characters also were examined.

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MATERIALS

Specimens used in this study were collected from seven localities in Thailand and from one locality in Malaysia. The number of specimens used for electrophoretic and morphological analyses was 155 and 247, respectively. Material data are given below. These specimens belong to the following institutions: Institute for Breeding Research, Tokyo University of Agriculture (IBRP), National Institute of Coastal Aquaculture, Thailand (NICA) and National Science Museum, Tokyo (NSMT). In the text, localities are indicated by the locality numbers given in Fig. 1, following the locality names.

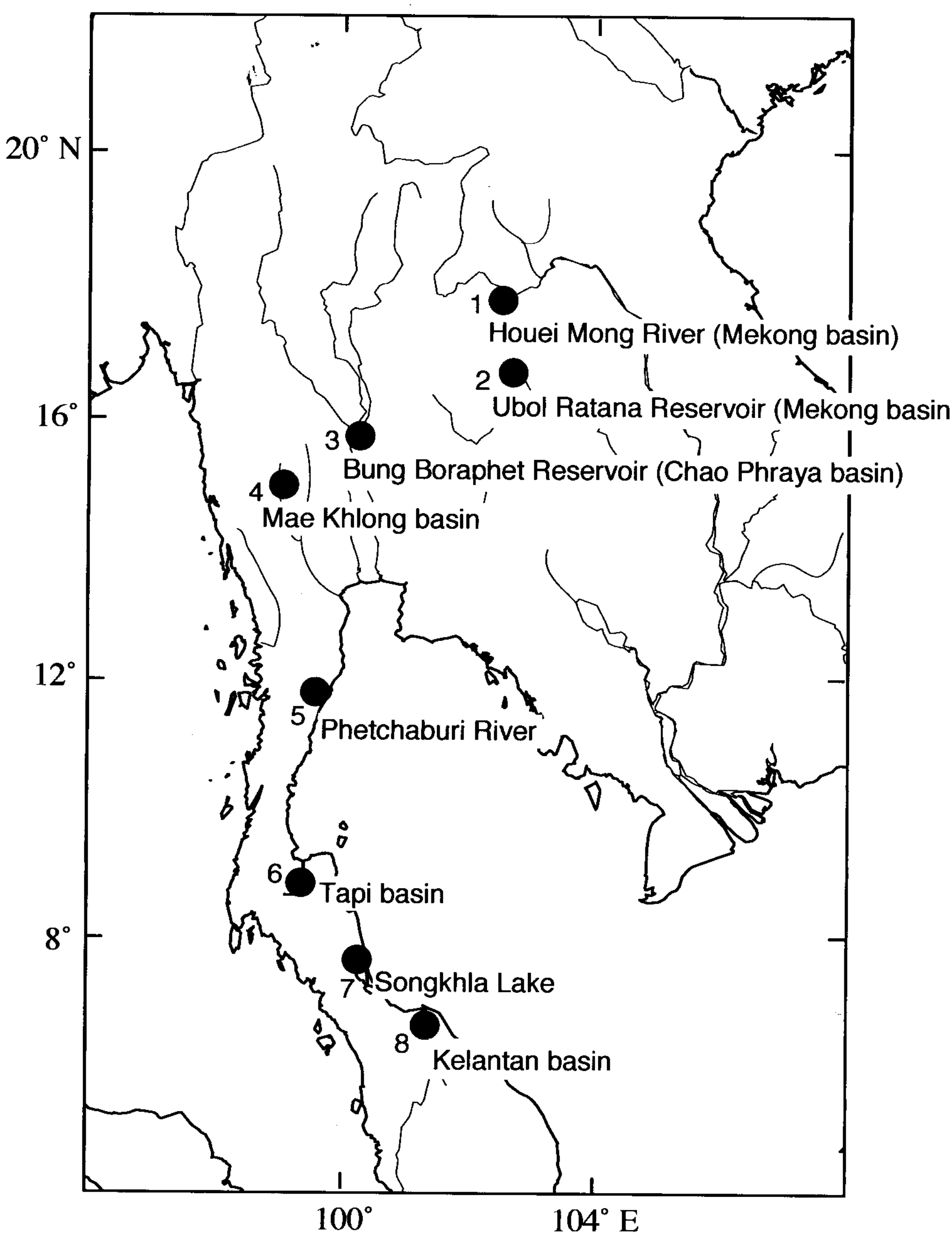

Fig. 1. Location of *Osteochilus hasseltii* sampling sites.

Materials for electrophoretic analyses. **- Houei Mong River (Mekong basin).** At Ban Dong Rai: NSMT-P 35830, 20 ex., 77.7-124.0mm SL, 13 Jul.1990. **Ubol Ratana Reservoir (Mekong basin).** At Khon Kaen: NSMT-P 25381, 17 ex., 96.0-107.0mm SL, 7 Nov.1989. **Bung Boraphet Reservoir (Chao Phraya basin).** Bung Boraphet Reservoir, Nakhon Sawan: NSMT-P 35832, 7 ex., 99.8-160.0mm SL, 29 Nov.1989; NSMT-P 35833, 13 ex., 96.0-167.6mm SL, 15 Aug.1990. **Mae Khlong basin.** Khao Laem Reservoir, at Jongoua, Kanchanaburi: NSMT-P 35913, 20 ex., 85.3-179.4mm SL, 17 Jul.1990. **Phetchaburi River.** Kang Krachan Reservoir, Phetchaburi: NSMT-P 35834, 20 ex., 64.8-140.5mm SL, 24 Aug.1990. **Tapi basin.** Near Ban Phun Phin, Surat Thani: NSMT-P 35835, 20 ex., 66.7-136.6mm SL, 31 Jul.1990. **Songkhla Lake.** Pond at Ranot, Songkhla: NSMT-P 35836, 20 ex., 106.7-138.6mm SL, 9 Aug.1990. **Kelantan basin.** Paloh Pintu Geag Garak, Kelantan: NSMT-P 35837, 18 ex., 88.5-155.0mm SL, 22 Nov.1989.

Materials for morphological examination. **- Houei Mong River (Mekong basin).** Houei Mong R. at its mouth on Mekong R., near Tha Bo: IBRP 5020, 3 ex., 63.0-70.0mm SL, 9 Dec.1970; IBRP 5682, 2 ex., 77.0-98.5mm SL, 6 May.1971. Houei Mong R. at Ban Dong Rai: NSMT-P 35820, 16 ex., 63.5-110.0mm SL, 13 Jul.1990. **Ubol Rat Reservoir (Mekong basin).** Ubol Rat Reservoir at Khon Kaen: NSMT-P 35802, 5 ex., 127.9-161.9mm SL, 5 Nov.1989; NSMT-P 35803, 20 ex., 83.5-119.0mm SL, 7 Nov.1989. **Bung Boraphet Reservoir (Chao Phraya basin).** Bung Boraphet Reservoir, Nakhon Sawan: NSMT-P 35805, 1 ex., 171.8mm SL, 29 Nov.1989; NSMT-P 35806, 6 ex., 161.4-195.7mm SL, 28 Nov.1989; NSMT-P 35807, 18 ex., 86.4-106.2mm SL, 27 Nov.1989; NSMT-P 35821, 16 ex., 104.1-137.8mm SL, 15 Aug.1990. **Mae Khlong basin.** Khao Laem Reservoir at Pausamton, Khanchanaburi: NSMT-P 35815, 40 ex., 72.2-257.1mm SL, 19 Jul.1990. **Phetchaburi River.** Kang Krachan Reservoir, Phetchaburi: NSMT-P 35819, 5 ex., 67.8-88.7mm SL, 24 Jul.1990; NSMT-P 35834, 14 ex., 66.7-136.6mm SL, 24 Jul.1990 (also used for isozyme analysis). **Tapi basin.** Rajjaprapa Dam, Surat Thani: NSMT-P 35812, 33 ex., 87.4-133.6mm SL, 1 Aug.1990. Tapi R. near Ban Phun Phin, Surat Thani: NSMT-P 35813, 3 ex., 85.1-92.8mm SL, 31 Jul.1990. Pumdong R. at Talingchan, Surat Thani: NSMT-P 35814, 1 ex., 122.3mm SL, 2 Aug.1990. **Songkhla Lake.** Thale Luang (northern section of Songkhla Lake): NICA 083, 1 ex., 129.5mm SL, date unknown; NICA 092, 1 ex., 127.3mm SL, date unknown; NICA 094, 1 ex., 118.1mm SL, date unknown; NICA 095, 1 ex., 119.7mm SL, date unknown; NICA 644, 2 ex., 95.9-103.5mm SL, date unknown; NSMT-P 35817, 8 ex., 70.0-137.6mm SL, date unknown. Pond at Ranot, Songkhla: NSMT-P 35816, 7 ex., 120.8-133.1mm SL, 9 Aug.1990; NSMT-P 35818, 5 ex., 108.5-130.6mm SL, 9 Aug.1990. **Kelantan basin.** Paloh Pintu Geag Garak, Kelantan: NSMT-P 35808, 12 ex., 107.5-173.5mm SL, 12 Dec.1989; NSMT-P 35810, 7 ex., 107.0-130.4mm SL, 13 Dec.1989. Pasir Puteh, Kotabaru, Kelantan: NSMT-P 35811, 2 ex., 114.3-140.7mm SL, 14 Dec.1989.

METHODS

Electrophoretic analyses

Liver and muscle tissues were extracted from live specimens. Individual tissue samples were mixed with an equal (for muscle) or double (for liver) quantity of distilled water, homogenized, and centrifuged at 16,000 rpm for 30 minutes. The supernatant was stored at -80°C until analyzed. Horizontal starch gel electrophoresis of supernatant fractions was performed following Kuroda et al. (1982). A tris-citrate (pH 8.0) buffer system (Selander et al., 1971) was employed to resolve products of 10 isozyme/protein loci, as shown in Table 1. Enzyme/protein staining procedures followed those described by Shaw & Prasad (1970), Selander et al. (1971) and Bell et al. (1982). Locus and allele terminologies followed Numachi et al. (1983). Genetic divergence between samples from different localities was expressed using Nei's (1978) index of genetic distance. A phenogram based on the matrix of genetic distances was produced according to the UPGMA method (Sneath & Sokal, 1972).

Morphological analyses

Methods for meristic counts and morphometric measurements followed Doi & Taki (1994). The Kruskal-Wallis test and Dunn's multiple comparison test were performed to analyze meristic counts. Principal component analysis was done based on the correlation matrices of log-transformed morphometric variables.

RESULTS

Electrophoresis

Isozyme variability. - Variations in 10 loci detected for six enzymes and one non-enzymatic protein (Table 1) were analyzed on 155 specimens collected from the Mekong (Nos. 1, 2), Bung Boraphet (No. 3) and Mae Khlong (No. 4) basins, and Malay Peninsula (Nos. 5-8). IDH-2 and LDH-1 were each fixed for one allele in all samples (Table 2). Other loci exhibited varying magnitudes of polymorphism, with marked geographic variations in many of them. The alleles at AAT, GPI-2 and SOD showed abrupt shifts in frequency between the Bung Boraphet (No. 3) and Mae Khlong (No. 4) basins. For each of GPI-1, IDH-1 and PGM, a single, identical allele predominated in samples from the Mekong (Nos. 1, 2) and Bung Boraphet (No. 3) basins, Songkhla Lake (No. 7) and Kelantan basin(No. 8), whereas samples from the Mae Khlong, Phetchaburi and Tapi basins (Nos. 4-6) showed divergent allele frequencies. For SP, Mekong and Chao Phraya samples (Nos. 1-3) exhibited complete or nearly complete fixation for allele b and Mae Khlong and Phetchaburi specimens (Nos. 4,5) for allele a, whereas both alleles were scored at nearly equal frequencies in Tapi, Songkhla and Kelantan specimens (Nos. 6-8). LDH-2 was monomorphic, being fixed for allele a, at all localities, except for Bung Boraphet (No. 3), where allele b was scored at a low frequency. Patterns of geographic variation in AAT, IDH-1, SOD and SP are illustrated in Fig. 2.

Table 1. List of enzymes and protein examined, shown by locus detected and tissues assayed.

Enzyme and protein	Locus	Tissue assayed
Aspartate aminotransferase (AAT: 2.6.1.1)	AAT	liver
Glucosephosphate isomerase (GPI: 5.3.1.9)	GPI-1	muscle
	GPI-2	muscle
Isocitrate dehydrogenase (IDH: 1.1.1.42)	IDH-1	liver, muscle
	IDH-2	liver
Lactate dehydrogenase (LDH: 1.1.1.27)	LDH-1	muscle
	LDH-2	muscle
Phosphoglucomutase (PGM: 2.7.5.1)	PGM	muscle
Superoxide dismutase (SOD: 1.15.1.1)	SOD	liver
Sarcoplasmic protein (SP)	SP	muscle

Genetic relationships. - Genetic distances between the eight samples examined were computed based on the allele frequencies at the 10 loci given in Table 2. Genetic distances between localities in the Mekong-Chao Phraya group (Nos. 1-3) and those in the Mae Khlong-Malay Peninsula group (Nos. 4-8) were small, being 0.0007-0.0099 and 0.0000-0.1125, respectively, whereas the distances between the two groups were much greater, ranging from 0.3706 to 0.5709 (Table 3).

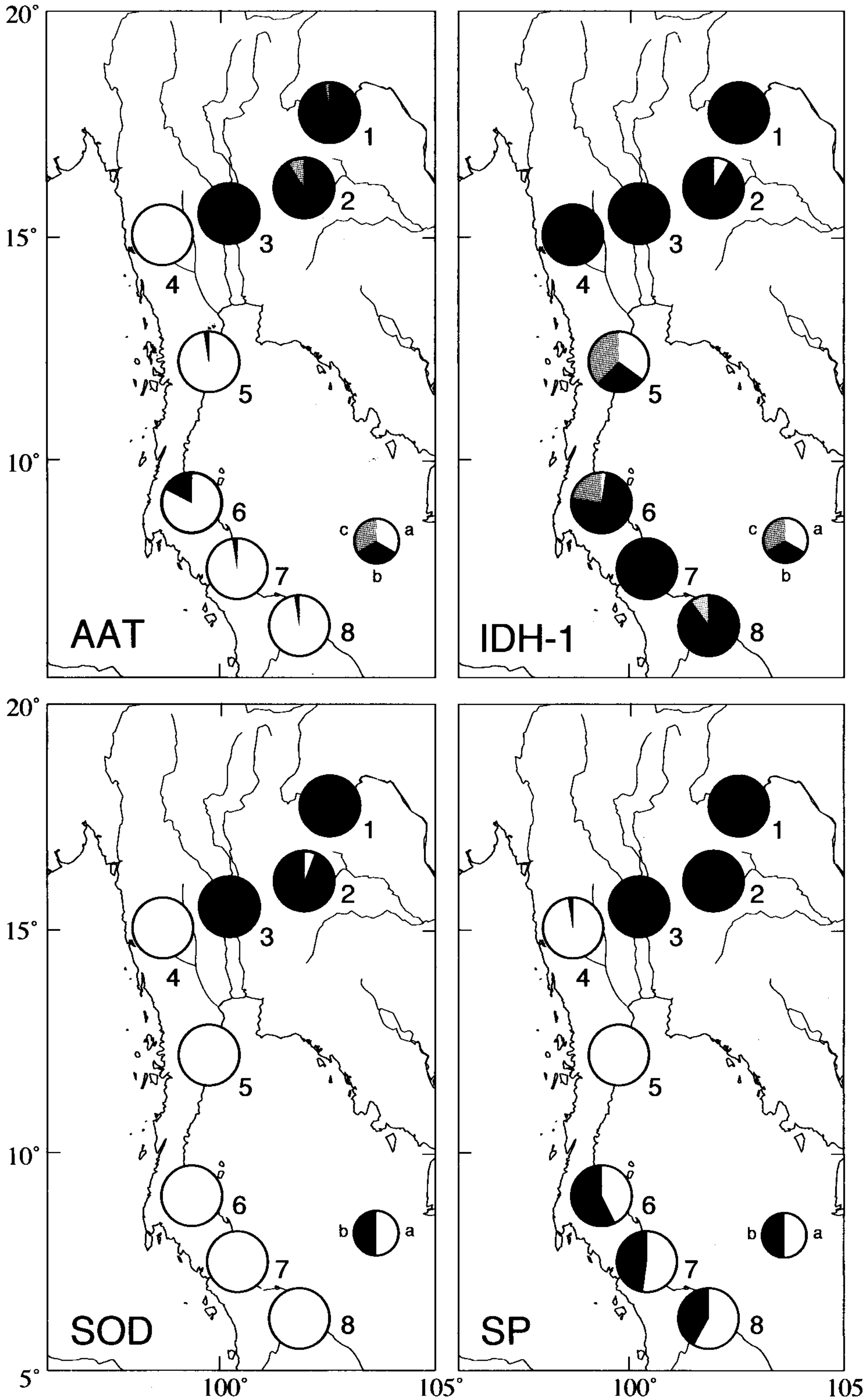

Fig. 2. Geographic distributions of alleles of *Osteochilus hasseltii*. For explanation of locality numbers and allelic frequencies, see Fig. 1 and Table 2, respectively.

Table 2. Allele frequencies and levels of isozyme variability in *Osteochilus hasseltii* collected from Thailand and Malaysia.

Locus	Allele	Houei Mong River (1)	Ubol Ratana Reservoir (2)	Bung Boraphet Reservoir (3)	Mae Khlong basin (4)	Phetcha-buri River (5)	Tapi basin (6)	Songkhla Lake (7)	Kelantan basin (8)
AAT	a	0.000	0.000	0.000	1.000	0.975	0.825	0.975	0.975
	b	0.975	0.912	1.000	0.000	0.025	0.175	0.025	0.025
	c	0.025	0.088	0.000	0.000	0.000	0.000	0.000	0.000
GPI-1	a	0.025	0.000	0.020	0.000	0.000	0.000	0.000	0.000
	b	0.000	0.000	0.000	0.425	0.025	0.125	0.000	0.000
	c	0.975	1.000	0.980	0.575	0.975	0.875	1.000	1.000
GPI-2	a	0.000	0.000	0.293	0.000	0.000	0.000	0.000	0.000
	b	1.000	1.000	0.707	0.000	0.000	0.025	0.025	0.000
	c	0.000	0.000	0.000	1.000	0.425	0.950	0.975	1.000
	d	0.000	0.000	0.000	0.000	0.575	0.025	0.000	0.000
IDH-1	a	0.000	0.088	0.000	0.000	0.350	0.025	0.000	0.000
	b	1.000	0.912	1.000	1.000	0.275	0.750	1.000	0.900
	c	0.000	0.000	0.000	0.000	0.375	0.225	0.000	0.100
IDH-2	a	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
LDH-1	a	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
LDH-2	a	1.000	1.000	0.960	1.000	1.000	1.000	1.000	1.000
	b	0.000	0.000	0.040	0.000	0.000	0.000	0.000	0.000
PGM	a	0.000	0.000	0.020	0.000	0.000	0.000	0.000	0.000
	b	1.000	1.000	0.980	1.000	1.000	0.650	0.875	0.944
	c	0.000	0.000	0.000	0.000	0.000	0.300	1.000	0.000
	d	0.000	0.000	0.000	0.000	0.000	0.050	0.025	0.056
SOD	a	0.000	0.059	0.000	1.000	1.000	1.000	1.000	1.000
	b	1.000	0.941	1.000	0.000	0.000	0.000	0.000	0.000
SP	a	0.000	0.000	0.000	0.975	1.000	0.425	0.525	0.583
	b	1.000	1.000	1.000	0.025	0.000	0.575	0.475	0.417

Parenthesized number indicates locality number given in Fig. 1.

Table 3. Matrix of genetic distance for *Osteochilus hasseltii* collected in eight localities, based on allel frequencies of 10 loci and Nei's (1978) formula.

Locus	Houei Mong River (1)	Ubol Rat Reservoir (2)	Bung Boraphet Reservoir (3)	Mae Khlong basin (4)	Phetcha-buri River (5)	Tapi basin (6)	Songkhla Lake (7)	Kelantan basin (8)
Ubol Rat Reservoir	0.0007							
Bung Boraphet Reservoir	0.0080	0.0099						
Mae Klong basin	0.5490	0.5339	0.5342					
Phetchaburi River	0.5709	0.5364	0.5562	0.0991				
Tapi basin	0.3894	0.3706	0.3729	0.0625	0.1125			
Sohkhla Lake	0.4013	0.3845	0.3856	0.0416	0.1088	0.0112		
Kelantan basin	0.4192	0.4010	0.4027	0.0359	0.0904	0.0136	0.0000	

Parenthesized number indicates locality number given in Fig. 1.

A UPGMA phenogram for the eight local samples was constructed based on the unbiased genetic distances between them. The phenogram indicated that the samples were grouped into two distinct clusters, one comprising samples from the Mekong-Chao Phraya group (Nos. 1-3) and the other, those from the Mae Khlong-Malay Peninsula group (Nos. 4-8) (Fig. 3). The genetic distance between the two clusters was 0.4539.

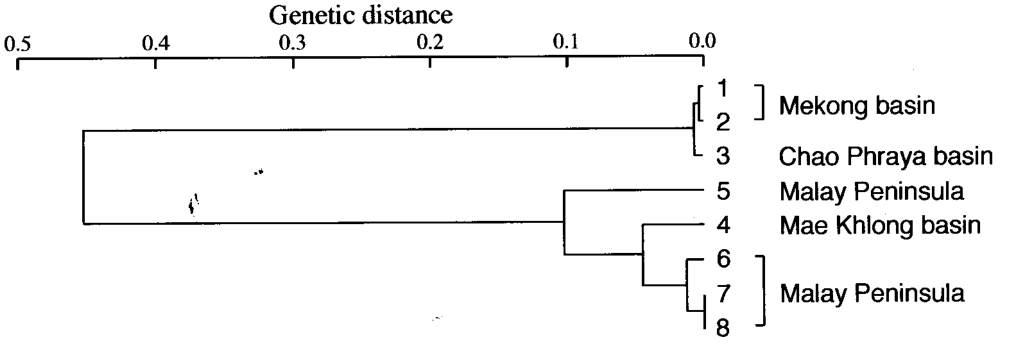

Fig. 3. Dendrogram of genetic distance of *Osteochilus hasseltii* from eight localities, based on allelic frequencies at 10 loci. For explanation of locality numbers, see Fig. 1.

Morphological Examination

Meristics. - Intraspecific variations were found in the number of total vertebrae, total lateral line scales, pre-dorsal scales, branched dorsal fin rays and pectoral fin rays (Kruskal-Wallis test: $p<0.001$, Table 4). Other meristic counts showed little or no variation: branched anal fin rays, 5; pelvic fin rays, 8; scales above lateral line to dorsal fin origin, 5.5 (rarely 4.5); scales below lateral line to anal fin origin, 4.5 (rarely 5.5); scales below lateral line to pelvic insertion, 4.5; and circumpeduncular scales, 16.

All localities were divided into two groups based on the significant differences found for the number of total vertebrae and total lateral line scales (Dunn's multiple comparison test: $p<0.05$), i.e., the Mekong-Chao Phraya group (Nos. 1-3) and the Mae Khlong-Malay Peninsula group (Nos. 4-8) (Table 5).

For the other characters, the following pairs showed significant difference (Dunn's multiple comparison test: $p<0.05$); pre-dorsal scales: Bung Boraphet specimens (No. 3) vs. Tapi specimens (No. 6); branched dorsal fin rays: Houei Mong specimens (No. 1) vs. Tapi specimens (No. 6) and Bung Boraphet specimens (No. 3) vs. Tapi specimens (No. 6), pectoral fin rays: Ubol Ratana specimens (No. 2) vs. Mae Khlong specimens (No. 4). These combinations agreed with the grouping, the Mekong-Chao Phraya group (Nos. 1-3) vs. the Mae Khlong-Malay Peninsula group (Nos. 4-8), based on significant differences of the number of total vertebrae and total lateral line scales.

Morphometrics. - Principal component analysis was carried out for the correlation matrices of 22 log-transformed morphometric variables. The first, second and third components accounted for 94.8%, 1.6% and 0.8% of total variance, respectively (Table 6). Clearly, the first component, in which the values of eigenvector were all positive and the score of the first component largely correlated with standard lengths ($r=0.994$), is indicative of size of the specimens. On the other hand, the second and third component, accounting for 30.8% and 15.4% of the remaining variation, respectively, indicated the difference in body shape.

Table 4. Frequencies of meristic characters in *Osteochilus hasseltii* collected from Thailand and Malaysia.

Character	Locality	Frequency						
Total vertebrae	Houei Mong River		32(1)	**33(18)**				
	Ubol Rat Reservoir		32(1)	**33(22)**	34(2)			
	Bung Boraphet Reservoir			**33(24)**	34(9)			
	Mae Klong basin	31(2)	**32(27)**	33(7)				
	Phetchaburi River		**32(9)**	33(3)	34(1)			
	Tapi basin	31(1)	**32(27)**	33(7)				
	Songkhla Lake	31(1)	**32(22)**	33(1)				
	Kelantan basin	31(1)	**32(17)**	33(1)				
Total line scales	Houei Mong River				34(1)	**35(12)**	36(4)	
	Ubol Rat Reservoir					**35(17)**	36(6)	
	Bung Boraphet Reservoir				34(1)	**35(21)**	36(17)	
	Mae Klong basin			33(2)	**34(22)**	35(12)	36(2)	
	Phetchaburi River			33(1)	**34(11)**	35(2)	36(1)	
	Tapi basin			33(2)	**34(25)**	35(9)		
	Songkhla Lake		32(1)	33(3)	**34(18)**	35(4)		
	Kelantan basin			33(3)	**34(10)**			
Pre-dorsal scales	Houei Mong River		**10(13)**	11(6)				
	Ubol Rat Reservoir	9(1)	**10(13)**	11(8)	12(1)			
	Bung Boraphet Reservoir	9(2)	10(15)	**11(19)**	12(2)	13(1)		
	Mae Klong basin	9(3)	**10(28)**	11(6)				
	Phetchaburi River	9(2)	10(3)	**11(4)**				
	Tapi basin	9(6)	**10(26)**	11(4)				
	Songkhla Lake	9(3)	**10(15)**	11(5)				
	Kelantan basin	9(2)	**10(8)**					
Branched dorsal fin rays	Houei Mong River			14(2)	**15(13)**	16(4)		
	Ubol Rat Reservoir			14(1)	15(10)	**16(11)**	17(2)	
	Bung Boraphet Reservoir			14(2)	**15(21)**	16(14)	17(1)	
	Mae Klong basin	12(1)		14(1)	15(13)	**16(21)**	17(2)	
	Phetchaburi River				15(7)	**16(10)**	17(2)	
	Tapi basin				15(8)	**16(21)**	17(6)	18(1)
	Songkhla Lake				15(7)	**16(18)**	17(1)	
	Kelantan basin				15(5)	**16(9)**	17(3)	
Pectoral fin rays	Houei Mong River			14(6)	**15(8)**	16(5)		
	Ubol Rat Reservoir		13(1)	14(3)	**15(12)**	16(7)	17(1)	
	Bung Boraphet Reservoir		13(1)	14(13)	**15(17)**	16(4)		
	Mae Klong basin			**14(25)**	15(11)			
	Phetchaburi River			**14(3)**	15(1)	16(1)		
	Tapi basin		13(1)	14(12)	**15(22)**	16(1)		
	Songkhla Lake	12(1)		14(12)	**15(14)**			
	Kelantan basin			14(3)	**15(7)**	16(4)		

Numbers of parentheses and bold indicate number of specimens and mode, respectively.

By the three components, the Mae Khlong-Malay Peninsula group (Nos. 4-8), which concentrated on positive value of second component, was wholly included in the Mekong-Chao Phraya group (Nos. 1-3) (Fig. 4).

DISCUSSION

Unbiased estimates of genetic distances made for small-sized samples (Nei, 1978), and used in this study, yield greater values than estimates made through the ordinary calculation

Table 5. Result of Dunn's multiple comparison test analysis of total vertebrae (upper right) and total lateral line scales (lower left) in *Osteochilus hasseltii* between all localities.

Locality	Houei Mong River (1)	Ubol Rat Reservoir (2)	Bung Boraphet Reservoir (3)	Mae Khlong basin (4)	Phetcha-buri River (5)	Tapi basin (6)	Songkhla Lake (7)	Kelantan basin (8)
Howei Mong River		–	–	*	*	*	*	*
Ubol Rat Reservoir	–		–	*	*	*	*	*
Bung Boraphet Reservoir	–	–		*	*	*	*	*
Mae Klong basin	*	*	*		–	–	–	–
Phetchaburi River	–	–	*	–		–	–	–
Tapi basin	*	*	*	–	–		–	–
Sohkhla Lake	*	*	*	–	–	–		–
Kelantan basin	*	*	*	–	–	–	–	

*, p<0.05; –, not significant; parenthesized number indicaes locality given in Fig. 1.

Table 6. Factor score coefficients for the principal component analysis for *Osteochilus hasseltii* using 22 log-transformed morphometric variables.

	PC 1	PC 2	PC 3
Log standard length	0.218	0.027	0.074
Log head length	0.216	-0.216	-0.116
Log head depth at occiput	0.214	-0.204	-0.173
Log head width	0.214	-0.219	-0.093
Log snout length	0.213	-0.215	-0.055
Log orbit diameter	0.197	0.508	-0.720
Log postorbital length	0.209	-0.420	-0.048
Log interorbital width	0.214	-0.237	-0.097
Log body depth	0.216	-0.048	-0.032
Log caudal peduncle length	0.209	0.019	0.408
Log caudal peduncle depth	0.217	0.019	0.004
Log predorsal length	0.217	-0.012	-0.024
Log preanal length	0.215	-0.029	0.035
Log prepectoral length	0.215	-0.197	-0.055
Log prepelvic length	0.217	-0.039	0.007
Log length of last simple dorsal ray	0.212	9.252	0.234
Log length of dorsal fin base	0.215	0.113	0.007
Log length of last simple anal ray	0.212	0.205	0.259
Log length of anal fin base	0.210	0.090	-0.165
Log pectoral fin legnth	0.215	0.176	0.119
Log pelvic fin length	0.214	0.155	0.196
Log length of upper lobe of caudal fin	0.210	0.318	0.187
Eigenvalue	20.853	0.358	0.173
Variance explained	94.8%	1.6%	0.8%
Correlation between score of PC and log-transformed SL	0.994	-0.220	0.254

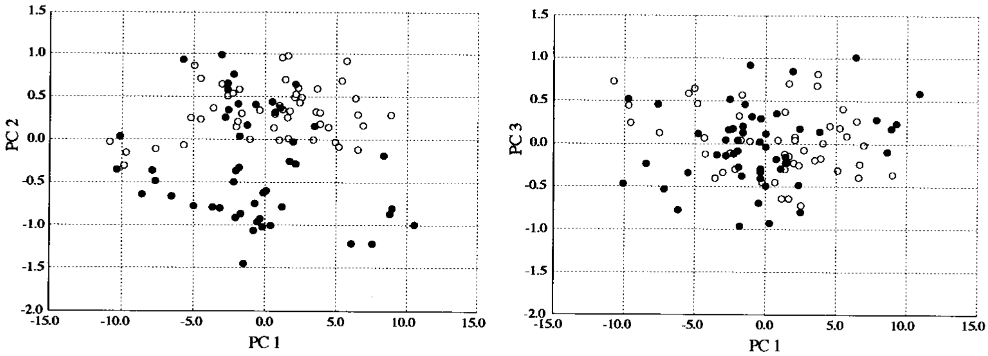

Fig. 4. *Osteochilus hasseltii* individuals distributed according to the scores given by the first (PC1) and second (PC2) (left) and the first (PC1) and third (PC3) (right) principal components derived from the principal component analysis based of 22 log-transformed morphometric variables. ●: Mekong-Chao Phraya group. ○: Mae Khlong-Malay Peninsula group.

method (Nei, 1972). Even so, the genetic distances of 0.3706-0.5709 between the Mekong-Chao Phraya and the Mae Khlong-Malay Peninsula groups of *Osteochilus hasseltii* are considerably greater than the distances generally found between conspecific cypriniform populations occurring in contiguous locations (e.g., 0.00-0.19 in eight populations of the cyprinid *Campostoma anomslum*, Buth & Burr, 1978; 0.004-0.036 in three populations of the catostomid *Hypentelium nigricans*, Buth, 1980; 0.001-0.254 in *Catostomus plebeius*, Ferris et al., 1982). Rather, the distance between the Mekong-Chao Phraya and Mae Khlong-Malay Peninsula groups is comparable with the values between Borneo, Palawan (Philippines) and Mindanao (Philippines) populations of *Puntius binotatus* (0.248-0.454, Kornfield & Carpenter, 1984). Kornfield & Carpenter (1984) concluded that the Borneo and Palawan populations (D=0.248) should be a distinct species from the Mindanao population (D= 0.404 with Borneo and 0.454 with Palawan). Based on these studies, the Mekong-Chao Phraya and Mae Khlong-Malay Peninsula groups (D=0.3706-0.5709) of *O. hasseltii* are considered to be distinct from each other.

In the morphological characters, the Mekong-Chao Phraya and the Mae Khlong-Malay Peninsula groups are distinguished solely by the number of total vertebrae and total lateral line scales. However, because of the wide overlaps in ranges of these characters, differences are not recognized at the individual level.

To summarize, *O. hasseltii* in Thailand consists of two population groups distinguished genetically, although this genetical differentiation are not accompanied by enough morphological differences.

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