

## Review of *Schismatogobius* (Gobiidae) from Japan, with the description of a new species

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**Abstract** Three species of *Schismatogobius* de Beaufort 1912, distinguished by their morphology and mitochondrial DNA sequences, were found in freshwater streams in the Ryukyu Archipelago, Japan. Although two species were previously known from Japan (*S. roxasi* Herre 1936 and *S. ampluvinculus* Chen, Shao, and Fang 1995), the taxonomy needs to be revised. To identify these species, the holotype morphology of *S. marmoratus* (Peters 1868), *S. bruyneri* de Beaufort 1912, and *S. roxasi*, originally described from the Philippines and Indonesia, were examined and re-described here, because relatively little information about their diagnostic characters was provided in the original descriptions. The three Japanese species were identified as *S. ampluvinculus*, *S. marmoratus*, and a new species. They were

distinguished from each other and from their congeners by the banding pattern of the body, markings on the pectoral fins, pigment patterns on ventral surface of the head and pelvic fin, body depth at the pelvic-fin origin, pre-anal length, and pectoral-fin ray counts. Although the new species had been regarded as *S. roxasi* in previous publications, we show that it is actually not *S. roxasi* and that it also differs from all other nominal species of *Schismatogobius*. This is described as a new species, *S. ninja*. Additionally, this is the first record of *S. marmoratus* from Japan.

**Keywords** *Schismatogobius* • Gobiidae • new species • mitochondrial genome • Ryukyu Archipelago

## **Introduction**

*Schismatogobius* de Beaufort 1912 is a genus of small, scaleless gobies, primarily found on sand, pebbles, and gravel substrates of tropical and subtropical, fast-flowing, freshwater streams (Jenkins and Boseto 2005). They are distributed in the Indo-Pacific from southern India to Samoa (Arunachalam et al. 2014; Keith et al. 2017). Details of their life histories are unknown, but they are believed to be amphidromous (Chen et al. 1995a, 2001; Maeda 2016). On Okinawa Island, in southern Japan, a small juvenile (9.8 mm SL) *Schismatogobius ampluvinculus* Chen, Shao, and Fang 1995 was captured (Maeda 2014) in a trap in a fishway at a weir just above the upper limit of tidal fluctuations. The trap was specifically set to collect ascending juveniles (Maeda unpublished data). Species of *Schismatogobius* display sexual dimorphism in regard to jaw length. Males have remarkably larger jaws than females, although lengths of the fins do not differ between males and females (Keith et al. 2004).

In total, 16 species are recognized in this genus, including seven species described in a recent review of this genus from Papua New Guinea to Samoa (Keith et al. 2017). Five species are known from islands in Southeast and East Asian regions (Japan, Taiwan, Philippines, and Indonesia): *Schismatogobius marmoratus* (Peters 1868), *Schismatogobius bruynisi* de Beaufort 1912, *Schismatogobius insignis* (Herre 1927), *Schismatogobius roxasi* Herre 1936, and *Schismatogobius ampluvinculus*. Four of these five species were described more than 80 years ago, and the diagnostic characters that are necessary to distinguish between species of *Schismatogobius*, such as the number of pectoral-fin rays, pigment patterns on the ventral surface of the head and pelvic fin, and markings on pectoral fins (Keith et al. 2017), were not provided. Consequently, these species have never been fully compared with others in the genus. Although Keith et al. (2017) provided dorsal-, anal-, and pectoral-fin ray counts, morphomeristic values, and the pigment patterns on ventral surfaces of the head and pelvic fin of *S. bruynisi*, they did not describe the morphology of the holotype, nor did they explain why they identified their material as *S. bruynisi*. Thus, examination of the type specimens of all these nominal species was necessary in order to review species of *Schismatogobius* from Japan.

Two species have been reported in Japan, recognized as *S. roxasi* and *S. ampluvinculus* (see Nakabo 2013). They were distinguished only on the basis of coloration of the body and fins (Nakabo 2013), even though coloration of these gobies varies among individuals and depends upon the environmental conditions. Therefore, identification is difficult and they are often misidentified. The Ryukyu Archipelago has the only suitable habitat for this genus in Japan (Nakabo 2013).

We collected three species of *Schismatogobius* in the Ryukyu Archipelago. Because some individuals had indeterminate markings on their bodies and fins, we identified them using their mitochondrial genome sequences as well as morphology. We also investigated

morphology of holotypes for three old species, *Gobiosoma marmoratum* Peters 1868 (= *S. marmoratus*), *S. bruynisi*, and *S. roxasi*. Because syntypes of another old species, *Gobiosoma insignum* Herre 1927 (= *S. insignus*), were destroyed at the Bureau of Science in Manila during World War II (Pietsch and Anderson 1997; Kottelat 2013), we compared characters of this species described by Herre (1927) with those of the other species. In this paper, we describe the morphology of the holotypes of the old species and we review the taxonomy of *Schismatogobius* from Japan with a description of one new species.

## **Materials and methods**

*Collection of samples.* Forty-five specimens of *Schismatogobius* were collected on Okinawa and Iriomote Islands in the Ryukyu Archipelago, using a hand net. After being anesthetized with 2-phenoxyethanol, their right pectoral fins were cut off and preserved in 99.5% ethanol for mitochondrial DNA analysis, and then gobies (without their right pectoral fins) were fixed in 10% formalin and preserved in 70% ethanol (except for one specimen, the entire body of which was fixed in 99.5% ethanol; URM-P 48699. It was used for mitochondrial DNA analysis, but not for morphological examination, as it had shrunk in 99.5% ethanol).

*Morphological examination.* Measurements and counts were taken from the left side of each fish. Measurements were made point-to-point to the nearest 0.1 mm using a vernier caliper or a divider under a stereomicroscope, and were expressed as a percentage of standard length (SL). Measurements and counts followed Nakabo (2002), with the following modifications: body depths were measured at the pelvic- and anal-fin origins; pre-anal-fin length was measured from tip of the snout to the origin of the anal fin, in addition to the pre-anal length (distance from the tip of the snout to the center of the anus). The first and second dorsal- and anal-fin lengths were measured from the origin of each fin to the farthest

point when the fin was adpressed. Caudal-fin length was measured from the midpoint of the caudal-fin base to the posteriormost part of the caudal fin. Symbolic codes used to represent collections and institutions follow Sabaj (2016).

In addition to the 45 Japanese specimens used for descriptions and/or mitochondrial DNA analysis, 54 specimens of *Schismatogobius* were examined to verify the taxonomy of Japanese species and to obtain distributional information; these 54 included specimens from URM and YCM collections and the holotypes of *G. marmoratum* (= *S. marmoratus*), *S. bruynisi*, and *S. roxasi*. Japanese specimens are listed in the “Taxonomy of Japanese species” section. Other materials are listed below:

*Comparative material. Schismatogobius bruynisi*: ZMA 111196, holotype, 31.6 mm SL, West Seram, Indonesia, 24 Feb. 1910.

*Schismatogobius marmoratus*: ZMB 6756, holotype, 33.3 mm SL, Samar Island, Philippines, July to Sep. 1860.

*Schismatogobius insignis*: CAS-SU 30968, holotype of *S. roxasi*, 45.9 mm SL, Panay Island, Philippines, Feb. 1926; URM-P 31459, 37.9 mm SL, Mindanao Island, Philippines, 25 July 1985; URM-P 31689, 21.6 mm SL, Mindoro Island, Philippines, 15 Aug. 1985.

*Mitochondrial DNA analysis.* Total genomic DNA of 45 specimens of *Schismatogobius* and three specimens of *Kraemeria cunicularia* Rofen 1958 (as an outgroup taxon) was extracted from right pectoral fins preserved in 99.5% ethanol, using DNeasy Blood & Tissue Kit (Quiagen, Hilden, Germany) or Maxwell RSC Blood DNA Kit (Promega, Fitchburg, Wisconsin, USA).

A whole genome shotgun sequencing library was prepared using a KAPA HyperPlus Kit, PCR-free (KAPA Biosystems, Wilmington, Massachusetts, USA). Extracted genomic DNA was enzymatically fragmented into pieces of 200–1000 bp. After repairing protruding ends and A-tailing, sequencing adaptors were ligated onto both ends of DNA fragments. Shotgun

libraries were then sequenced on an Illumina HiSeq 2500 sequencer (Illumina, San Diego, California, USA) in Rapid Run mode version 2 using a HiSeq Rapid Cluster Kit v2 – Paired-End (Illumina) and HiSeq Rapid SBS Kit v2 (Illumina) following manufacturer’s instructions.

Sequencing data from each library were assembled with IDBA\_UD assembler version 1.1.1 (Peng et al. 2012) with different kmer lengths (60, 80, 100). Identification of complete mitochondrial genomes from assembled contigs was performed by 1) comparing them with the complete *Stiphodon alcedo* mitochondrial genome (accession: AB613000.1) (BLASTN e-value  $\leq 1e-100$ ), and by 2) confirming that 100 bp of both head and tail DNA sequences of a contig were identical, indicating that the sequence was circular. Complete mitochondrial genomes were aligned using MAFFT v7.244 (Katoh and Standley 2013) and all positions with gaps were removed using trimAl (Capella-Gutierrez et al. 2009). We performed maximum likelihood (ML) analysis using RAxML version 8.2.3 (Stamatakis 2014) with 100 bootstrap replicates. All sequenced raw data is available in the DDBJ Sequence Read Archive under BioProject accession number PRJDB5763. The assembled mitochondrial genome sequences with gene annotations are available in the DDBJ under accession numbers: AP018053–AP018100. Accession numbers for each individual are shown in Table S1.

Collection data for outgroup specimens are as follows: *Kraemeria cunicularia* Rofen 1958, URM-P 48722–48724, 3 specimens, Tsuken Island in Okinawa Islands, Ryukyu Archipelago, Japan, 22 Sept. 2015, coll. K. Maeda. Nelson et al. (2016) recently included the genus *Kraemeria* into the family Gobiidae, although it formerly belonged to the family Kraemeriidae. Because *Kraemeria* is the most closely related taxon to the genus *Schismatogobius* among 40 goby genera analyzed in our preliminary research on phylogeny of gobies using mitochondrial genome sequences (Maeda et al. unpublished data), it was used as an outgroup taxon in the present study.

## Results

**Molecular phylogenetic analysis.** In the ML phylogenetic tree using the aligned 16,177 bp of mitochondrial genomes (Fig. 1), 45 specimens of *Schismatogobius* collected from the Ryukyu Archipelago were divided into three clades with high bootstrap supports. They are identified as *S. ampluvinculus*, *S. marmoratus*, and *S. ninja* sp. nov., respectively, and their morphology is described below. Among the three species, *S. marmoratus* and *S. ninja* sp. nov. are sister species and *S. ampluvinculus* is placed in a basal position. Although specimens of *S. ampluvinculus* and *S. ninja* sp. nov. were both collected in Okinawa and Iriomote Islands (located more than 400 km apart), the two species did not form geographic subclades (Fig. 1).

**Holotype morphology of *S. marmoratus*, *S. bruynisi*, and *S. roxasi*.** Counts and measurements are provided in Table 1.

Holotype of *Gobiosoma marmoratum* Peters 1868 (ZMB 6756, Loquilócum, Samar, Philippines, July to Sept. 1860, coll. F. Jagor; Fig. 2). A female (33.3 mm SL) with protuberant belly; mature oocytes visible. Snout steep (angle to body axis nearly 45 degrees). Body deep (body depth 19.8% of SL at pelvic-fin origin, 17.4% of SL at anal-fin origin). Pre-anal and pre-anal-fin lengths large (60.1 and 64.6% of SL, respectively). Second dorsal fin with one spine and eight soft rays (one spine and 10 soft rays in Peters 1868) [it must have been one spine and nine soft rays originally, because the interval between 4th and 5th soft rays was twice as wide as normal (probably the membrane regenerated without a soft ray, after being damaged). A radiograph provided by ZMB indicated the presence of a set of pterygiophores at this interval]. Anal fin with one spine and nine soft rays. Pectoral fin with 16 soft rays. Cephalic sensory pore system with B', D(S), F', K', L', M' and O'. Dorsal and

lateral surfaces of body above lateral midline having three transverse, brownish bands. Lower part of lateral trunk with five indefinite transverse, brownish bars. One brownish blotch proximally on caudal fin. Ventral surface of head pigmented, but gular region lacking dense pigmentation. Pelvic fin without dense pigmentation but a few melanophores on 3rd, 4th, and 5th soft rays. Pectoral fin with 1–3 brownish spots along each central ray. This species was originally described as a member of the genus *Gobiosoma* by Peters (1868), but it actually belongs to *Schismatogobius* (Herre 1953; Kottelat and Pethiyagoda 1989; Chen et al. 1995a, 2001; Keith et al. 2004, 2017; Jenkins and Boseto 2005).

Holotype of *Schismatogobius bruynisi* de Beaufort 1912 (ZMA 111196, Eme River at Honitetu, West Seram, Indonesia, 24 Feb. 1910, coll. L. F. de Beaufort; Fig. 3). A male (31.6 mm SL) with slender body (body depth 15.8% of SL at pelvic-fin origin, 12.3% of SL at anal-fin origin). Mouth large (upper jaw length 19.3% of SL). Snout rather pointed (angle to body axis less than 30 degrees). Pre-anal and pre-anal-fin lengths moderate (57.3 and 61.4% of SL, respectively). Second dorsal fin with one spine and nine soft rays. Anal fin with one spine and nine soft rays; the spine hardly visible but present (de Beaufort 1912 and 1913 described both second dorsal- and anal-fin ray counts as “9”, as number of total elements). Pectoral fin with 15 soft rays. Cephalic sensory pore system with B', D(S), F', K', L', M' and O'. Although pigmentation somewhat indistinct due to long preservation, some diagnostic markings visible: one broad transverse brownish band on body below posterior part of second dorsal fin and anterior part of caudal peduncle, not clearly separated into anterior and posterior bands; pectoral fin with a large dusky brown patch around distal part of 2nd to 7th rays (more distinct in posterior view of fin); pelvic fin with a few melanophores, but not dense. Illustrations of the holotype have been provided in de Beaufort (1913: black and white in fig. 6 on page 142, color in fig. 2 on Plate II).



Holotype of *Schismatogobius roxasi* Herre 1936 (CAS-SU 30968, San Jose, Panay Island, Philippines, Feb. 1926, coll. F. Reveche; Fig. 4). A male (45.9 mm SL) with robust body (body depth 19.4% of SL at pelvic-fin origin, 17.0% of SL at anal-fin origin). Head large (head length 33.8% of SL) with very large mouth (upper jaw length 23.5% of SL). Snout blunt (angle to body axis nearly 45 degrees). Pre-dorsal length large (41.4% of SL), but pre-anal and pre-anal-fin lengths moderate (56.6 and 61.7% of SL, respectively). Second dorsal fin with one spine and nine soft rays, and anal fin with one spine and eight soft rays (both corresponding to description in Herre 1936). Pectoral fin with 17 soft rays. Cephalic sensory pore system with B', D(S), F', K', L', M' and O'. Three transverse, mottled brown bands dorsally on body; anteriormost one below first dorsal fin, second one below middle of second dorsal-fin base, and third one on anterior part of caudal peduncle. Caudal fin preserved as a separate piece; with two brown rings aligned horizontally along midline and two vertical brown bands distally. Ventral surface of head, including gular region, pigmented. Pelvic fin with a few melanophores. Pectoral-fin rays (except upper- and lower-most rays) with 1–5 brownish spots forming bars. Black and white illustration of holotype provided in Herre (1936: Fig. 5 in Plate 2). Coloration observed corresponded closely to that described in the text and figure of Herre (1936).

**Status of *Schismatogobius insignis*.** *Gobiosoma insignum* Herre 1927 was described from 10 syntypes collected from Negros, Philippines, but the syntypes have been lost (Kottelat 2013). This species belongs to *Schismatogobius*, not to *Gobiosoma* (Kottelat and Pethiyagoda 1989; Chen et al. 1995a, 2001; Keith et al. 2004, 2017; Jenkins and Boseto 2005). Although Koumans (1940, 1953) considered *G. insignum* as a synonym of *S. bruynisi*, it clearly differs from *S. bruynisi*, as shown below: *G. insignum* has a deeper body (body depth 4–4.25 times in the length, = 23.5–25.0% of SL, in the text; body depth 18.0% of SL at pelvic-fin origin and 15.5% of SL at anal-fin origin extrapolated by measuring the illustration

of Herre (1927) vs. 15.8% of SL at pelvic-fin origin and 12.3% of SL at anal-fin origin in *S. bruynisi*), steeply descending snout (vs. rather pointed in *S. bruynisi*), two brown, transverse bands dorsally on posterior half of the body (vs. bands nearly combined into one broad band in *S. bruynisi*), and pectoral fin crossbarred by three or four rows of brown spots (vs. pectoral fin with a large, distal brown patch on the upper part in *S. bruynisi*). These characteristics are shared with *S. marmoratus* and *S. roxasi*, but not with *S. bruynisi*. The caudal fin of *G. insignum* has a proximal white spot surrounded by a brown ring and two brown vertical bands with a relatively wide interval in the middle and distal parts (Herre 1927). Similar caudal fin markings are seen in the *S. roxasi* holotype (though it has another white spot formed by two vertical brown bars on the middle part and another vertical band along the tip; Fig. 4), while the proximal brown blotch lacks a distinct white gap and the middle and distal parts have 3–4 thin, brown vertical bars in *S. marmoratus* (holotype and Japanese material examined in the present study). Fin markings usually vary depending on the individual, but we examined two other specimens from the Philippines (URM-P 31459 and 31689 from comparative material) having the same pattern of caudal markings as in the *S. roxasi* holotype. This pattern may be unique to this species.

In the original description of *G. insignum*, Herre (1927) mentioned four non-type specimens collected from Antique Province, Panay Island in February 1926 by F. Reveche. Because the holotype of *S. roxasi* described in Herre (1936) was also collected from Antique Province in February 1926 by F. Reveche, it could be one of the four specimens mentioned in Herre (1927). Herre (1936) considered that *S. roxasi* “is close to *Gobiosoma insignum*, but differs in the extraordinary development of the maxillary.” Larger males of *Schismatogobius* usually have larger jaws (Fig. 5). Therefore, Herre (1936) may have misidentified the largest male of *G. insignum* (= holotype of *S. roxasi*) as a new species. We tentatively regard here *Schismatogobius roxasi* Herre 1936 as a junior synonym of *Schismatogobius insignus* (Herre

1927), although extensive examination with a much greater number of specimens would be required to verify their status.

### **Taxonomy of Japanese species**

#### ***Schismatogobius ninja* Maeda, Saeki, and Satoh, sp. nov.**

(Japanese name: Eso-haze) (Figs. 5–8, 9a–d, 10, S1, Tables 2, 3)

*Schismatogobius* sp. 1: Suzuki and Senou 1981:157, pl. 2, figs. I and J

*Schismatogobius* sp.: Hayashi et al. 1981:13 “145”, pl. 10, fig. 144

*Schismatogobius roxasi* (not of Herre): Akihito in Masuda et al. 1984:253, pl. 245 fig. H (with new Japanese name “eso-haze”); Akihito et al. in Nakabo 1993:1047; Akihito et al. in Nakabo 2000 (same in Nakabo 2002):1211; Akihito et al. in Nakabo 2013:1439

**Material examined. Holotype.** NSMT-P 127395, male (30.4 mm SL), northern part of Okinawa Island, Ryukyu Archipelago, Japan, 16 Mar. 2015, coll. K. Maeda and T. Saeki.

**Paratypes.** 14 specimens (5 males, 5 females, and 4 individuals of unknown sex, 18.2–32.4 mm SL) collected from northern part of Okinawa Island: NSMT-P 127400, 127401, 1 female (19.6 mm SL) and 1 individual of unknown sex (18.2 mm SL), 16 Mar. 2015, coll. K. Maeda and T. Saeki; NSMT-P 127408, 127410, 127412, 1 male (21.4 mm SL), 1 female (21.8 mm SL), and 1 individual of unknown sex (19.7 mm SL), 7 Oct. 2015, coll. K. Maeda; URM-P 48279, 1 male (23.1 mm SL), 11 Oct. 2010, coll. K. Maeda; URM-P 48280–48282, 1 female (20.1 mm SL) and 2 individuals of unknown sex (18.3–19.7 mm SL), 16 Sep. 2014, coll. T. Saeki; URM-P 48700, 48701, 48705, 2 males (18.3–29.7 mm SL) and 1 female (32.4 mm SL), 16 Mar. 2015, coll. K. Maeda and T. Saeki; URM-P 48710, 48711, 1 male (21.5) and 1 female (26.9 mm SL), 7 Oct. 2015, coll. K. Maeda.

**Non-type material.** 35 specimens (16 males and 19 females, 15.1–31.1 mm SL) collected from Okinawa, Ishigaki, and Iriomote islands in the Ryukyu Archipelago. These specimens were not used for the description, but examined to verify taxonomic status and distribution. Okinawa Island: URM-P 44724, 19.8 mm SL, 3 Dec. 2008; URM-P 48730, 19.5 mm SL, 8 Apr. 2017. Ishigaki Island: URM-P 4889, 15.3 mm SL, 12 Sept. 1982; URM-P 48728, 48729, 2 specimens, 15.1–16.3 mm SL, 5 Mar. 2017; YCM-P 7646, 7683, 7 specimens, 18.2–21.7 mm SL, 2 Aug. 1980. Iriomote Island: URM-P 11731, 19.2 mm SL, 19 Aug. 1985; URM-P 34252, 19.3 mm SL, 5 Aug. 1995; URM-P 36455, 27.7 mm SL, 20 Aug. 1996; URM-P 48699, 21.5 mm SL, 16 Aug. 2012; YCM-SSP 8947, 11 specimens, 19.0–31.1 mm SL, 26 July 1979; YCM-SSP 8972, 8 specimens, 17.0–26.6 mm SL, 28 July 1979.

**Diagnosis.** The new species is distinguished by a combination of the following characters: pectoral-fin rays 15–16 (usually 15); body relatively slender (depth at pelvic-fin origin 16.9–19.5% of SL); trunk relatively short (pre-dorsal length 35.5–39.1% of SL; pre-anal length 53.2–56.7% of SL; pre-anal-fin length 58.4–61.0% of SL; length of second dorsal-fin base 26.5–29.0% of SL; length of anal-fin base 22.3–25.9% of SL); two dark brown, transverse bands on posterior half of the body; pectoral fin with 1–5 black spots along each ray; pectoral fin often with one large, black vertical blotch on upper part, but blotch not extending to distal part; isthmus and gular region almost cream or white, often with some melanophores but not densely pigmented; pelvic fin usually almost without pigmentation, but often lightly pigmented in larger males.

**Description.** Body slender, nearly cylindrical; body depth 16.9–19.5% of SL (17.8% in holotype) at pelvic-fin origin and 13.8–17.1% of SL (14.5% in holotype) at anal-fin origin (Fig. 5, Table 2). Caudal peduncle slender. Pre-dorsal, pre-anal, and pre-anal-fin lengths relatively short (Figs. 5, 6). Head cylindrical. Eyes located dorsolaterally and close to each other. Snout short, nearly equal to eye diameter; snout blunt with angle to body axis < 45

degrees. Mouth terminal and oblique with lower jaw usually slightly protruding beyond upper jaw, but tips of upper and lower jaws almost in same position in holotype. Posterior tip of upper jaw reaching below middle of pupil in females; and to below posterior margin of eye or further in males (reaching far posteriorly from posterior margin of the eye in holotype). Larger males having larger upper jaws (Fig. 5). Anterior nostril short and tubular, posterior nostril a pore.

Size and shape of fins not differing between males and females. First dorsal fin with six spines, second dorsal fin with one spine and nine soft rays, except one paratype individual with one spine and 10 soft rays. Posterior end of first dorsal-fin base close to origin of second dorsal fin, and often in contact, but not connected broadly. Anal fin with one spine and nine soft rays. Origin of anal fin located below between bases of first and second soft rays of second dorsal fin. Anal fin shorter than second dorsal fin. Caudal fin truncated or rounded (rounded in holotype), with 17 segmented rays including 10–13 (usually 12, holotype 13) branched rays. Pectoral fin with 14 ( $n = 3$ ) or 15 ( $n = 12$  including holotype) soft rays (Table 3). Pelvic fin with one spine and five soft rays; pelvic fins joined together; frenum developed. Urogenital papilla round, plump in females and somewhat flat in males.

Cephalic sensory pore system with B', D(S), F', K', L', M' and O' (Fig. 8). Oculoscaphular canal interrupted between pores F' and K'. Cutaneous sensory papillae developed over dorsal, lateral, and ventral surface of head (Fig. 8).

*Color in preservative.* Background of body cream. Head brownish, but isthmus and gular region usually cream, often with a dark brown blotch on gular region in both males and females (holotype with an indistinct blotch); isthmus and gular region often with scattered melanophores in males, but melanophores not dense (Fig. 9). Body with three dark brown transverse bands; second and third bands often nearly combined by a brown interval (clearly separated in holotype), but the bands still distinguishable from the interval being somewhat

paler. Antermost band located on trunk. Second band located below the bases of fourth and fifth soft rays of second dorsal fin. Third band located on anterior part of caudal peduncle. Dorsal half of body between transverse bands and posterior part of caudal peduncle brown mottled with cream spots. Transverse bands also often mottled with pale brown spots. One large dark brown blotch from caudal-fin base to proximal half of caudal fin; one white patch below the dark brown blotch on proximal part of caudal fin; two white, round patches arrayed vertically just posterior of the dark brown blotch; posterior one third of caudal-fin rays pale gray, but larger individuals having one or two grayish to dark brown vertical bands with white interval (two bands in holotype). First dorsal fin of male having a black band across middle of spines; fin also with black distal margin and black proximal blotches. First dorsal fin of female usually with a large black blotch on its anterior part; distal margin of fin usually black. Spine and soft rays in second dorsal fin with 1–5 black spots. Anal fin usually translucent without notable pigmentation, but with a few melanophores in one paratype male. Each pectoral-fin ray with 1–5 black spots except uppermost and lower 3–5 rays; proximal 1–2 spots often enlarged and forming one large, vertical black blotch proximally around second to seventh rays, but not extending to distal part of pectoral fin. Pelvic fin usually almost without pigmentation, but often lightly pigmented in larger males (including holotype) (Fig. 9).

*Color in life* (Fig. 10). Background whitish and somewhat translucent. Patterns of dark brown bands and markings similar to those in preservative, but occipital, temporal, and opercular regions of head, pectoral-fin base, and proximal part of pectoral fin usually white with pale pink tinge; snout usually reddish brown, gray, or pink; nape usually brown. Intervals between first and second dark brown transverse bands usually pale pink with scattered small bluish-white spots. Interval between second and third transverse bands gray or pale pink with scattered small bluish-white spots. Posterior part of caudal peduncle usually

pink or orange. Spines and soft rays in first and second dorsal and pectoral fins with white spots between each black spot, but those usually indistinct in smaller individuals (< ca. 20 mm SL). In larger males (including holotype), anal-fin membrane yellow; pelvic fin and distal part of first and second dorsal fins also becoming yellowish.

**Distribution.** All specimens described here with mitochondrial DNA sequences were collected from Okinawa Island in the Ryukyu Archipelago. Thirty-five non-type specimens collected from Okinawa, Ishigaki, and Iriomote islands were also identified as the new species, *S. ninja*. Many authors reported “eso-haze” (regarded as *S. roxasi*) in Japan and the range often included Amami-oshima Island, in addition to Okinawa, Ishigaki, Iriomote islands (Yoshigou 2014). These records need to be verified by reexamination of the specimens. This species seems to have often been misidentified as “*S. ampluvinculus*” (see Distribution section of *S. ampluvinculus*). In conclusion, verified localities of *S. ninja* in Japan are Okinawa, Ishigaki, and Iriomote islands, but it may be found on some other islands in the Ryukyu Archipelago.

**Ecology.** All specimens were collected from lower freshwater reaches of the streams, at 50 to 1,300 m from the upper limit of tidal fluctuations. This species was found on coarse sand and gravel bottoms of shallow areas (depth usually 5–30 cm) around rapids. Typical habitat was at the ends of the rapids (transitional areas from rapid to pool) with steady flow. These gobies usually stay on the bottom and often bury half of their body in the substratum (Fig. 10e). They prefer substrates with free gravel granules which can be moved easily by water flow, and they do not inhabit silty substrates. Body markings perfectly camouflage them on the gravel bottom of their habitats (Fig. 10e), therefore they are hard to detect if they remain motionless.

*Schismatogobius ninja* is often found with *S. ampluvinculus*, and sometimes with *S. marmoratus*. The most abundant syntopic species was *Rhinogobius nagoyae* Jordan and Seale

1906. Other gobies, such as *Stiphodon percnopterygionus* Watson and Chen 1998, *Sicyopterus lagocephalus* (Pallas 1770), *Glossogobius illimis* Hoese and Allen 2012, and *Luciogobius ryukyuensis* Chen, Suzuki, and Senou 2008 and amphidromous pipefish, *Microphis leiaspis* (Bleeker 1854) were also frequently observed with *S. ninja*.

**Etymology.** Although the new species shows attractive coloration when viewed against a simple background, they are very cryptic against the gravel substrates of their habitats. The stealthy capacity of this species reminds us of Japanese “ninja,” which were known as masters of camouflage. Therefore, the new species is named *Schismatogobius ninja*. The new specific name is a noun in apposition.

**Comparison.** The new species is distinguished from all other nominal species as follows: It differs from *S. ampluvinculus* in having the two dark brown, transverse bands on the posterior half of the body (vs. bands almost combined into one broad band), pectoral fin with 1–5 black spots along each ray (proximal 1–2 spots are often enlarged, forming one large, vertical blotch on the upper part, but this blotch does not extend to the distal part in *S. ninja*, vs. with a large, distal black patch on upper part extending nearly to the fin tip in *S. ampluvinculus*), shorter pre-dorsal, pre-anal, pre-anal-fin lengths (pre-dorsal length 35.5–39.1% vs. 37.7–40.5% of SL; pre-anal length 53.2–56.7% vs. 56.1–59.5% of SL; pre-anal-fin length 58.4–61.0% vs. 60.3–64.5% of SL; Figs. 5, 6), and longer second dorsal- and anal-fin bases (length of second dorsal-fin base 26.5–29.0% vs. 24.8–27.0% of SL except 28.3% in an individual irregularly with 10 soft rays; length of anal-fin base 22.3–25.9% vs. 20.0–23.1% of SL; Fig. 6); i.e. *S. ninja* has a shorter trunk and longer tail (body from anus to caudal-fin base) than *S. ampluvinculus*. *Schismatogobius ninja* (up to 32.4 mm SL) grows larger than *S. ampluvinculus* (up to 26.9 mm SL; Chen et al. 1995a) and the sexual dimorphism of jaw length begins at a greater SL than the latter (Fig. 5). *Schismatogobius*



*bruynisi* shares having one broad dark brown transverse band on the posterior half of the body and pectoral-fin marking with *S. ampluvinculus*, also distinguishing it from *S. ninja*.

*Schismatogobius ninja* differs from *S. tiola* Keith, Lord, and Larson 2017, *S. essi* Keith, Lord, and Larson 2017, and *S. mondo* Keith, Lord, and Larson 2017 in the pigmentation on the ventral surfaces of the head and pelvic fins (less pigmented in both males and females of *S. ninja*).

The new species appears to be most similar to *S. baibatag* Keith, Lord, and Larson 2017, although only three specimens are known of the latter (Keith et al. 2017). However, it differs from *S. baibatag* in having a shorter pre-anal length (53.2–56.7% vs. 56.0–58.6% of SL), longer second dorsal and caudal fins (length of second dorsal fin 32.7–37.5% vs. 29.0–32.4% of SL; length of caudal fin 20.1–24.2% vs. 18.3–21.3% of SL), and longer jaws in female (upper jaw length in female 9.2–10.2% vs. 7.5–7.7% of SL).

*Schismatogobius deraniyagalai* Kottelat and Pethiyagoda 1989 lacks the preopercular canal and associated pores in the cephalic sensory pore system (Keith et al. 2004; Arunachalam et al. 2014) which all other species of the genus possess. The new species is also distinguished from *S. deraniyagalai* by presence of the preopercular canal and pores M' and O'.

*Schismatogobius ninja* differs from *Schismatogobius vitiensis* Jenkins and Boseto 2005 by range and/or mode of second dorsal-, anal-, and pectoral-fin ray counts (soft rays in second dorsal fin usually 9 in *S. ninja* vs. mode 9 but often 10 in *S. vitiensis*; soft rays in anal fin always 9 vs. 8–9, mode 8; pectoral-fin rays 14–15, usually 15 vs. 15–16, mode 16).

Finally, *S. ninja* is distinguished from *S. fuligimentus* Chen, Séret, Pöllabauer, and Shao 2001 and *S. tuimanua* Keith, Lord, and Larson 2017 in having more pectoral-fin rays (usually 15 vs. 13–14), and from *S. marmoratus*, *S. insignis*, *S. vanuatuensis* Keith, Marquet, and

Watson 2004, *S. hoesei* Keith, Lord, and Larson 2017, and *S. alleni* Keith, Lord, and Larson 2017 in having fewer pectoral-fin rays (usually 15 vs. 16–17).

***Schismatogobius ampluvinculus* Chen, Shao, and Fang 1995**

(Japanese name: Shima-eso-haze) (Figs. 5, 6, 9e–h, 11, 12, 13a–c, S2, S3, Tables 2, 3)

*Schismatogobius* sp. 2: Suzuki and Senou 1981:157, pl. 2, figs. K and L

*Schismatogobius* sp.: Hayashi et al. 1981:13 “144”, pl. 10, fig. 145; Akihito in Masuda et al. 1984:253 (with new Japanese name “shima-eso-haze”); Akihito et al. in Nakabo 1993:1047

*Schismatogobius ampluvinculus* Chen, Shao, and Fang 1995:202 (type locality: Jinglun River, Taitung Country, Taiwan); Akihito et al. in Nakabo 2000 (same in Nakabo 2002):1211; Akihito et al. in Nakabo 2013:1439

**Examined material with mitochondrial DNA sequences.** 24 specimens (13 males and 11 females, 14.7–23.6 mm SL) collected from the Ryukyu Archipelago, Japan. NSMT-P 127398, 127399, 127402, 127403, 3 males (14.7–17.5 mm SL) and 1 female (16.6 mm SL), Okinawa Island, Japan, 16 Mar. 2015, coll. K. Maeda and T. Saeki; NSMT-P 127404–127412, 2 males (17.8–17.9 mm SL) and 4 females (17.7–20.0 mm SL), Okinawa Island, 7 Oct. 2015, coll. K. Maeda; URM-P 48706, 1 female (15.2 mm SL), Okinawa Island, 19 Sep. 2015, coll. K. Maeda; URM-P 48707–48709, 48712–48718, 7 males (15.5–19.2 mm SL) and 3 females (16.5–19.5 mm SL), Okinawa Island, 7 Oct. 2015, coll. K. Maeda; URM-P 48719–48721, 1 male (20.2 mm SL) and 2 females (15.3–23.6 mm SL), Iriomote Island, Japan, 10 Nov. 2015, coll. K. Maeda.

**Additional material examined.** 16 specimens (6 males and 10 females, 15.1–23.5 mm SL) collected from Iriomote Island in the Ryukyu Archipelago and 1 male specimen (19.7

mm SL) from the Philippines. These specimens were not used for the description, but were examined to verify taxonomic status and distribution: Iriomote Island: URM-P 34251, 6 specimens, 15.6–22.1 mm SL, 5 Aug. 1995; URM-P 35806, 2 specimens, 18.6–22.2 mm SL, 18 July 1996; URM-P 35848, 22.2 mm SL, 23 July 1996; URM-P 36456, 2 specimens, 21.7–23.5 mm SL, 20 Aug. 1996; YCM-SSP 8973, 9109, 5 specimens, 15.1–23.1 mm SL, 28 July 1979. Mindoro Island, Philippines: URM-P 31453, 19.7 mm SL, 15 Aug. 1985.

**Diagnosis.** Pectoral-fin rays 15–16; body relatively slender (depth at pelvic-fin origin 16.6–19.0% of SL); trunk relatively longer (pre-dorsal length 37.7–40.1% of SL; pre-anal length 56.1–59.5% of SL; pre-anal-fin length 60.3–64.5% of SL; length of second dorsal-fin base 24.8–28.3% of SL; length of anal-fin base 20.0–23.1% of SL); one dark brown, broad transverse band on posterior half of the body; pectoral fin with a large, obvious black patch on upper part extending nearly to the distal tip of fin; isthmus and gular region dark brown in larger males, but usually cream or white in females and smaller males; pelvic fin with many melanophores in larger males, but with few or no melanophores in females and smaller males.

**Description.** Body slender (body depth at pelvic- and anal-fin origins 16.6–19.0 and 13.3–16.9% of SL, respectively; Fig. 5), and nearly cylindrical. Caudal peduncle slender. Pre-dorsal, pre-anal, and pre-anal-fin lengths relatively long (Figs. 5, 6). Head cylindrical. Eyes located dorsolaterally and close to each other. Snout short, nearly equal to eye diameter; snout blunt with angle to body axis less than 45 degrees. Mouth terminal and oblique with lower jaw usually slightly protruding beyond upper jaw. Posterior tip of upper jaw reaching below middle of pupil in females and to below posterior margin of eye or further in males; larger males having larger upper jaws (Fig. 5). Anterior nostril short tubular, posterior nostril a pore.

Size and shape of fins not differing between males and females. First dorsal fin with six spines, second dorsal fin usually with one spine and nine soft rays, but one specimen having

one spine and 10 soft rays (second dorsal-fin base longer in this specimen, 28.3% of SL; Fig. 6). Posterior end of first dorsal-fin base close to origin of second dorsal fin, and often in contact, but not connected broadly. Anal fin usually with one spine and nine soft rays, but two specimens having one spine and eight soft rays. Origin of anal fin located below between bases of first and second soft rays of second dorsal fin. Anal fin shorter than second dorsal fin. Caudal fin truncated or somewhat rounded, with 17 segmented rays (but one specimen having 15) including 11–13 branched rays. Pectoral fin with 15 or 16 soft rays (Table 3). Pelvic fin with one spine and five soft rays; pelvic fins joined together; frenum developed. Urogenital papilla round, plump in females and somewhat flat in males.

Cephalic sensory pore system with B', D(S), F', K', L', M' and O' (Fig. 12).

Oculoscapular canal interrupted between pores F' and K'. Cutaneous sensory papillae developed over dorsal, lateral, and ventral surface of head (Fig. 12).

*Color in preservative.* Background of body cream. Head brown, but isthmus and gular region cream in females (Fig 9g, h); dark area on ventral surface of head usually reaching origin of pelvic fin in larger males (> 18 mm SL) (Fig. 9e), but smaller males usually having cream gular region (sometimes with a dark brown blotch) (Fig. 9f). Occipital region often paler brown. Body with two dark brown, broad transverse bands. Anterior one located on trunk. Posterior band located on body from base of third soft ray of second dorsal fin to middle of caudal peduncle; ventral half often darker. Dorsal half of body between anterior and posterior transverse bands and posterior part of caudal peduncle often mottled by pale brown spots. One large dark brown blotch from caudal-fin base to proximal half of caudal fin; one white patch below the dark brown blotch on proximal part of caudal fin; two white, round patches arrayed vertically just posterior to dark brown blotch; posterior one third of caudal-fin rays pale gray. Spines and soft rays in first and second dorsal fins usually having 1–4 black spots; fin elements in smaller specimens (< 17 mm SL) often totally brown without distinct

spots. First dorsal-fin membrane between first and fifth spines often having proximal black blotch. Anal fin translucent and usually with a few melanophores proximal to the sixth to ninth soft rays. Pectoral fin with a large, obvious black patch on dorsal part, with dorsalmost three or four rays and membrane between these rays almost totally black; black patch extending ventrally but gradually reducing and not extending to ventral third of the fin; middle rays often with black spots forming 1–3 vertical black bars. Pelvic-fin rays and frenum with many melanophores in larger males (> 19 mm SL; Fig. 9e), but with few or none in females and smaller males (Fig. 9f, g, h).

*Color in life* (Fig. 13a, b, c). Background whitish and somewhat translucent. Patterns of dark brown bands and markings similar to those in preservative, but snout, occipital, temporal, and opercular regions of head, pectoral-fin base, and proximal part of pectoral fin usually white with a tinge of pale pink. Interval between dark brown transverse bands and posterior part of caudal peduncle also white usually with a tinge of pale pink and scattered small bluish-white spots. Middle of posterior dark brown band often becoming paler gray or brown. Spines and soft rays in first and second dorsal and pectoral fins with white spots between each black spot; usually indistinct in smaller individuals (< ca. 19 mm SL).

**Distribution.** Japanese specimens examined were collected from Okinawa and Iriomote Islands in the Ryukyu Archipelago. Shima-eso-haze (*S. ampluvinculus*) has also been reported from other islands in the archipelago, including Tanegashima, Yakushima, Kume, and Ishigaki islands (Yoshigou 2014), but previous records need to be verified by reexamining the specimens. For example, Yonezawa and Shinomiya (2002) recorded *S. ampluvinculus* from Tanegashima and Yakushima islands, but the goby in an in situ photograph (Fig. 3 of Yonezawa and Shinomiya 2002) seems to be *S. ninja*, not *S. ampluvinculus*. Satou (2005) recorded “shima-eso-haze” from Kume Island; however, the goby in the picture may be *S. ninja* (eso-haze).

*Schismatogobius ampluvinculus* is also distributed in southeastern Taiwan (Chen et al. 1995a) and the Philippines (Mindoro Island; present study).

**Ecology.** *Schismatogobius ampluvinculus* was found in the lower freshwater reaches of streams, 50 to 800 m from the upper limit of tidal fluctuations. Their habitat is almost the same as that of *S. ninja* and they were often observed syntopically. *Schismatogobius marmoratus* was also found sometimes. Major syntopic fishes were also the same (see Ecology section of *S. ninja*).

This species also has camouflage coloration. These fish often spread the pectoral fins and flutter them (Fig. 13c). Although the proximal part of the large black patch on the pectoral fin is covered by white pigmentation on anterior side (Fig. 13a, b), it lacks white pigmentation on posterior side (Fig. 13c). Therefore, the black patch on the posterior side is conspicuous during the fluttering behavior. They may communicate with other individuals by this behavior, although its meaning is unknown.

**Comparison.** Morphology of specimens with sequenced mitochondrial genomes and described above correspond to *S. ampluvinculus* Chen, Shao, and Fang 1995, described in Chen et al. (1995a). According to the original description by Chen et al. (1995a), *S. ampluvinculus* has 16 pectoral fin rays. The mode of the count was 15 in the present study, but 16 rays were also common (15 rays, n=17; 16 rays, n=7; Table 3).

This species shares many characters, including a large, black patch on the dorsal part of pectoral fin and broad black transverse band on posterior half of the body, with *S. bruynisi*, but *S. bruynisi* seems to grow far larger than *S. ampluvinculus*. The largest specimen of *S. ampluvinculus* is 23.6 mm SL in the present study (among 41 specimens examined) and 26.9 mm SL in Chen et al. (1995a), while the holotype of *S. bruynisi* is far larger, 31.6 mm SL. *Schismatogobius ampluvinculus* mature at a small size, as the presence of a gravid female

19.5 mm SL indicates, and sexual dimorphism of jaw length is seen even in the smallest individual examined (14.7 mm SL; Fig. 5).

Chen et al. (1995a) wrote that body depth is smaller in *S. ampluvinculus* than in *S. bruynisi*. However, the holotype of *S. bruynisi* examined in the present study was slightly more slender than *S. ampluvinculus* (body depth at pelvic- and anal-fin origins 15.8 and 12.3% of SL in *S. bruynisi* vs. 16.6–19.0 and 13.3–16.9% of SL in *S. ampluvinculus*; Fig. 5). This may be due to shrinkage of the *S. bruynisi* holotype in preservative. In comparison with morphometric values of *S. bruynisi* given in Keith et al. (2017), *S. ampluvinculus* females have longer heads (27.4–29.0 vs. 25.0–27.9% of SL in *S. bruynisi*); however, both male and female *S. ampluvinculus* have deeper caudal peduncles (depth 8.5–10.2% vs. 6.8–8.8% of SL) and shorter anal fins (26.0–29.3% vs. 29.7–32.6% of SL) than *S. bruynisi*.

### ***Schismatogobius marmoratus* (Peters 1868)**

(New Japanese name: Kaeru-eso-haze) (Figs. 5, 6, 9i–k, 13d–f, 14, 15, S4, Tables 2, 3)

*Gobiosoma marmoratum* Peters 1868: 267 (type locality: Loquilócun, Samar, Philippines)

**Examined material with mitochondrial DNA sequences.** NSMT-P 127396, 127397, URM-P 48702–48704, 1 male (33.5 mm SL) and 4 females (31.1–41.9 mm SL), Okinawa Island, Japan, 16 Mar. 2015, coll. K. Maeda and T. Saeki.

**Diagnosis (including holotype data).** Pectoral-fin rays 16; body deep (body depth at pelvic-fin origin 19.8–21.9% of SL); trunk relatively long (pre-anal length 57.4–60.1% of SL; pre-anal-fin length 62.2–64.6% of SL); snout blunt and steep; two dark brown, transverse bands on posterior half of the body; pectoral fin with 1–8 black spots along each ray forming vertical bars; isthmus, gular region, and pelvic fins densely pigmented in male; isthmus and

gular region of female almost cream or white often with a brownish blotch on gular region; pelvic fin almost translucent, but a few melanophores along soft rays in female; one diamond-shaped dark brown blotch from caudal-fin base to proximal part of caudal fin; posterior half of branched caudal-fin rays having 3–5 small black spots forming thin vertical bars.

**Description (based on Japanese specimens).** Body nearly cylindrical, but remarkably deep at pelvic-fin origin (19.8–21.9% of SL; Fig. 5) than at anal-fin origin (15.5–17.7% of SL; Fig. 5), with slender caudal peduncle. Pre-dorsal length relatively short (36.9–38.3% of SL; Fig. 5), but pre-anal and pre-anal-fin lengths relatively long (57.4–59.4 and 62.2–64.6% of SL, respectively; Fig. 6). Head cylindrical. Eyes located dorsolaterally and close to each other. Snout short, nearly equal to eye diameter; snout blunt and steep with angle to body axis usually more than 45 degrees. Mouth terminal and oblique with lower jaw slightly protruding beyond upper jaw. Posterior tip of upper jaw reaching below posterior margin of pupil or posterior margin of eye in females; extending further posteriorly in male. Anterior nostril short tubular, posterior nostril a pore.

Size and shape of fins does not differ between male and females. First dorsal fin with six spines, second dorsal fin with one spine and nine soft rays. Posterior end of first dorsal-fin base contacting the origin of second dorsal fin, but not connected broadly. Anal fin with one spine and nine soft rays. Origin of anal fin located below between bases of first and second soft rays of second dorsal fin. Anal fin shorter than second dorsal fin. Caudal fin truncated or rounded, with 17 segmented rays including 12 or 13 branched rays. Pectoral fin with 16 soft rays (Table 3). Pelvic fin with one spine and five soft rays; pelvic fins joined together; frenum developed. Urogenital papilla round, plump in females and somewhat flat in male.

Cephalic sensory pore system with B', D(S), F', K', L', M' and O', but one individual also having pore N only in the left side (Fig. 15). Oculoscapular canal interrupted between



pores F' and K'. Cutaneous sensory papillae developed over dorsal, lateral, and ventral surface of head (Fig. 15).

*Color in preservative.* Background of body cream. Head brownish, but middle of upper and lower jaws and ventral surface of head cream in females (often with a black blotch on gular region); brownish area on ventral surface of head reaching pelvic-fin origin in male (Fig. 9). Dorsal half of body with three dark brown, transverse bands. Anteriormost band located below middle of first dorsal-fin base. Second band located below around bases of fourth and fifth soft rays of second dorsal fin. Third band located on anterior part of caudal peduncle. Dorsal half of body between transverse bands and posterior part of caudal peduncle brown mottled with cream spots. Transverse bands also often mottled with pale brown spots. Lateral surface of body below lateral midline with irregular dark brown blotches forming vermiculate patterns. One diamond-shaped dark brown blotch from caudal-fin base to proximal part of caudal fin; a brown, round patch anteriorly above and another below the dark brown blotch; a cream, round patch posteriorly above and another below the dark brown blotch. Posterior half of branched caudal-fin rays with 3–5 small black spots forming bars. First dorsal fin of male with a black band across middle of spines; fin also with black distal margin and black spots on proximal part of spines. In females, 3–5 black spots along each spine of first dorsal fin. Spine and soft rays in second dorsal fin having 4–7 black spots. Anal-fin membrane dusky gray in male; membrane almost translucent in females, but spine and soft rays with a few melanophores. Each pectoral-fin ray with 1–8 black spots forming bars, spots usually indistinct on lower 1–3 rays. Spines, soft rays, membrane, and frenum of pelvic fin densely pigmented in male (Fig. 9i); pelvic fin almost translucent but a few melanophores along soft rays in females (Fig. 9j, k).

*Color in life* (Fig. 13d, e, f). Background white. Patterns of dark brown bands and markings similar to those in preservative, but snout, occipital, temporal, and opercular regions

of head, pectoral-fin base, and proximal part of pectoral fin often whitish with a tinge of pale pink and mottled with small cream spots. Intervals between first and second dark brown transverse bands usually pale pink with scattered small bluish-white spots. Interval between second and third dark brown transverse bands orange to brown with scattered small bluish-white spots. Posterior part of caudal peduncle usually pale pink. Spines and soft rays in first and second dorsal and pectoral fins with white spots between each black spot. Distal part of first dorsal fin of male yellowish. Anal and pelvic fin also yellowish in male.

**Distribution and ecology.** *Schismatogobius marmoratus* has been found in the Philippines (Samar Island; Peters 1868) and in Japan (Okinawa Island; present study).

Japanese specimens of *S. marmoratus* were collected from lower freshwater reaches of a stream, 150 m from the upper limit of tidal fluctuations, with *S. ninja*. This species was also observed in two other streams on Okinawa Island. These sites were located 50 and 600 m from the upper limit of tidal fluctuations, respectively. They were found in rapids at depths of 5–30 cm. In one case, we observed an *S. marmoratus* that swam away in the middle of the water column against a strong current, although we have not seen such behavior in *S. ninja* and *S. ampluvinculus*.

**Comparison.** The morphology of the Japanese specimens described above corresponds to that of the *S. marmoratus* (Peters 1868) holotype from the Philippines.

The morphology of *S. insignis* (= *S. roxasi*) is similar to that of *S. marmoratus*, but it differs from the latter by having a longer head and jaws (head length 33.8% vs. 31.3% of SL in *S. marmoratus* male; upper jaw length 23.5% vs. 19.4% of SL; Fig. 5; but note that larger males usually have longer heads and jaws and the *S. roxasi* holotype is larger), longer pre-dorsal length (41.4% vs. 36.9–38.7% of SL; Fig. 5; this may be caused by the longer head), 17 rays in pectoral fin (vs. 16 rays; Table 3), one spine and eight soft rays in anal fin

(vs. one spine and nine soft rays in *S. marmoratus*: note that eight soft rays in *S. roxasi* may be irregular), and the markings on the caudal fin.

**Remarks.** Kottelat et al. (1993) included Sulawesi, the Philippines, and Japan in the distribution of *S. marmoratus* and showed two specimens identified as *S. marmoratus* in their plate 71. However, the specimen (32 mm SL) at the top of the figure looks like *S. insignis* (*S. roxasi*). Another specimen on the bottom is a juvenile (10 mm SL) and we hesitate to attempt to identify it. “*Schismatogobius roxasi*” reported from southeastern Taiwan by Chen et al. (1995b) may be actually *S. marmoratus*. These records require reexaminations of the specimens.

## Discussion

**Revision of Japanese species.** The presence of *Schismatogobius* in Japan was first reported by Suzuki and Senou (1981) and Hayashi et al. (1981). Suzuki and Senou (1981) reported two species of *Schismatogobius* as “*Schismatogobius* sp. 1” and “*Schismatogobius* sp. 2” based on 21 and eight specimens collected from Iriomote Island, respectively. Specimens (one male and one female for each species) shown in plate 2 of Suzuki and Senou (1981) are identified as *S. ninja* (for sp. 1) and *S. ampluvinculus* (for sp. 2) based on markings on the pectoral fin and band arrangement on the body. Hayashi et al. (1981) also reported both species as “*Schismatogobius* sp.” They listed eight specimens from Iriomote Island (same material as in Suzuki and Senou 1981) for “144. *Schismatogobius* sp.” and 32 specimens from Iriomote and Ishigaki islands (including 21 specimens used in Suzuki and Senou 1981) for “145. *Schismatogobius* sp.” These specimens were deposited at YCM (Suzuki and Senou 1981; Hayashi et al. 1981). As a result of reexamining five of the eight listed specimens that were available for “144. *Schismatogobius* sp.” and 26 of the 32 specimens listed for “145.

*Schismatogobius* sp.”, we identified the former specimens as *S. ampluvinculus* and the latter as the new species, *S. ninja*. Although Hayashi et al. (1981) provided a picture of an *S. ninja* female as number 144 and a picture of an *S. ampluvinculus* female as number 145, the numbers must be reversed.

Akihito in Masuda et al. (1984) identified these two species as “*Schismatogobius roxasi* Herre” and “*Schismatogobius* sp.” and provided new Japanese names, “eso-haze” for the former and “shima-eso-haze” for the latter. Based on characteristics described in the text (p. 253), “eso-haze *S. roxasi*” is identified as *S. ninja* and “shima-eso-haze *Schismatogobius* sp.” is identified as *S. ampluvinculus*. Pictures of the two species are shown on plate 245 of Masuda et al. (1984): “eso-haze *S. roxasi*” (plate 245 H) is identified as *S. ninja*, but we hesitate to identify “shima-eso-haze *Schismatogobius* sp.” (plate 245 I), because it is a small juvenile (8 mm). Kawanabe and Mizuno (1989), Akihito et al. in Nakabo (1993), and Masuda and Kobayashi (1994) also identified “eso-haze” as *S. roxasi* and “shima-eso-haze” as *Schismatogobius* sp., but a pictured individual shown as “shima-eso-haze” on p. 562 (picture M) of Kawanabe and Mizuno (1989) is *S. ninja*, not *S. ampluvinculus* (shima-eso-haze).

After Chen et al. (1995a) described *S. ampluvinculus* as a new species, Akihito et al. in Nakabo (2000, 2002) identified “shima-eso-haze” as *S. ampluvinculus*. Later Japanese publications always regarded the two species as “eso-haze *S. roxasi*” and “shima-eso-haze *S. ampluvinculus*” (e.g. Suzuki et al. 2004; Akihito et al. in Nakabo 2013).

All published pictures and illustrations of Japanese *Schismatogobius* present characteristics of *S. ninja* or *S. ampluvinculus*, and we could not find other species (e.g. *S. marmoratus* and *S. insignis* = *S. roxasi*) among them. We examined all available Japanese specimens of *Schismatogobius* in URM (n = 19, excluding materials used for descriptions in the present study) and YCM (n = 31) collections. They are all identified as *S. ninja* or *S. ampluvinculus*.

Therefore, “eso-haze” is actually a new species, *S. ninja*, not *S. roxasi* and “shima-eso-haze” is *S. ampluvinculus*. We found the third species, *S. marmoratus* in Japan, but it is rare here. As *S. marmoratus* did not have a Japanese name, we propose a new Japanese name “kaeru-eso-haze” for *S. marmoratus* described in the present study. The name is derived from its frog-like face. “Kaeru” means frog, “eso” means lizardfish (Synodontidae), and “haze” means goby.

**Key to species of Japanese *Schismatogobius***

- 1a. Body depth at pelvic-fin origin usually > 19.5% of SL; pectoral-fin rays usually 16  
 .....*S. marmoratus*
- 1b. Body depth at pelvic-fin origin usually < 19.5 % of SL; pectoral-fin rays 14–16  
 (mode 15).....2
- 2a. Posterior half of the body with two dark brown, transverse bands; pectoral fin with  
 1–5 black spots along each ray (proximal 1–2 spots are often enlarged and forming  
 into one large, vertical blotch on upper part, but this blotch does not extend to distal  
 part); pre-anal length 53–57% of SL; pre-anal-fin length 58–61% of SL.....*S. ninja*
- 2b. Posterior half of the body with one dark brown, broad transverse band; pectoral fin  
 with a large, distal black patch on upper part extending nearly into the tip; pre-anal  
 length 56–60% of SL; pre-anal-fin length 60–65% of SL.....*S. ampluvinculus*

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**Table 1** Counts and morphometrics of holotypes of *Gobiosoma marmoratum* Peters 1868 (= *S. marmoratus*), *Schismatogobius bruynisi* de Beaufort 1912, and *Schismatogobius roxasi* Herre 1936

|                                      | <i>G. marmoratum</i> | <i>S. bruynisi</i> | <i>S. roxasi</i> |
|--------------------------------------|----------------------|--------------------|------------------|
|                                      | ZMB 6756             | ZMA 111196         | CAS-SU 30968     |
|                                      | Female               | Male               | Male             |
| Standard length (mm)                 | 33.3                 | 31.6               | 45.9             |
| Counts                               |                      |                    |                  |
| D <sub>1</sub> -D <sub>2</sub>       | VI-I, 9              | VI-I, 9            | VI-I, 9          |
| A                                    | I, 9                 | I, 9               | I, 8             |
| P <sub>1</sub>                       | 16                   | 15                 | 17               |
| P <sub>2</sub>                       | I, 5                 | I, 5               | I, 5             |
| Measurements as % of standard length |                      |                    |                  |
| Head length                          | 28.3                 | 30.7               | 33.8             |
| Snout length                         | 6.3                  | 6.3                | 7.8              |
| Eye diameter                         | 6.0                  | 5.7                | 5.2              |
| Postorbital length of head           | 16.2                 | 19.3               | 21.8             |
| Upper-jaw length                     | 11.4                 | 19.3               | 23.5             |
| Body depth at P <sub>2</sub> origin  | 19.8                 | 15.8               | 19.4             |
| Body depth at A origin               | 17.4                 | 12.3               | 17.0             |
| Depth at caudal peduncle             | 8.4                  | 7.6                | 8.9              |
| Length of caudal peduncle            | 18.0                 | 19.0               | 20.3             |
| Predorsal length                     | 38.7                 | 38.9               | 41.4             |
| Preanal length                       | 60.1                 | 57.3               | 56.6             |
| Pre-anal-fin length                  | 64.6                 | 61.4               | 61.7             |
| Length of D <sub>2</sub> base        | 26.4                 | 25.0               | 26.6             |
| Length of A base                     | 19.5                 | 19.3               | 19.0             |

*D*<sub>1</sub> first dorsal fin, *D*<sub>2</sub> second dorsal fin, *A* anal fin, *C* caudal fin, *P*<sub>1</sub> pectoral fin, *P*<sub>2</sub> pelvic fin

**Table 2** Morphometrics of *Schismatogobius ninja* sp. nov., *S. ampluvinculus*, and *S. marmoratus* collected in Japan, expressed as a percentage of standard length

|                                     | <i>S. ninja</i> sp. nov. |           |           |              | <i>S. ampluvinculus</i> |           | <i>S. marmoratus</i> |           |
|-------------------------------------|--------------------------|-----------|-----------|--------------|-------------------------|-----------|----------------------|-----------|
|                                     | Holotype                 | Paratype  |           |              | Male                    | Female    | Male                 | Female    |
|                                     | Male                     | Male      | Female    | Unidentified |                         |           |                      |           |
| Number of specimens measured        | 1                        | 5         | 5         | 4            | 13                      | 11        | 1                    | 4         |
| Standard length (mm)                | 30.4                     | 18.3–29.7 | 19.6–32.4 | 18.2–19.7    | 14.7–20.2               | 15.2–23.6 | 33.5                 | 31.1–41.9 |
| Head length                         | 29.3                     | 27.6–30.3 | 25.9–28.1 | 27.4–28.4    | 28.3–30.2               | 27.2–30.3 | 31.3                 | 27.4–29.0 |
| Snout length                        | 7.6                      | 5.5–7.1   | 5.6–6.5   | 5.5–6.6      | 5.8–7.3                 | 5.9–6.7   | 6.9                  | 5.5–6.4   |
| Eye diameter                        | 5.6                      | 5.2–6.6   | 5.2–6.1   | 5.5–6.6      | 5.4–7.5                 | 5.7–6.7   | 5.4                  | 5.4–6.0   |
| Postorbital length of head          | 17.4                     | 16.4–18.2 | 15.1–16.5 | 15.8–17.8    | 16.3–18.8               | 15.1–17.0 | 19.4                 | 15.7–18.2 |
| Upper-jaw length                    | 19.1                     | 12.0–18.9 | 9.2–10.2  | 10.4–11.2    | 11.6–16.3               | 9.1–10.6  | 19.4                 | 12.2–13.1 |
| Body depth at P <sub>2</sub> origin | 17.8                     | 16.9–19.5 | 17.3–19.0 | 16.9–19.3    | 16.8–18.9               | 16.6–19.0 | 20.3                 | 19.8–21.9 |
| Body depth at A origin              | 14.5                     | 14.8–16.7 | 13.8–17.1 | 14.2–15.7    | 14.2–16.4               | 13.3–16.9 | 15.5                 | 15.8–17.7 |
| Depth at caudal peduncle            | 9.2                      | 8.7–9.8   | 8.7–9.5   | 8.7–9.6      | 8.9–10.2                | 8.5–9.7   | 9.0                  | 8.4–10.0  |
| Length of caudal peduncle           | 20.1                     | 18.9–20.3 | 18.9–20.2 | 18.6–19.3    | 18.7–20.8               | 17.8–21.2 | 18.5                 | 18.2–19.6 |
| Predorsal length                    | 37.2                     | 37.2–39.1 | 35.5–38.8 | 37.6–38.8    | 38.5–40.5               | 37.7–40.1 | 37.9                 | 36.9–38.3 |
| Preanal length                      | 55.9                     | 54.5–56.7 | 53.2–56.6 | 53.8–55.5    | 56.1–58.9               | 56.9–59.5 | 59.4                 | 57.4–59.4 |
| Pre-anal-fin length                 | 59.9                     | 58.5–60.5 | 58.7–61.0 | 58.4–59.9    | 60.3–62.6               | 61.2–64.5 | 63.9                 | 62.2–64.6 |
| Length of D <sub>1</sub> base       | 18.8                     | 17.8–19.0 | 17.9–20.4 | 16.5–19.3    | 16.8–19.0               | 17.6–20.0 | 20.0                 | 20.3–21.0 |
| Length of D <sub>2</sub> base       | 28.3                     | 26.8–28.6 | 26.5–29.0 | 26.9–28.0    | 25.2–26.9               | 24.8–28.3 | 26.9                 | 26.5–28.4 |
| Length of A base                    | 24.3                     | 23.4–24.6 | 22.3–24.9 | 23.1–25.9    | 20.3–23.1               | 20.0–22.7 | 21.2                 | 20.0–21.9 |

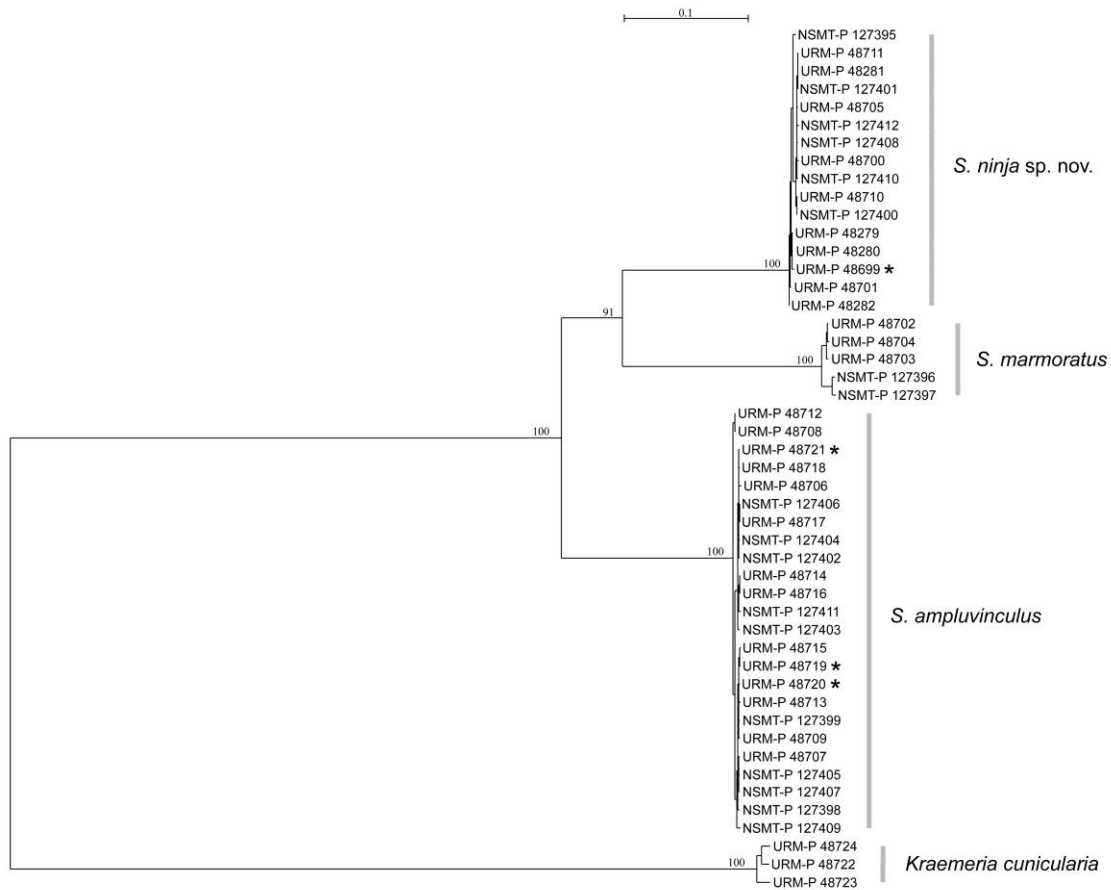
|                       |      |           |           |           |           |           |      |           |
|-----------------------|------|-----------|-----------|-----------|-----------|-----------|------|-----------|
| D <sub>1</sub> length | 18.8 | 17.8–20.2 | 17.9–20.4 | 17.6–19.3 | 16.8–19.0 | 17.6–20.0 | 20.0 | 20.3–21.0 |
| D <sub>2</sub> length | 37.5 | 33.9–37.4 | 32.7–37.2 | 33.3–34.5 | 31.3–33.7 | 31.4–34.3 | 33.7 | 33.8–35.2 |
| A length              | 32.9 | 31.1–32.7 | 30.1–31.7 | 29.7–31.0 | 27.3–29.3 | 26.0–29.3 | 28.7 | 26.0–29.3 |
| C length              | 23.4 | 21.5–24.2 | 20.1–22.9 | 22.3–23.0 | 20.3–23.2 | 20.0–23.5 | 20.6 | 20.3–22.8 |
| P <sub>1</sub> length | 21.7 | 21.0–25.1 | 21.0–25.0 | 22.3–24.7 | 20.5–24.0 | 21.0–24.6 | 22.1 | 21.5–23.9 |

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*D*<sub>1</sub> first dorsal fin, *D*<sub>2</sub> second dorsal fin, *A* anal fin, *C* caudal fin, *P*<sub>1</sub> pectoral fin, *P*<sub>2</sub> pelvic fin

**Table 3** Number of pectoral-fin rays of *Schismatogobius ninja* sp. nov., *S. ampluvinculus*, and *S. marmoratus* collected in Japan, compared with holotypes of *Gobiosoma marmoratum* Peters 1868 (= *S. marmoratus*), *Schismatogobius bruynisi* de Beaufort 1912, and *Schismatogobius roxasi* Herre 1936

|                                  | 14 | 15 | 16 | 17 |
|----------------------------------|----|----|----|----|
| <i>S. ninja</i> sp. nov. (Japan) | 3  | 12 | -  | -  |
| <i>S. ampluvinculus</i> (Japan)  | -  | 17 | 7  | -  |
| <i>S. marmoratus</i> (Japan)     | -  | -  | 5  | -  |
| <i>G. marmoratum</i> (holotype)  | -  | -  | 1  | -  |
| <i>S. bruynisi</i> (holotype)    | -  | 1  | -  | -  |
| <i>S. roxasi</i> (holotype)      | -  | -  | -  | 1  |



**Fig. 1** Maximum likelihood phylogenetic tree with 100 bootstrap using the aligned 16,177 bp of mitochondrial genomes of 45 specimens of *Schismatogobius* collected from the Ryukyu Archipelago with *Kraemeria cunicularia* as an outgroup taxon. The scale bar indicates 0.1 substitutions per site. Specimens of *Schismatogobius* with an asterisk were collected on Iriomote Island; all others except *K. cunicularia* were collected on Okinawa Island

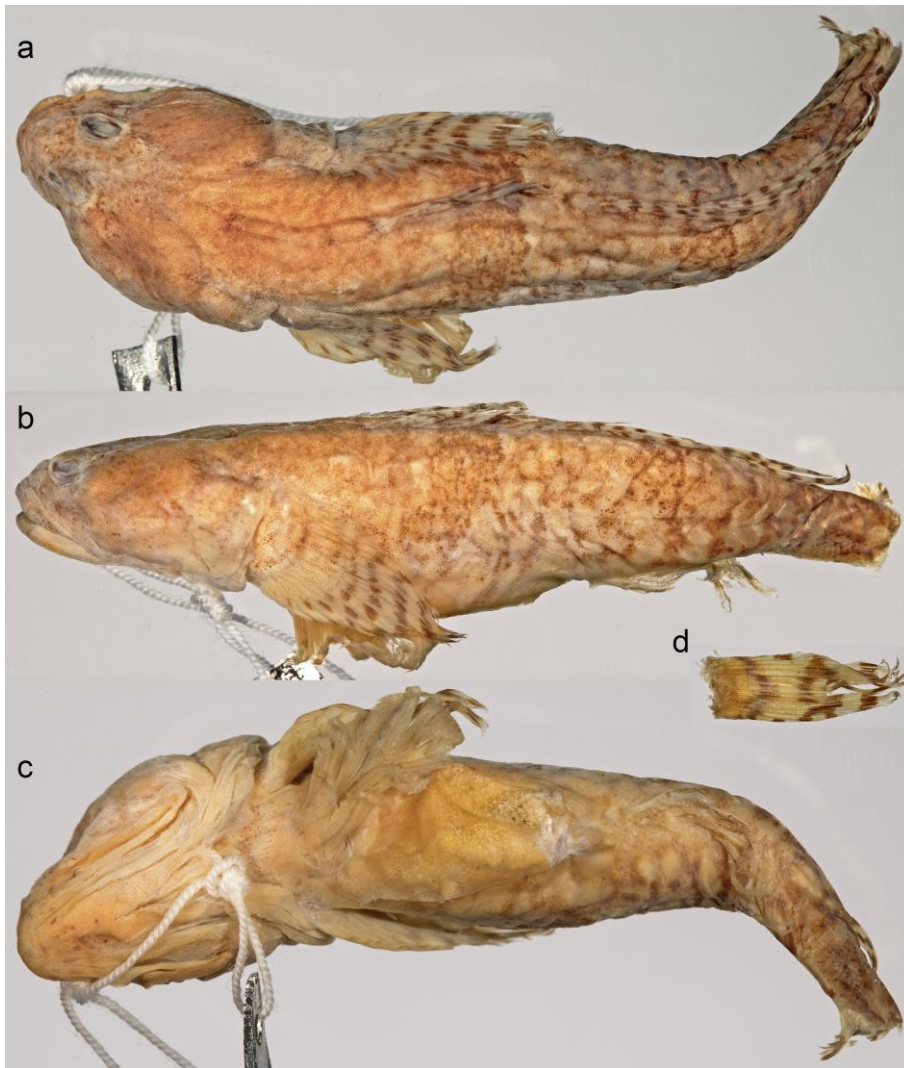


**Fig. 2** Holotype of *Gobiosoma marmoratum* Peters 1868 (ZMB 6756, female, 33.3 mm SL). **a** dorsal view, **b** lateral view, **c** ventral view (photo by K. Maeda)

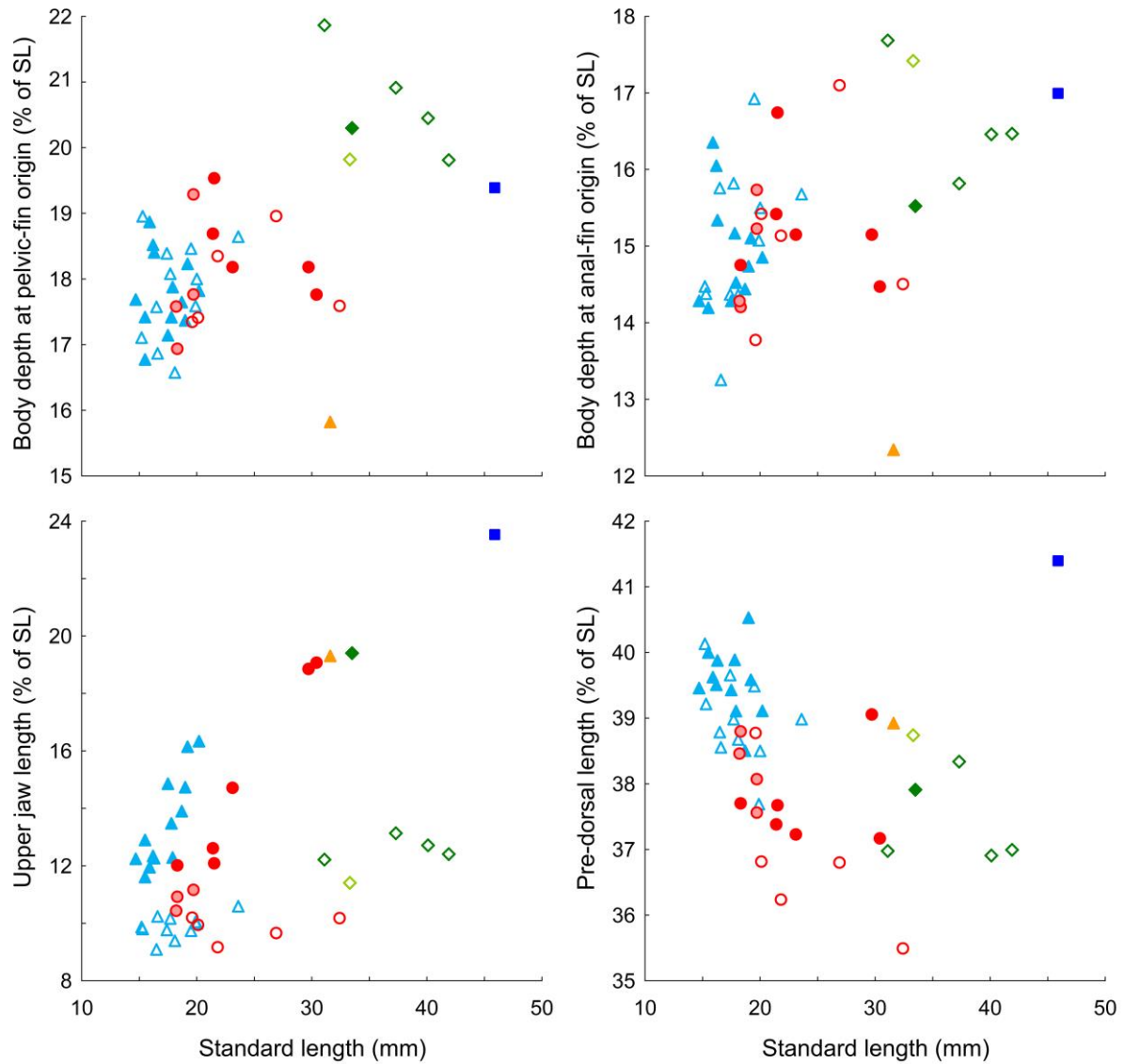


**Fig. 3** Holotype of *Schismatogobius bruynisi* de Beaufort 1912 (ZMA 111196, male, 31.6 mm SL). **a** dorsal view, **b** lateral view, **c** ventral view (photo by K. Maeda)

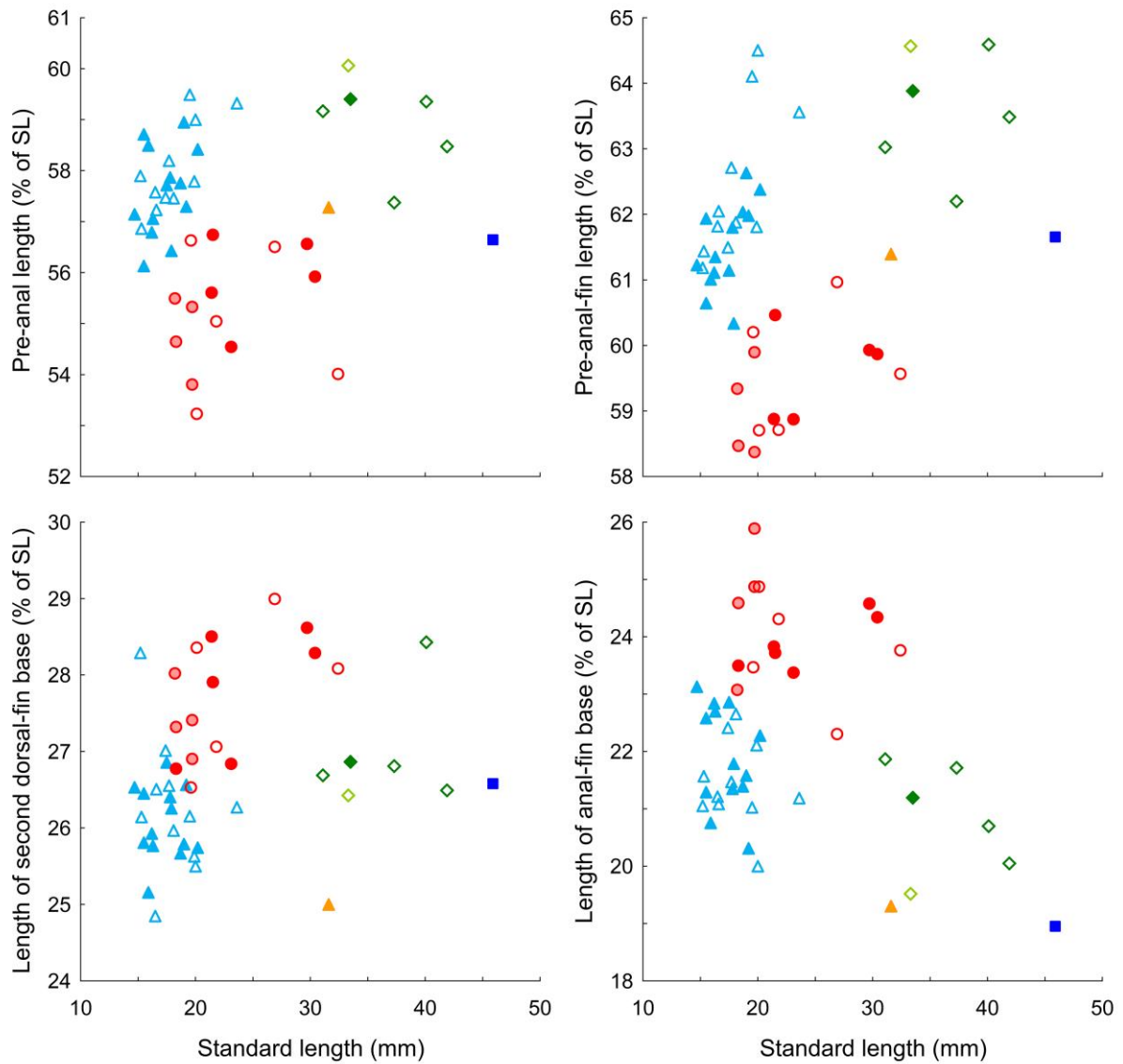




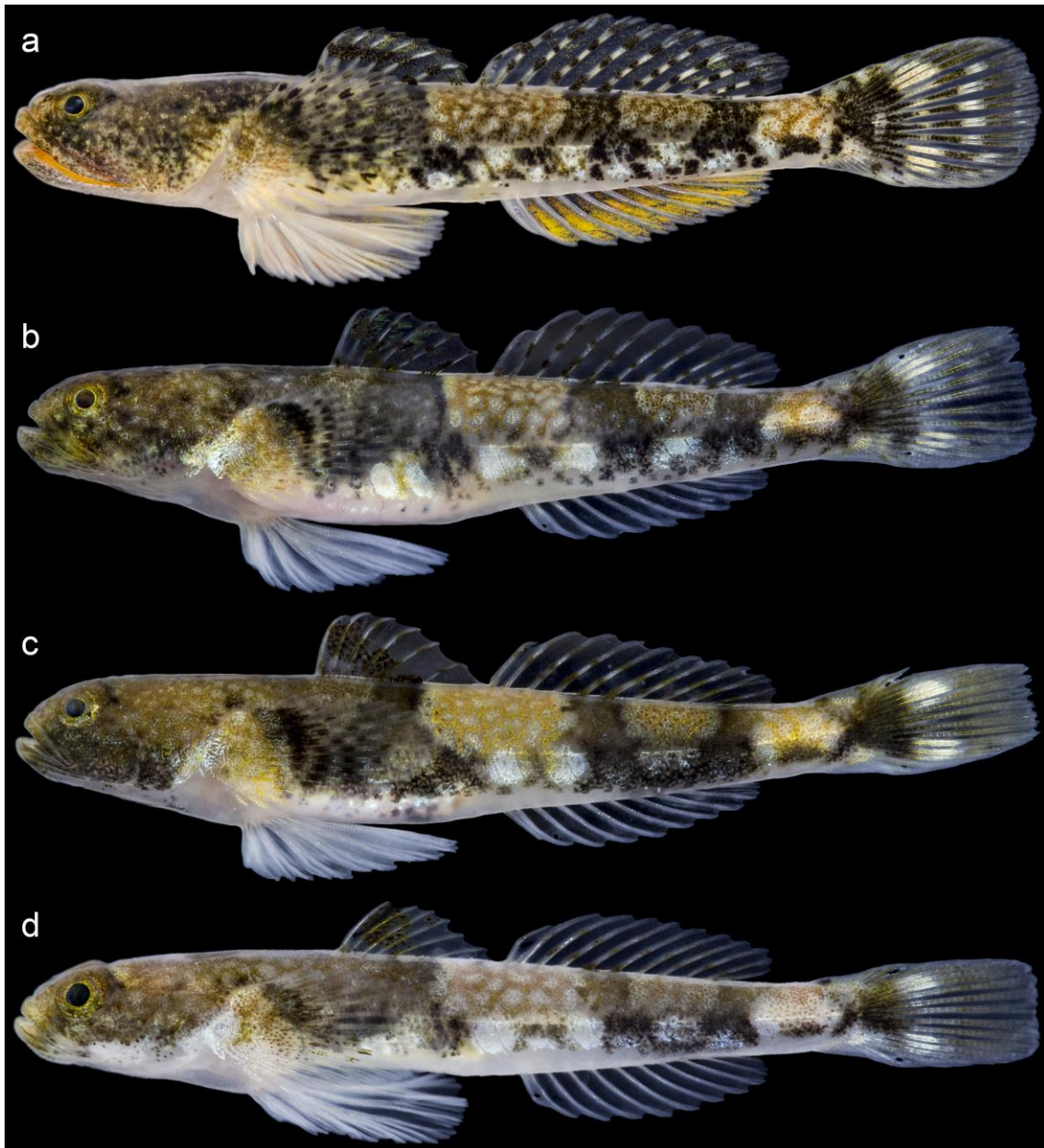
**Fig. 4** Holotype of *Schismatogobius roxasi* Herre 1936 (CAS-SU 30968, male, 45.9 mm SL). **a** dorsal view, **b** lateral view, **c** ventral view, **d** lateral view of caudal fin preserved as a separate piece (photo by K. Maeda)



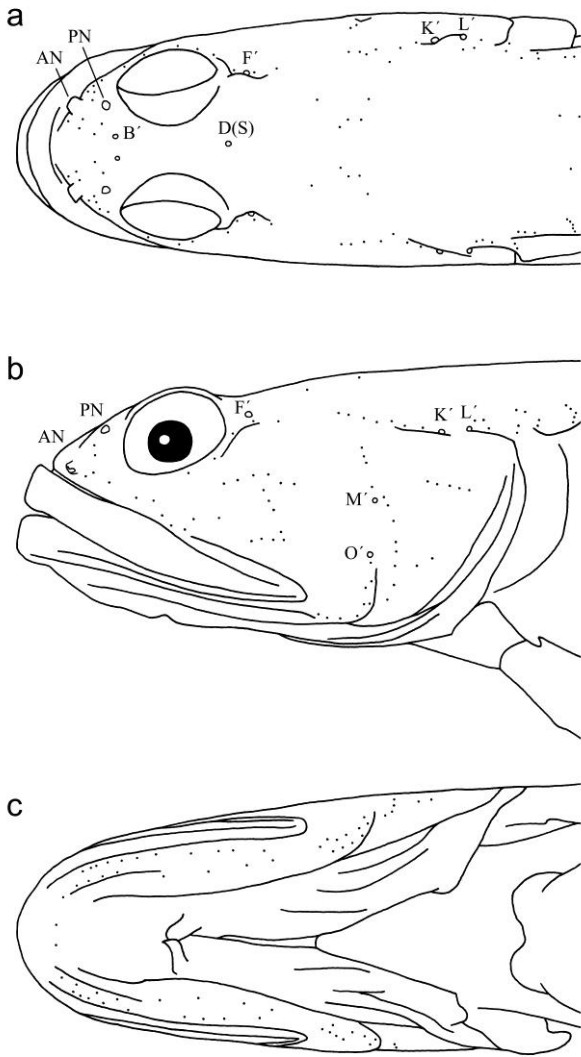
**Fig. 5** Body depths at pelvic- and anal-fin origins, upper jaw length, and predorsal length of *Schismatogobius ninja* sp. nov. (red circles), *S. ampluvinculus* (light blue triangles), *S. marmoratus* from Japan (deep green diamonds), *G. marmoratum* holotype (light green diamond), *S. roxasi* holotype (deep blue square), and *S. bruynisi* holotype (yellow triangle), expressed as a percentage of standard length (SL). Solid and open symbols represent males and females, respectively. Red circles with pink inside represent individuals of which sex is unknown



**Fig. 6** Preanal length, pre-anal-fin length, length of second dorsal- and anal-fin bases of *Schismatogobius ninja* sp. nov. (red circles), *S. ampluvinculus* (light blue triangles), *S. marmoratus* from Japan (deep green diamonds), *G. marmoratum* holotype (light green diamond), *S. roxasi* holotype (deep blue square), and *S. bruynisi* holotype (yellow triangle), expressed as a percentage of standard length (SL). Solid and open symbols represent males and females, respectively. Red circles with pink inside represent individuals of which sex is unknown



**Fig. 7** *Schismatogobius ninja* sp. nov. immediately after fixation. **a** NSMT-P 127395, holotype (male, 30.4 mm SL), **b** URM-P 48711, paratype (male, 21.5 mm SL), **c** NSMT-P 127408, paratype (female, 21.8 mm SL), **d** NSMT-P 127400, paratype (19.6 mm SL) (photo by K. Maeda)



**Fig. 8** Diagrammatic illustration of head showing arrangement of cephalic sensory pores and cutaneous sensory papillae in *Schismatogobius ninja* sp. nov. (URM-P 48701). **a** dorsal view, **b** lateral view, **c** ventral view, **AN** anterior nostril, **PN** posterior nostril



*Schismatogobius ninja*

male



female



*Schismatogobius ampluvinculus*

male



female



*Schismatogobius marmoratus*

male

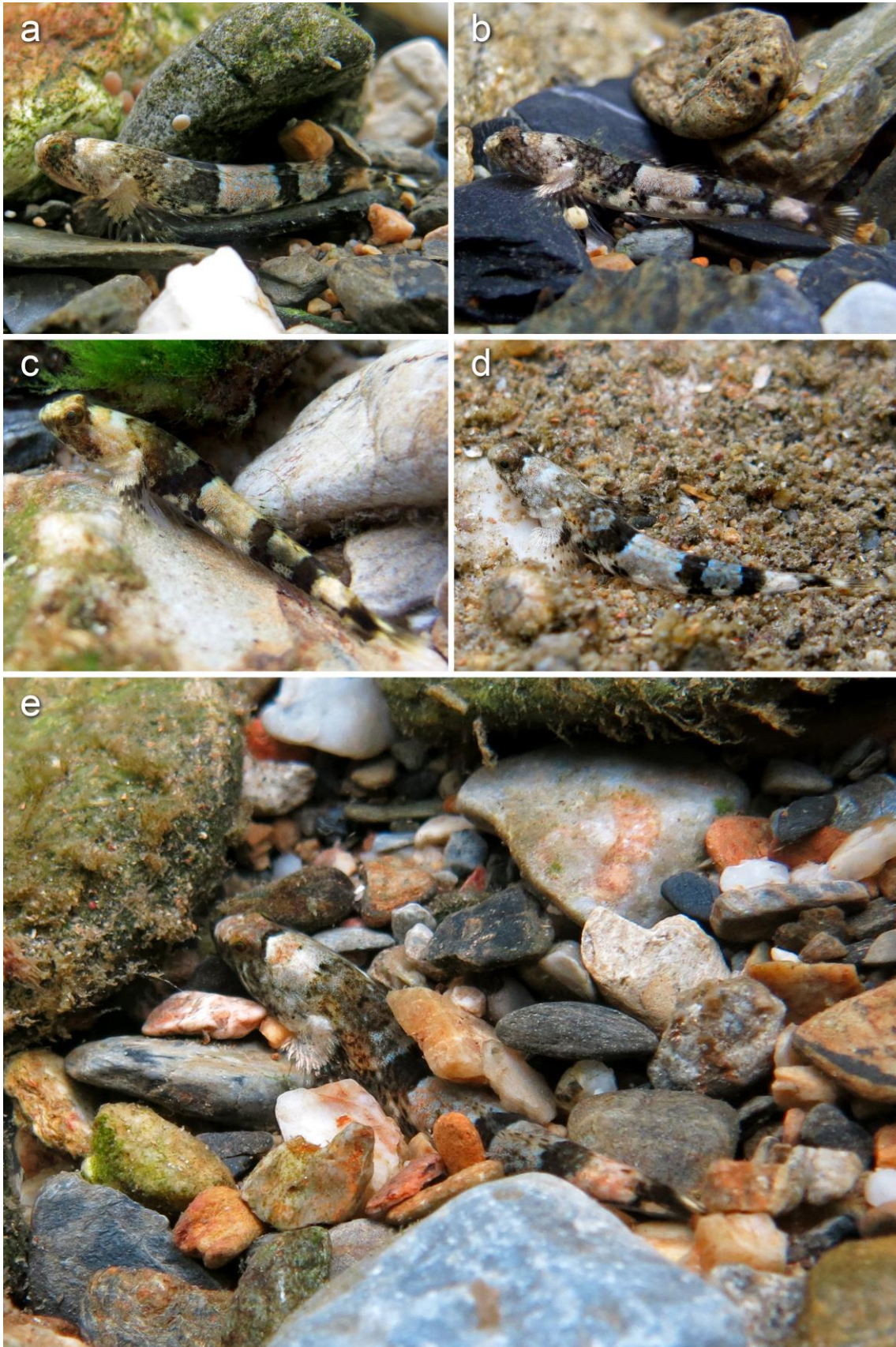


female



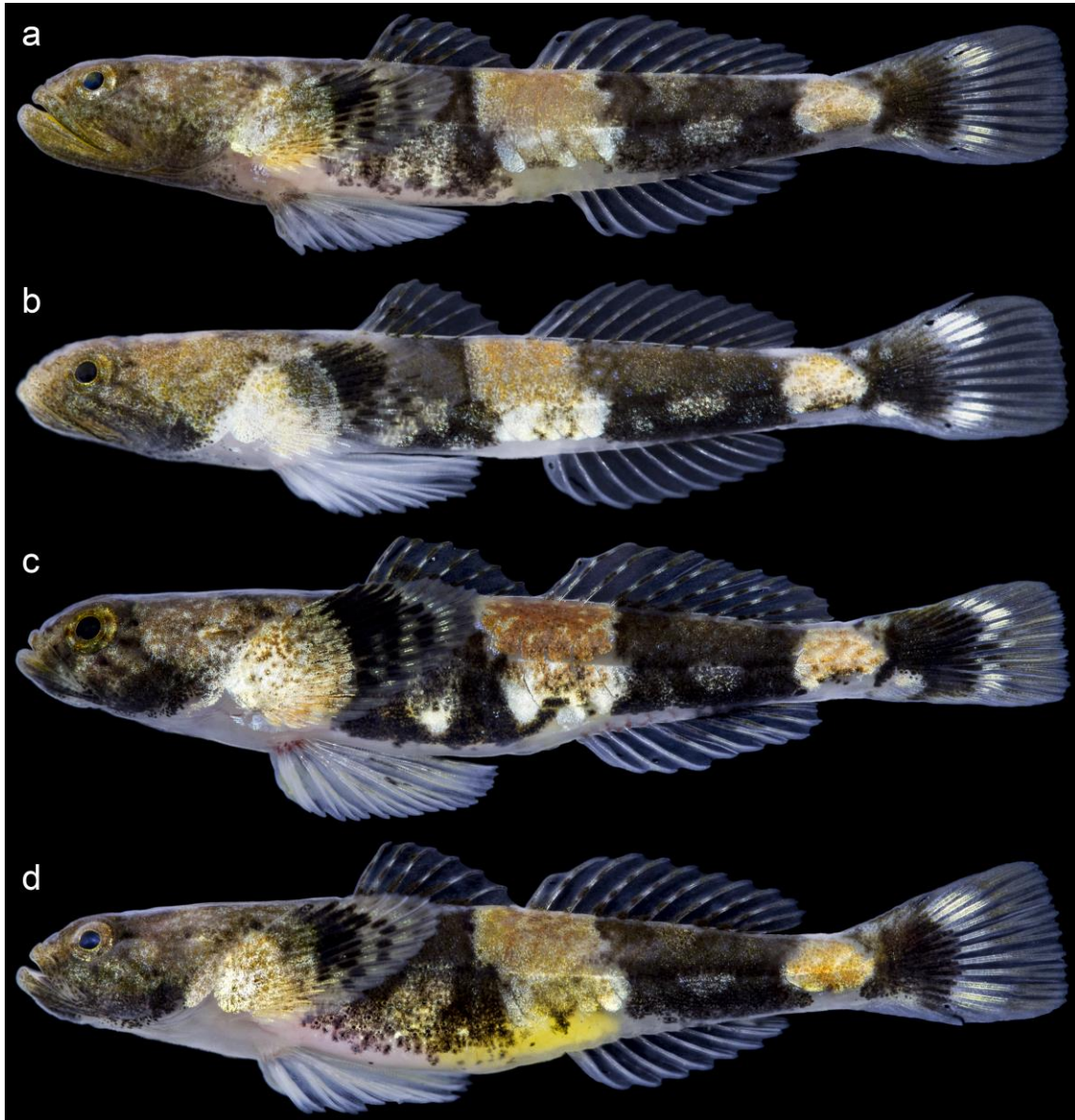
**Fig. 9** Ventral surface of head and trunk of *Schismatogobius ninja* sp. nov. (**a** NSMT-P 127395, 30.4 mm SL; **b** NSMT-P 127410, 21.4 mm SL; **c** URM-P 48710, 26.9 mm SL; **d** NSMT-P 127408, 21.8 mm SL), *S. ampluvinculus* (**e** URM-P 48720, 20.2 mm SL; **f** NSMT-P 127407, 17.9 mm SL; **g** URM-P 48719, 23.6 mm SL; **h** NSMT-P 127405, 19.9 mm SL), and *S. marmoratus* (**i** URM-P 48702, 33.5 mm SL; **j** NSMT-P 127396, 37.3 mm SL; **k** URM-P 48703, 41.9 mm SL), photographed after preservation (photo by K. Maeda)





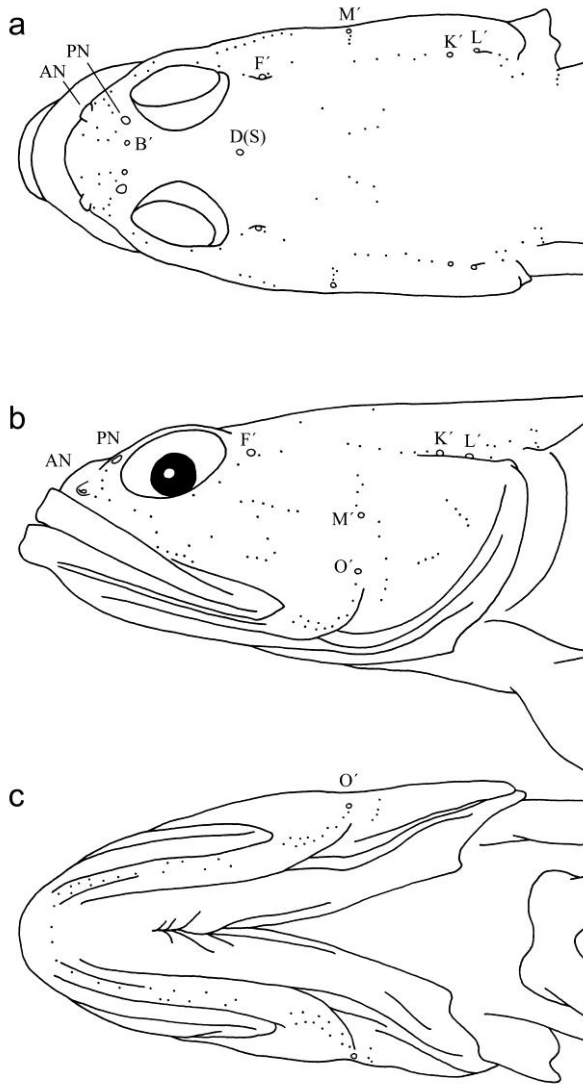
**Fig. 10** Live *Schismatogobius ninja* sp. nov. observed in streams on Okinawa Island (**a, d, e** 16 Mar. 2015; **b** 11 Oct. 2010; **c** 16 June 2008) (photo by K. Maeda)





**Fig. 11** *Schismatogobius ampluvinculus* immediately after fixation. **a** URM-P 48716 (male, 19.0 mm SL), **b** NSMT-P 127402 (male, 17.5 mm SL), **c** URM-P 48719 (female, 23.6 mm SL), **d** URM-P 48707 (19.5 mm SL) (photo by K. Maeda)





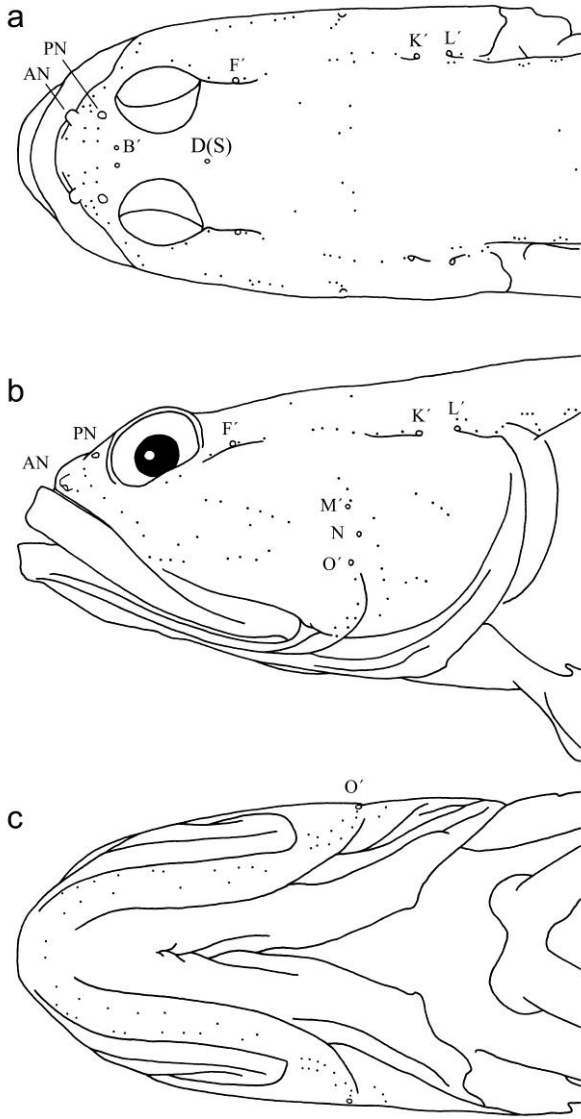
**Fig. 12** Diagrammatic illustration of head showing arrangement of cephalic sensory pores and cutaneous sensory papillae in *Schismatogobius ampluvinculus* (URM-P 48720). **a** dorsal view, **b** lateral view, **c** ventral view, **AN** anterior nostril, **PN** posterior nostril



**Fig. 13** Live *Schismatogobius amphuvinculus* observed in streams on Okinawa Island (**a** 16 June 2015, **c** 13 Aug. 2015) and Iriomote Island (**b** 10 Aug. 2006), and *Schismatogobius marmoratus* observed in streams on Okinawa Island (**d**, **e** 16 Mar. 2015; **f** 29 Aug. 2009) (photo by K. Maeda)



**Fig. 14** *Schismatogobius marmoratus* immediately after fixation. **a** URM-P 48702 (male, 33.5 mm SL), **b** NSMT-P 127396 (female, 37.3 mm SL), **c** URM-P 48703 (female, 41.9 mm SL) (photo by K. Maeda)



**Fig. 15** Diagrammatic illustration of head showing arrangement of cephalic sensory pores and cutaneous sensory papillae in *Schismatogobius marmoratus* (URM-P 48702). **a** dorsal view, **b** lateral view, **c** ventral view, **AN** anterior nostril, **PN** posterior nostril