

The Recognition and Molecular Phylogeny of *Mugilogobius mertonii* Complex (Teleostei: Gobiidae), with Description of a New Cryptic Species of *M. flavomaculatus* from Taiwan

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Shih-Pin Huang, I-Shiung Chen, Mana M. N. Yung, and Kwang-Tsao Shao (2016) *Mugilogobius mertonii* (Weber, 1911) is considered as a widely distributed species around the Indo- West Pacific region, and several nominal species are considered as junior synonyms of *M. mertonii*. However, in our recent study, several different morphological types of *M. mertonii* were observed, they were collected from Taiwan, Palau and Phuket Island. This study aimed to investigate the taxonomic status of those *M. mertonii*-like individuals, we also attempted to assess their phylogenetic relationship base on combined mitochondrial DNA ND5, Cyt *b* and D-loop sequences. The present morphological and molecular evidences suggested that the current *M. mertonii* could be regarded as a species complex, and several cryptic species might be included in *M. mertonii* complex. One of these which collected from Taiwan is described as a new species, *Mugilogobius flavomaculatus* n. sp. based on both morphological and molecular evidence in this study. The phylogenetic tree also revealed that *M. flavomaculatus* n. sp. is the closest to *M. mertonii*. *M. flavomaculatus* n. sp. and its sister species *M. mertonii* are found to have different niches in the same estuary. Moreover, stable morphological characters and nuclear gene RAG2 also clearly show that no hybridization is detected in between *M. flavomaculatus* n. sp. and *M. mertonii*. Except the present new species, taxonomic status of all junior synonyms refers to *M. mertonii* are also discussed.

Key words: *Mugilogobius*, New species, Taxonomy, Phylogeny, Mitochondrial DNA.

BACKGROUND

The genus *Mugilogobius* was established based on *Ctenogobius abei* Smitt 1900. This is a group of small sized benthic gobies, which mostly occur in blackish water of the Indo-West Pacific region. A few species of this genus can also be found in freshwater habitats (Larson and Lim 2005; Huang 2014). Generally, species of *Mugilogobius* can be recognized by following combination of unique features: head pore always absent, typical longitudinal sensory papillae pattern, papillae rows *c* and *c1* usually present, papillae row *p* always complete (Huang 2014). In addition to 25 species were regarded as valid (Larson 2001), recently,

a species, *M. hitam* Larson, Geiger, Hadiaty and Herder, 2014 also have described from Sulawesi (Larson et al. 2014).

Although several species of *Mugilogobius* have described in last two decades, however, molecular phylogeny of *Mugilogobius* is still poorly known, only a phylogenetic analysis based on partial Cytochrome Oxidase Subunit 1 (COI) gene sequences from 10 species of *Mugilogobius* have reported so far (Larson et al. 2014), most species of their targets belong to endemic species of Sulawesi.

In recent years, detailed study of *Mugilogobius* species had been carried out in Taiwanese waters. *M. abei*, *M. cavifrons*, *M. chulae*, *M. mertonii* and

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M. myxodermus were collected and recorded from Taiwan (Huang et al. 2013b). Among the genus *Mugilogobius*, *M. mertonii* was established by Weber in 1911, which was collected from eastern Indonesia. A total of four nominal species, including *Gobius durbanensis* Barnard, 1927, *Tamanka mindora* Herre, 1945, *Vaimosa layia* Herre, 1953 and *Stigmatogobius inhacae* Smith, 1959 were considered as junior synonyms of *M. mertonii* (Larson 2001). Among these nominal species, *Tamanka mindora* and *Vaimosa layia* were described from Philippines, *Gobius durbanensis* and *Stigmatogobius inhacae* described from South Africa and Mozambique, respectively, around the East Indian Ocean. In our recent study, several different morphological types of *M. mertonii* are observed, they are collected from Taiwan, Palau and Phuket Island of Thailand, respectively.

The molecular markers usually provide a good resolution for congeneric specific identification (Huang et al. 2013a). We herein attempt to resolve taxonomic problem of *M. mertonii* and other *M. mertonii*-like species based on combined molecular and morphological evidence. Moreover, a phylogenetic relationship of *M. mertonii* and other several valid species from Taiwan and Southeast Asia will also be assessed.

MATERIALS AND METHODS

Sample collection

All the examined specimens from Taiwan and Southeast Asia were collected by hand-net. Specimens tissues used for molecular analysis were preserved in 95% ethanol; specimens used for morphological studies were fixed in 10% formalin, followed by 70% ethanol for long-term preservation.

Morphological studies

The morphological measurements followed Miller (1988) and meristic counts follow Chen and Shao (1996) and Chen and Kottelat (2003). The terminology of cephalic sensory canals and free neuromast organ (sensory papillae) was based on Sanzo (1911), Miller (1988) and Chen and Shao (1998). All lengths were standard length (SL). All examined materials were deposited in National Taiwan Ocean University, Keelung, Taiwan (NTOUP) and Biodiversity Research Museum, Biodiversity Research Center, Academia Sinica,

Taipei, Taiwan (ASIZP).

Molecular phylogenetic studies

The phylogenetic relationships were studied using the mitochondrial DNA (mtDNA) sequence of full length of mitochondrial Cytochrome *b* (Cyt *b*), D-loop and partial NADH dehydrogenase subunit 5 (ND5). All DNA extractions of the samples used a High Pure Product Preparation kit (Roche, USA). ND5 region was amplified by PCR using the following two primers: (PGLeuD1: 5'-AAAGGATAACAGCTCATCCGTTGGTCT-3'; ND5MR: 5'-CCTATTTTKCGGATGTCYTG-3'). Cyt *b* region was amplified by PCR using the following two primers: (GGLuF: 5'-TAACCAGGACTARTGRCTTG-3'; GProR: 5'-GTTARAATCTCYTTCTTTG-3'). D-loop region was amplified by polymerase chain reaction (PCR) using the following two primers: (GTHR: 5'-TCAGC GCCAGAGCGCCGKTCTTGTA-3'; PGL5: 5'-CTA GGGYCTATCCTAACATCTTCA-3').

In order to strengthen the reliability of taxonomic conclusion, and attempt to include more species in the present molecular analysis, another molecular phylogenetic analysis using partial COI gene was performed, which include all examined species of *Mugilogobius* in the present study and Larson et al. (2014). COI region was amplified by PCR using the following two primers: (FishF2: 5'-TCGACTAATCATAAAGATATCGGCAC-3'; FishR2: 5'-ACTTCAGGGTGACCGAAGAATCAG AA-3'), which followed Ward et al. (2005).

On account of mitochondrial DNA was characterized by inherited exclusively from the female. Thus, a further molecular analysis between the present new species and its closely related species was carried out using a nuclear gene, recombination-activating gene 2 (RAG2) for affirming whether hybridization may occur in between two sister species. RAG2 region was amplified by PCR using the following two primers: (R-RAG2F: 5'-GTCTGAACCCCAACAATGAG-3'; R-RAG2R: 5'-GCTGTCTGTCATTCATGTG-3'), which followed Yamasaki et al. (2015).

PCR was done in a MODEL 2700 or 9700 thermal cycler (Perkin-Elmer) and 35-40 cycles were carried out. The 50 µL reaction volume contained 33.5 µL of sterile distilled water, 5 µL of 10X PCR buffer (Takara), 4 µL of dNTP (2.5 mM each), 3 µL of MgCl₂ (2.5 mM each), 1 µL of each primer, 0.5 µL of 0.5 unit Ex Taq (Takara) and 2 µL of template. The thermal cycler profile was as follow: denaturation at 94°C for 60 seconds,

annealing at 52–58°C for 60 seconds and extension at 72°C for 120 seconds. A negative control without template was carried out for each run of PCR. The PCR products were run on a 1.0% L 03 agarose gel (Takara) and stained with ethidium bromide for band characterization under ultraviolet trans-illumination.

Double-stranded PCR products were purified using a High Pure Product Purification kit (Roche), before undergoing direct cycle sequencing with dye-labeled terminators (ABI Big-Dye kit). All sequencing reactions were performed according to the manufacturer's instructions. Labeled fragments were analyzed using an ABI PRISM Model 377-64 DNA Automated sequencer (ABI). All sequencing works were carried out by National Center for Genome Medicine, Academia Sinica.

Nucleotide sequence alignment was verified manually after running through BIOEDIT version 5.9 (Hall 2001). The analysis of aligned mutation sites was conducted using Molecular Evolutionary Genetics Analysis (MEGA) version 5.05 (Tamura et al. 2011) for aligned mutation sites analysis. Neighbor-joining (NJ), maximum parsimony (MP) and Bayesian inference (BI) were employed for phylogenetic analysis in this study. The NJ analysis was carried out using PAUP* version 4.0B10 (Swofford 2003) using Neighbor Joining/UPGMA. The MP analysis is carried out using PAUP* version 4.0B10 (Swofford 2003) using heuristic search. Branch support for NJ and MP trees were established via bootstrap analysis (2000 replications). For the Bayesian inference (BI), the best-fitting model for sequence evolution was determined for mitochondrial and nuclear DNA sequences using jModelTest v.2.1.3 (Darriba et al. 2012). The BI analyses was performed using MrBayes 3.0 (Ronquist and Huelsenbeck 2003), a total of 2000000 times of replications were performed in BI analyses. The posterior probabilities of each node were computed from the remaining 75% of all sampled trees. *Rhinogobius brunneus* (Temminck and Schlegel 1845) was used as outgroup in all molecular analysis, which obtained from GenBank. This genus was regarded as an appropriate outgroup for assessing the relatedness of genus *Mugilogobius* (Larson et al. 2014).

RESULTS

Molecular phylogenetic analysis

The aligned ND5, Cyt *b* and D-loop sequence consisted of 21 haplotypes which were from eight species of *Mugilogobius* with 21 individuals. The sampling localities, OTU codes and accession numbers are listed in table 1. The total length of combined sequences of partial ND5 sequence, complete Cyt *b* and D-loop sequences was 3016–3025 bp (1035 bp in ND5, 1141 bp in Cyt *b* and 840–849 bp in D-loop). This alignment contained 1149 total mutations and 887 polymorphic sites. The molecular phylogenetic tree of NJ was based on the Kimura 2-parameter (K2P) model. The phylogenetic tree of BI analysis was based on GTR + I + G model. The result of MP analysis by the heuristic search only obtained one tree with minimum tree length 2295 with the consistency index (CI) of 0.6710, retention index (RI) of 0.8286, and homoplasy index (HI) of 0.3290.

The aligned partial COI sequence consisted of 17 haplotypes which were from 15 species of *Mugilogobius* with 17 individuals. The species, accession numbers and sources for COI gene analysis were listed in table 2. Total length of partial COI sequences was 563 bp. This alignment contained 288 total mutations and 191 polymorphic sites. MP and BI were employed for phylogenetic analysis. The phylogenetic tree of BI analysis was based on HKY+G model. The result of MP analysis by the heuristic search only obtained one tree with minimum tree length 574, with the CI of 0.5436, RI of 0.5428, and HI of 0.4564.

The aligned partial RAG2 sequence consisted of three haplotypes which were from *M. flavomaculatus* n. sp. and its related species, *M. mertoni* with six individuals. The total length of partial RAG2 gene was 683 bp. This alignment contained 21 total mutations and polymorphic sites, respectively. MP and BI were employed for phylogenetic analysis. The phylogenetic tree of BI analysis was based on GTR model. The result of MP analysis by the heuristic search only obtained one tree with minimum tree length 102 with the CI of 0.9804, RI of 0.7143, and HI of 0.0196.

For the phylogenetic analysis reconstructed by combined ND5, Cyt *b* and D-loop sequences, the BI, NJ and MP methods represented very similar tree topology (Fig. 1).

The phylogenetic tree was divided into three major clades. *M. tigrinus* formed a distant clade (clade I). Clade II consisted of four species: *M.*

Table 1. Sampling localities, OTU codes and accession numbers for molecular sequence analysis based on mitochondrial ND5, Cyt *b*, D-loop and nuclear RAG2 sequences

Code	Species	Locality	Accession Number			
			ND5	Cyt <i>b</i>	D-loop	RAG2
MABPZ1	<i>Mugilogobius abei</i>	Estuary of Puzi River, Taiwan	JX133909	KT203503	JX133897	-
MABKM1	<i>Mugilogobius abei</i>	Kinmen Island, Taiwan	JX133910	KT203504	JX133898	-
MABKM2	<i>Mugilogobius abei</i>	EKinmen Island, Taiwan	JX133910	KT203504	JX133898	-
MABHK1	<i>Mugilogobius abei</i>	Hong Kong, China	JX133910	KF929327	JX133898	-
MCAPZ1	<i>Mugilogobius cavifrons</i>	Estuary of Puzi River, Taiwan	JX133912	KF929321	KF779960	-
MCAZA1	<i>Mugilogobius cavifrons</i>	Estuary of Zhuan River, Taiwan	JX133912	KF929321	JX133902	-
MCAZA2	<i>Mugilogobius cavifrons</i>	Estuary of Zhuan River, Taiwan	JX133912	KF929321	JX133903	-
MCHZA1	<i>Mugilogobius chulae</i>	Estuary of Zhuan River, Taiwan	JX133911	KT203501	JX133900	-
MCHKM1	<i>Mugilogobius chulae</i>	Kinmen Island, Taiwan	JX133911	KT203500	JX133900	-
MCHHK1	<i>Mugilogobius chulae</i>	Hong Kong, China	JX133911	KT203502	JX133900	-
MMEPL1	<i>Mugilogobius mertoni</i>	Mangrove in Palau	KF958743	KF929318	KF779961	KX158192
MMEZA1	<i>Mugilogobius mertoni</i>	Estuary of Zhuan River, Taiwan	JX133914	KF929318	JX133904	KX158191
MMEZA2	<i>Mugilogobius mertoni</i>	Estuary of Zhuan River, Taiwan	JX133914	KF929318	JX133905	KX158191
MFLZA1	<i>Mugilogobius flavomaculatus</i> n. sp.	Estuary of Zhuan River, Taiwan	JX133915	KT203498	JX133906	KX158193
MFLZA2	<i>Mugilogobius flavomaculatus</i> n. sp.	Estuary of Zhuan River, Taiwan	JX133915	KT203498	JX133907	KX158193
MMYCL1	<i>Mugilogobius myxodermus</i>	Yangliao River, Taiwan	JX133913	KF929325	JX133901	-
MMYHJ1	<i>Mugilogobius myxodermus</i>	Hanjiang River basin, China	JX133913	KF929326	JX133901	-
MSPPK1	<i>Mugilogobius</i> sp.	Phuket Island, Thailand	KT203495	KT203499	KT203492	-
MSPPK2	<i>Mugilogobius</i> sp.	Phuket Island, Thailand	KT203495	KT203499	KT203493	-
MTIML1	<i>Mugilogobius tigrinus</i>	Matang mangrove, Malaysia	KT203494	KT203496	KT203490	-
MTIML2	<i>Mugilogobius tigrinus</i>	Matang mangrove, Malaysia	KT203494	KT203497	KT203491	-
RBRUN1	<i>Rhinogobius brunneus</i>		KT601096	KT601096	KT601096	AB988580

Table 2. Species, accession numbers and sources for molecular sequence analysis based on COI sequences

Species	Code	Accession Number	Sources
<i>Mugilogobius abei</i>	MABPZ1	KX056131	This study
<i>Mugilogobius adeia</i>		KM887179	Larson et al. 2014
<i>Mugilogobius cavifrons</i>	MCAZA1	KX056132	This study
<i>Mugilogobius chulae</i>	MCHZA1	KX056133	This study
<i>Mugilogobius flavomaculatus</i> n. sp.	MFLZA1	KX056134	This study
<i>Mugilogobius hitam</i>		KM887181	Larson et al. 2014
<i>Mugilogobius latifrons</i>		KM887182	Larson et al. 2014
<i>Mugilogobius lepidotus</i>		KM887165	Larson et al. 2014
<i>Mugilogobius mertoni</i>	MMEZA1	KX056135	This study
<i>Mugilogobius mertoni</i> 1		KM887185	Larson et al. 2014
<i>Mugilogobius mertoni</i> 2		KM887180	Larson et al. 2014
<i>Mugilogobius myxodermus</i>	MMYCL1	KX056136	This study
<i>Mugilogobius rexi</i>		KM887183	Larson et al. 2014
<i>Mugilogobius sarasinorum</i>		KM887184	Larson et al. 2014
<i>Mugilogobius</i> sp.		KM887168	Larson et al. 2014
<i>Mugilogobius</i> sp.	MSPPK1	KX056137	This study
<i>Mugilogobius tigrinus</i>	MTIML1	KX056138	This study
<i>Rhinogobius brunneus</i>		KT601096	Park et al. 2015 (unpublished)

cavifrons, *M. mertoni*, *M. flavomaculatus* n. sp. and *M. sp.*, among this clade, *M. mertoni* and *M. flavomaculatus* n. sp. were in a closely related sister group. Clade III consisted of *M. chulae*, *M. myxodermus* and *M. abei*, which were sister to clade II. Except for one node on the branch which divided *M. cavifrons* and *M. mertoni*-*M. flavomaculatus* n. sp. with low bootstrap supports, as 50 and 51 in NJ and MP trees, respectively. Most specific level of nodes are generally with highly support of bootstrap values (69-100 in NJ tree, 78-100 in MP tree), and posterior probabilities generally as high as 0.91-1.00 in BI tree.

Compared to combined ND5, Cyt *b* and D-loop tree, the COI phylogenetic tree revealed a different grouping result. The phylogenetic tree was divided into five major clades (Fig. 2). The tree topology revealed that the *M. sp.* (from Phuket Island) was the earliest offshoot (clade I), *M. cavifrons* formed the clade II, *M. mertoni* and *M. flavomaculatus* n. sp. formed a sister pair (clade III), six species from Sulawesi formed a closely related group (clade IV), five species from Taiwan and Southeast Asia formed another related group (clade V). Most specific level of nodes are generally supported by high posterior probabilities, as 0.88-1.00 in BI tree. However, most bootstrap

supports are less than 50 at several inter-clade and interspecific level of nodes. Inter-specific nodes between the herein described *M. flavomaculatus* and its sister species, *M. mertoni* were supported by high bootstrap values (reaching 100) in MP and posterior probabilities (as high as 1.00) in BI tree.

A molecular tree based on nuclear gene RAG2 revealed that *M. flavomaculatus* n. sp. was well separated from its sister species, *M. mertoni*, and interspecific nodes with high posterior probabilities as high as 1.00 in BI tree, and bootstrap support also reached 93 in MP tree (Fig. 3).

In comparison with genetic distance of interspecific relationship among all species of *Mugilogobius* from Taiwan and Southeast Asia is the range from 5.5-19.6; 7.8-19.0 and 5.2-14.8% for ND5, Cyt *b* and D-loop sequences, respectively based on K2P model. The range of the interspecific genetic distance of *M. flavomaculatus* n. sp. and other 7 species is 5.5-18.1; 7.8-17.8 and 5.2-13.3% for ND5, Cyt *b* and D-loop sequences, respectively. The range of genetic distance of *M. flavomaculatus* n. sp. and *M. mertoni* is 5.5-6.4; 7.8-7.9 and 5.2-6.0% for ND5, Cyt *b* and D-loop sequences, respectively.

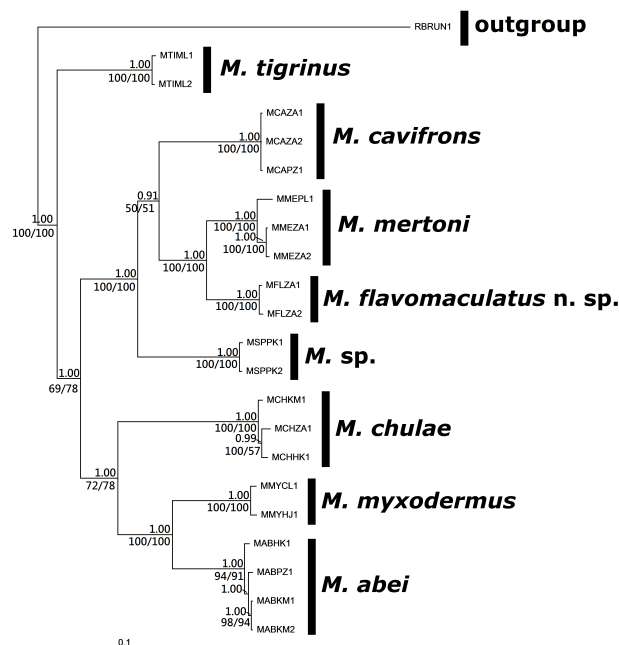


Fig. 1. Molecular phylogenetic tree of *Mugilogobius flavomaculatus* n. sp. and other 7 species of *Mugilogobius* from Taiwan and Southeast Asia reconstructed based on combined ND5, Cyt *b* and D-loop sequences by Bayesian inference method (values above the branch: posterior probability). The similar topology for bootstrap consensus tree by neighbor-joining method (anterior value) and maximum parsimony method (posterior value) list only the bootstrap (value below the branch: bootstrap number, 2000 replications). The bootstrap support > 50 was shown.

SYSTEMATIC

***Mugilogobius flavomaculatus* n. sp.**

urn:lsid:zoobank.org:act:D0318D27-2C4A-4B31-9694-FEF9304A0FD1

Material examined: Holotype: ASIZP0078393, 33.0 mm SL, male, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and M.T. Chou, 6 October, 2012. Paratypes: NTOUP 2012-03-129, 3 specimens, 35.3-40.0 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and M.T. Chou, 30 November, 2009. NTOUP 2013-10-107, 3 specimens, 25.7-28.1 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and M.T. Chou, 4 November, 2013.

Diagnosis: *Mugilogobius flavomaculatus* n. sp. can be well distinguished from other congeners by the combination of following features: (1) second dorsal fin rays modally 1/8, anal fin rays 1/8, pectoral fin rays modally 15. Longitudinal scales 34-35, predorsal scales 18-21; (2) first dorsal fin rounded, without filamentous spinous ray; and (3) body with seven distinct black stripes, cheek and operculum with blackish brown network surrounding of 5-7 rounded, bright yellow spots,

first dorsal fin with a somewhat horizontally broad black blotch, basal region of caudal fin membrane with a vertical black bar or two separate brownish black spots.

Description: Body elongated, subcylindrical anteriorly and compressed posteriorly. Head large. Snout slightly prominent than the lower lip. Eye rather large. Mouth medium, maxillary extending to the vertical of anterior margin of pupil. Anterior nasal as short tube, posterior nasal as round hole. Gill slit rather restricted, extending ventrally, reaching the middle vertical line of the operculum. The morphological measurements are given in table 3. VC 10 + 16 = 26 (in 3 individuals).

Fins: D1 VI; D2 I/7-8 (modally 8); A I/8; P 14-16 (modally 15) (Table 4). First dorsal fin rounded, without filamentous spinous ray (Fig. 4). Second to fifth spinous rays usually longest. Second dorsal and anal fin low. Anal fin inserted below first branched rays of second dorsal fin. Pelvic fin large and rounded. Caudal fin rounded.

Scales: LR 34-35 (modally 34); TR 10; PreD 18-21 (modally 19); SDP 9 (Table 4). Body covered with moderate size ctenoid scales. Predorsal region covered with small sized cycloid scales. Belly covered with small sized cycloid scales. Cheek naked. Upper region of operculum covered with small sized cycloid scales.

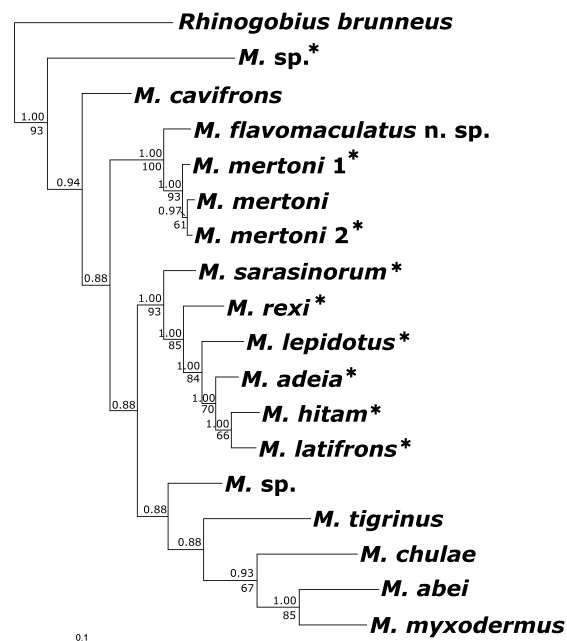


Fig. 2. Molecular phylogenetic tree of 15 species of *Mugilogobius* from Taiwan and Southeast Asia reconstructed based on partial COI sequences by Bayesian inference method (values above the branch: posterior probability). The similar topology for bootstrap consensus tree by maximum parsimony method list only the bootstrap (value below the branch: bootstrap number, 2000 replications). The bootstrap support > 50 was shown. * means sequences from Larson et al. (2014).

Head lateral-line system (Fig. 5): **Head pores**-Head pore absent. **Sensory papillae**- Row *a* short, about half of orbit diameter. Row *b* long, about equal to eye diameter, and with densely-set papillae, starting from vertical of rear margin of pupil. Rows *c* and *c1* short, length almost equal to pupil diameter. Row *cp* rather long, almost equal to eye diameter, and starting from vertical of anterior margin of orbit, extending to vertical of rear margin of orbit. Row *d* longer than eye diameter. Row *s* has three rows papillae. Row *p* completed. Opercular papillae with rows *oi*, *os* and *ot*. Rows *ot* and *oi* well separated. Row *f* has a pair papillae.

Coloration in fresh specimen (Fig. 6): Head and body yellow or yellowish brown, body with seven distinct vertical black cross-bands, first and second stripes somewhat oblique dorsally. Upper and lower lips blackish brown. Cheek and operculum with blackish brown network

surrounding of 5-7 rounded, bright yellow spots. Nape generally yellow or yellowish brown with anterior two blackish brown cross-bands radiated narrowly from anterior dorsal part curved to both lateral regions. First dorsal fin yellow, with a somewhat horizontally broad black blotch, and a distal narrow creamy yellow blotch above black blotch. First dorsal fin have rather thin black margin. Lower region of second dorsal fin membrane yellow and distal region with few black bars. Pectoral fin membrane pale grayish white, middle region of the base with a black mark. Anal fin membrane pale grayish white with deep brown spine and rays. Caudal fin membrane somewhat translucent and grayish, and its fin base usually yellow in adult males, but usually grayish white in females, basal region of caudal fin membrane with a vertical black bar in large adult individual, but usually two separate oval, brownish black marks

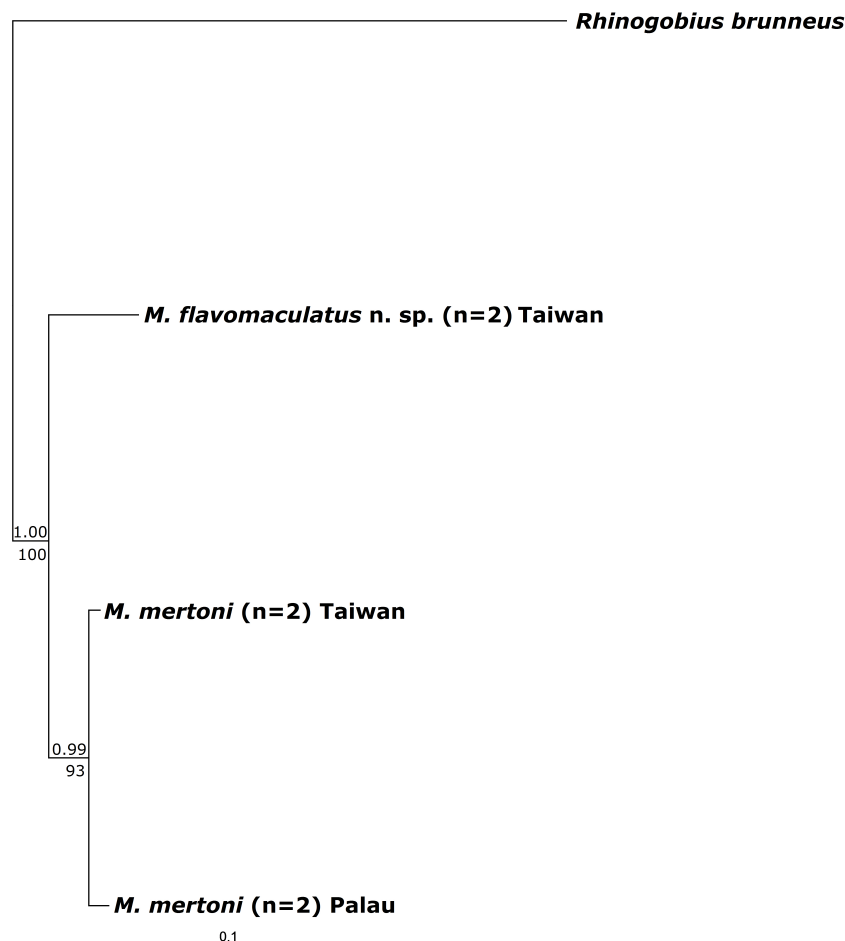


Fig. 3. Molecular phylogenetic tree of *M. flavomaculatus* n. sp. and its sister species, *M. mertoni* from Taiwan and Palau reconstructed based on nuclear gene RAG2 by Bayesian inference (values above the branch: posterior probability). The similar topology for bootstrap consensus tree by maximum parsimony method list only the bootstrap (value below the branch: bootstrap number, 2000 replications). The bootstrap support > 50 was shown.

in small size individuals (less than 3.0 mm SL). Upper region of caudal fin base with a horizontal black bar sometimes connected with the last black cross-band of caudal peduncle. Generally, no obvious sexual dimorphism existing in this new species.

Coloration in long preserved specimen: After the long preservation, all bright yellow marks faded into pale brown or grayish white, other dark coloration patterns still similar to fresh specimens. Head and body brown or gray, body side with seven vertical black cross-bands, first and second stripes somewhat oblique dorsally. Upper and lower lips blackish brown. Cheek and operculum with black network surrounding of 5-7 rounded, grayish white spots. Nape brown with two oblique black cross-bands. Belly pale brown. First dorsal

fin gray, with a somewhat horizontally broad black blotch, and a distal narrow gray blotch above black blotch. Second dorsal fin, pectoral fin, anal fin, and caudal fin membranes always grayish brown. Basal region of caudal fin membrane with a vertical black bar or two separate oval, black marks. Upper region of caudal fin base with a horizontal black bar sometimes connected with the last black cross-band of caudal peduncle.

Distribution: *M. flavomaculatus* n. sp. is a rare species. So far, it has been found only in low salinity waters (0.1-0.4 psu) in the estuaries of northeast Taiwan (Fig. 7).

Etymology: This new specific name, *flavomaculatus*, refers to its diagnostic coloration on cheek and operculum: the presence of 5-7 rounded, bright yellow spots, derived from Latin

Table 3. Morphometric measurements of *Mugilogobius flavomaculatus* n. sp. from Taiwan

Character	<i>Mugilogobius flavomaculatus</i> n. sp.		
	Holotype	Paratypes	
	Male	Male	Female
n		2	1
Percent standard length (%)			
Head length	27.2	26.6 - 27.1 (26.9)	25.0
Predorsal length	37.9	36.3 - 37.6 (36.9)	35.0
Snout to 2nd dorsal origin	57.7	57.9 - 58.1 (58.0)	56.9
Snout to anus	52.8	53.8 - 55.4 (54.6)	54.9
Snout to anal fin origin	57.1	58.0 - 58.6 (58.3)	58.2
Prepelvic length	29.7	27.0 - 28.0 (27.5)	28.3
Caudal peduncle length	27.3	26.0 - 26.6 (26.3)	26.0
Caudal peduncle depth	12.5	11.7 - 13.3 (12.5)	13.0
1st dorsal fin base	11.1	10.8 - 11.9 (11.4)	10.7
2nd dorsal fin base	19.6	20.0 - 20.5 (20.2)	18.1
Anal fin base	19.5	17.7 - 18.0 (17.9)	17.4
Caudal fin length	25.2	24.3 - 24.9 (24.6)	22.4
Pectoral fin length	22.4	20.6 - 21.9 (21.3)	20.2
Pelvic fin length	17.4	16.4 - 16.5 (16.5)	15.3
Body depth at pelvic fin origin	18.5	16.8 - 17.4 (17.1)	18.0
Body depth at anal fin origin	16.5	16.5 - 17.8 (17.2)	19.2
Body width at anal fin origin	14.6	14.0 - 14.2 (14.1)	13.2
Pelvic fin origin to anus	25.4	26.9 - 27.2 (27.1)	31.6
Percent head length (%)			
Snout length	36.5	35.4 - 36.5 (36.0)	33.8
Eye diameter	28.2	26.1 - 26.9 (26.5)	30.2
Cheek depth	30.7	28.7 - 30.1 (29.4)	27.8
Postorbital length	56.2	55.8 - 56.5 (56.2)	56.6
Head width in maximum	78.6	79.1 - 83.0 (81.0)	80.1
Head width in upper gill	55.2	56.2 - 58.3 (57.3)	59.2
Bony interorbital width	22.2	22.8 - 23.7 (23.2)	25.4
Fleshy interorbital width	38.5	37.8 - 38.5 (38.1)	39.3
Lower jaw length	45.2	45.1 - 46.5 (45.8)	43.3

words, *flavus* (yellow) and *maculata* (spot).

Remarks: *Mugilogobius flavomaculatus* n. sp. can be well distinguished from all 26 valid species by the combined morphological features. This new species can be immediately distinguished from nine valid species, including *M. abei*, *M. chulae*, *M. fasciatus*, *M. filifer*, *M. fuscus*, *M. lepidotus*, *M. myxodermus*, *M. rivulus* and *M. tigrinus* by all spinous rays never filamentous versus first spinous dorsal ray elongated and filamentous in adult males.

Mugilogobius flavomaculatus n. sp. can also be well distinguished from *M. amadi* and *M.*

platynotus by meristic features. This new species can be immediately distinguished from *M. amadi* by having fewer second dorsal fin rays (I/7-8 versus to I/9-10) and fewer anal fin rays (I/8 versus to I/10-12); and it can also be separated from *M. platynotus* by having fewer longitudinal scales 34-35 versus to more longitudinal scales 49-59. *M. flavomaculatus* n. sp. can also be well distinguished from *M. rexi* by having different sensory types (typical longitudinal sensory papillae versus to transverse papillae below the eye).

Compared to remaining following 13 valid species, including: *M. adeia*, *M. cagayanensis*,

Table 4. Frequency distribution of meristic counts of *Mugilogobius flavomaculatus* n. sp. and other comparative materials from Taiwan and Southeast Asia

	D1			D2					A				P						
	VI	VII	x	I/6	I/7	I/8	I/9	x	I/6	I/7	I/8	x	13	14	15	16	17	x	
<i>M. flavomaculatus</i> n. sp.	7	-	6.0	-	1	6	-	7.9	-	-	7	8.0	-	2	8	1	-	14.9	
<i>M. abei</i>	15	-	6.0	-	-	15	-	8.0	-	-	15	8.0	-	-	2	19	9	16.2	
<i>M. cavifrons</i>	9	1	6.1	-	-	9	1	8.1	-	1	9	7.9	-	-	5	13	1	15.8	
<i>M. chulae</i>	25	-	6.0	2	22	1	-	7.0	1	24	-	7.0	2	38	10	-	-	14.2	
<i>M. mertonii</i>	10	-	6.0	-	10	-	-	7.0	-	10	-	7.0	-	2	12	3	-	15.6	
<i>M. myxodermus</i>	9	1	6.1	-	1	9	-	7.9	-	2	8	7.8	-	-	14	3	1	15.3	
<i>M. tigrinus</i>	5	-	6.0	-	5	-	-	7.0	5	-	-	6.0	-	4	6	-	-	14.6	
<i>M. sp.</i>	17	-	6.0	-	17	-	-	7.0	-	17	-	7.0	-	-	8	18	4	15.9	

	LR															TR									
	28	29	30	31	32	33	34	35	36	37	~	44	45	46	47	48	x	8	9	10	11	~	14	15	x
<i>M. flavomaculatus</i> n. sp.	-	-	-	-	-	-	11	3	-	-	-	-	-	-	-	-	34.2	-	-	5	2	-	-	-	10.3
<i>M. abei</i>	-	-	-	-	-	-	-	18	9	3	-	-	-	-	-	-	35.5	-	-	15	-	-	-	-	10.0
<i>M. cavifrons</i>	-	-	-	-	-	-	-	-	-	-	-	2	7	8	2	1	45.7	-	-	-	-	-	7	3	14.3
<i>M. chulae</i>	9	28	13	-	-	-	-	-	-	-	-	-	-	-	-	-	29.1	3	22	-	-	-	-	-	8.9
<i>M. mertonii</i>	-	-	12	8	-	-	-	-	-	-	-	-	-	-	-	-	30.4	-	3	7	-	-	-	-	9.7
<i>M. myxodermus</i>	-	-	-	-	-	8	10	2	-	-	-	-	-	-	-	-	33.7	-	-	10	-	-	-	-	10.0
<i>M. tigrinus</i>	4	3	3	-	-	-	-	-	-	-	-	-	-	-	-	-	28.9	5	-	-	-	-	-	-	8.0
<i>M. sp.</i>	-	-	14	17	3	-	-	-	-	-	-	-	-	-	-	-	30.7	-	3	14	-	-	-	-	9.8

	PreD															SDP							VC				
	10	11	12	13	14	15	16	17	18	19	20	21	22	23	x	7	8	9	10	11	12	13	x	25	26	27	x
<i>M. flavomaculatus</i> n. sp.	-	-	-	-	-	-	-	-	2	3	1	1	-	-	19.1	-	1	6	-	-	-	-	8.9	-	3	-	26.0
<i>M. abei</i>	-	-	-	-	2	1	5	5	1	1	-	-	-	-	16.3	-	-	7	8	-	-	-	9.5	1	5	-	25.8
<i>M. cavifrons</i>	-	-	-	-	-	-	-	-	-	1	-	4	3	2	21.5	-	-	-	-	3	5	2	11.9	-	6	-	26.0
<i>M. chulae</i>	-	3	10	10	2	-	-	-	-	-	-	-	-	-	12.4	5	20	-	-	-	-	-	7.8	-	6	-	26.0
<i>M. mertonii</i>	-	-	2	4	3	1	-	-	-	-	-	-	-	-	13.3	-	5	5	-	-	-	-	8.5	-	4	-	26.0
<i>M. myxodermus</i>	-	-	-	-	2	1	4	3	-	-	-	-	-	-	15.8	-	-	10	-	-	-	-	9.0	-	9	1	26.1
<i>M. tigrinus</i>	4	1	-	-	-	-	-	-	-	-	-	-	-	-	10.2	4	1	-	-	-	-	-	7.2	-	3	-	26.0
<i>M. sp.</i>	-	-	5	9	3	-	-	-	-	-	-	-	-	-	13	5	11	1	-	-	-	-	7.8	-	9	-	26.0

M. cavifrons, *M. fuscus*, *M. hitam*, *M. latifrons*, *M. littoralis*, *M. notospilus*, *M. platystomus*, *M. rambaiae*, *M. sarasinorum*, *M. stigmaticus* and *M. wilsoni* by having different color patterns. For their specific differentiation as coloration patterns of caudal fin, this new species can be easily distinguished from two of these species by representing spotless caudal fin membrane except a basal vertical black bar and upper horizontal black bar versus to more than six vertical thin wavy dark brown stripes in *M. cavifrons* and *M. rambaiae*; two basal brownish black spots in *M. notospilus*; radiated diffuse dark bars or stripes in *M. littoralis*; and several coalescing waving dark lines in *M. cagayanensis* and *M. latifrons*.

For the specific differentiation as coloration patterns of body, this new species can be well

distinguished from *M. fuscus*, *M. hitam* and *M. stigmaticus* by having yellow or yellowish brown body, and with seven distinct vertical black cross-bands versus to having irregular small dark short bars in *M. fuscus*; versus to body dark brown to blackish, without obvious stripes in *M. hitam*; and versus to oblique bars and square blotches on

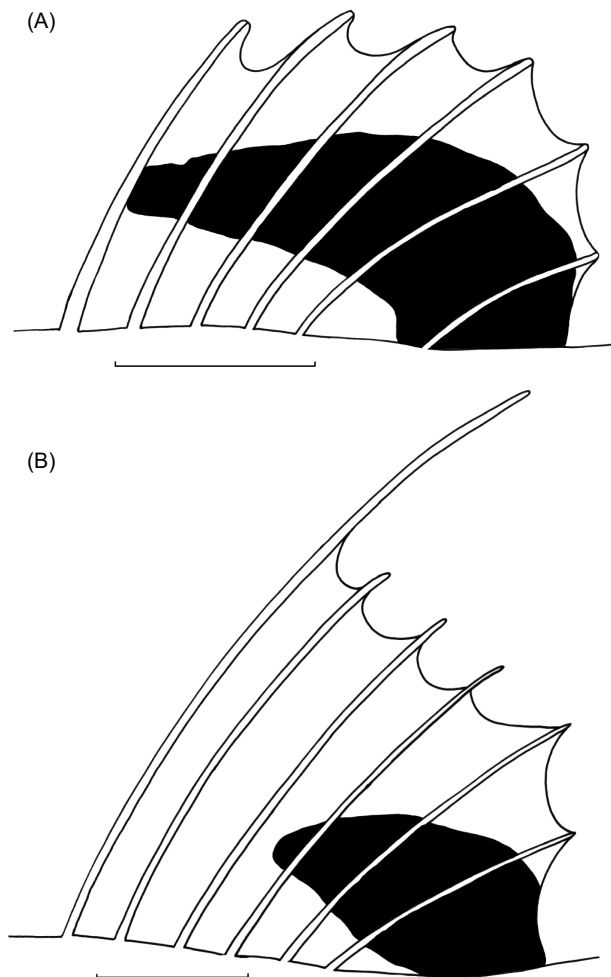


Fig. 4. The comparison of first dorsal fin features of adult male individuals of A, *Mugilogobius flavomaculatus* n. sp. ASIZP0078393, and B, *Mugilogobius mertoni*, NTOUP 2010-08-433. Scale bar = 1 mm. Drawing by Shih-Pin Huang.

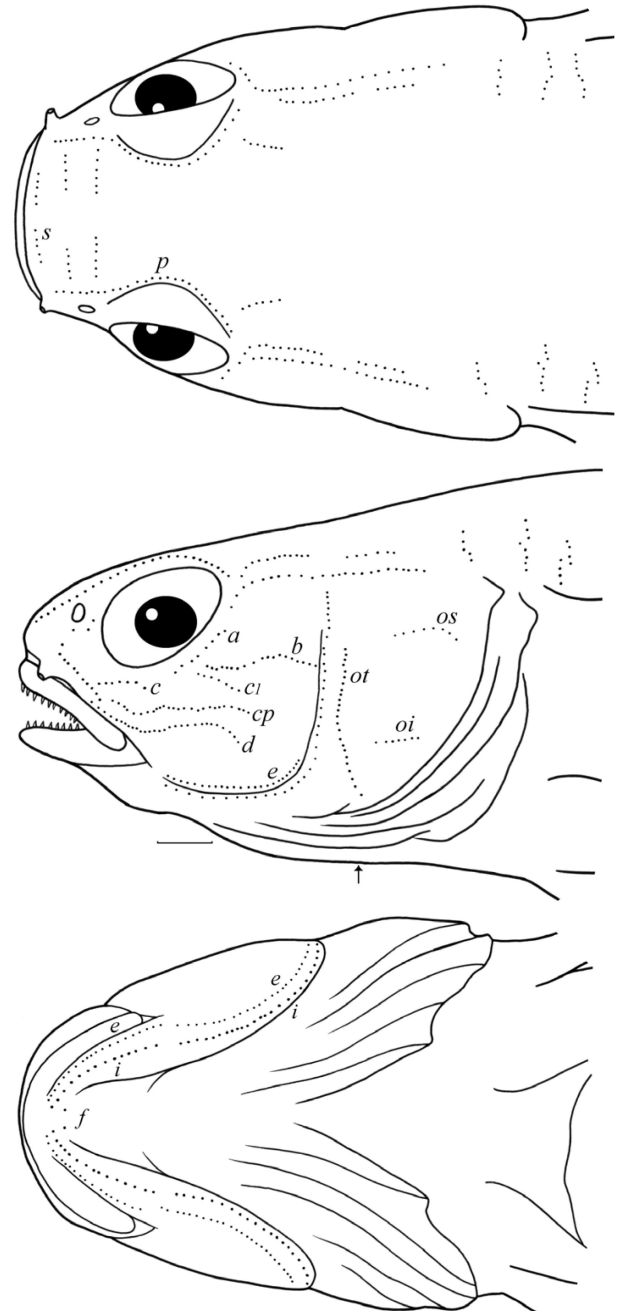


Fig. 5. Head lateral-line system of *Mugilogobius flavomaculatus* n. sp. from Taiwan, ASIZP0078393, holotype, 33.0 mm SL. Scale bar = 1 mm. Drawing by Shih-Pin Huang.

body side in *M. stigmaticus*.

For the specific differentiation in coloration patterns on cheek and operculum, *M. flavomaculatus* n. sp. can be well distinguished from *M. adeia*, *M. platystomus* and *M. wilsoni* by cheek and operculum having blackish brown network surrounding of 5-7 rounded, bright yellow spots versus to having oblique black stripes in *M. adeia*; and two horizontal stripes in *M. platystomus* and *M. wilsoni*). For the specific differentiation as coloration patterns on first dorsal fin, this new species can be well separated from *M. sarasinorum* by having a somewhat horizontally broad black blotch (versus to without obvious blotch in *M. sarasinorum*).

Mugilogobius flavomaculatus n. sp. is most similar to *M. mertoni*, both share the similar body and caudal fin base color patterns. However, this new species still can be immediately distinguished from *M. mertoni* by (1) first dorsal fin without filamentous spines in adult male versus to first spinous ray elongated and filamentous; (2) anal fin rays I/8 versus to I/7; (3) cheek and operculum with blackish brown network surrounding of 5-7 rounded, bright yellow spots versus to having two horizontal stripes; and (4) first dorsal fin with a somewhat horizontally broad black blotch versus to an oval black blotch (Fig. 4).



Fig. 6. The fresh specimen photographs of eight species of *Mugilogobius* from Taiwan and Southeast Asia. (A) *M. flavomaculatus* n. sp., ASIZP0078393, holotype, male, 33.0 mm SL. (B) *M. abei*, NTOUP 2010-11-586, male, 22.4 mm SL; (C) *M. cavifrons*, NTOUP 2012-02-114, male, 42.9 mm SL. (D) *M. chulae*, NTOUP 2010-08-429, male, 29.5 mm SL. (E) *M. mertoni*, NTOUP 2010-08-433, male, 24.5 mm SL. (F) *M. myxodermus*, NTOUP 2010-08-435, male, 32.9 mm SL. (G) *M. tigrinus*, NTOUP 2011-05-002, male, 17.5 mm SL. (H) *M. sp.*, NTOUP 2012-11-171, male, 23.2 mm SL.

DISCUSSION

The first dorsal fin of gobiids is considered as an important character for sexual selection or courtship behavior (Kvarnemo et al. 1995; Takahashi and Yanagisawa 1999; Takahashi and Kohda 2004), diverse types of first dorsal fin are also considered as a diagnostic feature for interspecific identification (Huang et al. 2013a). In the present study, a great differentiation between *Mugilogobius mertonii*, *M. flavomaculatus* n. sp. and *M. sp.* are strongly supported by the present morphological and molecular evidences, it also shows that the first dorsal fin type could be regarded as a useful diagnostic feature within genus *Mugilogobius*.

Except for *M. flavomaculatus* n. sp. we herein described as a new species, all junior synonyms refers to *M. mertonii* will also be reassessed. Another related species, *M. sp.* from Phuket Island is found to have similar first dorsal fin types while comparing to *M. flavomaculatus* n. sp.; however, they still can be immediately separated by different meristic features (A I/8 vs. I/7; LR 34-35 vs. 30-32; PreD 18-21 vs. 12-14), and different color patterns (Cheek and operculum with blackish brown network surrounding of 5-7 rounded, bright yellow

spots versus to two longitudinal black stripes).

Among all junior synonyms of *M. mertonii*, both *Tamanka mindora* and *Vaimosa layia* should be regarded as junior synonyms of *M. mertonii* by the very similar first dorsal fin type and meristic features, it is consistent with Larson's (2001) taxonomic treatments. Compared to *M. mertonii*, we suggest that *Mugilogobius durbanensis* (= *Gobius durbanensis*) could be regarded as a valid species based on different first dorsal fin type and meristic features, we consider this species may be restrictedly distributed around the Indian Ocean region, moreover, *Stigmatogobius inhacae* could be regarded as a junior synonym of *M. durbanensis* based on very similar meristic features. Furthermore, although the present evidences strongly support *M. sp.* as an independent species, however, the *M. sp.* is found to be similar to *M. durbanensis* which is also recorded from the Indian Ocean. Further detailed comparison between *M. durbanensis* and *M. sp.* are still needed in the future.

The *M. mertonii* complex herein we used are referring to several related species which are considered as junior synonyms of *M. mertonii* by Larson (2001). We suggest that *M. mertonii* complex include at least the following three

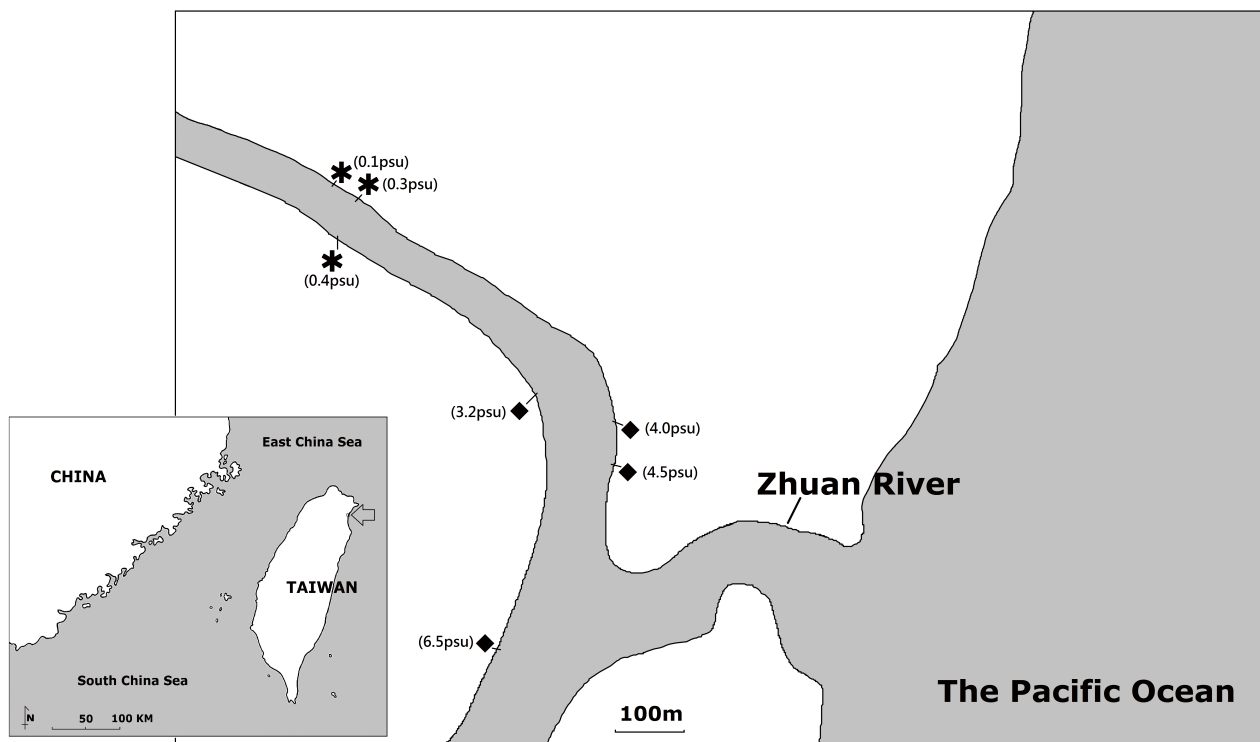


Fig. 7. Sampling sites of *M. flavomaculatus* n. sp. (*) and its closely related species, *M. mertonii* (◆) in the Zhuan River estuary, northeastern Taiwan. Salinities were shown behind the symbols.

species, *M. mertoni*, *M. flavomaculatus* n. sp., and *M. durbanensis*. Among these, we herein described *M. flavomaculatus* n. sp. as a new species, and considered *M. durbanensis* could be a valid species.

The members of *M. mertoni* complex are found to share the following morphological features: (1) Body with about seven vertical black cross-bands; (2) first dorsal fin with a large sized black blotch; (3) cheek and operculum with blackish brown stripes; and (4) caudal fin base with a vertical black bar, but without any vertically aligned lines. Although members of *M. mertoni* complex are found to share the similar color patterns, diagnostic meristic features and first dorsal fin types in adult males are beneficial for identifying these closely related species.

The genetic distance is considered as a diagnostic molecular evidence for species identification in gobiids (Huang et al. 2013a). Compared to other gobiids, the range of genetic distance of *M. flavomaculatus* n. sp. and other species is higher than Japanese freshwater gobies, *Rhinogobius* spp. for ND5 sequence (5.5–18.1 % vs. 4.0–4.8 %) (Mukai et al. 2005) based on K2P model. The range of genetic distance of *M. flavomaculatus* n. sp. and other species is higher than three valid species of brackish water goby, *Hemigobius* for D-loop sequence (5.2–13.3% vs. 3.4–9.4%) (Huang et al. 2013a).

Although *M. flavomaculatus* n. sp. and *M. mertoni* are found in the Zhuan river estuary, they occupy different habitats; the former species occurs in very low salinity areas (0.1–0.4 psu), whereas the latter occurs in higher salinity areas (3.2–6.5 psu). This sampling information revealed that two closely related species utilize different habitats even though they occur in the same estuary.

In one case, hybrid offspring derived from two species of monkey goby *Neogobius* could be identified based on morphological features (Lindner et al. 2013). In our present study, stable meristic features and color patterns could be clearly identified in both *M. flavomaculatus* n. sp. and its sympatric sister species *M. mertoni*, this result also strongly supports the validity of *M. flavomaculatus* n. sp. Moreover, a further molecular analysis based on the nuclear gene, RAG2 also clearly shows that no hybridization is detected in between *M. flavomaculatus* n. sp. and *M. mertoni*.

In recent years, the COI sequences are used for species identification (Ward et al. 2005). The present COI tree topology (Fig. 2) revealed that

each species could be well recognized. However, compared to the combined three mtDNA genes tree, the COI tree showed obviously low bootstrap support at most inter-clade level of nodes, and also revealed that a different grouping result, that indicated the partial COI sequences could provide a good marker for species identification, but may not always be useful for estimating the relatedness of *Mugilogobius*, it is probably due to insufficient sequence length for phylogenetic analysis.

CONCLUSIONS

Our present molecular and morphological evidence strongly support *Mugilogobius flavomaculatus* n. sp. is a valid species, this new species can also be well distinguished from *M. mertoni*. Stable morphological characters and a further molecular analysis based on nuclear gene, RAG2 clearly shows that no hybridization is detected in between *M. flavomaculatus* n. sp. and its related species *M. mertoni*. Except for this species, taxonomic status of other member within *M. mertoni* complex, such as *M. durbanensis*, is still required to resolve.

Overall, the employment of molecular markers has yielded rather useful genetic information for resolving the closely related specific identification such as *M. mertoni* complex. The molecular phylogenetic analysis not only confirms the validity of these possible undescribed species of *Mugilogobius* but also could reconstruct the phylogenetic aspect for further evolutionary history among all species of *Mugilogobius*.

A diagnostic key to all species of *Mugilogobius* from Taiwan

- 1a. Caudal peduncle with two distinct horizontal stripes *M. abei*
- 1b. Caudal peduncle without horizontal stripes 2
- 2a. Longitudinal scale series 44–48 *M. cavifrons*
- 2b. Longitudinal scale series less than 43 3
- 3a. Body side with indistinct irregular thin oblique orange or brown lines when joined forming indistinct X or V-shapes; cheek with five irregular oblique stripes *M. myxodermus*
- 3b. Body side with distinct oblique or transverse black cross-bands 4
- 4a. Predorsal scale series 18–21; cheek and operculum with blackish brown network surrounding of 5–7 rounded, bright yellow spots *M. flavomaculatus* n. sp.
- 4b. Predorsal scale series less than 17; cheek and operculum with horizontal black stripe 5
- 5a. Body side with seven oblique black cross-bands; two round or oval black spots aligned on caudal fin base *M. chulae*
- 5b. Body side with seven irregular transverse black cross-

bands; caudal fin base with a vertical black bar, the front of the bar, with two vertically aligned yellow spots
 *M. mertoni*

List of abbreviations

A; anal fin.
 D1; first dorsal fin.
 D2; second dorsal fin.
 LR; longitudinal scale series.
 P; pectoral fin.
 PreD; predorsal scales.
 SDP; scale series from origin of first dorsal fin to upper pectoral origin.
 TR; transverse scale series.
 VC; vertebral count.

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Other comparative materials

Mugilogobius abei (Jordan and Snyder, 1901)

NTOUP 2010-10-519, 10 specimens, 16.4–33.3 mm SL, estuary of Xinwu River, Taoyuan County, Taiwan, coll. S.P. Huang and H.M. Huang, 28 July, 2010; NTOUP 2012-02-103, 8 specimens, 25.9–31.6 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and Y.H. Kung, 15 February, 2012; NTOUP 2012-02-104, 3 specimens, 23.6–26.0 mm SL, estuary of Puzi River, Dongshi Township, Chiayi County, Taiwan, coll. S.P. Huang, 30 July, 2010; NTOUP 2012-02-105, 2 specimens, 27.4–29.0 mm SL, estuary of Chienpu River, Kinmen Island, Taiwan, coll. S.P. Huang, 22 November, 2011; NTOUP 2012-02-106, 4 specimens, 18.9–24.5 mm SL, Chingyuan Lake, Lieyu Island, Kinmen, Taiwan, coll. S.P. Huang, 24 November, 2011; NTOUP 2012-02-109, 1 specimen, 22.5 mm SL, mangrove of Hong Kong, China, coll. I-S. Chen, 22 November, 2011.

Mugilogobius cavifrons (Weber, 1909)

NTOUP 2010-11-570, 1 specimen, 42.6 mm SL, estuary of Puzi River, Dongshi Township, Chiayi County, coll. S.P. Huang, 2 March, 2010; NTOUP 2010-11-585, 2 specimens, 19.4–19.7 mm

SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang, 22 March, 2010; NTOUP 2012-02-114, 5 specimens, 34.6–42.9 mm SL, Kouhu Township, Yunlin County, Taiwan, coll. S.P. Huang, 5 May, 2010; NTOUP 2012-02-118, 4 specimens, 24.2–27.1 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and Y.H. Kung, 15 February, 2012.

Mugilogobius chulae (Smith, 1932)

NTOUP 2010-08-425, 35 specimens, 16.2–30.0 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and H.M. Huang, 15 January, 2010; NTOUP 2010-08-428, 35 specimens, 20.7–30.6 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and H.M. Huang, 22 April, 2010; NTOUP 2010-08-429, 21 specimens, 20.3–29.5 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and H. M.Huang, 13 May, 2010; NTOUP 2010-08-431, 26 specimens, 20.5–31.9 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and H.M. Huang, 15 July, 2010. NTOUP 2012-02-112, 6 specimens, 18.3–21.3 mm SL, estuary of Chienpu River, Kinmen Island, Taiwan, coll. S.P. Huang, 22 November, 2011; NTOUP 2012-02-113, 3 specimens, 13.3–17.6 mm SL, Chingyuan Lake, Lieyu Island, Kinmen, Taiwan, coll. S.P. Huang, 24 November, 2011; NTOUP 2012-02-109, 1 specimen, 24.7 mm SL, mangrove of Hong Kong, coll. I-S. Chen, 18 July, 2012.

Mugilogobius mertoni (Weber, 1911)

NTOUP 2010-02-102, 3 specimens, 12.9–17.0 mm SL, Palau, coll. I-S. Chen and J.T. Chen, 17 November, 2006; NTOUP 2010-08-432, 2 specimens, 19.3–21.8 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and H.M. Huang, 25 February, 2010; NTOUP 2010-08-433, 1 specimen, 24.5 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and H.M. Huang, 22 March, 2010; NTOUP 2010-08-434, 1 specimen, 26.0 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S. P. Huang and H.M. Huang, 22 April, 2010; NTOUP 2012-02-101, 5 specimens, 18.3–19.6 mm SL, estuary of Zhuan River, Toucheng Township, Yilan County, Taiwan, coll. S.P. Huang and Y.H. Kung,

15 February, 2010.

***Mugilogobius myxodermus* (Herre, 1935)**

NTOUP 2010-08-435, 1 specimen, 32.9 mm SL, lower reach of Yangliao River, Xinwu Township, Taoyuan County, Taiwan, coll. S.P. Huang and H.M. Huang, 9 July, 2010; NTOUP 2010-08-436, 1 specimen, 29.6 mm SL, lower reach of Xinwu River, Xinwu Township, Taoyuan County, Taiwan, coll. S.P. Huang and H.M. Huang, 28 July, 2010; NTOUP 2010-08-437, 8 specimens, 17.0-19.6 mm SL, a pond near Zhongli City, Taoyuan County, Taiwan, coll. S.P. Huang and H.M. Huang, 28 July, 2010; NTOUP 2012-02-110, 1 specimen, 32.1 mm SL, lower reach of Chienpu River, Kinmen Island, Taiwan, Coll. S.P. Huang, 19 May, 2010; NTOUP 2012-02-111, 2 specimens, 21.6-27.1 mm SL, Shuangli Lake, Kinmen Island, Taiwan, coll. S.P. Huang, 20 May, 2010. NTOUP 2010-08-438, 1 specimen, 24.3 mm SL, a stream near Meizhou City, Guangdong Province, Hanjiang River, China, coll. S.P. Huang and M. Chang, 27 July, 2005.

***Mugilogobius tigrinus* Larson, 2001**

NTOUP 2011-05-008, 5 specimens, 14.4-19.6 mm SL, Matang mangrove, Malaysia, coll. I-S. Chen and S.P. Huang, 20 April, 2011.

***Mugilogobius* sp.**

NTOUP 2012-11-171, 23.4 mm SL, male, Sai Yuan, Phuket Island, coll. S.P. Huang, 23 November, 2012. NTOUP 2012-11-162, 13 specimens, 18.9-26.9 mm SL, Sai Yuan, Phuket Island, Thailand, coll. S.P. Huang, 23 November, 2012. NTOUP 2012-11-166, 3 specimens, 13.5-25.5 mm SL, Cherngtalay, Phuket Island, Thailand, coll. S.P. Huang, 23 November, 2012.

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