

Tonlesapia tsukawakii, a new genus and species of freshwater dragonet (Perciformes: Callionymidae) from Lake Tonle Sap, Cambodia

Hiroyuki Motomura* and Takahiko Mukai**

Tonlesapia tsukawakii, new genus and species, is described from the northwestern part of Lake Tonle Sap, Cambodia. *Tonlesapia tsukawakii* is distinguished from all known genera and species of Callionymidae by the following combination of characters: first dorsal fin absent; 8 dorsal-fin rays; 7 anal-fin rays; all dorsal- and anal-fin rays, except last ray, divided into 2 at base, unbranched; opercular membrane fused with body, not forming a free skin flap. *Tonlesapia tsukawakii* is the only species in the family which lives in freshwater throughout its entire life cycle.

Introduction

The family Callionymidae (Teleostei: Perciformes: Callionymidei), a large family of benthic fishes, comprises 182 recent valid species worldwide (Fricke, 2002). The family is characterized by having the gill opening much reduced, the preopercle with a strong spine, the opercle and subopercle without spines, the lateral line continuous on the body, the eyes oriented dorsally and no scales (Nelson, 1994; Randall, 1999). Although callionymid fishes are primarily marine, two euryhaline species are known to enter and live in freshwater rivers (Fricke, 1983; Nelson, 1994; Randall, 1999).

Since 1980, a number of taxonomic and sys-

tematic papers on Callionymidae have been published by Fricke, including a review of the Indo-Pacific species (Fricke, 1983) and a checklist of Callionymidae and Draconettidae, with a review of generic classification (Fricke, 2002). Over the same period, Nakabo (e.g. 1982, 1983, 2002) published a number of papers on generic classification and reviews of the Japanese and Korean members of Callionymidae, but without detailed reference to Fricke's classification. Although Nakabo (1982) recognized 19 genera in Callionymidae, Fricke (2002) recognized only 10. Because his generic classification was based on examinations of all known species in the Indo-Pacific and has been followed by many authors (e.g. Randall et al., 1990; Randall, 1999; Randall

* The Kagoshima University Museum, 1-21-30 Korimoto, Kagoshima 890-0065, Japan.
E-mail: motomura@kaum.kagoshima-u.ac.jp

** Faculty of Regional Studies, Gifu University, 1-1 Yanagito, Gifu 501-1193, Japan.
E-mail: tmukai@cc.gifu-u.ac.jp

& Earle, 2000; Hutchins, 2001; Allen & Adrim, 2003), Fricke's (1983, 2002) concept of callionymid genera is largely adopted here.

During a series of ichthyological surveys in Cambodia (e.g. Motomura et al., 2002), an unidentified callionymid, collected from the north-western part of Lake Tonle Sap, near Siem Reap, were purchased at a market in Phsar Krom, Siem Reap by the authors. These specimens were identified as a new genus and species, following comparisons with all known genera based mainly on characters previously regarded as generic differences in the family. The new genus and species is described here as *Tonlesapia tsukawakii*.

Material and methods

Specimens were fixed in 10 % formalin; small pieces of skeletal muscle were removed from the right side of the specimens for DNA analysis before fixation. Measurements followed Fricke (1983: 7-10), with the following additional measurements. Head width was measured between the preopercular spine bases. Postorbital length was measured from the posteriormost bony edge of the orbit to the gill opening. Dorsal- and anal-fin base lengths were measured from the origins of first rays to the posterior ends of the last ray bases. Longest dorsal- and anal-fin ray lengths were measured from the origins to tips of the longest rays. Terminology of snout length, orbit diameter and upper-jaw length used in this study are equivalent to preorbital length, eye diameter and maxillary length, respectively, defined by Fricke (1983). Measurements were made with needle-point calipers under a dissecting microscope, on the left side wherever possible. Standard and head lengths are abbreviated as SL and HL, respectively. In the description section, data for the holotype are presented first, followed by paratype data (if different) in parentheses. The last two soft rays of the dorsal and anal fins were counted as single rays, each pair being associated with a single pterygiophore. Osteological characters were confirmed from X-ray photos taken of all specimens. The sex of the types was established by dissection of the abdomen on the right side. The following institutional abbreviations are used in the text: AMS, Ichthyology, Australian Museum, Sydney; and CSIRO, Division of Marine Research, Commonwealth Science and Industrial

Research Organization, Hobart. A total DNA was extracted from 99 % ethanol preserved muscle tissue using the ChargeSwitch gDNA Mini Tissue Kit (Invitrogen, USA). Partial regions of the 12S ribosomal RNA (rRNA) and 16S rRNA genes (about 730 bp and 570 bp, respectively) were amplified and sequenced following Mukai et al. (2004).

Tonlesapia, new genus

Type species. *Tonlesapia tsukawakii*, new species.

Diagnosis. *Tonlesapia tsukawakii* differs from all other members of the family Callionymidae, except *Draculo mirabilis* (= *Eleutherochir mirabilis*), by the absence of the first dorsal fin (vs. presence). *Tonlesapia tsukawakii* differs from *Draculo mirabilis* in having 8 dorsal-fin rays (vs. 12-14 rays), 7 anal-fin rays (vs. 12-14 rays), all anal-fin rays, except the last, unbranched (vs. all rays branched) and the opercular membrane fused with the body (vs. not fused with body). *Tonlesapia* is distinguished from all other genera of the family by the following combination of characters: first dorsal fin absent; 8 dorsal-fin rays; 7 anal-fin rays; all dorsal- and anal-fin rays, except last ray divided into 2 at base, unbranched; first pelvic-fin soft ray not elongate, not separated from second soft ray; i 6 i or ii 6 ii caudal-fin rays; lower lip without papillae; snout length (19.2-22.9 % HL) shorter than orbit diameter (34.9-37.2 % HL); gill opening positioned dorsally; preopercular spine with an incurved tip, 5-7 inwardly directed spinules along inner side of main spine and without an antorse spine at base of main spine or spinules along its outward margin; all spinules on fully laid back preopercular spine fitting inside a groove on lateral surface of head; opercular membrane fused with body, not forming a free skin flap; supraocular tentacles and tentacles on lateral surface of body absent; lower lateral part of body without longitudinal skin fold; lateral line on trunk without transverse upward branches; lateral line of each side connected by a transverse branch across upper part of caudal peduncle, other dorsally transverse branches on body absent; lateral line not extending onto caudal-fin membrane; infraorbital sensory canals not extending below eyes; mandibular sensory canals absent.

Remarks. Fricke's (1983) review of Indo-Pacific species of Callionymidae recognized nine valid genera: *Anaora*, *Callionymus*, *Dactylopus*, *Diplogrammus*, *Draculo*, *Eleutherochir*, *Neosynchiropus*, *Paracallionymus*, and *Synchiropus*. Later, *Neosynchiropus* was considered as a junior synonym of *Synchiropus* by Fricke (2002). *Neosynchiropus* Nakabo, 1982 is an objective homonym of *Neosynchiropus* Nalbant, 1979 and thus invalid.

Tonlesapia is very similar to *Draculo* and *Eleutherochir* in overall body appearance, the three genera sharing a relatively poorly-developed first dorsal fin (0-4 short spines), compared with the well-developed first dorsal fin in other genera (Fricke, 1983). However, *Tonlesapia* clearly differs

from *Draculo* and *Eleutherochir* in having the opercular membrane fused with the body, and not forming a free flap of skin (Fig. 1; vs. membrane not fused with body and forming a free flap of skin in the latter genera). In addition, *Draculo* and *Eleutherochir* are characterized by having large and small papillae on the lower lip, respectively (Fricke, 1983), whereas *Tonlesapia* lacks such papillae (Fig. 2a). Nakabo (1982) synonymized *Draculo* with *Eleutherochir* because they shared the presence of papillae on the lower lip. Fricke (1983) regarded *Draculo* as valid, separating it from *Eleutherochir* by the distance between the eyes, dorsal- and caudal-fin structures, and the size of the papillae on the lower lip.

Table 1. Morphometric characters of *Tonlesapia tsukawakii*.

	holotype	paratypes			mean
	AMS I. 43624-001	AMS I. 43625-001	AMS I. 43625-002	CSIRO H 6249-01	
Standard length (mm)	33.6	29.5	29.7	25.4	
Total length (mm)	42.1	37.3	37.1	31.2	
Percentage of standard length					
Head length	25.1	24.9	24.9	23.6	24.6
Head width	20.9	23.5	22.5	21.1	22.0
Body depth	14.8	14.5	15.0	14.3	14.7
Body width	20.3	20.8	21.5	18.1	20.2
Snout length	5.8	5.6	5.2	4.5	5.3
Orbit diameter	9.2	8.8	8.7	8.8	8.9
Interorbital width	2.0	2.0	1.9	1.7	1.9
Postorbital length	10.8	9.8	10.6	10.2	10.3
Upper-jaw length	6.4	6.6	6.7	6.3	6.5
Preopercular spine length	10.0	11.3	11.8	10.1	10.8
Predorsal-fin length	47.9	48.7	50.9	46.0	48.4
Prepelvic-fin length	27.2	26.8	25.1	24.7	25.9
Preanal-fin length	53.6	50.1	54.4	52.9	52.8
Dorsal-fin base length	25.9	26.1	27.6	26.5	26.5
Anal-fin base length	25.8	26.1	27.5	27.4	26.7
Pectoral-fin length	20.2	21.6	21.9	19.9	20.9
Longest dorsal-fin ray length	12.0	13.3	11.4	11.8	12.1
Longest pelvic-fin length	23.4	25.0	24.8	23.3	24.1
Longest anal-fin ray length	10.3	7.9	10.5	9.1	9.4
Caudal-peduncle length	19.1	17.0	18.1	18.7	18.2
Caudal-peduncle depth	6.3	5.8	5.7	5.1	5.7
Caudal-fin length	24.5	23.5	26.0	25.0	24.7
Percentage of head length					
Head width	83.3	94.6	90.4	89.3	89.4
Snout length	22.9	22.3	21.0	19.2	21.4
Orbit diameter	36.6	35.4	34.9	37.2	36.0
Interorbital width	7.8	7.9	7.7	7.0	7.6
Postorbital length	43.1	39.2	42.6	43.2	42.0
Upper-jaw length	25.5	26.4	27.1	26.7	26.4
Preopercular spine length	39.8	45.6	47.4	42.8	43.9



Fig. 1. *Tonlesapia tsukawakii*, holotype, AMS I. 43624-001, 33.6 mm SL; Cambodia: Lake Tonle Sap. a, lateral view; b, dorsal view; c, ventral view.

Tonlesapia tsukawakii and *Draculo mirabilis* (*Eleutherochir mirabilis* in Nakabo, 1982) are the only species in the family lacking the first dorsal fin (Fricke, 1983; Randall, 1999), all other Indo-Pacific callionymids, including other members of *Draculo*, having two separate dorsal fins. In addition, *T. tsukawakii* differs from *D. mirabilis* in having 8 dorsal-fin rays (vs. 12-14 rays in the latter), 7 anal-fin rays (vs. 12-14), all anal-fin rays, except the last, unbranched (vs. all rays branched) and the infraorbital sensory canals not extending below the eye (vs. extending well below). Data on *D. mirabilis* are based on Fricke (1983: 538-540) and Nakabo (1983: 252-253). Furthermore, *D. mirabilis* is distributed in cold temperate marine waters, where it ranges from northern Japan to the Yellow Sea and Pohai Sea (Nakabo, 2002), whereas *T. tsukawakii* occurs in tropical freshwater (Lake Tonle Sap, Cambodia).

In addition to the absence of the first dorsal fin, *Tonlesapia* can be easily distinguished from *Diplogrammus* by lacking the longitudinal skin fold along the lower lateral surface of the body (Fig. 1; vs. uniquely present in the latter; Fricke,

1983, 2000); from *Anaora* by lacking tentacles on the lateral surface of the body (Fig. 1; vs. many being uniquely present in the latter); from *Paracallionymus* by having a simple lateral line on the trunk (vs. a unique arrangement of many long, upward transverse branches in the latter); and from *Dactylopus* by having the first pelvic-fin soft ray neither elongate nor separated from the second soft ray, both conditions occurring uniquely in *Dactylopus*.

Continuing differences of opinion regarding the generic/subgeneric status of species included in *Callionymus* and *Synchiropus* by Fricke (1983, 2000, 2002), Fricke & Zaiser (1983) and Zaiser & Fricke (1985), and Nakabo (1982, 1987, 2002) and Nakabo et al. (1987), have resulted in stalemate, still requiring resolution. *Tonlesapia* clearly differs from Fricke's (1983, 2000) *Callionymus* and *Synchiropus* and Nakabo's (1982) 14 genera, which were reassigned to the two genera by Fricke (1983, 2000, 2002), in having the following combination of characters: first dorsal fin absent; dorsal-fin (equivalent to the second dorsal fin in other genera) and anal-fin rays, except posteriormost,

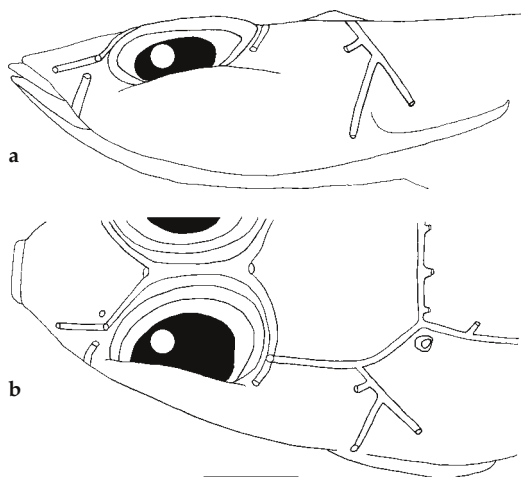


Fig. 2. *Tonlesapia tsukawakii*, holotype, AMSI. 43624-001, 33.6 mm SL; sensory pores and canals of cephalic lateralis system. **a**, lateral view of head; **b**, dorsal view of head. Scale bar 2 mm.

unbranched; gill opening a small dorsally-positioned hole (Fig. 2b); snout length (19.2-22.9 % HL) shorter than orbit diameter (34.9-37.2 % HL); preopercular spine with an incurved tip, 5-7 inwardly directed spinules along the inner side of the main spine and without an antrorse spine at the base of the main spine (Fig. 3); all spinules on the fully laid back preopercular spine fitting inside a groove on the lateral surface of the head (Fig. 2); lateral line of each side connected by a transverse branch across the upper part of the caudal peduncle, other dorsally transverse branches on body absent; lateral line not extending onto the caudal-fin membrane; infraorbital sensory canals not extending to below the eyes (Fig. 2a); and mandibular sensory canals absent (Fig. 2a).

In addition to the above-mentioned genera occurring in the Indo-Pacific, two genera (*Chalinops*, *Protogrammus*) have been recognized that are distributed only in the Atlantic Ocean (Fricke, 1985, 2002; *Protogrammus* also occurs in the Southwest Pacific, R. Fricke, pers. comm.). Although Nakabo (1982) synonymized *Chalinops* with *Diplogrammus*, Fricke (2002) regarded it as valid. In either case, *Tonlesapia* can be distinguished from these three nominal genera by the lack of a longitudinal skin fold along the lower lateral surface of the body (vs. presence).

Etymology. *Tonlesapia* is named for Lake Tonle Sap in Cambodia. Gender feminine.

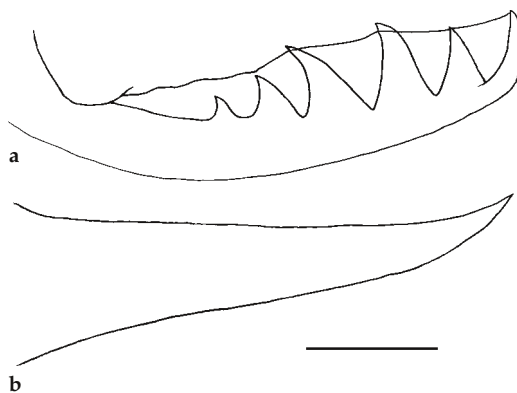


Fig. 3. *Tonlesapia tsukawakii*, holotype, AMSI. 43624-001, 33.6 mm SL; preopercular spine. **a**, dorsal view; **b**, lateral view. Note: most spinules along main spine covered with thin translucent membrane, and all spinules fitting inside a groove on lateral surface of head when preopercular spine fully laid back. Scale bar 1 mm.

Tonlesapia tsukawakii, new species
(Tonle Sap dragonet)
(Figs. 1-3)

Holotype. AMSI. 43624-001, mature male, 33.6 mm SL; Cambodia: northwestern part of Lake Tonle Sap, near Siem Reap; water depth less than 1 m; purchased at a market in Phsar Krom, Siem Reap; T. Mukai & H. Motomura, 28 May 2005.

Paratypes. AMS I. 43625-001, mature male, 29.5 mm SL; AMS I. 43625-002, mature male, 29.7 mm SL; CSIRO H 6249-01, probable male, 25.4 mm SL; same data as holotype, 30 May 2005.

Diagnosis. Characters differentiating the taxon are listed in generic diagnosis.

Description. Proportional measurements are given in Table 1. First dorsal fin absent. Dorsal fin with 8 rays; all rays, except last ray, unbranched; last ray divided into 2 at base, each branch not divided at tip; second (first in one paratype) ray longest, but first to third rays subequal in length; third to seventh rays progressively shorter; eighth ray slightly longer than seventh ray; lengths of rays less than body depth; inter-ray membrane strongly incised, not reaching tips of rays, ray tips forming short filaments; posterior branch of last ray not joined by mem-

brane to dorsal caudal peduncle; base of fin not covered with fleshy skin.

Anal fin with 7 rays; all rays, except last ray, unbranched; last ray divided into 2 at base, then each branch divided into 2 at tip; second (third in two paratypes) ray longest, but second to last rays subequal length; first ray shortest; lengths of rays less than body depth and length of longest dorsal-fin ray; inter-rayed membrane strongly incised, not reaching tips of rays, ray tips forming short filaments; posterior branch of last ray not joined by membrane to ventral caudal peduncle; base of fin not covered with fleshy skin.

Pectoral fin with 16 rays; uppermost ray and lower 6 (5 or 6) rays unbranched, remaining rays branched; sixth ray longest, sixth to lowermost rays progressively shorter; length of fin slightly shorter than head length, but greater than body depth; inter-rayed membrane slightly incised, but reaching near ray tips; base of fin covered with fleshy skin.

Pelvic fin with 1 soft spine and 5 rays; all rays branched, divided into 5 or 6 branches distally; length of spine shorter than that of first ray; first ray shortest, about half length of fourth ray; first to fourth progressively longer; fourth ray longest, subequal to head length; fifth ray longer than first to third rays; inter-ray membrane weakly incised, ray tips not forming filaments; posterior margin of last ray broadly joined by membrane to pectoral-fin base; base of fin covered with fleshy skin.

Caudal fin with 6 branched rays medially; 1 (2 in two paratypes) dorsal series and 1 (2 in two paratypes) ventral series of segmented unbranched rays, each series located between branched and procurrent rays; 2 (1 in one paratype) dorsal series and 2 (1 in one paratype) ventral series of procurrent rays; inter-ray membrane weakly incised, tips of branched rays forming short filaments; posterior margin of fin rounded. 20 vertebrae. Swimbladder absent.

Body elongate and depressed; highest body depth between dorsal-fin origin and above pectoral-fin base; highest body depth less than body width at pectoral-fin base, depth 1.7 (1.6-1.7, mean 1.7) in head length; body depth progressively narrower from dorsal-fin origin to caudal-fin base; caudal-peduncle depth 3.0 (3.0-3.7, mean 3.2) in caudal-peduncle length. Head strongly depressed; head wide in dorsal view, head width at base of preopercular spine 1.2 (1.1, mean 1.1) in head

length; dorsal profile of head nearly straight. Mouth small, protrusible, subterminal; posterior margin of maxilla just reaching a vertical through anterior margin of orbit; upper-jaw length 3.9 (3.7-3.8, mean 3.8) in head length, 1.4 (1.3-1.4, mean 1.4) in orbit diameter; lower jaw lacking a symphyseal knob; lips on upper and lower jaws well developed, upper lip covering entire maxilla; lateral surface of maxilla without ridges or spines; lower lip lacking fleshy papillae. Symphyseal gap between premaxillary teeth bands absent; upper jaw with a band of villiform teeth; all teeth directed inward. Lower jaw with a band of short, conical teeth; teeth directed slightly inward; lengths of most teeth longer than those of upper jaw; tooth band of lower jaw wider than that of upper jaw; underside of lower jaw smooth, without ridges. Vomerine and palatine plates without teeth.

A single nostril on each side of snout, located at just front of anterior margin of orbit; nostril opening oval, without a short membranous tube or tentacle. Eye large, oriented dorsally; orbit diameter 2.7 (2.7-2.9, mean 2.8) in head length, 1.2 (1.1-1.2, mean 1.2) in postorbital length; orbit diameter greater than 1.5 times of snout length. Interorbital space very narrow, nearly flat; width less than pupil diameter, and snout and upper-jaw lengths; interorbital width 4.7 (4.5-5.3, mean 4.7) in orbit diameter. Occipital region with bump of parietal bone on each side; distance between parietal bumps more than twice interorbital width; parietal bump located at midway between posterior margin of pupil and gill opening. Opercle without a free flap of skin; opercular membrane fused with body. Gill opening a small hole with a short membranous tube; gill opening located above opercle, directed upward. Preopercular spine long, strongly developed; its length greater than orbit diameter; length 2.5 (2.1-2.3, mean 2.3) in head length; preopercular spine with an incurved tip and 5 and 6 (5 and 6, 6 and 6, or 6 and 7) inwardly directed spinules on left and right sides of head, respectively; main tip and spinules occurring along inner side of preopercular spine; main tip and spinules inwardly directed and tips antorsely curved; all spinules fitting inside a groove on lateral surface of head when preopercular spine fully laid back; thin skin completely covering all spinules; base and outside of main preopercular spine smooth, without spines.

Lateral line extending to caudal-fin base, not extending to caudal-fin membrane; lateral line of each side connected by a transverse branch across posterior margin of occiput and one upper part of caudal peduncle. Sensory pores and canals of cephalic lateralis system poorly developed, indistinct (Fig. 2); mandibular sensory canals absent; infraorbital sensory canals not extending below eyes. No supraocular tentacles; no tentacles on lateral surface of body. Urogenital papilla small, not elongate.

Posterior tip of preopercular spine extending beyond a vertical through base of third pelvic-fin soft rays. Posterior tip of pectoral fin extending well beyond a vertical through anus and origin of dorsal fin. Posterior tip of depressed pelvic fin just reaching anus. Origin of dorsal fin anterior to that of anal fin; posterior end of dorsal-fin base anterior to that of anal-fin base.

Color in fresh. Based on color transparencies of holotype (AMS I. 43624-001): Upper half of head and body deep reddish orange with scattered deep brown blotches averaging half of pupil diameter in size. Longitudinal deep brown blotches along lateral line. A series of blackish blotches, averaging pupil diameter in size, below lateral line on lateral surface of body. Lower one-third of head and body dull yellowish orange. Dorsal-fin rays yellowish orange; membranes transparent. Pectoral- and anal-fin rays and membranes yellowish orange with poorly defined blackish blotches. Anal-fin rays white; membranes transparent. Caudal-fin rays and membranes white and transparent, respectively, with yellowish orange basally and upper half of fin.

Color of preserved specimens. Upper half of head and body grayish-white with scattered blackish blotches averaging one-third of pupil diameter in size, blotches extending onto lateral line. A series of blotches, averaging pupil diameter in size, below lateral line on lateral surface of body, each blotch formed from a cluster of scattered large melanophores. Underside of head, including upper and lower jaws white, without melanophores. Abdomen yellow, other part of ventral surface of body white, without melanophores. Indistinct small melanophores on dorsal-fin rays, and on membranes along anterior edge of rays, remaining parts of membranes transparent. Pectoral- and anal-fin rays white with scattered indistinct melanophores; membranes trans-

parent without melanophores. Anal-fin membranes and rays white, without melanophores. Caudal-fin rays and membranes white and transparent, respectively, with a small number of scattered melanophores.

Molecular data. Partial nucleotide sequences of mitochondrial 12S and 16S rRNA gene were determined from the four specimens (Appendixes). The nucleotide sequences from holotype were deposited in GenBank (<http://www.insdc.org/>), EMBL (<http://www.ebi.ac.uk/embl/>) and DDBJ (<http://www.ddbj.nig.ac.jp/>) as accession AB236192 and AB236193. Those from paratypes were accession No. AB236194-AB236198. A few genetic variations among the four specimens were observed. There were three nucleotide substitutions in 12S rRNA gene (pair-wised sequence differences were 0-0.4 %) (Appendix 1), and four nucleotide substitutions and two insertions (or deletions) in 16S rRNA gene (pair-wised sequence differences, except inserted position, were 0-0.7 %) (Appendix 2).

Distribution. Currently known only from the northwestern part of Lake Tonle Sap, Cambodia.

Ecological notes. The type specimens of *T. tsukawakii* were collected by small nets by a local fisherman in Siem Reap with the following species of fishes (all registered at AMS collection): *Coilia lindmani* (Engraulidae), *Clupeoides borneensis* (Clupeidae), *Albulichthys albuloides*, *Cyclocheilichthys armatus*, *Labiobarbus leptocheila*, *Thyrsocypris tonlesapensis* (Cyprinidae), *Parambassis siamensis*, *Parambassis wolffii* (Ambassidae), *Polynemus aquilonaris* (Polynemidae), *Boesemania microlepis* (Sciænidae), *Glossogobius aureus* (Gobiidae), and *Hyporhamphus limbatus* (Hemiramphidae). Of these, the dominant species were *Coilia lindmani* and *Polynemus aquilonaris*, accounting for close to 90 % of the whole catch. These two species occur offshore and have never been observed inshore, nor in channels or ponds scattered around the lake. This suggests that *T. tsukawakii* was likely collected offshore too. The substrate of the lake is mud.

Etymology. Named for Dr. Shinji Tsukawaki of Kanazawa University, in acknowledgment of his kind and invaluable assistance during our ichthyological surveys in Cambodia.

Discussion

Dissections of the abdomen on the right side of all type specimens of *T. tsukawakii* showed that the three larger specimens, including the holotype, had remarkably expanded gonads, indicating that it is a small-sized species (the smallest probable mature male was 29.5 mm SL). The ventral surface of the abdomen in the three specimens was expanded by the well-developed gonads, the yellowish color of the latter being discernible through the white abdominal skin (see Fig. 1c). Sexual dimorphism of the species is unknown because all of the adult specimens examined in this study were males.

Although Lake Tonle Sap is presently connected to the South China Sea through the Tonle Sap and Mekong Rivers, the collection locality of *T. tsukawakii* at northwestern extremity of the lake is over 500 km distant from the sea. The specimens of *T. tsukawakii* collected had well-developed, expanded gonads, suggesting that the species reproduces in the lake. They also have simplified fin ray structures, including unbranched dorsal and anal-fin rays and lack of the first dorsal fin, and a strongly depressed head and body, suggesting that the species does not have the strong swimming ability to migrate to the sea. Accordingly, *T. tsukawakii* is most likely to live in freshwater throughout its entire life cycle, although all other Indo-Pacific species of Callionymidae, except for two euryhaline species (Randall, 1999), live in marine waters.

Acknowledgments

We are specially indebted to S. Tsukawaki (Kanazawa University) for his kind assistance at various stages during our fieldwork and making all arrangements for our stay in Cambodia for over 5 years. The first author wishes to thank all staff of the Fish Section, Australian Museum, Sydney and Y. Motomura for their generous support during this study, C. Bent (AMS) for taking photographs of the holotype, and G. S. Hardy (Ngunguru, New Zealand) for reviewing the manuscript. Material was collected under the Evaluation of Mechanisms Sustaining the Biodiversity of Lake Tonle Sap Programme (Japan Society for the Promotion of Science, Grant-in-aid for International Scientific Research, no. 15405004, 2003-2005) and UNESCO Japanese Funds-in-Trust (MAB-IHP Joint Programme). We thank members of the programme, A. Ohtaka, T. Narita, H. Katakura, T. Ishikawa, Y. Araki, H. Oyagi and K. Sakai, for their

assistance during our fieldwork and S. Sieng (Ministry of Industry, Mines and Energy, Cambodia) and T. Murayama (JICA, Phnom Penh) for their help with logistics. The Authority for the Protection and Management of Angkor and the Region of Siem Reap, Cambodia, provided permission for fish sampling in Siem Reap area.

Literature cited

- Allen, G. R. & M. Adrim. 2003. Coral reef fishes of Indonesia. *Zoological Studies*, 42: 1-72.
- Fricke, R. 1983. Revision of the Indo-Pacific genera and species of the dragonet family Callionymidae (Teleostei). Cramer, Braunschweig, x + 774 pp.
- 1985. *Protogrammus*, a new genus of callionymid fishes, with a redescription of *P. sousai* from the eastern Atlantic. *Japanese Journal of Ichthyology*, 32: 294-298.
- 2000. Callionymidae of New Caledonia, with remarks on related species and descriptions of 10 new species from New Caledonia, Australia, New Guinea, and Hawaii (Teleostei). *Stuttgarter Beiträge zur Naturkunde, Serie A (Biologie)*, 617: 1-81.
- 2002. Annotated checklist of the dragonet families Callionymidae and Draconettidae (Teleostei: Callionymoidei), with comments on callionymid fish classification. *Stuttgarter Beiträge zur Naturkunde, Serie A (Biologie)*, 645: 1-103.
- Fricke, R. & M. J. Zaiser. 1983. A new callionymid fish, *Synchiropus kiyoeae*, from the Izu Islands, Japan. *Japanese Journal of Ichthyology*, 30: 122-128.
- Hutchins, J. B. 2001. Checklist of the fishes of Western Australia. Records of the Western Australian Museum, Supplement, 63: 9-50.
- Motomura, H., S. Tsukawaki & T. Kamiya. 2002. A preliminary survey of the fishes of Lake Tonle Sap near Siem Reap, Cambodia. *Bulletin of the National Science Museum, Tokyo, Series A (Zoology)*, 28: 233-246.
- Mukai, T., T. Suzuki & M. Nishida. 2004. Genetic and geographical differentiation of *Pandaka* gobies in Japan. *Ichthyological Research*, 51: 222-227.
- Nakabo, T. 1982. Revision of genera of the dragonets (Pisces: Callionymidae). *Publications of the Seto Marine Biological Laboratory*, 27: 77-131.
- 1983. Revision of the dragonets (Pisces: Callionymidae) found in the waters of Japan. *Publications of the Seto Marine Biological Laboratory*, 27: 193-259.
- 1987. A new species of the genus *Foetorepus* (Callionymidae) from southern Japan with a revised key to the Japanese species of the genus. *Japanese Journal of Ichthyology*, 33: 335-341.
- 2002. Callionymidae. Pp. 1125-1138 in T. Nakabo (ed.), *Fishes of Japan with pictorial keys to the species*, English edition. Tokai University Press, Tokyo.

- Nakabo, T., S.-R. Jeon & S.-Z. Li. 1987. A new species of the genus *Repomucenus* (Callionymidae) from the Yellow Sea. Japanese Journal of Ichthyology, 34: 286-290.
- Nelson, J. S. 1994. Fishes of the World. Third edition. Wiley, New York, xvii + 600 pp.
- Randall, J. E. 1999. Review of the dragonets (Pisces: Callionymidae) of the Hawaiian Islands, with descriptions of two new species. Pacific Science, 53: 185-207.
- Randall, J. E., G. R. Allen & R. C. Steene. 1990. Fishes of the Great Barrier Reef and Coral Sea. Crawford House Press, Bathurst, xx + 507 pp.
- Randall, J. E. & J. L. Earle. 2000. Annotated checklist of the shore fishes of the Marquesas Islands. Occasional Papers, Bernice P. Bishop Museum, 66: 1-39.
- Zaiser, M. J. & R. Fricke. 1985. *Synchiropus moyeri*, a new species of dragonet (Callionymidae) from Miyakejima, Japan. Japanese Journal of Ichthyology, 31: 389-397.

Received 13 October 2005
 Revised 21 December 2005
 Accepted 27 December 2005

Appendix 1. Aligned DNA sequences from 743 bp of the 12S rRNA genes. Identity with first sequence denoted by dots.

AMS I 43624-001	TTGCCAGCCACCGCGTTATACGAGCGACCCAAATTGAAAGAAAACGGCGTAAAGGGTGGTTATAAATTTTTTAAA
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	AATAGAGCCGAACCTTACTCCTGACTGTTATACGTTACGAAATAAAGAAGAACAATACGAAAGTAGCTCTAAATA
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	TTTGAACCCACGAAAGTTAGGAACAACACTGGGATTAGATACCCCACTATGCCTAACCATAAACCCTGATAATTT
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	AATACATCCATTATCCGCCAGGGTACTACGAGCGTAAGCTTAAAACCCAAAGGACTTGGCGGTGCTTTAGACCCC
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	CCTAGAGGAGCCTGTCTATAACCGATAATCCCGTTAAACCTCACCTCTTTTGCCAAATCCGCCTATATACCA
AMS I 43625-001
AMS I 43625-002C.....
CSIRO H 6249-01
AMS I 43624-001	CCGTCGTAGCTTACCCCTTGAAGGCTTACTAGTAAGCATAATTGGTTTAAACCTAACGTCAGGTCGAGGTGCA
AMS I 43625-001T.....
AMS I 43625-002T.....
CSIRO H 6249-01T.....
AMS I 43624-001	GTGAATAAGAGGGGAAGAAATGGGCTACATTTACTAAAAGTAAATACGAAAATGTACTGAAATGTACTTTA
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	AAAGGAGGATTTAGTAGTAAGGAATGAATAGAGTGCATTTCTGAAACTGGCCCTGAAGCGCGCACACCCGCCCG
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	TCACTCTCCCCACAAAATAAACTCAGACTTCCATAAAACCAAAAACATATTTATTAGGGGAGGCAAGTCGTAACA
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	TGGTAAGCGTACCGGAAGGTGCTCTTGAAAACAGAGTGTAGCTAAATAGTAAGCCTCTCTCTTAC
AMS I 43625-001C.....
AMS I 43625-002C.....
CSIRO H 6249-01C.....

Appendix 2. Aligned DNA sequences from 579 bp of the 16S rRNA genes. Identity with first sequence denoted by dots. Insertions/deletions of specific nucleotides indicated by dashes.

AMS I 43624-001	AAACATCGCCTCTTGTACCAATTACATAAGAGGTCGCCGCTGCCAGTGACTAAATGTTTAACGGCCGCGGTATT
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01C.....
AMS I 43624-001	TTAACCGTGCAAAGGTAGCGTAATCACTTGTCTTTTAAATGAAGACCTGTATGAATGGCATCACGAGGGCTTAAC
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	TGTCTCCTTTTCTAATCAATTAATTGATCTCCCGTGCAGAAGCTGGGATTTAACATAAGACGAGAAGACCC
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	TATGGAGCTTTAAACATATCAAGCATACGTCAAGTACTTTAAAATAAAAAATGAACTAAGTATCTTTCTTGTA
AMS I 43625-001
AMS I 43625-002--.....
CSIRO H 6249-01A.....A.....
AMS I 43624-001	TTGTTTTGGTTGGGGCGACCACGGAAGAATAAATATCTCCCATGCTAAAAAGCACTAATGCTTTTAAACTAAG
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	ACCACCAAGTCTAAGTCATAGAAACTTAAAAATGATCCGGCCAGCCGATCAACGAAACGAGTTACCCTAGGGAT
AMS I 43625-001G.....
AMS I 43625-002
CSIRO H 6249-01G.....
AMS I 43624-001	AACAGCGCAATCCCCTTTAAGAGCCCTTATCAACAAGGGGTTTACGACCTCGATGTTGGATCAGGACATCCTAA
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01
AMS I 43624-001	TGGTGTAGCCGTTATTAAGGGTTCGTTTGTTCACGATTAAGTCCTACGTGAT
AMS I 43625-001
AMS I 43625-002
CSIRO H 6249-01