Systematic revision of the Japanese freshwater snail *Semisulcospira decipiens* (Mollusca: Semisulcospiridae): Implications for diversification in the ancient Lake Biwa

Naoto Sawada[A*] and Yusuke Fuke[A]

*Correspondence to: Naoto Sawada
Department of Zoology, Graduate School of Science, Kyoto University, Kyoto 606-8502, Japan
Email: sawada@zoo.zool.kyoto-u.ac.jp

Running title
Systematic revision of *Semisulcospira decipiens*

urn:lsid:zoobank.org:pub:A83B99F4-8709-4295-86B3-A6C595D65DA0

ABSTRACT

Semisulcospira is a freshwater snail genus highly divergent in the ancient Lake Biwa, Japan, with a history of approximately four million years. Although the shell morphology, karyotype, and molecular phylogeny of the genus have been well studied, the systematic status of several non-monophyletic species remains uncertain. In this study, we have evaluated the taxonomic accounts of the species previously identified as *Semisulcospira decipiens*, *S. habei*, and their relatives. We examined their genetic relationships using genome-wide SNP data and elucidated morphological variation among them using Random Forest classification. Morphological relationships between the name-bearing type of *S. decipiens* and the newly collected specimens were also evaluated. Morphological characteristics effectively discriminated between the nine genetic clusters, and the correlation among morphology and the substrates was elucidated. Taxonomic accounts of *S. decipiens*, *S. habei*, *S. arenicola*, *S. nakasekoae*, and *S. ourensis* were revised in the present systematics with synonymization of *S. multigranosa*, *S. habei yamaguchi*, and *S. dilatata* under *S. decipiens* and *S. fluvialis* under *S. nakasekoae*. We also described two new species, *Semisulcospira elongata* sp. nov. and *Semisulcospira cryptica* sp. nov. and redefined two phylogroups of the lacustrine species as the *Semisulcospira niponica*-group and the *Semisulcospira nakasekoae*-group. Traits of the species examined exhibiting intraspecific variation in the different substrates and flow velocity may indicate their morphological and trophic adaptations. The habitat-related variation has certainly caused the taxonomic confusion of the lacustrine species. Lake drainage contributes to increasing the species diversity of the genus, generating ecological isolation between the riverine and lacustrine habitats.

Keywords: adaptive radiation, ancient lake, Caenogastropoda, MIG-seq, morphology, next-generation sequencing, Random Forest, taxonomy, type specimen, intraspecific variation

Short summary

The systematic status of several *Semisulcospira* species has been uncertain despite their importance in elucidating the adaptive radiation of freshwater gastropods in ancient lakes. We used the genome-wide SNP-based population genetics and the Random Forest classification of the shell morphological traits to clarify the species diversity and delimitation of the genus in Lake Biwa, Japan. Based on the nine genetic clusters being well morphologically discriminated, our systematics successfully arranged taxonomic accounts of 11 known and two new species. The intraspecific variation in their shell and radula morphology highlights their plastic adaptation to various diet, substrates and flow velocities.

Introduction

Semisulcospira Boettger, 1886 is a freshwater snail genus that is widely distributed in Japan, Korea, Taiwan, and China (Davis 1969; Du *et al.* 2019). The genus is the most derived in the family Semisulcospiridae and has been characterized by the synapomorphic trait of the viviparous reproductive mode (Strong and Köhler 2009). The genus has radiated in Lake Biwa, which is the largest lake in Japan with a history of approximately four million years (Setoguchi 2020). Nineteen of the 31 extant species are endemic to the lake, exhibiting significant interspecific diversity in terms of the teleoconch morphology and karyotypes (*e.g.* Society for the Study of Aquatic Life 1989; MolluscaBase 2022; Sawada and Fuke 2022).

The genus is the most speciose mollusc taxa in Lake Biwa, and their high endemicity in the lake, where no other aquatic organism has undergone adaptive radiation, has received considerable scientific attention (*e.g.* Nishino and Watanabe 2000; Tabata *et al.* 2016; Lopes-Lima *et al.* 2020). According to nuclear DNA phylogeny, the lacustrine species has been divided into two phylogroups, the *S. habei*- and the *S. decipiens*-groups (Nomoto 2001; Miura *et al.* 2019). The members of the *S. habei*-group generally possess a more globose teleoconch, and another group has a shell that is more elongated (Watanabe and Nishino 1995).

The past ecological niche differentiation associated with lake expansion has accelerated adaptive radiation of *Semisulcospira* (Miura *et al.* 2019). Members of the genus have been spread to vast shallow sandy beaches, offshore muddy bottoms, and scattered rocky coasts and islands. Different species have advanced into each environment in the lake and the drainage (Watanabe 1984; Watanabe and Nishino 1995). Past genetic introgression and insufficient variation in the allozyme loci have hampered the clarification of their phylogenetic relationships (Kamiya *et al.* 2011; Köhler 2016; Miura *et al.* 2020). However, recent genome-wide SNP analyses have shown that the previous shell morphology-based species delimitation is concordant with the nuclear phylogeny (Miura *et al.* 2019; Sawada and Fuke 2022).

Semisulcospira decipiens (Westerlund, 1883) is the third oldest lacustrine species described after *S. niponica* (Smith, 1876) and *S. biwae* (Kobelt, 1879). *Semisulcospira decipiens* was initially treated as a variation of *S. niponica* with more (5–7) spiral cords on the teleoconch surface (Smith 1876; Kobelt 1879). Westerlund (1883) then described *S. decipiens* as "*Melania niponica* var. *decipiens*" based on a specimen collected by the Swedish Vega Expedition in 1878–1880 from Lake Biwa. The exact type locality of the species is likely to be near the boundary between the north and south lake basin at a water depth of 9 m (Mano in Fig. 1) (Takigawa *et al.* 2020). Three years later, Boettger (1886) described *S. multigranosa* (Boettger, 1886) from a variation of *S. niponica*, questioning its identity with *S. decipiens*. This was supported by subsequent studies (Pilsbry 1902; Annandale 1916), and *S. decipiens* was treated as a species inquirenda (Kuroda 1929). Kuroda (1941) indicated that the *S. multigranosa* may be identical to *S. decipiens*, and then he synonymized *S. multigranosa* under *S. decipiens* (Kuroda 1962).

Taxonomic accounts of the Japanese *Semisulcospira*, including the two species, were arranged by the first comprehensive examination of the genus (Davis 1969). Davis used roundness of the teleoconch for species identification and redefined the two species. Consequently, *S. decipiens sensu* Davis 1969 was characterized by an elongated teleoconch with more spiral cords and a medium-sized protoconch. *Semisulcospira multigranosa* (currently *S. davisi* Sawada & Nakano, 2021) featured similar teleoconch morphology to *S. decipiens sensu* Davis 1969 and a substantially large embryo. He also described *S habei* Davis, 1969 and *S. habei yamaguchi* Davis, 1969, as distinguishable by a more globose teleoconch with many spiral cords. Several species whose teleoconch and protoconch morphology resembles *S. decipiens sensu* Davis 1969 or *S. habei* were later described (Watanabe and Nishino 1995).

Those comparative studies have established taxonomic diagnoses for the lacustrine species. However, the recent revision of the type specimens has also amended accounts of several older species described in the 1800s (Sawada and Nakano 2021; Sawada and Fuke 2022). Broad sampling and genetic analyses have also been used to identify new species from a geographic variation of the known species. Although the type specimen of *S. decipiens* was figured by Habe (1984), its morphological examination has not yet been conducted. Moreover, Matsuoka (1981) pointed out that *S. decipiens* may be identical to *S. habei yamaguchi*. It has also been suggested that the specimens identified as *S. decipiens sensu* Davis 1969 may not belong to a monophyletic group (Miura *et al.* 2019).

In this study, we revisited the systematic status of *S. decipiens*, *S. decipiens sensu* Davis 1969, *S. habei*, and their relatives. In addition to morphological examination of the name-bearing types, we conducted investigations on the teleoconch, protoconch, radula, and the genitalia morphology, and the population genetic structure of the newly collected specimens. The present analyses have clarified the genetic relationships among the nine valid species and elucidated the systematic status of the 13 nominal taxa. The two phylogroups in the lake were also redefined as the *Semisulcospira niponica*- and the *S. nakasekoae*-groups. The present results provide new insights into the phylogeny, morphology, and biogeography of the lacustrine *Semisulcospira*.

Materials and methods

Samples

A total of 628 *Semisulcospira* specimens were newly collected by the first author via snorkel and dredging from 29 localities in Lake Biwa, central Japan including the *S. niponica*-group from 17 sites and the *S. nakasekoae*-group from 21 sites (Fig. 1). The specimens were morphologically identified following Boettger (1886), Davis (1969), and Watanabe and Nishino (1995): *S. multigranosa* from creaks at Ebie and Lake Matsunoki, *S. habei* from the Uji River, *S. habei yamaguchi* from all sites in Lake Biwa and the upstream of the Seta River except at Ebie, Imazu Beach, Lake Matsunoki, and Iso, *S. dilatata* Watanabe & Nishino, 1995 from Iso, *S. rugosa* Watanabe & Nishino, 1995 from Kitafunaki and Imazu Beach, *S. reticulata* Kajiyama & Habe, 1961 from Mano (Fig. 1a); *S. nakasekoae* (Kuroda, 1929) from the Uji and Yodo Rivers and the Lake Biwa Canal, *S. decipiens sensu* Davis 1969 from Minamihama to Otsu Port in Lake Biwa, *S. arenicola* Watanabe & Nishino, 1995 *sensu stricto* from Satsuma and Tamura, *S. fluvialis* Watanabe & Nishino 1995 from Nango, *S. ourensis* Watanabe & Nishino 1995 (previously *S. ourense*; see Systematics) from Oura and Sugaura (Fig. 1b). The *S. nakasekoae*-group specimens that could not be identified to the known species were also obtained at several sites. The snails were collected from rocky, piled rock, sandy, and muddy bottoms and concrete blocks around the lakeside, islands, drainage, and the canal at a water depth of 0–12 m (Table 1, S1–S2). The Seta, Uji, and Yodo Rivers are the names of the specific sections of the sole contiguous lake drainage (Nakamura *et al.* 2020).

The following specimens were used for morphological examination: 117 mature females, 40 males, and four juveniles of *S. decipiens* (including *S. multigranosa*, *S. habei yamaguchi*, and *S. dilatata*; see Systematics); 40 females and five males of *S. habei*; 24 females and one male of *S. rugosa*; four females, four males, and two juveniles of *S. reticulata*; 106 females, 31 males, and six juveniles of *S. arenicola* (including *S. decipiens sensu* Davis 1969 from outside the northern part of Lake Biwa); 86 females and 33 males of *S. nakasekoae* (including *S. fluvialis*); 31 females and 11 males of *S. ourensis* (including *S. decipiens sensu* Davis 1969 from the northern part of Lake Biwa); 29 females, four males, and three juveniles of *S. elongata* sp. nov.; 30 females and seven males of *S. cryptica* sp. nov.; and five females and five males of putative hybrids between *S. arenicola* and *S. nakasekoae*. Among them, 49 of *S. decipiens*, 15 of *S. habei*, six of *S. rugosa*, three of *S. reticulata*, 106 of *S. arenicola*, 33 of *S. nakasekoae*, 14 of *S. ourensis*, 21 of *S. elongata* sp. nov., 16 of *S. cryptica* sp. nov., and four of the putative hybrids were used for the genetic analyses.

Sexual dimorphism and allometric growth have been recorded in the teleoconchs of the genus, and examination of the mature females is reliable (Sawada and Nakano, 2022; Sawada and Fuke 2022). The mature females and males and juveniles were examined separately, and only the females were used for the present morphological analyses. The teleoconchs, protoconchs, radulae, and the reproductive organs were separated, cleaned, and observed following the method described by Sawada and Nakano (2021) and Sawada *et al.* (2021). The foot tip was cut off and preserved in 99% ethanol for the genetic analyses. The newly collected specimens were deposited in the Zoological Collection of Kyoto University (KUZ).

Morphological examinations were also conducted for the holotype of *S. decipiens* preserved in the Invertebrate Collections at the Swedish Museum of Natural History (SMNH), the lectotype of *S. multigranosa* in the Malacological Collection at Senckenberg Naturmuseum, Frankfurt (SMF), the holotypes of *S. habei* and *S. habei yamaguchi* in the Mollusk Collection at University of Michigan Museum of Zoology (UMMZ), the holotypes of *S. dilatata*, *S. arenicola*, *S. fluvialis*, and *S. ourensis* in the Lake Biwa Museum (LBM). Type material of *S. nakasekoae* described by Kuroda (1929) could not be found by the first author's investigation at the malacological collection of the National Museum of Nature and Science, Tokyo (NSMT), Nishinomiya Shell Museum (NSM), and Kyoto University Museum (KUM), where the type specimens of the species may have been preserved (Kikuchi *et al.* 1996; Kikuchi *et al.* 1997; Callomon 2017).

Genetic analyses

Extraction of the genomic DNA and library preparation, sequencing, SNP detection, and estimation of the population structure were conducted for 209 snails following the methods described by Sawada and Fuke (2022), using Multiplexed ISSR Genotyping by sequencing (MIG-seq) analyses (Suyama and Matsuki 2015). Pooled libraries were outsourced to Novogene for 150 bp paired-end sequencing using Illumina NovaSeq 6000. The raw MIG-seq data were deposited in the DDBJ Sequence Read Archive (accession number: DRA014667).

Demultiplexing the raw data was conducted using the "process_shortreads" programme in Stacks v2.59 (Rochette et al., 2019). Low-quality bases (< *Q* 30) and the adapter sequences were removed using fastp v0.20.1 (Chen et al., 2018) and the read length was trimmed to 109 bp to match the shorter Read 1. SNP detection was performed on quality-controlled reads using the "Denovo_map.pl" pipeline of Stacks with the following settings: "paired-end" mode; the minimum depth of coverage was set to five ($m = 5$), and the maximum allowable number of substitutions between stacks was set to three ($M = 3$). SNP filtering and output were conducted using populations with the following settings: only one SNP from a locus ("--write-single-snp") common to more than 60% of all samples ($R = 0.6$) retained; SNPs with heterozygosity greater than 75% ("--max-obs-het" = 0.75) and minor alleles less than two ("--min-mac" = 2) excluded. All other parameters were set to the default setting.

Population genetic structure was estimated by a principal component analysis (PCA) conducted for all the specimens. Subsequently, a PCA was conducted respectively for the *S. niponica*- and the *S. nakasekoae*-groups, because the first PCA separated the specimens into the two phylogroups (see Results). The PCA was performed using PLINK v1.90b6.24 (Purcell *et al.* 2007). Individual admixture proportions were also calculated via the likelihood model-based clustering with ADMIXTURE v1.3.0 (Alexander *et al.* 2009) with the following setting: the number of genetic populations (*K*) was set to 1–10; the convergence criterion (*C*) was set 0.0001. These analyses were repeated 100 times with random seeds, and the optimal K-value was estimated based on the lowest mean cross-validation (CV) error value for each K calculated in the ADMIXTURE. The estimated admixture proportions were visualized using the seed value for $K = 2–5$, where all the analyses estimated lower CV error values (see Results). After separating the two phylogroups, the PCA and the ADMIXTURE analyses were first performed for all the specimens of each phylogroup. The second analyses were conducted for species with unclear population genetic structures due to proximity PC scores within and/or among the groups.

Morphological analyses

The sample numbers of teleoconchs, protoconchs, radulae, and reproductive organs from specimens from each locality used for the morphological analyses are shown in Table 2 and S1–S2. Teleoconch protoconch, radula, and genitalia morphology were examined following the methods in Sawada and Nakano (2021). Reproductive organs were observed under a Leica M125C stereoscopic microscope. After the dissection, radulae were extracted by soaking oral tissues in 1 M sodium hydroxide solution for a day. Extracted radulae were photographed with a Hitachi TM1000 scanning electron microscope.

In addition, sculpture types of teleoconch ("Sculpture Type") defined by Sawada and Fuke (2022, fig. 2) were split based on the dominant type on the penultimate whorl for the Random Forest (RF) analysis below: node type, granulate rib type, smooth rib type, spiral cord type, and smooth type. The "Node Number" and "Spiral Cord Type" of protoconch were newly determined (Fig. 2). The Node Number on the body whorl was counted as one to three in the protoconchs with granulated rib (Fig. 2a) and node (Fig. 2b), and as zero in the ribbed ones (Fig 2c). The Spiral Cord Type were identified as prominent (Fig. 2a), weak (Fig. 2b), and absent (Fig 2c). Measurements of morphological characters were obtained with ImageJ v1.51 (Schneider *et al.* 2012).

Abbreviations of morphological characters examined are as follows: Teleoconch: AH, aperture height; AL, aperture length; ASR, aperture slenderness ratio (the proportion of aperture length to fourth aperture width); AW, aperture width; BCN, basal cord number; BWL, body whorl length; FWL, fourth whorl length; PWL, penultimate whorl length; RN, longitudinal rib number of penultimate whorl; SA, spire angle; SCN, spiral cord number of penultimate whorl; SH, shell height; SW, shell width; TWL, third whorl length; WER, whorl elongation ratio (the proportion of aperture height to fourth whorl length); WN, whorl number. Protoconch: PN, number of protoconchs; RNP, longitudinal rib number on body whorl of the largest protoconch; SHP, shell height of the largest protoconch; SWP, shell width of the largest protoconch; WNP, whorl number of the largest protoconch.

After separating the specimens into the *S. niponica*- and the *S. nakasekoae*-groups, morphological variation among the groups discriminated by the present genetic analyses were explored. The differences in the teleoconchs and protoconchs were detected with the RF classification using the package randomForest v4.6-14 (Andy and Matthew 2002) for R v3.6.1 (R Development Core Team 2019). The RF is a machine learning algorithm using tree predictors generated by bootstrap samplings and useful for the classification using data with categorical variables, such as the current dataset (Breiman 2001). The specimen numbers used for the RF analyses are shown in Table 3 and 4. The following 15 characters were used for the classification: ASR, BCN, BWL, RN, SA, SCN, WER, WN, Sculpture Type, PN, SHP, RNP, WNP, Node Number, and Spiral Cord Type. Intraspecific morphological variation among the substrates was also examined in *S. decipiens*, *S. arenicola*, and *S. cryptica* sp. nov., in which multiple specimens were obtained from both rocky and sandy to muddy substrates. A total of 100,000 trees were generated, given that the out-of-bag (OOB) error rate fully decreased with the large number of trees. The missing values were replaced with the population average. The proximities among individuals were converted to Euclidean distances to visualize the morphological relationships among the groups. The putative hybrids and *S. arenicola* from Yokoehama, where intermediate genetic structures were detected, were not used in the RF analysis (see Results).

The morphological similarity of the juvenile shell of the holotype of *S. decipiens* to the juveniles collected from Mano [presumed type locality of *S. decipiens* (Takigawa *et al.* 2020)] was also evaluated. For the comparison, only specimens with BWL close to the holotype were used because the correlations between body size and diagnoses have been revealed in the genus (Sawada and Nakano 2022).

Results

Genetic analyses

All specimens. A total of 394 SNPs were obtained from the 209 snails. The first PCA generated twenty principal component (PC)s based on all the SNPs, and the PC 1 and 2 explained 65.26% and 4.72% of the total variation, respectively (Fig. S1). The first PC separated the *S. niponica*- and the *S. nakasekoae*-group species. In the *S. niponica*-group, *S. reticulata* and the three other species were divided by the second component.

***Semisulcospira niponica*-group.** 628 SNPs were obtained from the 72 snails and the first PCA generated twenty PCs based on all the SNPs. The first two PCs explained 32.62% of the total variation (Fig. 3). The first and second PCs separated the specimens into four groups, *S. decipiens*, *S. habei*, *S. rugosa*, and *S. reticulata*. The first ADMIXTURE analysis found low mean cross-validation (CV) error values for 1–3 genetic populations, while the optimal number of clusters was two (Table S3). The analysis divided *S. decipiens*, *S. habei*, and a cluster including *S. rugosa* and *S. reticulata* at $K = 3$, and *S. rugosa* and *S. reticulata* were discriminated at $K = 5$ (Fig. 4).

The second PCA and ADMIXTURE analysis was performed for *S. decipiens* and *S. habei* with 622 SNPs. The PC 1 and 2 elucidated 16.65% and 6.88% of the total variation, respectively (Fig. S2). The second PCA segmentalized the specimens into *S. decipiens* and *S. habei* as with the first analysis. The second ADMIXTURE analysis showed low mean CV error values for 1–3 clusters, and the optimal number was detected to be two (Table S4). The clustering from the second ADMIXTURE analysis at $K = 2$ corresponded with the result of the second PCA (Fig. S3).

The specimens of *S. decipiens* from Lake Biwa and the Seta River, which were identified in advance with morphological traits as *S. multigranosa*, *S. habei yamaguchi*, and *S. dilatata*, were not discriminated by the genetic analyses. The present results show that *S. decipiens* are distributed at lake coasts, offshore, islands, and upstream of drainage. The specimens from downstream of the drainage belonged to *S. habei*. The snails from a single population were estimated to originate from a single species at most sites, whereas *S. decipiens* were found sympatrically with *S. rugosa* at Kitafunaki and with *S. reticulata* at Mano.

***Semisulcospira nakasekoae*-group.** 804 SNPs were obtained from the 137 *S. nakasekoae*-group snails. Among the 20 PCs generated, the first and second PCs explained 26.01% of the total variation (Fig. 5). The first two PCs approximately discriminated the specimens into three groups, *S. nakasekoae*, *S. cryptica* sp. nov., and a group comprising *S. arenicola*, *S. ourensis*, and *S. elongata* sp. nov. The ADMIXTURE analysis found low mean CV error values for 2–5 of the genetic populations. Three clusters were predicted to be optimal (Table S5). The analysis separated *S. nakasekoae*, a cluster including *S. elongata* sp. nov. and *S. cryptica* sp. nov., and one comprising *S. arenicola* and *S. ourensis* at $K = 3$ (Fig. 6). The cluster including two new species and another group was divided into independent populations at *K* = 4 and 5, respectively. The first analyses identified intermediate genetic structures between *S. arenicola* and *S. nakasekoae* for the specimens obtained from Araizeki and Nango. Multiple ancestries were also detected in some specimens of *S. arenicola* and *S. elongata* sp. nov. from Okude, Yokoehama, Horikiri Port, and Mano at *K* = 4 and 5.

The second PCA and ADMIXTURE analysis were executed for *S. arenicola*, *S. ourensis*, and *S. elongata* sp. nov. based on 781 SNPs. The first two PCs explained 21.12% of the total variation, identifying three species (Fig. 7). The second ADMIXTURE analysis showed the optimal number of clusters to be one (Table S6). The analysis separated *S. elongata* sp. nov. and a group including *S. arenicola* and *S. ourensis* at *K* = 2 (Fig. 8). The group was almost divided into independent clusters at *K* = 4. The specimens from Yokoehama were composed of multiple ancestry components in *K* = 2 to 5.

The results of the genetic analyses elucidated similar genetic structures of the snails identified morphologically as *S. decipiens sensu* Davis 1969 and *S. arenicola sensu stricto*. The specimens of *S. nakasekoae* and *S. fluvialis* were not distinguished by the analyses. The analyses also identified genetic proximity between *S. ourensis* and the sympatric *S. decipiens sensu* Davis 1969. The distribution of *S. arenicola* and *S. elongata* sp. nov. are predicted to be widespread on lake coasts and offshore, whereas the ranges of *S. ourensis* and *S. cryptica* sp. nov. are restricted to the northern coasts and an island. The drainage and the Lake Biwa Canal are inhabited by *S. nakasekoae*. Several *S. nakasekoae*-group species were found sympatrically: *S. ourensis*, *S. elongata* sp. nov., and *S. cryptica* sp. nov. at Okude; *S. ourensis* and *S. cryptica* sp. nov. at Chikubu-shima Island; *S. arenicola* and *S. elongata* sp. nov at Kitafunaki and Mano.

Morphological analyses

***Semisulcospira niponica*-group.** Morphological characteristics obtained from the teleoconch, protoconch, radula, and the genitalia are shown in Table 2, S1, and S2. The first RF analyses exploring the interspecific variation correctly classified 94.6% of the specimens into four species discriminated by the present genetic analyses. Bootstrap samplings identified 100% of *S. decipiens*, 92.5% of *S. habei*, and 75.0% of *S. rugosa* and *S. reticulata*. The Gini coefficients of the Node Number, Spiral Cord Type, and the RN were larger, significantly contributing to the classification (Table 3). These three characters were important for the morphological discrimination of each species: Node Number for *S. decipiens* and *S. habei*; Spiral Cord Type for *S. rugosa*; Spiral Cord Type and RN for *S. reticulata*. The measurements of RN were fewer in *S. rugosa*, intermediate in *S. decipiens*, slightly larger in *S. habei*, and substantially larger for *S. reticulata* (Table 2). Most specimens possessed one node on the protoconchs in *S. decipiens* and *S. rugosa*, one or two nodes in *S. reticulata*, and two or three nodes in *S. habei*. The dominant spiral cord type of the protoconchs was prominent in *S. decipiens* and *S. habei*, weak in *S. rugosa*, and absent in *S. reticulata*. The Euclidean distances generated from proximities among individuals visualized the morphological similarities of the teleoconch and protoconch among the four species (Fig. 9). The distances overlapped partially between *S. decipiens* and *S. rugosa* and slightly among the other species.

The second RF analysis was conducted for 117 *S. decipiens* obtained from the different substrates. It classified 84.6% of all the specimens. A total of 97.2% rocky, 42.1 % sandy, and 80.8% muddy substrate snails were correctly identified. The characters for BWL, SHP, and WNP showed significant variation among the different substrates (Table S7). The measurements for BWL were smaller on the sandy bottoms, larger in the muddy lakebeds, and variable in the rocky areas (Table S1). The two protoconch characters had smaller values for the sandy areas, larger values for the muddy areas, and intermediate values for the rocky areas.

Considerable intraspecific variation was detected in the number of dental cusps and the proportion of denticle lengths of the radulae. However, a flat tip of the large central cusp of the lateral teeth discriminated *S. rugosa* from the other three species. Pointed tips of the small central denticle of the rachidian and the lateral teeth were characteristic in *S. reticulata*. The central cusp shape of *S. decipiens* was variable among the different substrates. The rachidian are mostly rounded to flat in the rocky habitats and pointed in sandy to the muddy habitats, while the lateral teeth are mostly flat in the rocky areas and rounded in the sandy and muddy areas. No significant interspecific and intraspecific variations were identified in the genitalia morphology of the four species.

***Semisulcospira nakasekoae*-group.** The first RF analyses among the five species correctly distinguished 87.8% of the specimens. Bootstrap samplings correctly sorted 94.7% of *S. arenicola*, 98.9% of *S. nakasekoae*, 54.8% of *S. ourensis*, 69.0% of *S. elongata* sp. nov. and 86.7% of *S. cryptica* sp. nov. Characters of WER, SA, SCN, and BWL effectively contributed to the classification (Table 4). Most of the five species were identified using the four characters and RNP: WER for *S. arenicola*; WER and SA for *S. nakasekoae*; RN and SCN for *S. ourensis*; RNP for *S. elongata* sp. nov.; BWL for *S. cryptica* sp. nov. The measurements of WER and SA were small to intermediate in *S. arenicola*, intermediate in *S. ourensis*, *S. elongata* sp. nov., and *S. cryptica* sp. nov., and large in *S. nakasekoae* (Table 2). The number of spiral cords was fewer in *S. arenicola* and *S. ourensis*, intermediate in *S. elongata* sp. nov. and *S. cryptica* sp. nov., and larger in *S. nakasekoae*. The BWL measurements were slightly smaller in *S. arenicola*, prominently smaller or intermediate in *S. nakasekoae*, intermediate in *S. ourensis*, intermediate to larger in *S. elongata* sp. nov., and larger in *S. cryptica* sp. nov.

The Euclidean distances overlapped largely between *S. arenicola* and *S. ourensis* and partially between *S. arenicola* and *S. cryptica* sp. nov. (Fig. 10). The distances of *S. elongata* sp. nov. were intermediate among *S. arenicola* and *S. cryptica* sp. nov. and significantly overlapped those of *S. cryptica* sp. nov. *Semisulcopira nakasekoae* was found to be distinguishable from the other four species.

The second RF analysis examining morphological variation of *S. arenicola* among the different substrates identified 86.3% of all the specimens and 88.2% rocky, 100 % sandy, and 21.4% muddy snails. The number of longitudinal ribs showed significant variation among the different substrates (Table S8). It was fewer in the rocky lakebeds, larger in the sandy and muddy lakebeds (Table S2).

The intraspecific variation of *S. cryptica* sp. nov. was also examined. The analysis correctly separated all the specimens, and the characters of RN, WN, RNP, and ASR were identified to be important (Table S9). The measurement of RN and RNP was fewer on the rocky bottoms and larger on the sandy ones, while the WN and ASR values had the opposite tendency (Table 2).

A flat tip on the large central cusp of the lateral teeth discriminated *S. nakasekoae* from the other three species. Pointed tips of the central denticle of the rachidian and the lateral teeth were also characteristic of *S. elongata* sp. nov. The central denticle of rachidian is pointed and that of the lateral teeth is rounded in *S. cryptica* sp. nov.

Interspecific and intraspecific variations were not detected in the reproductive organ morphology of the five *S. nakasekoae*-group species.

Type specimen of *Semisulcospira decipiens*. The juveniles of four species, *S. decipiens*, *S. reticulata*, *S. arenicola*, and *S. elongata* sp. nov., were obtained from the presumed type locality of *S. decipiens*. The newly collected specimens exhibited larger interspecific variation in the measurements of SA, WN, and RN (Table 5). The SA measurements were larger in the holotype of *S. decipiens*, the newly collected *S. decipiens* and *S. reticulata*, while they were smaller in the two other species. The juveniles of *S. reticulata* possessed the fewer WN than the other specimens. The RN measurements of the holotype of *S. decipiens* were intermediate between the newly collected *S. decipiens* and *S. reticulata*. According to the combination of the SA and the WN, it has been estimated that the newly collected specimens of *S. decipiens* are most similar morphologically to its type specimen.

Discussion

Genetic relationships and the biogeographical implications

The present genetic and morphological study revealed the species diversity and delimitation of the *Semisulcospira niponica*- and the *S. nakasekoae*-groups. The PCA detected the four *S. niponica*-group and the five *S. nakasekoae*-group clusters. The results of the ADMIXTURE analyses almost corresponded with the PCA result in $K = 2$ to 5. Although the optimal numbers of clusters estimated by the ADMIXTURE analysis were less than the number of groups identified by the PCA, the nine groups detected by the genetic analyses were also highly distinct in their traits for the teleoconch, protoconch, and radula.

The present investigation identified sympatric occurrences of several populations within the same phylogroup: *S. decipiens* and *S. rugosa* at Kitafunaki; *S. decipiens* and *S. reticulata* at Mano; *S. ourensis*, *S. elongata* sp. nov., and *S. cryptica* sp. nov. at Okude; *S. ourensis* and *S. cryptica* sp. nov. at Chikubu-shima Island; *S. arenicola* and *S. elongata* sp. nov. at Kitafunaki and Mano. Maintenance of their genetic identity suggests reproductive isolation among the groups. *Semisulcospira arenicola* and *S. nakasekoae* likely represent parapatric distribution forming a hybrid zone upstream of the drainage. The low fitness of hybrids in the lacustrine habitat of *S. arenicola* and the riverine ones of *S. nakasekoae* may have caused the outbreeding depression among them.

Closely related species, *S. decipiens* and *S. habei* represented allopatric distribution in the drainage. The allopatry was also observed between *S. arenicola* and *S. ourensis* in the northern lake. The evidence of the reproductive isolation within the two pairs could not be obtained in this study. However, they could be distinguished genetically and morphologically, and the difference in habitat preferences was observed between *S. decipiens* and *S. habei*. According to the genetic isolation and potential ecological isolation discussed above, we consider the present nine genetic groups to be independent species.

The results of the genetic analyses were highly consistent with those of the previous genome-wide SNP analysis based on the RAD-seq analysis (Miura *et al.* 2019). The study showed polyphyly of "*S. decipiens*" and "*S. habei*". Given that the study performed species identification following Davis (1969), the clades, which is composed of "*S. decipiens*" and "*S. arenicola*" from the central to the northern part of the lake, correspond to *S. arenicola* and *S. ourensis* in this study. The "*S. decipiens*" from Otsu and Nango is likely to be *S. nakasekoae* or hybrids between *S. arenicola* and *S. nakasekoae*. In the polyphyletic clade comprising "*S. habei*" from the north coast and the Uji River, the former can be identified here as *S. decipiens,* and the latter are *S. habei*.

The karyotypic relationships among the species strongly corresponded with the present genetic results. Karyotypes of most of the lacustrine species were reported by Burch and Davis (1967), Society for the Study of Aquatic Life (1989), and Takami (2013, 2019). The uniqueness of the karyotypes for *S. rugosa* ($2n$ = 22) and *S. reticulata* ($2n$ = 26), and the commonality of *S. habei yamachi* and *S. dilatata* ($2n$ = 18–20), which could not be genetically distinguished here, were noted by Society for the Study of Aquatic Life (1989) and Takami (2013). The commonality in *S. decipiens sensu* Davis 1969 and *S. arenicola sensu stricto* ($2n$ = 24–26) and the distinctiveness of *S. ourensis* ($2n$ = 28) have also been shown in the present study. However, the karyotypes of *S. nakasekoae* differ significantly among the studies: $2n$ = 26 (*S. nakasekoae sensu stricto*) by Burch and Davis (1967), $2n$ = 38 (*S. nakasekoae sensu stricto*) and $2n$ = 26 (*S. fluvialis*) by Society for the Study of Aquatic Life (1989), $2n$ = 26 (*S. fluvialis*) by Takami (2013), $2n$ = 22 (*S. nakasekoae sensu stricto*) by Takami (2019). Whereas *S. nakasekoae* can exhibit considerable intraspecific variation, artefacts may be included in the previously reported karyotypes. Accordingly, further research is required to elucidate the karyotypic variation in *S. nakasekoae* and other congeners.

Hybridization may occur infrequently in the lacustrine *Semisulcospira*, as suggested by Sawada and Fuke (2022). This is because the putative hybrids were only found at the boundary between the parapatric distribution of *S. arenicola* and *S. nakasekoae*. The results of the ADMIXTURE analysis also suggest gene flows between *S. nakasekoae* and the hybrids. Although the population from Yokoehama was clearly identified by the PCA as being *S. arenicola*, the ADMIXTURE analysis revealed that the genetic structure of the population comprises multiple ancestry components. The genetic relationships between the population and the others should be elucidated.

The present investigation found that *S. decipiens* and *S. arenicola* are widely distributed in Lake Biwa across different substrates, while their sympatric occurrences with closely related species are restricted at several sites. *Semisulcospira niponica* and its relatives infrequently form sympatric distributions, suggesting the possibility of species-specific microhabitat differences or competitive exclusion (Sawada and Fuke 2022). These factors may also contribute to distributional patterns among the species examined in this study.

Different species were distributed in both the *S. niponica*- and the *S. nakasekoae*-groups in the lake and downstream of the drainage. A similar pattern has also been observed between loach subspecies indigenous to the water system (Nakajima 2012). Differences between the lacustrine and the riverine habitats may have caused ecological isolation among the species and contributed to increasing species diversity of *Semisulcospira*.

Morphology

The nine species examined in this study could be distinguished using a combination of morphological traits of the teleoconch, protoconch, and radula. However, significant variation was observed in several characters of *S. decipiens*, *S. arenicola*, and *S.*

cryptica sp. nov. on the different substrates. The previous phylogenetic study supported morphology-based species delimitation in the genus (Miura *et al.* 2019), and the characteristics of the teleoconch discriminated closely related genetic clusters better than the protoconch and radula (Sawada and Fuke 2022). In contrast, this was not the case for the present species. The several teleoconch characters represent variation in the different substrates, and the protoconch and radula morphology were more reliable for species discrimination among the several species. The RF analysis also revealed differences in the morphological diversification patterns between the present *S. niponica*- and the *S. nakasekoae*-group species. Protoconchs were more useful in the *S. niponica*-group species, whereas teleoconchs were more important in the *S. nakasekoae*-group species.

The teleoconch morphology of freshwater gastropods can diversify in response to the predation pressure and calcium availability (Covich 2010). Substrate differences have been suggested to play a role in the variation of the teleoconch sculpture and the radula morphology (Rintelen *et al.* 2004). Despite the presence of fish, turtles, and crustaceans, which are potential predators of freshwater gastropods, the density of semisulcospirids in Lake Biwa is substantially high (Yusa *et al.* 2006; Nishino and Tanida 2018; Scientific Committee for Research into the Wildlife in Shiga Prefecture 2021). The calcium content of the lake water is uniformly low (Negoro 1957). Therefore, in the lacustrine *Semisulcospira*, relationships between the species composition and the substrates rather than other factors have been noted (Nishino and Watanabe 2000; Miura *et al.* 2019). The difference in substrates, in addition to the genetic background, affects the frequency of longitudinal ribs in the riverine *Semisulcospira* (Urabe 2000). Correlation between strong water flow and teleoconchs with a larger aperture and lower spires has been clarified in riverine *S. reiniana* (Brot in Kobelt, 1876) (Urabe 1998). As discussed below, relationships between environmental factors and shell and radula morphology were observed among the populations examined in this study.

The longitudinal ribs on the teleoconch were coarser and more pronounced in the rocky areas and finer and weaker on the muddy lakebed areas in *S. arenicola*, *S. ourensis*, and *S. cryptica* sp. nov. In the sandy substrates, the ribs of *S. arenicola* were further indistinct, and some snails did not have any longitudinal ribs. This is likely to be a general pattern in lacustrine species, given that this trend has been observed in other species (Watanabe 1984; Sawada and Nakano 2021; Sawada and Fuke 2022): rugged sculptures in rupicolous *S. niponica*, *S. watanabei* Sawada in Sawada & Fuke, 2022, *S. salebrosa* Sawada in Sawada & Fuke, 2022, *S. nakanoi* Sawada in Sawada & Fuke, 2022, and *S. morii* Watanabe, 1984; fine ribs in muddy *S. reticulata* and *S. davisi*. While elimination of the ribs was also observed in *S. decipiens* from the sandy area, a significant difference was not observed in the rib intensity between the rocky and muddy lakebeds. Both the smooth types of *S. decipiens* (described as *S. dilatata*) and *S. arenicola* (*S. arenicola sensu stricto*) are found on the shallow sandy beaches, where the snails are exposed to rough waves. Given that snails with smooth shell surfaces possess higher resistance to water currents (Holomuzki and Biggs 2006), wave-induced sculpture dissipation may have occurred in parallel in the two phylogroups.

Substantial differences were detected in several characters of shell roundness (SA), size (BWL), and the growth rate (WER) in *S. nakasekoae* among the sites. In freshwater gastropods, intense water currents have been suggested to be associated with a more rounded shell with a larger aperture (Urabe 1998) and a larger foot size (Verhaegen *et* *al.* 2019). The teleoconch roundness of *S. nakasekoae* is likely to be related to flow velocity given that more globose shells occurred at Uji, where the water current was strong (Kihira *et al.* 2009), and greatly elongated types were found in the muddy, stagnant water area at Fushimi. The population with the intermediate SA values and smooth shell surfaces have been morphologically discriminated as *S. fluvialis*. The SA and WER values for *S. nakasekoae* and *S. decipiens* decreased downstream in the Uji and Yodo Rivers. Although further investigation into the relationship between the shell characters, water flow, and the genetic gradient is needed, the observed tendency may indicate similar selections that the two phylogroups have undergone.

We identified a small-sized population of *S. nakasekoae* in the Lake Biwa Canal, where construction was completed in 1890 (The Lake Biwa Canal Promotion Council 2022). The species seems to migrate into the new habitat from the Uji River and/or Lake Biwa with a reduction in body size. It has been known that in the genus that the number and the size of the protoconchs correlate with the teleoconch size (Takami 1994; Sawada and Nakano 2022). Accordingly, the smaller PN and SHP observed at Higashiyama in the canal are likely to be related to the smaller teleoconchs.

Correlations between the radula morphology and substrates were observed in *S. decipiens*. The radula shape has been suggested to be associated with the substrate and trophic morphology in *Tylomelania* Sarasin & Sarasin 1897, which have radiated in ancient lakes of Southeast Asia (Rintelen *et al.* 2004). As in *Semisulcospira*, it has been shown that rupicolous *S. niponica* and its relatives possess flat tips and *S. davisi* in muddy lakebeds exhibits pointed cusps (Sawada and Nakano 2021; Sawada and Fuke 2022). The present specimens of *S. decipiens* possessed flat to rounded tips in the rocky substrates and pointed to rounded tips in the muddy lakebed areas, exhibiting a similar trend within the species. On the other hand, those relationships could not be ascertained among the snails from the different substrates in *S. arenicola* and *S. cryptica* sp. nov. Therefore, diversification patterns of radula morphology and food habitat may be different between the *S. niponica*- and the *S. nakasekoae*-groups.

According to the morphological variation above, it has been estimated that the presently examined species have expanded to different environments, exhibiting habitat-related variation in their teleoconchs and radulae. The characteristics of shell sculptures and SA were important for the species discrimination in *S. niponica* and its relatives (Sawada & Fuke 2022), whereas these traits were plastic in the present species among the different substrates and flow velocities. The fact suggests that species boundaries have appeared for different characters among the assemblages of closely related species through their different diversification patterns. Their unique radiation patterns have likely caused the historical taxonomic confusion of the lacustrine species.

The protoconchs of *S. decipiens, S. habei*, and *S. rugosa* were similar with rounded to slightly elongated shells and surface nodes. Given that a sister group comprising the three species has been supported by Miura *et al.* (2019), the characteristics of the protoconchs are shared traits of the group. Elongated protoconchs with longitudinal ribs were common in *S. arenicola*, *S. ourensis*, *S. elongata* sp. nov., and *S. cryptica* sp. nov. Although the phylogenetic relationships among the four species should be clarified, the protoconch traits may also be shared among them.

The putative hybrids between *S. arenicola* and *S. nakasekoae* collected from Araizeki and Nango showed the intermediate SA and WER values between the two species. The shell roundness of *S. nakasekoae* may be affected by the flow velocity.

However, the observed morphological differences between sympatric *S. nakasekoae* and the hybrids are likely to reflect their genetic differences.

Systematic status

The type specimen of *S. decipiens* was collected during the Vega Expedition in 1878–1880, and its type locality has been predicted to be around Mano (Takigawa *et al.* 2020). The present investigation collected four *Semisulcospira* species there: *S. decipiens* (previously *S. habei yamaguchi*), *S. reticulata*, *S. arenicola* (*S. decipiens sensu* Davis 1969), and *S. elongata* sp. nov. Although the specimen number was relatively small, the combination of the SA and WN characteristics has estimated that the newly collected *S. decipiens* are most similar to its type specimen. Based on this and the results of the genetic analyses, the systematic status of *S. decipiens sensu stricto*, *S. decipiens sensu* Davis 1969, and the 11 nominal taxa have been established here.

Although *S. decipiens* and *S. arenicola* can be clearly distinguished by their teleoconch roundness (SA) and the protoconch morphology, the original description of *S. decipiens* lacks these traits, and they were first used in the 1960s (Kajiyama and Habe 1961; Davis 1969). *Semisulcospira elongata* sp. nov. was included in the type series of *S. multigranosa* examined by Boettger (1886) [SMF 359900, identified as "*S. decipiens*" by Sawada and Nakano (2021)]. Brief descriptions in the 1800s based on the species delimitation different from the present, and the lack of examination of the type materials seem to have caused confusion in the taxonomic account of the older species.

Systematics

Several studies have proposed supra-specific groups or ranks for the lacustrine *Semisulcospira* species. Davis (1969) introduced the "*Semisulcospira niponica* species group" for six species and one subspecies which can be discriminated from other riverine congeners by a small number of chromosomes, BCN, and PN. The group was raised to the genus "*Biwamelania*" by Habe (1978) without type species designation and a description of the diagnosis. Subsequently, the subgenus "*Biwamelania*" was established by Matsuoka and Nakamura (1981) and was redefined by Matsuoka (1985) because the former study lacked a diagnosis for the subgenus. Nomoto (2001) indicated the non-monophyly of the genus "*Biwamelania*" and proposed the "*Biwamelania habei* species group" and the "*Biwamelania decipiens* species group" under the genus for the two phylogroups detected. Although the "*B. habei* species group" was further split into the "*S.* (*B.*) *habei* group" and the "*S.* (*B.*) *niponica* group" by Kamiya *et al.* (2011), Miura *et al.* (2019, 2020) have followed Nomoto (2001). The subgenus "*Biwamelania*" has not been received by several publications due to its non-monophyly and invalid description (Köhler 2016; Köhler 2017; Sawada and Nakano 2021). Sawada and Fuke (2022) also addressed an assemblage comprising *S. niponica* and its relatives as the "*S. niponica*-group".

Therefore, the delimitation of the phylogroups with independent evolutionary histories has been fluid, and they have not been circumscribed with morphological characteristics. The name "*B. decipiens* species group" is no longer suitable because the present systematics revealed that *S. decipiens* is a member of the "*B. habei* species group". To resolve the confusion in the delimitation and nomenclature of the phylogroups, we have proposed alternative names for the two phylogroups identified by Nomoto (2001). The alternative names are derived from the earliest-named member of each phylogroup following the Article 6.2 and its example of the Code (International Commission on Zoological Nomenclature [ICZN] 1999).

The *Semisulcospira niponica*-group corresponds to the "*Biwamelania habei* species group" introduced by Nomoto (2001). The group can be generally discriminated by globose to slightly elongated teleoconchs (SA of approximately more than 16 degrees) and protoconchs with pointed nodes. The group consists of 10 species: *S. niponica*, *S. decipiens*, *S. reticulata*, *S. kurodai* Kajiyama & Habe, 1961, *S. habei*, *S. rugosa*, *S. fuscata*, *S. watanabei*, *S. nakanoi*, *S. salebrosa*. No characteristics which distinguish the *S. niponica*-group from another group have been detected because the teleoconch and protoconch morphology has been considerably diversified among the species, and the radula and genitalia morphology has been almost preserved within the genus (Sawada and Fuke 2022). However, teleoconchs of the members of the *S. niponica*-group are wider than the other group, except for *S. nakasekoae* and *S. morii* (Watanabe and Nishino 1995). The *S. niponica*-group species also possess pointed nodes on their protoconchs except for *S. reticulata*. Therefore, the group can be discriminated from the other by the combination of these characteristics. This group includes at least two assemblages of close relatives: one composed of *S. niponica*, *S. watanabei*, *S. nakanoi*, *S. salebrosa* and *S. fuscata*; another comprising *S. decipiens*, *S. habei*, and *S. rugosa*.

The alternative name for the "*B. decipiens* species group" is defined as the *Semisulcospira nakasekoae*-group. Moderately to strongly elongated teleoconchs (approximately less than 15 degrees SA) and protoconchs with or without rounded nodes distinguish most of the species in the group. This group comprising nine species: *S. nakasekoae*, *S. morii*, *S. arenicola*, *S. ourensis*, *S. shiraishiensis* Watanabe & Nishino, 1995, *S. takeshimensis* Watanabe & Nishino, 1995, *S. davisi*, *S. elongata* sp.

nov, *S. cryptica* sp. nov. The teleoconchs of the *S. nakasekoae*-group members are narrower than those of the *S. niponica*-group species except for *S. nakasekoae* and *S. morii* (Watanabe and Nishino 1995). The *S. nakasekoae*-group species possess rounded nodes or longitudinal ribs without nodes on their protoconchs except for *S. shiraishiensis* and *S. takeshimensis* (Watanabe and Nishino 1995). The *Semisulcospira nakasekoae*-group can be discriminated from the *S. niponica*-group by the combination of these characteristics. As with the *S. niponica*-group, the *S. nakasekoae*-group is likely to include a species assemblage comprising *S. arenicola*, *S. ourensis*, *S. nakasekoae*, *S. elongata* sp. nov., and *S. cryptica* sp. nov.

The present analyses have clarified genetic and morphological differences among the nine valid species. However, the sample sizes of *S. rugosa* and *S. reticulata* were restricted and therefore, we consider that further examinations are required for the species. Taxonomic accounts of the other seven valid species have been established below.

Family SEMISULCOSPIRIDAE Morrison, 1952

Genus *Semisulcospira* Boettger, 1886

Type species: *Melania libertina* Gould, 1859 by subsequent designation (Wenz 1939).

The genus was originally erected as the subgenus below the genus *Melania* Lamarck, 1799.

Semisulcospira decipiens (Westerlund, 1883)

[Japanese name: Ibo-kawanina Iwakawa 1919]

(Table 2, S1; Fig. 11a–ax, 12a–j)

urn:lsid:zoobank.org:act:ABBCFE14-77DC-439F-9ED5-B73B4CBBA16F

Melania niponica Smith, 1876: 123–124 (part); Brot 1877: 338–339, pl. 34, fig. 10a (part); Kobelt 1879: 131, pl. 19, figs 6, 7, 11, 13, 14 (part).

Melania niponica var. *decipiens* Westerlund, 1883: 56–57 (original description; OD).

Melania (*Semisulcospira*) *multigranosa* Boettger, 1886: 7–8 (part).

Melania multigranosa – Pilsbry 1902: 120 (taxonomic account unknown; TAU); Iwakawa 1919: 82 (TAU); Annandale 1916: 44–45 (part).

Melanoides (*Semisulcospira*) *multigranosa* – Kuroda 1929: 186, 189, pl. 5, figs 34, 35 (part).

Semisulcospira multigranosa – Fukuoka 1933: 114, 117, fig. 4 (part); Sawada and Nakano 2021: 3–6, fig. 3; Sawada and Fuke 2022: fig. S1K, L.

Semisulcospira decipiens – Hirase and Taki, 1951: pl. 82, fig. 14; Kuroda 1962: 86, 89 (part); Habe and Kosuge 1967: 28, pl. 11, figs 19, 20.

Semisulcospira habei yamaguchi – Burch and Davis 1967: 37 (unavailable).

Semisulcospira sp. – Burch 1968: 7–8, fig. 2 (part).

Semisulcospira habei yamaguchi Davis, 1969: 240–243, pl. 3, figs 4–5, pl. 9, figs 11–15 (part); Higo and Goto 1993: 97; Goto and Poppe, 1996: 204; Köhler 2016: fig. 4A.

Biwamelania habei – Habe 1978: 94 (part); Nomoto 2001: 33 (part); Nomoto *et al.* 2001: 418; Nishino and Tanida 2018: 50, 247 (part).

Biwakomelania decipiens – Habe 1984: 306; Kubo 1985: 48.

Semisulcospira (*Biwamelania*) *habei yamaguchi* – Matsuoka 1985: 190.

Semisulcospira habei – Society for the Study of Aquatic Life 1989: 18–19, 49–50, figs 14, 31, 42 (part); Sawada and Fuke 2022: fig. 8E.

Semisulcospira type C – Society for the Study of Aquatic Life 1989: 26–27, 53, figs 18, 32-3, 45.

Semisulcospira (*Biwamelania*) *habei* – Nishino 1991: 11, fig. 10, unnumbered figures; Watanabe and Nishino 1995: fig. 5f, appendix pl. 1, figs 9, 10, appendix pl. 2, figs 24, 25; Nishino and Watanabe 2000: fig. 2-9; Urabe 2007: 80, 84; Kihira *et al.* 2009: 23, unnumbered figures (part); Kamiya *et al.* 2011: 25; Miura *et al.* 2019: fig. S1a (part); Nishino 2021: 620 (part).

Semisulcospira (*Biwamelania*) sp. 2. – Nishino 1991: 17, fig. 16, unnumbered figures.

Semisulcospira (*Biwamelania*) *dilatata* Watanabe and Nishino, 1995: 6, pl. 1, figs d–f, pl. 3, figs b, c, fig. 5i; Nishino and Watanabe 2000: fig. 2-15; Kihira *et al.* 2009: 29, unnumbered figures; Miura *et al.* 2019: fig. S1j–i; Nishino 2021: 607.

Semisulcospira (*Biwamelania*) *decipiens* – Kihira *et al.* 2009: 17, unnumbered figures (part); Nishino 2021: 628.

Semisulcospira (*Biwamelania*) *multigranosa* – Kihira *et al.* 2009: 22, unnumbered figures (part).

Semisulcospira (“*Biwamelania*”) *habei* – Sawada *et al.* 2020: fig. 2 A–B, AN–AO.

Semisulcospira (“*Biwamelania*”) *dilatata* – Sawada *et al.* 2020: fig. 2 D–E, AQ–AR.

Semisulcospira dilatata – Sawada and Fuke 2022: fig. S1Q, R.

Material examined

Holotype: SMNH-Type-1614, juvenile, sex undetermined, collected from “Japan, Honshu, Lake Biwa” in 1878–1880 by the Vega Expedition.

Other type materials of synonymized names: Lectotype of *Melania* (*Semisulcospira*) *multigranosa*, SMF 225654, 1 adult, sex undetermined, from “Reisfeldern am Biwa-See, Japan” (rice field near Lake Biwa, Japan) in 1885 by B. Schmacker. Holotype of *Semisulcospira habei yamaguchi*, UMMZ 228801, 1 adult female, from Lake Biwa, “Shiga Prefecture, north of Shina-naka harbour off Kusatsu City,” (Shinanaka-cho, Kusatsu City, Shiga Prefecture) in 1965 by G. M. Davis. Holotype of *Semisulcospira dilatata*, LBM 13-3, 1 adult female, from “Lake Biwa. Iso, Hikone City, Shiga, Japan” (Lake Biwa, Iso, Maibara City, Shiga Prefecture) on 13 August 1986 by N. Watanabe.

Additional materials: KUZ Z4208, 14 females, Z4273, 3 males, collected from Hannoura on 7 November 2021; KUZ Z4209, 7females, from Oura Port on 28 November 2021; KUZ Z4210, 13 females, Z4274, 7 males, from Ebie on 2 February

2021; KUZ Z4211, 2 females, from Chikubu-shima Island on 9 September 2020; KUZ Z2513, 1 female, Z4212, 6 females, on 4 September 2017, Z4213, 2 females on 23 June 2019 from Kitafunaki; KUZ Z4214, 13 females, Z4275, 5 males, from Lake Matsunoki on 6 February 2021; KUZ Z4215, 2 females, Z4276, 6 males, on 12 January 2017, Z4216, 3 females on 14 August 2017, Z4217, 9 females on 23 February 2020 from Iso; KUZ Z4218, 6 females, Z4277, 7 males, from Kitakomatsu on 9 January 2022; KUZ Z4219, 10 females, Z4278, 2 males, from Oki-shima Island on 10 August 2019; KUZ Z4220, 4 juveniles, from Mano on 12 October 2021; KUZ Z4221, 11 females, Z4279, 3 males, from Katata Port on 28 November 2021; KUZ Z4222, 8 females, Z4280, 4 males, from Otsu Port on 23 June 2020; KUZ Z4223, 10 females, Z4281, 3 males, from Araizeki on 3 November 2021.

Amended diagnosis

Viviparous. Teleoconch large in the genus [SH 32.9 ± 5.1 (mean ± SD) (female), 32.2 ± 5.1 (male) mm; BWL 18.6 ± 3.0, 18.1 ± 2.4 mm], moderately elongated (SA 19.4 ± 2.4, 19.5 ± 3.1 degrees); color in beige to dark brown background; outer lip of aperture simple, smooth; 4.0 ± 1.0, 4.0 ± 1.1 BCN; 16.7 ± 2.3, 15.3 ± 2.0 longitudinal ribs slightly to moderately granulated on penultimate whorl; 6.0 ± 0.9, 5.8 ± 0.8 SCN; 1.7 ± 0.1, 1.7 ± 0.1 ASR; 2.8 ± 0.2, 2.9 ± 0.3 WER. Protoconch medium-sized in the genus (SHP 2.4 ± 0.4 mm, WNP 3.0 ± 0.4), with pointed nodes in 1 row on distinct longitudinal ribs; prominent spiral cords present; color in beige to dark beige, with or without 1–3 thin brown bands.

Description of holotype (SMNH-Type-1614; Fig. 11a–c)

Teleoconch: AH 6.6 mm, AL 6.6 mm, BCN 4, BWL 10.6 mm, FWL 2.2 mm, PWL 3.5 mm, RN 15, SA 22.0 degrees, SH 20.2 mm, SW 7.1 mm, TWL 2.8 mm, WER 3.05, WN 7.50; shell elongated; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, smooth; longitudinal ribs distinct, smooth, oblique, slightly to moderately curved, almost opthocline on upper whorls, moderately opisthocyrt to opisthocline on lower whorls; spiral cord absents on penultimate whorl, indistinct on body whorl; apex of shell eroded; colored olive, without color band; without operculum.

Variation

Teleoconchs: Lectotype of *S. multigranosa*, SMF 225654 (Fig. 11d–f) designated by Sawada & Nakano (2021): AH 9.1 mm, AL 8.8 mm, ASR 1.66, AW 5.3 mm, BCN 3, BWL 14.4 mm, FWL 3.1 mm, PWL 5.4 mm, RN 17, SA 19.0 degrees, SCN 5, SH 27.3 mm, SW 9.3 mm, TWL 4.1 mm, WER 3.07, WN 5.00; shell elongated, suture slightly undulating, whorls moderately convex; outer lip of aperture simple, smooth; longitudinal ribs oblique, slightly to moderately curved, opthocline on upper whorls, opisthocyrt on lower whorls, partly granulated with spiral cords; ribs fade in body whorl; apex of shell eroded; shell surface colored brown to blackish brown with deposits; without operculum.

Holotype of *S. habei yamaguchi*, UMMZ 228801 (Fig. 11g–i): AH 8.4 mm, AL 8.6 mm, ASR 1.70, AW 5.1 mm, BCN 3, BWL 14.6 mm, PWL 5.6 mm, RN 22, SA 22.1 degrees, SCN 6, SH 18.7 mm, SW 9.6 mm, TWL 4.4 mm, WN 2.00; shell elongated, suture slightly undulating, whorls slightly convex; outer lip of aperture simple, smooth; longitudinal ribs oblique, slightly to moderately curved, opisthocyrt on lower whorls, weakly granulated with spiral cords; ribs fade in body whorl; apex of shell broken artificially [see Davis (1969)]; shell color faded to beige; without operculum.

Holotype of *S. dilatata*, LBM 13-3 (Fig. 11j–l): AH 12.6 mm, AL 13.3 mm, ASR 1.86, AW 7.1 mm, BCN 5, BWL 20.1 mm, FWL 3.7 mm, PWL 6.7 mm, SA 23.6 degrees, SCN 6, SH 33.1 mm, SW 13.0 mm, TWL 5.3 mm, WER 3.38, WN 4.50; shell nearly triangular, suture slightly undulating, whorls slightly convex; outer lip of aperture simple; smooth shell surface almost smooth, longitudinal rib absent, spiral cords indistinct; apex of shell eroded; shell colored brown; without operculum.

Newly collected specimens (Fig. 11m, p, s, v, y, ab, ae, ah, ak, ap, as, av): Measurements and counts shown in Table 2 and S1. Body whorl size larger on muddy substrates (BWL 21.4 ± 2.5, 20.1 ± 1.9 mm), smaller on rock (17.9 ± 2.8, 17.0 ± 2.1 mm) and sand (17.3 ± 2.1, 18.8 ± 0.5 mm) in the species; shell slightly to moderately elongated, sometimes nearly triangular; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, smooth; longitudinal ribs distinct, oblique, slightly to moderately curved, opthocline to prosocline on upper whorls, opisthocyrt to opisthocline on lower whorls, granulated with spiral cords, fade in end of body whorl, rarely smooth or absent; apex of shell eroded; colored beige to brown, without color bands, dark brown band rarely present on lower whorl, shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 11n, q, t, w, z, ac, af, ai, al, ao, aq, at, aw): 4.4–9.8 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls; nucleus subcentral.

Protoconchs (Fig. 11 o, r, u, x, aa, ad, ag, aj, am, ar, au, ax): Measurements and counts shown in Table 2 and S1. Shell size and whorl number larger on muddy bottoms

(SHP 2.7 ± 0.3 mm; WNP 3.3 ± 0.2), medium on rock (SHP 2.4 ± 0.4 mm; WNP 3.0 ± 0.4), smaller on sand (SHP 2.1 ± 0.4 mm; WNP 2.8 ± 0.4) in the species; shell globose to slightly elongated; suture moderately undulating, or prominently depressed by discrepancy between adjacent whorls; longitudinal ribs, distinct, with pointed nodes in 1 row, on central part of whorls; spiral cords distinct, on upper and/or lower part of whorls; shell colored light beige to light brown in background, sometimes 1–3 dark, thin, rarely thick brown bands on upper and lower part of each whorl and on basal part of shell.

Radulae (Fig. 12 a–j): Taenioglossa. Rachidian roughly triangular, with central denticle and 2–3 small pointed triangular cusps on each side; central denticle tip of rachidian mostly rounded to flat in rocky substrate, pointed on sand to mud, approximately regular triangular, about 2.0 to 4.0 times longer than other triangular cusps. Lateral teeth with large central denticle, 1–3 inner and outer pointed cusps; central cusp of lateral teeth mostly flat on rock, rounded on sand to mud, irregular triangular, about 2.0 to 4.5 times longer. Interior and exterior marginal teeth spoon-shaped, with 4–6 rounded denticles.

Reproductive organs (Fig. 13): Female: Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; long, rarely short protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Distribution and ecology

Semisulcospira decipiens is one of the most widespread species in Lake Biwa and upstream of the drainage (Fig. 1; Watanabe and Nishino, 1995; fig. 5f). The species was found on the coastal rock, piled rock, sandy, and the muddy bottoms, and the insular rocky bottoms at a depth of 0–12 m. *Semisulcospira decipiens* was collected with four *S. niponica*-group species: *S. niponica* at Hannoura, Oura, Iso, Kitakomatsu, Oki-shima Island, Katata Port, Otsu Port; *S. nakanoi* at Chikubu-shima Island; *S. rugosa* at Kitafunaki, Mano; *S. reticulata* at Hannoura, Kitafunaki, Mano. Seven *S. nakasekoae*-group species inhabit with *S. decipiens*: *S. arenicola* at Kitafunaki, Iso, Kitakomatsu, Oki-shima Island, Mano, Katata Port, Otsu Port; *S. ourensis*, and *S. cryptica* sp. nov. at Chikubu-shima Island; *S. morii* at Hannoura, Chikubu-shima Island; *S. elongata* sp. nov. at Kitafunaki and Mano; *S. davisi* at Hannoura and Kitafunaki; *S. nakasekoae* at Araizeki. At Ebie, *S. decipiens* was collected with *S. reiniana*.

Remarks

Semisulcospira decipiens have been identified as *S. habei yamaguchi* or *S. habei* since Davis (1969). The three species, *S. multigranosa* described from creeks around Lake Biwa, *S. habei yamaguchi* from the south basin of the lake, and *S. dilatata* from Iso in the north basin have been synonymized under *S. decipiens* here. The characteristics of the teleoconch size, the size and whorl number of the protoconch, and the cusp shape of the radula of *S. decipiens* represent correlations with substrates. However, the species can be distinguished from other congeners by an elongated teleoconch with a smaller number of granulated longitudinal ribs on the shell surface and medium-sized, granulated protoconchs. Although the species resembles *S. habei* and *S. rugosa*, *S. decipiens* tends to possess a medium number of axial ribs. Prominent spiral cords and nodes in one row on the protoconch surface also discriminate *S. decipiens* from the two congeners.

Semisulcospira habei Davis, 1969

[Japanese name: Habe-kawanina Habe 1970]

(Table 2, S1; Fig. 11ay–bj, 12k–m)

urn:lsid:zoobank.org:act:CF35A610-45E0-4194-A52E-F4A2DF369ECB

Semisulcospira multigranosa – Fukuoka 1933: 114, 117, fig. 4 (part).

Semisulcospira habei habei – Burch and Davis 1967: 37 (unavailable).

Semisulcospira sp. – Burch 1968: 7–8, fig. 2 (part).

Semisulcospira habei Davis, 1969: 237–240, pl. 3, figs 1–3, pl. 9, figs 6–10 (OD); Society for the Study of Aquatic Life 1989: 18–19, 49–50, figs 14, 31, 42 (part); Higo and Goto 1993: 97; Takami 1994: 202; Goto and Poppe 1996: 204; Takami 2013: 97, fig. 2B, fig. 4; Sawada and Fuke 2022: fig. S1O, P.

Biwamelania habei – Habe 1978: 94 (part); Nomoto 2001: 33 (part); Nishino and Tanida 2018: 50, 247 (part).

Semisulcospira (*Biwamelania*) *habei yamaguchi* – Matsuoka 1985: 190.

Semisulcospira (*Biwamelania*) *multigranosa* – Kihira *et al.* 2009: 22, unnumbered figures (part).

Semisulcospira (*Biwamelania*) *habei* – Kihira *et al.* 2009: 23, unnumbered figures (part); Miura *et al.* 2019: fig. S1b, c (part); Nishino 2021: 620 (part).

Material examined

Holotype: UMMZ 220236, adult female collected from “Kyoto administrative district, Uji City, Uji River” (Uji River, Uji, Uji City, Kyoto Prefecture) in central Honshu Island, Japan in 1965 by G. M. Davis.

Additional materials: KUZ Z4224, 14 females, Z4282, 2 males, collected from Uji on 16 November 2019; KUZ Z4225, 13 females, Z4283, 1 male, from Fushimi on 9 March 2021; KUZ Z4226, 13 females, Z4284, 2 males, from Yawata on 11 February 2021.

Amended diagnosis

Viviparous. Teleoconch medium sized in the genus [SH 29.5 ± 2.6, 24.8 ± 3.5 mm; BWL 17.2 ± 1.2, 14.7 ± 2.3 mm], slightly elongated (SA 18.4 ± 2.5, 18.4 ± 3.4 degrees); color in dark light brown to dark olive background; outer lip of aperture simple, smooth; 3.8 ± 0.8, 3.4± 0.6 BCN; 19.7 ± 1.8, 16.8 ± 3.0 longitudinal ribs moderately granulated on penultimate whorl; 6.5 ± 0.8, 6.0 SCN; 1.7 ± 0.1, 1.8 ± 0.1 ASR; 2.9 ± 0.3, 2.9 ± 0.2 WER. Protoconch medium sized in the genus (SHP 2.7 ± 0.4 mm, WNP 3.2 ± 0.4), with pointed nodes in 2–3 rows on distinct longitudinal ribs; prominent spiral cords present; color in beige to dark beige, with or without 1–3 thin brown bands.

Description of holotype (UMMZ 220236; Fig. 11ay–ba)

Teleoconch: AH 9.8 mm, AL 9.8 mm, ASR 1.71, AW 5.7 mm, BCN 4, BWL 16.1 mm, FWL 3.0 mm, PWL 5.5 mm, RN 19, SA 22.5 degrees, SCN 6, SH 24.9 mm, SW 10.0

mm, TWL 3.9 mm, WER 3.32, WN 4.00; shell elongated; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, almost smooth; longitudinal ribs oblique, slightly to moderately curved, prosocline on upper whorls, opisthocyrt on lower whorls, moderately granulated with spiral cords, fade in body whorl; apex of shell eroded; shell color faded to beige, without operculum.

Variation

Teleoconchs (Fig. 11bb, be, bh): Measurements and counts shown in Table 2 and S1. Shell slightly to moderately elongated, sometimes nearly triangular; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, almost smooth; longitudinal ribs distinct, straight to oblique, slightly to moderately curved, orthocline to prosocline on upper whorls, opisthocyrt on lower whorls, granulated with spiral cords, fade in end of body whorl; apex of shell eroded; shell colored light brown to dark olive, without color bands, dark brown band rarely present on lower whorl, shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 11bc, bf, bi): 4.9–7.0 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls; nucleus subcentral.

Protoconchs (Fig. 11bd, bg, bj): Measurements and counts shown in Table 2 and S1. Shell globose; suture moderately undulating, or prominently depressed by discrepancy between adjacent whorls; longitudinal ribs, distinct, with pointed nodes in 2–3 rows, rarely in 1 row, on central part of whorls; spiral cords distinct, on upper and/or lower part of whorls; shell colored light beige to light brown in background, sometimes 1–3 thin or thick dark brown bands on upper and lower part of each whorl and on basal part of shell.

Radulae (Fig. 12k–m): Taenioglossa. Rachidian roughly triangular, with central denticle and 2–3 small pointed triangular cusps on each side; central denticle tip of rachidian mostly pointed, rarely rounded or flat, approximately regular triangular, about 2.0 to 4.0 times longer than other triangular cusps. Lateral teeth with large central denticle, 2–3 inner and outer pointed cusps; central cusp of lateral teeth largely flat, sometimes pointed or rounded, irregular triangular, about 2.0 to 4.5 times longer. Interior and exterior marginal teeth spoon-shaped with 4–6 rounded denticles.

Reproductive organs (Fig. 13): Female: Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; long protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Distribution and ecology

Semisulcospira habei is distributed downstream of the drainage of Lake Biwa (Fig. 1). The species was found on the piled rock and sandy bottoms and the concrete blocks at a depth of 0–0.5 m. *Semisulcospira habei* was collected with *S. nakasekoae* and *S. reiniana* at all sites.

Remarks

Semisulcospira habei can be distinguished from other congeners by an elongated teleoconch with a medium number of granulated longitudinal ribs on the shell surface and medium-sized, granulated protoconchs. Although the species resembles *S. decipiens* and *S. rugosa*, *S. habei* tends to possess a greater number of axial ribs. Prominent spiral cords and nodes in 2–3 rows on the protoconch surface also discriminate *S. habei* from two other congeners.

Semisulcospira arenicola Watanabe and Nishino, 1995

[Japanese name: Tatehida-kawanina (Habe 1968)]

(Table 2, S2; Fig. 14a–ad, 15a–i)

urn:lsid:zoobank.org:act:04C7756C-91EB-449E-B474-6C3F48151C00

Melania niponica Smith, 1876: 123–124 (part); Kobelt 1879: 131, pl. 19, figs 10, 12 (part).

Melania multigranosa – Annandale 1916: 44–45, pl. 3, fig. 2A, C (part).

Semisulcospira decipiens – Kajiyama and Habe 1961: 171, figs 4, 4a; Kuroda 1941: 184; Kuroda 1962: 86, 89 (part); Burch and Davis 1967: 37; Burch 1968: 11, fig. 1A; Davis 1969: 246–248, pl. 4, fig. 6, pl. 10, figs 6–9 (part); Watanabe 1970: 93; Society for the Study of Aquatic Life 1989: 13–14, 48–49 figs 11, 39 (part); Goto and Poppe 1996: 204; Köhler 2016: fig. 4B, C, E, K (part); Sawada and Fuke 2022: fig. 8D, S1U, V.

Semisulcospira habei yamaguchi Davis, 1969: 240–243, pl. 3, figs 6 (part).

Semisulcospira multigranosa – Davis 1969: 255, 262, pl. 7, figs 2, 4, pl. 11, fig. 5 (part).

Biwamelania decipiens – Habe 1978: 94; Nomoto 2001: 33; Prozorova and Rasshepkina 2006: 130; Nishino and Tanida 2018: 43, 243.

Semisulcospira (*Biwamelania*) *decipiens* – Matsuoka 1985: 190; Nishino 1991: 12, fig. 11, unnumbered figures; Watanabe and Nishino 1995: fig. 5c, appendix pl. 1, figs 5, 6, appendix pl. 2, figs 18, 19 (part); Nishino and Watanabe 2000: fig. 2-11; Urabe 2007: 80; Kihira *et al.* 2009: 17, unnumbered figures (part); Kamiya *et al.* 2011: 25; Miura *et al.*, 2019: fig. S1w, x (part); Nishino 2021: 620.

Semisulcospira type I – Society for the Study of Aquatic Life 1989: 38–39, 56, figs 24, 29, 51.

Semisulcospira (*Biwamelania*) sp. 8. – Nishino 1991: 23, fig. 22, unnumbered figures.

Semisulcospira decipens – Higo and Goto 1993: 97.

Semisulcospira (*Biwamelania*) *arenicola* Watanabe and Nishino, 1995: 11, pl. 2, figs s–u, pl. 3, figs l, m, fig. 5o (OD); Nishino and Watanabe 2000: fig. 2-13; Kihira *et al.* 2009: 29, unnumbered figures; Miura *et al.* 2019: fig. S1y–aa; Nishino 2021: 612.

Biwamelania arenicola – Nomoto 2001: 33; Nishino and Tanida 2018: 41, 242.

Biwamelania decipience – Kurozumi 2007: 63.

Semisulcospira (“*Biwamelania*”) *decipiens* – Sawada *et al.* 2020: fig. 2 AB–AD, BL–BM.

Semisulcospira arenicola – Sawada and Fuke 2022: fig. S1AC, AD.

Material examined

Holotype: LBM 13-8, adult female collected from “Lake Biwa. Satsuma, Notogawa-cho, Shiga, Japan” (Lake Biwa, Satsuma-cho, Hikone City, Shiga Prefecture) in 1986 by N. Watanabe.

Additional materials: KUZ Z4231, 12 females, Z4287, 3 males, collected from Minamihama on 31 October 2021; KUZ Z4232, 13 females, Z4288, 7 males, from Tamura on 9 May 2021; KUZ Z4233, 13 females, Z4289, 1 male, from Kitafunaki on 28 August 2021; KUZ Z4234, 11 females, from Yokoehama on 1 August 2021; KUZ Z4235, 13 females, Z4290, 2 males, from Satsuma on 9 May 2021; KUZ Z4236, 12 females, Z4291, 6 males, from Horikiri Port on 7 November 2021; KUZ Z4237, 13 females, Z4292, 5 males, from Wani Beach on 28 August 2021; KUZ Z4238, 14 females, Z4293, 3 males, Z4239, 7 juveniles, from Mano on 12 October 2021; KUZ Z4240, 5 females, Z4294, 4 males, from Otsu Port on 23 June 2021.

Amended diagnosis

Viviparous. Teleoconch medium-sized in the genus [SH 27.0 ± 3.3, 24.5 ± 2.4 mm; BWL 14.6 ± 1.5, 12.9 ± 1.3 mm], greatly elongated (SA 13.5 ± 1.7, 12.9 ± 2.6 degrees); color in beige to light brown background; outer lip of aperture simple, smooth; 3.8 ± 0.8, 2.3 ± 0.7 BCN; 19.7 ± 1.8, 14.3 ± 3.4 longitudinal ribs slightly to moderately granulated, sometimes indistinct on penultimate whorl; 7.4 ± 0.8, 7.1 ± 0.9 SCN; 1.6 ± 0.1, 1.6 ± 0.1 ASR; 2.3 ± 0.2, 2.3 ± 0.2 WER. Protoconch medium-sized in the genus (SHP 2.6 ± 0.4 mm, WNP 3.4 ± 0.3), with or without rounded nodes in 1 row, prominent or weak spiral cords present; color in light beige to light brown, without thin brown bands.

Description of holotype (LBM 13-8; Fig. 14a–c)

Teleoconch: AH 7.3 mm, AL 7.0 mm, ASR 1.63, AW 4.3 mm, BCN 3, BWL 12.7 mm, FWL 3.2 mm, PWL 5.0 mm, SA 12.9 degrees, SH 24.0 mm, SW 7.5 mm, TWL 4.2 mm, WER 2.28, WN 4.50; shell greatly elongated; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, smooth; longitudinal rib absent; spiral cords indistinct; apex of shell eroded; shell colored brown; without operculum.

Variation

Teleoconchs (Fig. 14g, j, m, p, s, v, y, ac): Measurements and counts shown in Table 2 and S2. Shell greatly elongated; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, smooth, aperture rounder on muddy substrates (ASR 1.54 ± 0.07, 1.60 ± 0.04), more elongated on rock (1.62 ± 0.09, 1.63 ± 0.11) and sand (1.61 ± 0.07, 1.60 ± 0.06) in the species. Longitudinal ribs oblique, slightly to moderately curved, opisthocline to prosocline on upper whorls, opisthocyrt to opisthocline on lower whorls, weakly granulated with spiral cords, distinct, almost straight; lower number on rock (RN 12.1 ± 1.4, 11.4 ± 1.9); weak or absent, larger number on mud (17.2 ± 2.6, 15.3 ± 4.0) and sand (17.1 ± 1.8, 16.3 ± 2.7), fade in end of body whorl. Apex of shell eroded; shell colored dark beige to brown, without color bands, dark olive band rarely present on medium to lower part of whorl; shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 14e, h, k, n, q, t, w, z, ad): 3.9–6.5 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls; nucleus subcentral.

Protoconchs (Fig. 14f, i, l, o, r, u, x, aa, ae): Measurements and counts shown in Table 2 and S2. Shell mildly elongated; suture moderately undulating, or prominently depressed by discrepancy between adjacent whorls; longitudinal ribs prominent, with or without nodes rounded in 1 row, on central part of whorls; spiral cords distinct, on upper and/or lower part of whorls; shell colored light beige to light brown in background, rarely 1–3 thick dark brown bands on upper and lower part of each whorl and on basal part of shell.

Radulae (fig. 15a–i): Taenioglossa. Rachidian roughly triangular, with central denticle and 2–3 small pointed triangular cusps on each side; central denticle tip of rachidian mostly pointed, rarely rounded, approximately regular triangular, about 2.5 to

5.0 times longer than other triangular cusps. Lateral teeth with large central denticle, 1–3 inner and outer pointed cusps; central cusp of lateral teeth pointed or rounded, irregular triangular, about 1.5 to 5.5 times longer. Interior and exterior marginal teeth spoon-shaped with 4–7 rounded denticles.

Reproductive organs (Fig. 13): Female: Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; long or short protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Distribution and ecology

Semisulcospira arenicola shows widespread distribution in Lake Biwa and upstream of the drainage (Fig. 1; Watanabe and Nishino, 1995; fig. 5c, 5o). The species was found on the coastal rock, piled rock, sandy, and the muddy bottoms at a depth of 0–12 m. *Semisulcospira arenicola* was collected with *Semisulcospira elongata* sp. nov. at Kitafunaki and Mano. *Semisulcospira arenicola* inhabits with five *S. niponica*-group species: *S. decipiens* at all sites; *S. watanabei* at Horikiri Port; *S. niponica* at Otsu Port; *S. rugosa* at Kitafunaki; *S. reticulata* at Kitafunaki and Mano.

Remarks

Semisulcospira arenicola has been treated as *S. decipiens* since Davis (1969). The intraspecific variation in the teleoconch sculpture has been used to discriminate between *S. arenicola* and *S. decipiens sensu* Davis 1969 from outside the northern part of Lake Biwa. The characteristics of the longitudinal ribs and the aperture roundness exhibit variation associated with the substrate differences, while the cusp shape of the radula did not appear to be correlated with substrates. *Semisulcospira arenicola* can be distinguished from other congeners by a medium-sized, greatly elongated teleoconch with a lower number of weakly granulated longitudinal ribs and medium-sized protoconchs with nodes and spiral cords. The species resembles *S. ourensis* and *S. elongata* sp. nov., and *S. cryptica* sp. nov. However, the teleoconch size and the number of axial ribs of *S. arenicola* are greater than that of *S. ourensis*, while they are smaller than *S. elongata* sp. nov. The body whorl length of *S. arenicola* is also lower than *S. cryptica* sp. nov.

Semisulcospira nakasekoae (Kuroda, 1929)

[Japanese name: Nakaseko-kawanina Kuroda 1929]

(Table 2, S2; Fig. 14ae–be, 15j–p, 16a)

urn:lsid:zoobank.org:act:D59D6844-CF44-4612-93BD-AFEBBF944C5F

Melanoides (*Semisulcospira*) *nakasekoae* Kuroda, 1929: 186, 189, pl. 5, figs 37–41 (OD).

Semisulcospira nakasekoae – Fukuoka 1933: 114, 117, figs 7, 8; Kuroda 1962: 86, 89; Burch and Davis 1967: 37; Habe and Kosuge 1967: 28, pl. 11, fig. 18; Burch 1968: 7, fig. 1C; Davis 1969:

235–237, pl. 2, figs 4–6, pl.9, figs 1–5; Kobayashi 1986: 127, fig. 1D, fig. 2D, fig. 6; Oniwa and Kimura 1986: 503; Society for the Study of Aquatic Life 1989: 93; Higo and Goto 1993: 97; Takami 1994: 202; Goto and Poppe 1996: 204; Köhler 2016: fig. 4AI; Takami 2019: 37, fig. 1B, fig. 3.

Semisulcospira decipiens nakasekoae – Kajiyama and Habe 1961: 167; Kuroda and Habe 1965: 57.

Biwamelania (*decipiens*) *nakasekoae* – Habe 1978: 94.

Semisulcospira (*Biwamelania*) *nakasekoae* – Matsuoka and Nakamura 1981: 113; Matsuoka 1985: 190; Nishino 1991: 24, unnumbered figures; Watanabe and Nishino 1995: appendix pl. 1, fig. 13; Nishino and Watanabe 2000: fig. 2-25; Kihira *et al.* 2009: 18–21, unnumbered figures; Kamiya *et al.* 2011: 25; Miura *et al.* 2019: fig. S1ae–ag; Nishino 2021: 609.

Semisulcospira type H – Society for the Study of Aquatic Life 1989: 36–37, 56, figs 23, 35, 50.

Semisulcospira (*Biwamelania*) sp. 7. – Nishino 1991: 22, fig. 21, unnumbered figures.

Semisulcospira (*Biwamelania*) *fluvialis* Watanabe and Nishino, 1995: 10, pl. 2, figs p–r, pl. 3, figs i–k, fig. 5n; Nishino and Watanabe 2000: fig. 2-24; Kihira *et al.* 2009: 29, unnumbered figures; Miura *et al.* 2019: fig. S1ab–ad; Nishino 2021: 610.

Semisulcospira (*Biwamelania*) *decipiens* – Watanabe and Nishino 1995: fig. 5c (part).

Biwamelania fluvialis – Nomoto 2001: 33; Nishino and Tanida 2018: 47, 245.

Biwamelania nakasekoae – Nomoto 2001: 33; Nishino and Tanida 2018: 55, 250.

Semisulcospira fluvialis – Takami 2013: 97, fig. 2C, fig. 5; Sawada and Fuke 2022: fig. S1AE, AF.

Semisulcospira ("*Biwamelania*") *multigranosa* – Sawada *et al.* 2020: fig. 2 G–H, AS–AT.

Semisulcospira nakasekoae – Sawada and Fuke 2022: fig. S1W, X.

Material examined

Type material of synonymized name: LBM 13-16, adult female collected from Nango, Otsu City, Shiga, Japan" (Seta River, Nango, Otsu City, Shiga Prefecture) in 1987 by N. Watanabe.

Additional materials: KUZ Z4241, 3 females, Z4295, 3 males, collected from Araizeki on 3 November 2021; KUZ Z4242, 19 females, Z4296, 7 males, from Nango on 3 November 2021; KUZ Z4243, 13 females, Z4297, 7 males, from Uji on 16 November 2019; KUZ Z4244, 12 females, Z4298, 4 males, from Fushimi on 11 February 2021; KUZ Z4245, 14 females, Z4299, 5 males, from Higashiyama on 1 April 2022; KUZ Z4246, 12 females, Z4300, 3 males, from Yawata on 11 February 2021; KUZ Z4247, 13 females, Z4301, 4 males, from Neyagawa on 11 February 2021.

Emended diagnosis

Viviparous. Teleoconch medium-sized in the genus [SH 21.7 ± 4.0, 19.3 ± 3.7 mm; BWL 15.0 ± 2.4, 13.6 ± 2.5 mm], globose to moderately elongated (SA 22 ± 5.5, 22.9 ± 8.9 degrees); color in beige to dark olive background; outer lip of aperture simple, smooth; apex of shell greatly eroded; 3.8 ± 0.8, 3.9 ± 1.3 BCN; 19.7 ± 1.8, 16.1 ± 3.3 longitudinal ribs moderately granulated on penultimate whorl; 9.2 ± 1.1, 8.7 ± 1.4 SCN; 1.7 ± 0.1, 1.6 ± 0.1 ASR; 3.6 ± 1.3, 4.1 ± 1.5 WER. Protoconch medium-sized to large in the genus (SHP 3.0 ± 0.5 mm, WNP 3.3 ± 0.4), pear-shaped, with prominent longitudinal ribs without node on surface; color in light beige to dark brown, with or without 1–3 thick brown bands. Radula with large, flat tip of central cusp of lateral teeth.

Type specimen

The number and voucher of the type specimens of *S. nakasekoae* were not specified by the original description (Kuroda 1929). The type series could not be found in the malacological collection of the NSMT, NSM, and the KUM (see Materials and methods). A neotype should be designated in the following situations according to the Article 75.1 of the Code: 1) no name-bearing type specimen is believed to be extant, and 2) a name-bearing type is considered to be necessary to define the nominal taxon objectively (ICZN 1999). The nomenclatural status of *Semisulcospira nakasekoae* does not apply the condition 2) above because no other congener with a rounded teleoconch, which is consistent with the original description, is distributed in the candidates for its type locality (the Seta and Uji Rivers and the Lake Biwa Canal). Therefore, we consider the identity and the nomenclatural status of the species to be unquestionable and have not designated a neotype for *S. nakasekoae* here, although its type specimen is considered to be missing.

Description

Teleoconchs: Holotype of *S. fluvialis*, LBM 13-16 (Fig. 14ae–ag): AH 9.7 mm, AL 10.1 mm, ASR 1.76, AW 5.7 mm, BCN 4, BWL 17.0 mm, FWL 3.5 mm, PWL 6.6 mm, SA 21.6 degrees, SH 25.9 mm, SW 10.2 mm, TWL 4.5 mm, WER 2.74, WN 3.50; shell slightly elongated; suture slightly undulating; whorls moderately convex; outer lip of aperture simple, smooth; longitudinal ribs oblique, almost straight, prosocline on upper whorls, absent on lower whorl; spiral cords indistinct on penultimate whorl; apex of shell greatly eroded; shell surface colored light brown; without operculum.

Newly collected specimens (Fig. 14ah, ak, an, aq, at, aw, az, bc): Measurements and counts shown in Table 2 and S2. Shell globose at Uji, slightly to moderately elongated at other sites; suture slightly to strongly undulating; whorls slightly to moderately convex; outer lip of aperture simple, smooth; longitudinal ribs indistinct, oblique, slightly curved, opthocline to opisthocyrt on upper whorls, almost orthocline to opisthocyrt on lower whorls; ribs rarely distinct, weakly granulated with spiral cords, fade in end of body whorl; apex of shell largely eroded; shell colored dark beige to dark brown, without color bands, sometimes dark olive band present on medium to lower part of whorl; shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 14ai, al, ao, ar, au, ax, ba, bd): 3.8–7.3 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls; nucleus subcentral.

Protoconchs (Fig. 14aj, am, ap, as, av, ay, bb, be): Measurements and counts shown in Table 2 and S2. Shell globose, pear-shaped, rarely mildly elongated; suture moderately undulating, or prominently depressed by discrepancy between adjacent whorls; longitudinal ribs prominent, without nodes, on central part of whorls; ribs rarely indistinct; spiral cords prominent, weak, or absent, on upper and/or lower part of whorls; shell colored light beige to dark brown in background, sometimes 1–3 thick dark brown bands on upper and lower part of each whorl and on basal part of shell.

Radulae (Fig. 15j–p, 16a): Taenioglossa. Rachidian roughly triangular, with central denticle and 2–4 small pointed triangular cusps on each side; central denticle tip of rachidian largely pointed, sometimes rounded, rarely flat, approximately regular triangular, about 2.5 to 4.0 times longer than other triangular cusps. Lateral teeth with large central denticle, 1–3 inner and outer pointed cusps; central cusp of lateral teeth rounded or flat, prominently large, irregular triangular, about 2.5 to 4.5 times longer. Interior and exterior marginal teeth spoon-shaped with 3–7 rounded denticles.

Reproductive organs (Fig. 13): Female: Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; long, sometimes short protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Distribution and ecology

Semisulcospira nakasekoae is distributed downstream of the drainage of Lake Biwa and the Lake Biwa Canal (Fig. 1). The species was found on the piled rock and sandy bottoms and the concrete blocks at a depth of 0–0.5 m. *Semisulcospira nakasekoae* was found with two *S. niponica*-group species: *S. decipiens* at Araizeki; *S. habei* at all sites except at Higashiyama. *Semisulcospira nakasekoae* was also collected with *S. reiniana* at all sites in the Uji and Yodo Rivers.

Remarks

The present genetic analyses revealed that *S. fluvialis* and *S. decipiens sensu* Davis 1969 from the upstream of the Seta River are the geographic variation of *S. nakasekoae* and therefore, the two former species have been synonymized under *S. nakasekoae* here. The characteristics of shell size and roundness (ASR, BWL, SA, SH and WER) of *S. nakasekoae* present significant variation among populations. However, the SA and WER values of the species are greater than other congeners, and the species can be discriminated with a medium-sized, globose to moderately elongated teleoconch with a medium number of longitudinal ribs and a greater number of spiral cords. The species can also be distinguished by medium to large-sized, pear-shaped protoconchs with prominent, smooth nodes and radulae with large, flat tips on the central cusp of lateral teeth.

Semisulcospira ourensis Watanabe and Nishino, 1995

[Japanese name: Oura-kawanina Watanabe and Nishino 1995]

(Table 2, S2; Fig. 16d–h, 17a–u)

urn:lsid:zoobank.org:act:21812638-E68C-477C-8190-5FB4CC2AFF6D

Semisulcospira decipiens – Society for the Study of Aquatic Life 1989: 13–14, 48–49 figs 11, 39 (part).

Semisulcospira type D – Society for the Study of Aquatic Life 1989: 30–31, 54, figs 20, 33-2, 47.

Semisulcospira (*Biwamelania*) *decipiens* – Nishino 1991: 12, fig. 11, unnumbered figures; Miura *et al.* 2019: fig. S1v (part).

Semisulcospira (*Biwamelania*) sp. 4. – Nishino 1991: 19, fig. 18, unnumbered figures.

Semisulcospira (*Biwamelania*) *ourense* Watanabe and Nishino, 1995: 7–8, pl. 1, figs j, k, pl. 3, fig. e, fig. 5k (OD); Nishino and Watanabe 2000: fig. 2-17; Kihira *et al.* 2009: 29, unnumbered figures; Kamiya *et al.* 2011: 25; Nishino 2021: 605.

Biwamelania ourense – Nishino and Tanida 2018: 58, 252.

Semisulcospira ourense – Sawada and Fuke 2022: fig. S1AA, AB.

Material examined

Holotype: LBM 13-7, adult female collected from “Lake Biwa. Oura, Nishiazai-cho, Shiga, Japan” (Lake Biwa, Oura, Nagahama City, Shiga Prefecture) in 1986 by N. Watanabe.

Additional materials: KUZ Z4248, 6 females, Z4302, 5 males, collected on 25 July 2021, Z4249, 8 females on 1 May 2021 from Oura; KUZ Z4250, 1 female, Z4303, 2 males, from Okude on 25 July 2021; KUZ Z4251, 13 females, Z4304, 4 males, from Sugaura on 1 May 2021; KUZ Z4252, 3 females, from Chikubu-shima Island on 9 September 2020.

Amended diagnosis

Viviparous. Teleoconch medium-sized in the genus [SH 25.7 ± 3.3, 23.9 ± 4.4 mm; BWL 14.4 ± 1.2, 13.3 ± 1.5 mm], greatly elongated (SA 13.4 ± 1.8, 14.0 ± 2.3 degrees); color in beige to light brown background; outer lip of aperture simple, smooth; 2.6 ± 0.7, 2.6 ± 0.7 BCN; 13.7 ± 2.3, 11.3 ± 1.5 longitudinal ribs smooth or weakly granulated; 6.9 ± 0.7, 6.5 ± 0.5 SCN; 1.6 ± 0.1, 1.7 ± 0.1 ASR; 2.4 ± 0.2, 2.5 ± 0.3 WER. Protoconch small to medium-sized in the genus (SHP 2.3 ± 0.6 mm, WNP 3.2 ± 0.5), with or without rounded nodes in 1 row; prominent or weak spiral cords present; color in light beige to dark brown, without color bands.

Description of holotype (LBM 13-7; Fig. 17a–c)

Teleoconch: AH 8.8 mm, AL 8.9 mm, ASR 1.62, AW 5.5 mm, BCN 2, BWL 15.1 mm, FWL 3.8 mm, PWL 5.9 mm, RN 11, SA 13.7 degrees, SH 29.7 mm, SW 10.1 mm, TWL 4.3 mm, WER 2.32, WN 5.50; shell greatly elongated; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, smooth; longitudinal ribs smooth, oblique, slightly curved, opisthocyrt to opisthocline; spiral cords absent; apex of shell eroded; shell colored beige to light brown; without operculum.

Variation

Teleoconchs (Fig. 17d, g, j, m, p, s): Measurements and counts shown in Table 2 and S2. Shell greatly elongated; suture slightly to moderately undulating; whorls slightly convex on rocky bottom, moderately on mud; outer lip of aperture simple, smooth; longitudinal ribs distinct, straight to oblique, slightly to moderately curved, opisthocyrt to opisthocline, smooth, or weakly granulated with spiral cords, fade in end of body whorl; more ribs present on mud (RN 18, 15) than rock (13.6 ± 2.2, 10.9 ± 0.8); apex of shell eroded; shell colored dark beige to dark brown in background, dark olive band sometimes present on upper and/or lower part of whorl; shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 17e, h, k, n, q, t): 4.1–6.8 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls; nucleus subcentral.

Protoconchs (Fig. 17f, i, l, o, r, u): Measurements and counts shown in Table 2 and S2. Shell mildly elongated; suture moderately undulating, or prominently depressed by discrepancy between adjacent whorls; longitudinal ribs prominent, with or without rounded nodes in 1 row, on central part of whorls; spiral cords weak or absent, on upper and/or lower part of whorls; shell colored light beige to dark brown, rarely with 1–3 thick brown color bands. Shell rarely small sized, globose; longitudinal ribs absent.

Radulae (Fig. 16d–h): Taenioglossa. Rachidian roughly triangular, with central denticle and 2–3 small pointed triangular cusps on each side; central denticle tip of rachidian largely pointed, sometimes rounded, rarely flat, approximately regular triangular, about 2.0 to 4.5 times longer than other triangular cusps. Lateral teeth with large central denticle, 1–3 inner and outer pointed cusps; central cusp of lateral teeth pointed or rounded, irregular triangular, about 2.0 to 4.5 times longer. Interior and exterior marginal teeth spoon-shaped with 4–6 rounded denticles.

Reproductive organs (Fig. 13): Female: Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; long or short protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Distribution and ecology

The distribution of *S. ourensis* is restricted to the northern coasts and Chikubu-shima Island in Lake Biwa (Fig. 1; Watanabe and Nishino, 1995; fig. 5k). The species was found on the coastal rock, piled rock, and muddy bottoms and the insular rocky bottoms at a depth of 0–6 m. *Semisulcospira ourensis* was collected with three *S. nakasekoae*-group species: *Semisulcospira morii* at Chikubu-shima Island; *S. elongata* sp. nov. at Okude; *S. cryptica* sp. nov. at Okude and Chikubu-shima Island. Four *S. niponica*-group species cooccur with *S. ourensis*: *S. decipiens* at Oura and Chikubu-shima Island; *S. nakanoi* at Chikubu-shima Island; *S. watanabei* and *S. fuscata* at Oura. *Semisulcospira ourensis* was found with *S. reiniana* at Okude.

Remarks

Semisulcospira ourensis was originally described as *Semisulcospira* “*ourense*” (Watanabe & Nishino 1995). The genus *Semisulcospira* was distinguished from *Sulcospira* Troschel, 1858 by Boettger (1886) and its name consists of the Latin masculine noun “sulcus” and the Ancient Greek feminine noun “spira” with the connecting vowel “o” and the Latin prefix “semi-”. According to the final noun, the gender of *Semisulcospira* is feminine. Articles 31.2 and 34.2 of the Code prescribe that the gender of a Latin adjective used as a species-group name must agree with that of the generic name it is combined with (ICZN 1999). However, the gender of the specific name “*ourense*” is discordant with that of *Semisulcospira* because the neutral suffix “-ense” is combined to the stem “our-”, which means the type locality of the species. In the present systematics, therefore, the specific name has been changed to feminine “*ourensis*” to agree in gender with *Semisulcospira*.

In accordance with the original description, the present specimens collected from Oura and Sugaura were identified morphologically as *S. ourensis sensu stricto* (Fig. 13d–f, m–o) or *S. decipiens sensu* Watanabe and Nishino 1995 (Fig. 13g–I, p–r). *Semisulcospira ourensis sensu stricto* has been characterized by few, small, rounded protoconchs and was rarely found in the present investigation. However, significant differences were not detected between the genetic structures of the two sympatric species. The specimen number of *S. ourensis* used in the original description was considerably smaller than other species (Watanabe and Nishino 1995). These facts suggest that the traits of smaller PN and SHP, which infrequently appear in *S. ourensis*, were treated as its diagnoses in the original description.

The characteristics of longitudinal ribs represent correlations with substrates. *Semisulcospira ourensis* can be distinguished from other congeners by a small, greatly elongated teleoconch with a lower number of smooth to weakly granulated longitudinal ribs and medium-sized protoconchs with nodes and spiral cords. Although the species resembles *S. arenicola*, the teleoconch size and the number of axial ribs of *S. ourensis* are smaller than *S. arenicola*.

Semisulcospira elongata Sawada sp. nov.

[New Japanese name: Kesho-kawanina]

(Table 2, S2; Fig. 16i–m, 17v–at)

urn:lsid:zoobank.org:act:662976A7-F55B-495F-B152-1786AB8836D7

Melania multigranosa – Boettger 1886: 7–8 (part); Annandale 1916: 44–45, pl. 3, fig. 2E (part).
Semisulcospira decipiens – Davis 1969: 246–248, pl. 4, figs 4–5 (part).

Material examined

Holotype: KUZ Z4305, adult female collected from Lake Biwa, Kitafunaki on 28 August 2021 by the first author.

Paratypes: KUZ Z4306–Z4308, 3 adult females, collected with holotype.

Additional materials: KUZ Z4309, 4 females, collected with Holotype; KUZ Z4310, 3 females, from Okude on 25 July 2021; KUZ Z4311, 7 females, Z4312, 3 males, from Imazu Beach on 21 March 2022; KUZ Z4313, 11 females, Z4314, 1 male, Z4315, 3 juveniles, from Mano on 12 October 2021.

Diagnosis

Viviparous. Teleoconch large-sized in the genus [SH 32.3 ± 4.3, 31.1 ± 4.1 mm; BWL 16.5 ± 1.8, 16.9 ± 2.7 mm], greatly elongated (SA 13.6 ± 2.7, 13.9 ± 0.1 degrees); color in beige to dark brown background; outer lip of aperture simple, smooth; 2.8 ± 0.7, 2.5 ± 0.6 BCN; 20.0 ± 3.8, 20.7 ± 2.5 longitudinal ribs smooth or weakly granulated, strongly curved; 7.9 ± 1.2, 7.0 ± 1.4 SCN; 1.6 ± 0.1, 1.6 ± 0.1 ASR; 2.4 ± 0.2, 2.5 ± 0.2 WER. Protoconch medium-sized in the genus (SHP 3.2 ± 0.5 mm, WNP 3.5 ± 0.3), 12.1 ± 1.6 longitudinal ribs, with or without rounded nodes in 1 row; prominent or weak spiral cords present; color in light beige to dark brown, rarely with 1–3 thick brown bands. Radula with pointed tip of central cusp of rachidian and lateral teeth.

Description of holotype (KUZ Z4305; Fig. 16i, 17v–ab)

Teleoconch: AH 11.2 mm, AL 11.0 mm, ASR 1.63, AW 6.7 mm, BCN 2, BWL 19.0 mm, FWL 5.3 mm, PWL 7.4 mm, SA 11.1 degrees, SH 43.2 mm, SW 11.6 mm, TWL 7.1 mm, WER 2.12, WN 6.25; shell greatly elongated; suture slightly undulating on upper whorls, strongly on lower ones; whorls slightly convex on upper whorls, moderately on lower whorls; outer lip of aperture simple, smooth; longitudinal ribs oblique, greatly curved, opthocline to opisthocyrt, slightly granulated on upper whorl, faded on body to penultimate whorls; apex of shell eroded; shell colored dark beige in background, with 1 thick dark olive band on middle to lower parts of whorls.

Operculum: 6.2 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls. Nucleus subcentral.

Protoconchs: PN 35, RNP 10, SHP 2.7, SWP 1.6, WNP 3.50; shell mildly elongated; suture prominently undulating; ribs remarkable without nodes on middle part of whorls, 1 strong and weak spiral cords on upper and lower ones, respectively; shell colored light beige in background, without color band.

Radula: Taenioglossa. Rachidian roughly triangular, with large central denticle and 2–3 minor pointed triangular cusps on each side; central denticle tips of rachidian pointed, approximately regular triangular, about 3.0 to 3.5 times longer than other triangular cusps. Lateral teeth with large central denticle, 2–3 inner and outer pointed cusps; central denticle rounded, irregular triangular, about 3.0 times longer. Interior and exterior marginal teeth spoon-shaped with 4–6 rounded denticles.

Reproductive organs (female): Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; long protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Variation

Teleoconchs (Fig. 17c, af, ai, ak, an, aq): Measurements and counts shown in Table 2 and S2. Suture slightly to moderately undulating; whorls slightly to moderately convex; longitudinal ribs distinct on penultimate whorl, moderately curved, smooth or weakly granulated with spiral cords; shell colored dark beige to dark brown in background, dark olive band sometimes present on upper and/or lower part of whorl; shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 17ad, ag, aj, al, ao, ar): 4.6–8.0 mm in long diameter.

Protoconchs (Fig. 17ae, ah, am, ap, as): Measurements and counts shown in Table 2 and S2. Suture prominently depressed by discrepancy between adjacent whorls; longitudinal ribs with rounded nodes in 1 row, spiral cords prominent or weak on upper and/or lower part of whorls; shell colored light beige to dark brown, 1–3 thick dark brown bands rarely on upper and lower part of each whorl and on basal part of shell.

Radulae (Fig. 16j–m): Rachidian roughly triangular, with central denticle and 2–3 small pointed triangular cusps on each side; central denticle tip of rachidian pointed, about 2.5 to 4.5 times longer than other triangular cusps. Lateral teeth with large central denticle, 1–3 inner and outer pointed cusps; central cusp of lateral teeth mostly pointed, about 1.5 to 3.5 times longer. Interior and exterior marginal teeth spoon-shaped with 4–7 rounded denticles.

Reproductive organs (Fig. 13): Female: Long, rarely short protrusions on surface of seminal receptacle. Eggs colored beige to orange

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Etymology

The specific name is a participle referring to the greatly elongated teleoconch of the new species.

Distribution and ecology

Semisulcospira elongata sp. nov. was collected at four distant localities in Lake Biwa (Fig. 1). The new species was found only on the coastal sandy and the muddy bottoms at a depth of 0–12 m. *Semisulcospira elongata* sp. nov. was found with four *S. nakasekoae*-group species: *S. ourensis* and *S. cryptica* sp. nov at Okude; *S. arenicola* at Kitafunaki and Mano; *S. davisi* at Kitafunaki. The new species was observed with three *S. niponica*-group species: *S. reticulata* at Okude and Kitafunaki, Mano; *S. decipiens* at Kitafunaki, Mano; *S. rugosa* at Kitafunaki.

Remarks

The new species can be distinguished from other congeners by a large, greatly elongated teleoconch with a larger number of smooth to weakly granulated longitudinal ribs and medium-sized protoconchs with nodes and spiral cords. Although the species represents significant variation in its shell morphological characteristics and resembles *S. arenicola* and *S. cryptica* sp. nov., the teleoconch size of the *S. elongata* sp. nov. is greater than *S. arenicola,* and the number of longitudinal ribs on teleoconchs and protoconchs of the new species is greater than two other species. The new species can also be distinguished from other congeners by the radulae with the pointed tip of the central cusp of the rachidian and the lateral teeth.

Semisulcospira cryptica Sawada sp. nov.

[New Japanese name: Shinobi-kawanina]

(Table 2, S2; Fig. 16n–p, 17au–bk)

urn:lsid:zoobank.org:act:7B994126-888E-4542-9339-2C12CFE1F9BE

Material examined

Holotype: KUZ Z4316, adult female collected from Lake Biwa, Chikubu-shima Island on 9 September 2020 by the first author.

Paratypes: KUZ Z4317–Z4319, 2 adult females, 1 male collected with holotype.

Additional materials: KUZ Z4320, 14 females, collected with Holotype; KUZ Z4321, 13 females, Z4322, 6 males from Okude on 25 July 2021.

Diagnosis

Viviparous. Teleoconch large-sized in the genus [SH 33.7 ± 3.3, 29.9 ± 2.5 mm; BWL 17.7 ± 0.9, 16.1 ± 0.8 mm], greatly elongated (SA 14.0 ± 2.0, 14.8 ± 2.4 degrees); color in beige to dark brown background; outer lip of aperture simple, smooth; 2.9 ± 0.9, 2.6 ± 0.8 BCN; 15.2 ± 5.4, 19.7 ± 5.2 longitudinal ribs smooth or weakly granulated, almost straight or strongly curved; 7.5 ± 0.5, 7.7 ± 1.0 SCN; 1.6 ± 0.1, 1.6 ± 0.1 ASR; 2.2 ± 0.2, 2.2 ± 0.2 WER. Protoconch medium-sized in the genus (SHP 2.8 ± 0.4 mm, WNP 3.4 ± 0.4), with or without rounded nodes in 1 row; prominent spiral cords present; color in light beige to dark brown, with or without 1–3 thick brown bands. Radula with pointed tip of rachidian central cusp and rounded tip of central cusp of lateral teeth.

Description of holotype (KUZ Z4316; Fig. 16n, 17au–ba)

Teleoconch: AH 10.8 mm, AL 11.4 mm, ASR 1.79, AW 6.4 mm, BCN 3, BWL 18.3 mm, FWL 4.3 mm, PWL 7.0 mm, RN 14, SA 15.7 degrees, SH 36.9 mm, SW 11.7 mm, TWL 5.6 mm, WER 2.49, WN 5.75; shell greatly elongated; suture slightly undulating; whorls slightly convex; outer lip of aperture simple, smooth; longitudinal ribs slightly oblique, almost straight, orthocline to opisthocline, weakly granulated, faded on body whorls apex of shell eroded; shell colored dark beige in background, with 1 thick dark olive band on upper and lower parts of whorls.

Operculum: 7.2 mm in long diameter; nearly egg-shaped subcircular, paucispiral, comprising around 3 whorls. Nucleus subcentral.

Protoconchs: PN 75, RNP 12, SHP 2.8, SWP 1.8, WNP 3.75; shell slightly elongated; suture strongly undulating; ribs remarkable on middle part of whorls, without node; spiral cords weak, on upper and lower parts of whorls; shell colored dark brown in background without color band.

Radula: Taenioglossa. Rachidian roughly triangular, with large central denticle and 2–3 minor pointed triangular cusps on each side; central denticle tips of rachidian pointed; central denticle of rachidian approximately regular triangular, about 3.5 times longer than other triangular cusps. Lateral teeth with large central denticle, 2–3 inner and outer pointed cusps; central denticle tips rounded, irregular triangular, about 3.0 to 3.5 times longer. Interior and exterior marginal teeth spoon-shaped with 4–5 rounded denticles.

Reproductive organ (female): Renal oviduct long, narrow, entering pallial oviduct near seminal receptacle on ventral side of soft body; short protrusions on surface of seminal receptacle. Sperm gutter extending from spermatophore bursa toward mantle cavity, curved inward along whorls. Brood pouch elongated, on dorsal side of spermatophore bursa and sperm gutter, inflated dorsally, separated into many chambers, including eggs and embryos; eggs colored beige to orange; eggs and embryos developing radially from base of brood pouch near seminal receptacle; embryos more developed in anterior or dorsal chambers.

Variation

Teleoconchs (Fig. 17bb, bd, bg, bi): Measurements and counts shown in Table 2 and S2. Suture slightly undulating on rocky substrates, moderately on mud; whorls slightly convex on rock, moderately on mud; aperture rounder on mud; longitudinal ribs distinct, slightly curved, orthocline to opisthocline on rock, strongly, opisthocline to opisthocyrt on mud, greater number on mud; apex of whorl more preserved on rock; shell colored dark beige to dark brown in background, with or without dark olive band present on upper and/or lower part of whorl; shell surface colored brown to blackish brown with deposits before shell cleaning.

Opercula (Fig. 17be, bh, bj): 4.9–7.7 mm in long diameter.

Protoconchs (Fig. 17bc, bf, bk): Measurements and counts shown in Table 2 and S2. Suture prominently depressed by discrepancy between adjacent whorls; longitudinal ribs with rounded nodes in 1 row; spiral cords mostly prominent, rarely weak, on upper and/or lower part of whorls; rib number greater on muddy bottom (SHP 2.9 ± 0.5; WNP 3.5 ± 0.4) than rock (SHP 2.7 ± 0.4; WNP 3.4 ± 0.4) in the species; shell colored light beige to dark brown, 1–3 thick dark brown bands rarely on upper and lower part of each whorl and on basal part of shell.

Radulae (Fig. 16o–p): Rachidian roughly triangular, with central denticle and 2–3 small pointed triangular cusps on each side; central denticle of rachidian about 3.0 to 4.0 times longer than other triangular cusps. Lateral teeth with large central denticle, 2–3 inner and outer pointed cusps; central denticle of lateral teeth about 2.5 to 3.5 times longer. Interior and exterior marginal teeth spoon-shaped with 4–6 rounded denticles.

Reproductive organs (Fig. 13): Female: Long or short protrusions on surface of seminal receptacle. Eggs colored dark beige to orange.

Male: Reproductive organs consisting of testes, vas deferens, and prostate without penis. Prostate elongated, inflated in posterior ventral part, with deep groove, forming U-shape in transverse section, anterior narrowly opening to mantle cavity.

Etymology

The specific name is an adjective indicating the cryptic features of the new species, which are the restricted distribution and morphological similarity to other congeners.

Distribution and ecology

Semisulcospira cryptica sp. nov. was collected at two localities on the northern side of Lake Biwa (Fig. 1). The new species was found only on the muddy coastal bottom and the insular rocky bottom at a depth of 0–6 m. The new species was found with four *S. nakasekoae*-group species: *S. elongata* sp. nov at Okude; *S. ourensis* at Okude and Chikubu-shima Island; *S. morii* at Chikubu-shima Island. Two *S. niponica*-group species, *S. decipiens* and *S. nakanoi* inhabit the new species at Chikubu-shima Island. The new species coexist with *S. reticulata* at Okude.

Remarks

The characteristics of longitudinal ribs, aperture roundness, and WN of teleoconch and RNP represent correlations with substrates. The new species can be distinguished from other congeners by a large, greatly elongated teleoconch with a medium number of smooth to weakly granulated longitudinal ribs and medium-sized protoconchs with nodes and spiral cords. Although the species resembles *S. arenicola* and *S. elongata* sp. nov., the teleoconch size of the *S. cryptica* sp. nov. is greater than *S. arenicola,* and the number of longitudinal ribs of the new species is fewer than *S. elongata* sp. nov. The new species can also be distinguished from other congeners by the radulae with the pointed tip of the rachidian central cusp and rounded tip of the central cusp of the lateral teeth.

Supplementary material

Supplementary material is available online at ###.

Data availability. The raw data that support this study will be shared upon reasonable request to the corresponding author.

Conflicts of interest. The authors declare that there are no conflicts of interest.

Declaration of funding. This study was partly supported by the Research Institute of Marine Invertebrates Grant Number IKU 2021-01 and JSPS KAKENHI Grant Number JP21J22917.

Acknowledgements. The authors are very grateful to Anna Persson (SMNH), Taehwan Lee (UMMZ), Sigrid Hof (SMF), and Shigefumi Kanao (LBM) for their kind support and for providing photographs of the type specimens. We are also grateful to Shiga Prefecture for providing permission for dredging in the sample collection. We also thank Katsutoshi Watanabe (Kyoto University; KU), Osamu Miura (Kochi University), Taku Okamoto (KU), Tomoki Kadokawa (KU), and Minoru Tamura (KU) for supporting genetic and morphological analyses. We appreciate Misako Urabe (The University of Shiga Prefecture), Ryoji Takada (NSM), Kazunori Hasegawa (NSMT), Masaharu Motokawa (KUM), Hiroshi Fukuda (Okayama University), So Ishida (Osaka Museum of Natural History), Akihiro Takami, and Tadashi Okoba for providing information. The first author is grateful to Toshiaki Nishimura, Yoshiharu Sasaki (Shiga Prefectural Fisheries Experiment Station), Yukiko Goda (KU), Tetsuji Akatsuka (KU), Takuto Miyai, and Kaoru Sawada for assisting with sample collection. We also appreciate the constructive comments of Takafumi Nakano (KU). This study was supported by the Center for Ecological Research, Kyoto University, a Joint Usage / Research Center. We thank Editage (www.editage.com) for English language editing.

Author affiliations. Department of Zoology, Graduate School of Science, Kyoto University, Kyoto 606-8502, Japan

References

Alexander DH, Novembre J, Lange K (2009) Fast model-based estimation of ancestry in unrelated individuals. *Genome Research* **19**, 1655–1664. doi:10.1101/gr.094052.109

Andy L, Matthew W (2002) Classification and regression by randomForest. *R News* **2**, 18–22.

Annandale N (1916) Zoological results of a tour in the Far East. The Mollusca of Lake Biwa, Japan. *Records of the Indian Museum* **6**, 41–74. doi:10.5962/bhl.title.46524

Boettger O (1886) Zur Kenntniss der Melanien Chinas und Japans. *Jahrbücher der Deutschen Malakozoologischen Gesellschaft* **13**, 1–16.

Breiman L (2001) Random Forests. *Machine Learning* **45**, 5–32. doi:10.1023/A:1010933404324

Brot A (1877) Die Melaniaceen (Melanidae) in Abbildungen nach der Natur mit Beschreibunge. In 'Systematisches Conchylien-Cabinet von Martini und Chemnitz, Ersten Bandes, Vierundzwanzigste Abtheilung (1) 24 (259)'. (Eds HC. Küster, W Kobelt.) pp. 193–352. (Bauer & Raspe: Nürnberg, Germany)

Burch JB (1968) Cytotaxonomy of some Japanese *Semisulcospira* (Streptoneura: Pleuroceridae). *Journal de Conchyliologie* **107**, 3–51.

Burch JB, Davis GM (1967) A taxonomic study of some species of the freshwater snail genus *Semisulcospira* in Japan. *Annual Reports* **for 1967**, 36–38.

Callomon P (2017) Research on the collections of the Museum 1. Hirase Malacological collection. *The Kyoto University Museum Newsletter* **41**, 8–9.

Chen S, Zhou Y, Chen Y, Gu J (2018) fastp: an ultra-fast all-in-one FASTQ preprocessor. *Bioinformatics* **34**, i884–i890. doi:10.1093/bioinformatics/bty560

Covich AP (2010) Winning the biodiversity arms race among freshwater gastropods: competition and coexistence through shell variability and predator avoidance. *Hydrobiologia* **653**, 191–215. doi:10.1007/s10750-010-0354-0

Davis GM (1969) A taxonomic study of some species of *Semisulcospira* in Japan (Mesogastropoda: Pleuroceridae). *Malacologia* **7**, 211–294.

Du L-N, Guo-Hua Y, Jun-xing Y (2019) Systematic relationships of Chinese freshwater semisulcospirids (Gastropoda, Cerithioidea) revealed by mitochondrial sequences. *Zoological Research* **40**, 1–11. doi:10.24272/j.issn.2095-8137.2019.033

Fukuoka G (1933) Ecological distribution of the freshwater snail *Semisulcospira* in the environs of Kyôto. *Venus* **4**, 114–118. doi:10.18941/venusomsj.4.2_114

Goto Y, Poppe G (1996) 'A listing of living mollusca. Part II. Volume I'. (L'Informatore Piceno: Ancona, Italy)

Gould AA (1859) Descriptions of shells collected in the North Pacific Exploring Expedition under Captain Ringgold and Rodgers. *Proceedings of the Boston Society of Natural History* **7**, 40–48. doi:10.1017/CBO9781107415324.004

Habe T (1968) 'Color Books 157) Japanese shellfish'. (Hoikusha Publishing Co. , Ltd.: Osaka, Japan)

Habe T (1970) Papers on the taxonomy of the Japanese *Semisulcospira*. *Venus* **28**, 191–192. doi:10.18941/venusjjm.28.4_191

Habe T (1978) Fresh water molluscs in Korea and Japan as the intermediate hosts of trematodes. *Yonsei Reports on Tropical Medicine* **9**, 91–96.

Habe T (1984) Type specimens of the Japanese land and freshwater molluscs collected during the Vega Expedition and housed in the Swedish Natural History Museum in Stockholm. *Venus* **43**, 305–313. doi:10.18941/venusjjm.43.4_305

Habe T, Kosuge S (1967) 'Common Shells of Japan in Color. 21th Edition'. (Hoikusha Publishing Co., Ltd.: Osaka, Japan)

Higo S, Goto Y (1993) 'A Systematic List of Molluscan Shells from the Japanese Is. and the Adjacent Area'. (Elle Scientific Publications, Yao, Japan)

Hirase S, Taki I (1951) 'An Illustrated Handbook of Shells in Natural Colors from the Japanese Islands and Adjacent Territory'. (Bunkyokaku: Tokyo, Japan)

Holomuzki JR, Biggs BJF (2006) Habitat-specific variation and performance trade-offs in shell armature of New Zealand mudsnails. *Ecology* **87**, 1038–1047. doi:10.1890/0012-9658(2006)87[1038:HVAPTI]2.0.CO;2

International Commisson on Zoological Nomenclature (1999) 'International Code of Zoological Nomenclature'. (International Trust for Zoological Nomenclature: London, UK)

Iwakawa T (1919) 'Catalogue of Japanese Mollusca in the Natural History Department, Tokyo Imperial Museum'. (Tokyo Imperial Museum: Tokyo, Japan)

Kajiyama H, Habe T (1961) Two new forms of the Japanese melanians, *Semisulcospira*. *Venus* **21**, 167–176.

Kamiya S, Shimamoto M, Hashimoto T (2011) Allozyme analysis of Japanese *Semisulcospira* species (Gastropoda: Pleuroceridae) reveals that Lake Biwa endemic species are not monophyletic. *American Malacological Bulletin* **29**, 23–36. doi:10.4003/006.029.0213

Kihira H, Matsuda M, Uchiyama R (2009) 'Guide of Japanese Freshwater Shellfishes (1). Freshwater Shellfishes in Lake Biwa and Yodo River, Revised Edition'. (Pieces Publishers: Yokohama, Japan)

Kikuchi N, Ohara K, Otani Y, Katori H (1996) 'Catalogue of the shellfish type specimens described by the late Dr. Tokubei Kuroda in the possession of Nishinomiya City'. (Nishinomiya City: Nishinomiya, Japan)

Kikuchi N, Ohara K, Otani Y, Katori H (1997) 'Catalogue of the shellfish type specimens described by the late Dr. Tokubei Kuroda except the possession of Nishinomiya City'. (Nishinomiya City: Nishinomiya, Japan)

Kobayashi T (1986) Karyotypes of four species of the genus *Semisulcospira* in Japan. *Venus* **45**, 127–137. doi:10.18941/venusjjm.45.2_127

Kobelt W (1876) Conchologische Miscellen. *Jahrbücher der Deutschen Malakozoologischen Gesellschaft* **3**, 275–278.

Kobelt W (1879) 'Fauna molluscorum extramarinorum Japoniae. Nach den von Professor Rein Gemachten Sammlungen'. (Christian Winter: Frankfurt, Germany) doi:10.5962/bhl.title.12881

Köhler F (2016) Rampant taxonomic incongruence in a mitochondrial phylogeny of *Semisulcospira* freshwater snails from Japan (Cerithioidea: Semisulcospiridae). *Journal of Molluscan Studies* **82**, 268–281. doi:10.1093/mollus/eyv057

Köhler F (2017) Against the odds of unusual mtDNA inheritance, introgressive hybridisation and phenotypic plasticity: systematic revision of Korean freshwater gastropods (Semisulcospiridae, Cerithioidea). *Invertebrate Systematics* **31**, 249–268. doi:10.1071/IS16077

Kubo H (1985) A sinistral specimen of *Biwakomelania decipiens* from Lake Biwa. *Chiribotan* **16**, 48.

Kuroda T (1929) On Japanese Melanians. *Venus* **1**, 179–193. doi:10.18941/venusomsj.1.5_179

Kuroda T (1941). A catalogue of molluscan shells from Taiwan (Formosa), with description of new species. *Memoirs of the Faculty of Science and Agriculture, Taihoku Imperial University* **22**, 65–216.

Kuroda T (1962) Mollusca in Lake Biwa. *Venus* **22**, 86–91.

Kuroda T, Habe T (1965) *Semisulcospira decipiens*. In 'New Illuscrated Encyclopedia of the Fauna of Japan'. (Ed. K, Okada) p. 57. (Hukuryu-kan Publishing Co. Ltd.: Tokyo, Japan)

Kurozumi T (2007) Shellfish. In 'Survey Report on Ecological Diversity of Fuji Five Lakes in the Northern Area of Mt. Fuji, Central Japan'. (Ed Fuji-hokuroku Ecological Study Group) pp. 61–71. (Fuji-hokuroku Ecological Study Group, Fujiyosida, Japan)

Lamarck JBPA (1799) Prodrome d'une nouvelle classification des coquilles, comprenant une rédaction appropriée des caractères génériques, et l'établissement d'un grand nombre de genres nouveaux. *Mémoires de la Société d'Histoire Naturelle de Paris* **1**, 63–91.

Lopes-Lima M, Hattori A, Kondo T, Hee Lee J, Ki Kim S, Shirai A, Hayashi H, Usui T, Sakuma K, Toriya T, Sunamura Y, Ishikawa H, Hoshino N, Kusano Y, Kumaki H, Utsugi Y, Yabe S, Yoshinari Y, Hiruma H, Tanaka A, Sao K, Ueda T, Sano I, Miyazaki J-I, Gonçalves D V, Klishko OK, Konopleva ES, Vikhrev I V, Kondakov A V, Yu. Gofarov M, Bolotov IN, Sayenko EM, Soroka M, Zieritz A, Bogan AE, Froufe E (2020) Freshwater mussels (Bivalvia: Unionidae) from the rising sun (Far East Asia): phylogeny, systematics, and distribution. *Molecular Phylogenetics and Evolution* **146**, 106755. doi:10.1016/j.ympev.2020.106755

Matsuoka K (1981) Research history of *Semisulcospira decipiens* and its relatives in Lake Biwa. *Kakitsubata* **7**, 3–6.

Matsuoka K (1985) Pliocene freshwater gastropods from the Iga formation of the Kobiwako group, Mie Prefecture, central Japan. *Transactions and proceedings of the Paleontological Society of Japan* **139**, 180–195.

Matsuoka K, Nakamura T (1981) Preliminary report of the freshwater molluscs from the Pleistocene Katata Formation of the Kobiwako Group in Shiga Prefecture, Japan. *Bulletin of the Mizunami Fossil Museum* **8**, 105–126.

Miura O, Urabe M, Mori H, Chiba S (2020) Ancient drainage networks mediated a large-scale genetic introgression in the East Asian freshwater snails. *Ecology and Evolution* **10**, 8186–8196. doi:10.1002/ece3.6523

Miura O, Urabe M, Nishimura T, Nakai K, Chiba S (2019) Recent lake expansion triggered the adaptive radiation of freshwater snails in the ancient Lake Biwa. *Evolution Letters* **3**, 43–54. doi:10.1002/evl3.92

MolluscaBase (2022) *Semisulcospira*. Available at https://www.molluscabase.org/aphia.php?p=taxdetails&id=741120

Morrison JPE (1952) World relations of the melanians. *American Malacological Union. News Bulletin and Annual Report* **1951**, 6–9.

Nakajima J (2012) Taxonomic study of the *Cobitis striata* complex (Cypriniformes, Cobitidae) in Japan. *Zootaxa* **3586**, 103–130. doi:10.11646/zootaxa.3586.1.11

Nakamura M, Wada K, Taki K, Hirayama N (2020) Evolving issues toward improvement of the Lake Biwa–Yodo River Basin governance In 'Lake Biwa: Interactions between Nature and People. Second Edition. (Eds H Kawanabe, M Nishino, M Maehata.) pp. 485–528. (Springer International Publishing: Cham, Switzerland) doi:10.1007/978-3-030-16969-5_6

Negoro K. (1957) The water quality of Lake Biwa-ko. *Japanese journal of limnology* 19, 1–22. doi: 10.3739/rikusui.19.1

Nishino M (1991) 'Benthos in Lake Biwa. Life in watersides'. (Lake Biwa Research Institute, Otsu, Japan)

Nishino M (2021) *Semisulcospira* (*Biwamelania*) *ourensis*, *Semisulcospira* (*Biwamelania*) *rugosa*, *Semisulcospira* (*Biwamelania*) *dilatata*, *Semisulcospira* (*Biwamelania*) *nakasekoae*, *Semisulcospira* (*Biwamelania*) *fluvialis*, *Semisulcospira* (*Biwamelania*) *arenicola*, *Semisulcospira* (*Biwamelania*) *decipiens*, *Semisulcospira* (*Biwamelania*) *habei*. In 'Red data book of Shiga Prefecture 2020'. (Ed Scientific Committee for Research into the Wildlife in Shiga Prefecture) pp. 605, 607, 609, 610, 612, 620. (Sunrise Publishing Co., Ltd., Otsu, Japan)

Nishino M, Tanida K (2018) 'Illustration of Lake Biwa benthic fauna. Second Edition'. (Incorporated Administrative Agency Japan Water Agency Lake Biwa Development Integrated Operation & Maintenance Office: Otsu, Japan)

Nishino M, Watanabe NC (2000) Evolution and endmism in Lake Biwa, with special reference to its gastropod mollusc fauna. *Advances in Ecological Research* **31**, 151–180. doi:10.1016/s0065-2504(00)31011-x

Nomoto Y (2001) Studies on the explosive radiation in freshwater snails of the genus *Biwamelania*. PhD thesis. The University of Tokyo, Japan.

Nomoto Y, Hirai M, Ueshima R (2001) Cloning of molluscan telomere DNA with (TTAGGG) n repeat and its chromosomal location in the freshwater snail *Biwamelania habei*. *Zoological Science* **18**, 417–422. doi:10.2108/zsj.18.417

Oniwa K, Kimura M (1986) Genetic variability and relationships in six snail species of the genus *Semisulcospira*. *The Japanese journal of genetics* **61**, 503–514. doi:10.1266/jjg.61.503

Pilsbry HA (1902) Revision of Japanese Viviparidæ, with notes on *Melania* and *Bithynia*. *Proceedings of the Academy of Natural Sciences of Philadelphia* **54**, 115–121.

Prozorova LA, Rasshepkina AV (2006) On the radula and pallial gonoduct morphology of the gastropod *Biwamelania decipiens* and *B. multigranosa* (Cerithioidea: Pleuroceridae: Semisulcospirinae). *Bulletin of the Russian Far East Malacological Society* **10**, 130–132.

Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MAR, Bender D, Maller J, Sklar P, de Bakker PIW, Daly MJ, Sham PC (2007) PLINK: A Tool Set for Whole-Genome Association and Population-Based Linkage Analyses. *The American Journal of Human Genetics* **81**, 559–575. doi:10.1086/519795

R Development Core Team (2019) R for Windows, Version 3.6.1. R Foundation for Statistical Computing. Available at https://www.R-project.org/

Rintelen T von, Wilson AB, Meyer A, Glaubrecht M (2004) Escalation and trophic specialization drive adaptive radiation of freshwater gastropods in ancient lakes on Sulawesi, Indonesia. *Proceedings of the Royal Society of London. Series B: Biological Sciences* **271**, 2541–2549. doi:10.1098/rspb.2004.2842

Rochette NC, Rivera-Colón AG, Catchen JM (2019) Stacks 2: Analytical methods for paired-end sequencing improve RADseq-based population genomics. *Molecular Ecology* **28**, 4737–4754. doi:10.1111/mec.15253

Sarasin P, Sarasin F (1897) Über die Molluskenfauna der großen Süßwasser-Seen von Central-Celebes. *Zoologischer Anzeiger* **539**, 308–320.

Sawada N, Nakano T (2021) Revisiting a 135-year-old taxonomic account of the freshwater snail *Semisulcospira multigranosa*: designating its lectotype and describing a new species of the genus (Mollusca: Gastropoda: Semisulcospiridae). *Zoological Studies* **60**, 7. doi:10.6620/ZS.2021.60-07

Sawada N, Fuke Y (2022) Diversification in ancient Lake Biwa: integrative taxonomy reveals overlooked species diversity of the Japanese freshwater snail genus *Semisulcospira* (Mollusca: Semisulcospiridae). *Contributions to Zoology*. doi: 10.1163/18759866-bja10035

Sawada N, Nakano T (2022) Insights into sex-related size and shape dimorphism in *Semisulcospira niponica* (Caenogastropoda: Cerithioidea: Semisulcospiridae). *Journal of Molluscan Studies* **88**, eyac013. doi: 10.1093/mollus/eyac013

Sawada N, Toyohara H, Miyai T, Nakano T (2020) Further records of introduced *Semisulcospira* snails in Japan (Mollusca, Gastropoda): implications for these snails' correct morphological identification. *BioInvasions Records* **9**, 310–319. doi:10.3391/bir.2020.9.2.16

Sawada N, Toyohara H, Nakano T (2021) A New cleaning method for accurate examination of freshwater gastropod shell specimens covered with iron-rich deposits. *Species Diversity* **26**, 217–224. doi:10.12782/specdiv.26.217

Schneider CA, Rasband WS, Eliceiri KW (2012) NIH Image to ImageJ: 25 years of image analysis. *Nature Methods* **9**, 671–675. doi:10.1038/nmeth.2089

Scientific Committee for Research into the Wildlife in Shiga Prefecture (2021) 'Red data book of Shiga Prefecture 2020'. (Sunrise Publishing Co., Ltd: Hikone, Japan)

Setoguchi Y (2020) Geological History of paleo- and present Lake Biwa. In 'Lake Biwa: Interactions between Nature and People. Second Edition'. (Eds H Kawanabe, M Nishino, M Maehata.) pp. 17–24. (Springer International Publishing: Cham, Switzerland) doi:10.1007/978-3-030-16969-5_1

Smith EA (1876) Descriptions of some new species of land and freshwater shells, and remarks on other species found in Japan. *Quarterly Journal of Conchology* **1**, 118–127.

Society for the Study of Aquatic Life (1989) 'Report on research commissioned by the Lake Biwa Research Institute. Studies on the taxonomic reconsideration of the major benthic fauna'. (Lake Biwa Research Institute, Otsu, Japan)

Strong EE, Köhler F (2009) Morphological and molecular analysis of '*Melania*' *jacqueti* Dautzenberg and Fischer, 1906: From anonymous orphan to critical basal offshoot of the Semisulcospiridae (Gastropoda: Cerithioidea). *Zoologica Scripta* **38**, 483–502. doi:10.1111/j.1463-6409.2008.00385.x

Suyama Y, Matsuki Y (2015) MIG-seq: an effective PCR-based method for genome-wide single-nucleotide polymorphism genotyping using the next-generation sequencing platform. *Scientific Reports* **5**, 16963. doi:10.1038/srep16963

Tabata R, Kakioka R, Tominaga K, Komiya T, Watanabe K (2016) Phylogeny and historical demography of endemic fishes in Lake Biwa: the ancient lake as a promoter of evolution and diversification of freshwater fishes in western Japan. *Ecology and Evolution* **6**, 2601–2623. doi:10.1002/ece3.2070

Takami A (1994) The number and size of newborns in *Semisulcospira nakasekoae* and *S. habei* (Prosobranchia: Pleuroceridae). *Venus* **53**, 201–209. doi: 10.18941/venusjjm.53.3_201

Takami A (2013) Karyotypes of three lotic species of the genus *Semisulcospira* (Neotaenioglossa: Pleuroceridae) in Japan. *Venus* **71**, 97–103. doi:10.18941/venus.71.1-2_97

Takami A (2019) Reexamination of the karyotypes of three species of the genus *Semisulcospira* (Neotaenioglossa: Pleuroceridae). *Venus* **77**, 37–43. doi:10.18941/venus.77.1-4_37

Takigawa Y, Kato S, Nakano T, Nakai K, Tomikawa K, Ishiwata S, Fujita T, Hosoya K, Kawase S, Senou H, Yoshino T, Nishino M (2020) The Vega collection at the end of the nineteenth-century survey of Lake Biwa. In 'Lake Biwa: Interactions between Nature and People. Second Edition'. (Eds H Kawanabe, M Nishino, M Maehata.) pp. 248–257. (Springer International Publishing: Cham, Switzerland) doi:10.1007/978-3-030-16969-5_2

The Lake Biwa Canal Promotion Council (2022) The Lake Biwa Canal. Available at: https://biwakososui.city.kyoto.lg.jp/en/story/

Troschel FH (1856–1863) 'Das Gebiss der Schnecken zur Begründung einer natürlichen Classification. Vol. 1.' (Nicolaische Verlagsbuchhandlung: Berlin, Germany)

Urabe M (1998) Contribution of genetic and environmental factors to shell shape variation in the lotic snail *Semisulcospira reiniana* (Prosobranchia: Pleuroceridae). *Journal of Molluscan Studies* **64**, 329–343. doi:10.1093/mollus/64.3.329

Urabe M (2000) Phenotypic modulation by the substratum of shell sculpture in *Semisulcospira reiniana* (Prosobranchia: Pleuroceridae). *Journal of Molluscan Studies* **66**, 53–60. doi:10.1093/mollus/66.1.53

Urabe M (2007) Note on *Melania niponica* var. *minor* (Gastropoda: Pleuroceridae). *Chiribotan* **38**, 80–87.

Verhaegen G, Herzog H, Korsch K, Kerth G, Brede M, Haase M (2019) Testing the adaptive value of gastropod shell morphology to flow: a multidisciplinary approach based on morphometrics, computational fluid dynamics and a flow tank experiment. *Zoological Letters* **5**, 5. doi:10.1186/s40851-018-0119-6

Watanabe NC (1970) Studies on three species of *Semisulcospira* in Lake Biwa : i) Comparative studies of shell form and habitat. *Venus* **29**, 13–30. doi:10.18941/venusjjm.29.1_13

Watanabe NC (1984) Studies on taxonomy and distribution of the freshwater snails, genus *Semisulcospira* in the three islands inside Lake Biwa. *Venus* **45**, 194–203. doi:10.3739/rikusui.45.194

Watanabe NC, Nishino M (1995) A study on taxonomy and distribution of the freshwater snail, genus *Semisulcospira* in Lake Biwa, with descriptions of eight new species. *Lake Biwa Study Monographs* **6**, 1–36.

Wenz W (1939) Gastropoda. Teil 3: Prosobranchia. In 'Handbuch der Paläozoologie Band 6 Lieferung 4'. (Ed. OH, Schindewolf.) pp. 481–720. (Bornträger: Berlin, Germany)

Westerlund A (1883) Von der Vega-expedition in Asien gesammelte Binnen mollusken. *Nachrichtsblatt der Deutschen Malakozoologischen Gesellschaft.* **15**, 48–59.

Yusa Y, Sugiura N, Wada T (2006) Predatory potential of freshwater animals on an invasive agricultural pest, the apple snail *Pomacea canaliculata* (Gastropoda: Ampullariidae), in southern Japan. *Biological Invasions* **8**, 137–147. doi:10.1007/s10530-004-1790-4

Figure legends

Fig. 1. Map of collection sites of nine *Semisulcospira* species showing 29 sampling localities. (a) *Semisulcospira niponica*-group: blue, *S. decipiens*; orange, *S. habei*; red, *S. rugosa*; purple, *S. reticulata*. (b) *S. nakasekoae*-group: blue, *S. ourensis*; orange, *S. arenicola*; green, *S. elongata* sp. nov.; red, *S. cryptica* sp. nov.; purple, *S. nakasekoae*; black, putative hybrid between *S. arenicola* and *S. nakasekoae*.

Fig. 2. Protoconchs representing the criteria for the Node Number and the Spiral Cord Type of the protoconchs in this study. (a) Granulated ribs (3 nodes) and a prominent spiral cord. (b) Nodes and a weak spiral cord. (c) Ribs without spiral cord.

Fig. 3. Results of the first principal components analysis based on 628 SNPs conducted for the four *Semisulcospira niponica*-group species.

Fig. 4. Results of the first ADMIXTURE analysis based on 628 SNPs conducted for the four *Semisulcospira niponica*-group species.

Fig. 5. Results of the first principal components analysis based on 804 SNPs conducted for the five *Semisulcospira nakasekoae*-group species.

Fig. 6. Results of the first ADMIXTURE analysis based on 804 SNPs conducted for the five *Semisulcospira nakasekoae*-group species.

Fig. 7. Results of the second principal components analysis based on 781 SNPs conducted for the three *Semisulcospira nakasekoae*-group species.

Fig. 8. Results of the second ADMIXTURE analysis based on 781 SNPs conducted for the three *Semisulcospira nakasekoae*-group species.

Fig. 9. Results of the Random Forest analyses conducted for the four *Semisulcospira niponica*-group species. Euclidean distances generated from proximities among individuals are plotted.

Fig. 10. Results of the Random Forest analyses conducted for the five *Semisulcospira nakasekoae*-group species. Euclidean distances generated from proximities among individuals are plotted.

Fig. 11. Shells of the four *Semisulcospira niponica*-group species. (a–ax), *S. decipiens*; (ay–bj), *S. habei*; (bk–bp), *S. rugosa*; (bq–bu), *S. reticulata*. (a–l, ay–ba), Vouchered specimens; (m–ax, bb–bu), Newly collected specimens. (a–c), Holotype of *S. decipiens*, SMNH-Type-1614; (d–f), Lectotype of *S. multigranosa*, SMF 225654; (g–i), Holotype of *S. habei yamaguchi*, UMMZ 228801; (j–l), Holotype of *S. dilatata*, LBM 13-3; (m–o), Hannoura, KUZ Z4208; (p–r), Oura Port, KUZ Z4209; (s–u), Ebie, KUZ Z4210; (v–x), Chikubu-shima Island, KUZ Z4211; (y–aa, bn–bp), Kitafunaki, KUZ Z2513, Z2494; (ab–ad), Lake Matsunoki, KUZ Z4214; (ae–ag), Iso, KUZ Z4216; (ah–aj), Kitakomatsu, KUZ Z4218; (ak–am), Oki-shima Island, KUZ Z4219; (an, ao, bq–bu), Mano, KUZ Z4220, Z4229, Z4230; (ap–ar), Katata Port, KUZ Z4221; (as–au), Otsu Port, KUZ

Z4222; (av–ax), Araizeki, KUZ Z4223; (ay–ba), Holotype of *S. habei*, UMMZ 220236; (bb–bd), Uji, KUZ Z4224; (be–bg), Fushimi, KUZ Z4225; (bh–bj), Yawata, KUZ Z4226; (bk–bm), Imazu Beach, KUZ Z4228. Scale bars: 10 mm, (d–l, m, p, s, v, y, ab, ae, ah, ak, ap, as, av, ay–bb, be, bh, bk, bn, bq), adult, (a–c, an, bt), juvenile, (n, q, t, w, z, ac, af, ai, al, ao, aq, at, aw, bc, bf, bi, bl, bo, br, bu), operculum; 1 mm, (o, r, u, x, aa, ad, ag, aj, am, ar, au, ax, bd, bg, bj, bm, bp, bs), protoconch. Newly collected specimens were treated with 3% sodium hypochlorite.

Fig. 12. Radulae of the four *Semisulcospira niponica*-group species. (a–j), *S. decipiens*; (k–m), *S. habei*; (n, o), *S. rugosa*; (p), *S. reticulata*. (a), Hannoura, KUZ Z4208; (b), Oura Port, KUZ Z4209; (c), Ebie, KUZ Z4210; (d), Chikubu-shima Island, KUZ Z4211; (e), Lake Matsunoki, KUZ Z4214; (f), Iso, KUZ Z4216; (g), Kitakomatsu, KUZ Z4218; (h), Oki-shima Island, KUZ Z4219; (i), Katata Port, KUZ Z4220; (j), Araizeki, KUZ Z4223; (k), Uji, KUZ Z4224; (l), Fushimi, KUZ Z4225; (m), Yawata, KUZ Z4226; (n), Imazu Beach, KUZ Z4228; (o), Kitafunaki, KUZ Z2499; (p), Mano, KUZ Z4229. Scale bars: 100 μm.

Fig. 13. Schematic drawings indicating generalized features of reproductive organs of *Semisulcospira* species nov. (a), Female; (b), male. Abbreviations: bp, brood pouch; eg, egg; em, embryo; ov, oviduct; pr, prostate; pt, protrusions in the seminal receptacle; rcs, seminal receptacle; sg, sperm gutter; spb, spermatophore bursa; vd, vas deferens. Scale bars: 1 mm.

Fig. 14. Shells of *Semisulcospira arenicola* (a–ad), *S. nakasekoae* (ae–be), and their putative hybrids (bf–bk). (a–c, ae–ag), Vouchered specimens; (d–ae, ah–bk), Newly collected specimens. (a–c), Holotype of *S. arenicola*, LBM 13-8; (d–f), Minamihama, KUZ Z4231; (g–i), Tamura, KUZ Z4232; (j–l), Kitafunaki, KUZ Z4233; (m–o), Yokoehama, KUZ Z4234; (p–r), Satsuma, KUZ Z4235; (s–u), Horikiri Port, KUZ Z4236; (v–x), Wani Beach, KUZ Z4237; (y–ab), Mano, KUZ Z4238, Z4239; (ac–ae), Otsu Port, KUZ Z4240; (ae–ag), Holotype of *S. fluvialis*, LBM 13-16; (ah–aj, bf–bh), Araizeki, KUZ Z4241, Z4253; (ak–ap, bi–bk), Nango, KUZ Z4242, Z4254; (aq–as), Uji, KUZ Z4243; (at–av), Fushimi, KUZ Z4244; (aw–ay), Higashiyama, KUZ Z4245; (az–bb), Yawata, KUZ Z4246; (bc–be), Neyagawa, KUZ Z4247. Scale bars: 10 mm, (a–d, g, j, m, p, s, v, y, ac, ae–ah, ak, an, aq, at, aw, az, bc, bf, bi), adult, (ab), juvenile, (e, h, k, n, q, t, w, z, ad, ai, al, ao, ar, au, ax, ba, bd, bg, bj), operculum; 1 mm, (f, i, l, o, r, u, x, aa, ae, aj, am, ap, as, av, ay, bb, be, bh, bk), protoconch. Newly collected specimens were treated with 3% sodium hypochlorite.

Fig. 15. Radulae of *Semisulcospira arenicola* (a–i), *S. nakasekoae* (j–p). (a), Minamihama, KUZ Z4231; (b), Tamura, KUZ Z4232; (c), Kitafunaki, KUZ Z4233; (d), Yokoehama, KUZ Z4234; (e), Satsuma, KUZ Z4235; (f), Horikiri Port, KUZ Z4236; (g), Wani Beach, KUZ Z4237; (h), Mano, KUZ Z4238; (i), Otsu Port, KUZ Z4240; (j), Araizeki, KUZ Z4241; (k, l), Nango, KUZ Z4242; (m), Uji, KUZ Z4243; (n), Fushimi, KUZ Z4244; (o), Higashiyama, KUZ Z4245; (p), Yawata, KUZ Z4246. Scale bars: 100 μm.

Fig. 16. Radulae of the four *Semisulcospira nakasekoae*-group species and putative hybrids between *S. arenicola* and *S. nakasekoae*. (a), *S. nakasekoae*; (b, c), putative hybrids; (d–h), *S. ourensis*; (i–m), *S. elongata* sp. nov.; (n–p), *S. cryptica* sp. nov. (a), Neyagawa, KUZ Z4247; (b), Araizeki, KUZ Z4253; (c), Nango, KUZ Z4254; (d, e), Oura, KUZ Z4248; (f, k, p), Okude, KUZ Z4250, Z4312, Z4321; (g), Sugaura, KUZ Z4251; (h, n, o), Chikubu-shima Island, KUZ Z4252, Z4216, Z4217; (i, j), Kitafunaki, KUZ Z4305, Z4309; (l), Imazu Beach, KUZ Z4311; (m), Mano, KUZ Z4313. Scale bars: 100 μm.

Fig. 17. Shells of the three *Semisulcospira nakasekoae*-group species. (a–u), *S. ourensis*; (v–at), *S. elongata* sp. nov.; (au–bk), *S. cryptica* sp. nov. (a–c), Vouchered specimens; (d–bk), Newly collected specimens. (a–c), Holotype of *S. ourensis*, LBM 13-7; (d–i), Oura, KUZ Z4248; (j–l, ak–am, bi–bk), Okude, KUZ Z4250, Z4310, Z4321; (m–r), Sugaura, KUZ Z4251; (s–u) Chikubu-shima Island, KUZ Z4252 (v–ab), Holotype of *S. elongata* sp. nov. from Kitafunaki, KUZ Z4305; (ac–aj), Paratypes of *S. elongata* sp. nov. from Kitafunaki, KUZ Z4306–Z4308; (an–ap), Imazu Beach, KUZ Z4311; (aq–at), Mano KUZ Z4313, Z4315; (au–ba), Holotype of *S. cryptica* sp. nov. from Chikubu-shima Island, KUZ Z4316; (bb–bh), Paratypes of *S. cryptica* sp. nov. from Chikubu-shima Island, KUZ Z4317–Z4319. Scale bars: 10 mm, (a–d, g, j, m, p, s, v–x, ac, af, ai, ak, an, aq, au–aw, bb, bd, bg, bi), adult, (at), juvenile, (e, h, k, n, q, t, y, ad, ag, aj, al, ao, ar, ax, be, bh, bj), operculum; 1 mm, (f, i, l, o, r, u, z–ab, ae, ah, am, ap, as, av, ay–ba, bc, bf, bk), protoconch. Newly collected specimens were treated with 3% sodium hypochlorite.

Table 1. Specimen list of the *Semisulcospira* species with the voucher numbers, collection localities, and the DDBJ Sequence Read Archive (DRA) accession numbers for the specimens used for the phylogenetic analysis.

Vouncher number	Collection locality	DRA accession number
Semisulcospira decipiens		
SMNH-Type-1614 (holotype)	Lake Biwa, Japan (Westerlund, 1883)	
KUZ Z4208, Z4273	Lake Biwa, Hannoura, Nagahama City, Shiga Prefecture, Japan	DRR398459–DRR398463
KUZ Z4209	Lake Biwa, Oura Port, Oura, Nagahama City, Shiga Prefecture, Japan	DRR398588, DRR398589
KUZ Z4210, Z4274	Creak flows into Lake Biwa, Ebie, Nagahama City, Shiga Prefecture, Japan	DRR398445–DRR398449
KUZ Z4211	Lake Biwa, Chikubu-shima Island, Nagahama City, Shiga Prefecture, Japan	DRR398436, DRR398437
KUZ Z2513, 4212, Z4213	Lake Biwa, Kitafunaki, Takashima City, Shiga Prefecture, Japan	DRR398488
KUZ Z4214, Z4275	Lake Matsunoki, Yotsugawa, Takashima City, Shiga Prefecture, Japan	DRR398506–DRR398510
KUZ Z4215–Z4217, Z4276	Lake Biwa, Iso, Maibara City, Shiga Prefecture, Japan	DRR398477–DRR398480
KUZ Z4218, Z4277	Lake Biwa, Kitakomatsu, Otsu City, Shiga Prefecture, Japan	DRR398501–DRR398505
KUZ Z4219, Z4278	Lake Biwa, Oki-shima Island, Okishima-cho, Omihachiman City, Shiga Prefecture, Japan	DRR398555–DRR398557
KUZ Z4220	Lake Biwa, Mano, Otsu City, Shiga Prefecture, Japan	DRR398523, DRR398524
KUZ Z4221, Z4279	Lake Biwa, Katata Port, Honkatata, Otsu City, Shiga Prefecture, Japan	DRR398481–DRR398484
KUZ Z4222, Z4280	Lake Biwa, Otsu Port, Hamaotsu, Otsu City, Shiga Prefecture, Japan	DRR398578–DRR398582
KUZ Z4223, Z4281	Seta River, around Araizeki, Nango, Otsu City, Shiga Prefecture, Japan	DRR398426–DRR398430
Semisulcospira multigranosa		
SMF 225654 (lectotype)	rice field near Lake Biwa, Japan	
Semisulcospira habei yamaguchi		
UMMZ 228801 (holotype)	Lake Biwa, Shina-naka Port, Shina-naka, Kusatsu City, Shiga Prefecture, Japan	
Semisulcospira dilatata		
LBM 13-3 (holotype)	Lake Biwa, Iso, Maibara City, Shiga Prefecture, Japan	
Semisulcospira habei		
UMMZ 220236 (holotype)	Uji River, Uji City, Kyoto Prefecture, Japan	
KUZ Z4224, Z4282	Uji River, Oshima, Uji City, Kyoto Prefecture, Japan	DRR398605–DRR398609
KUZ Z4225, Z4283	Uji River, Yokoohji-shimomisu-higashinokuchi, Fushimi-ku, Kyoto City, Kyoto Prefecture, Japan	DRR398450–DRR398454
KUZ Z4226, Z4284	Uji River, Yawata-zaiohji, Yawata City, Kyoto Prefecture, Japan	DRR398620–DRR398624
Semisulcospira rugosa		
KUZ Z4227, Z4228, Z4285	Lake Biwa, Imazu Beach, Hamabun, Takashima City, Shiga Prefecture, Japan	DRR398476
KUZ Z2493–Z2502, Z2504–Z2506	Lake Biwa, Kitafunaki, Takashima City, Shiga Prefecture, Japan	DRR398496–DRR398500
Semisulcospira reticulata		

KUZ Z4229, Z4230, Z4286	Lake Biwa, Mano, Otsu City, Shiga Prefecture, Japan	DRR398530–DRR398532
Semisulcospira arenicola		
LBM 13-8 (holotype)	Lake Biwa, Satsuma-cho, Hikone City, Shiga Prefecture, Japan	
KUZ Z4231, Z4287	Lake Biwa, Minamihama-cho, Nagahama City, Shiga Prefecture, Japan	DRR398533–DRR398537
KUZ Z4232, Z4288	Lake Biwa, Tamura-cho, Maibara City, Shiga Prefecture, Japan	DRR398600–DRR398604
KUZ Z4233, Z4289	Lake Biwa, Kitafunaki, Takashima City, Shiga Prefecture, Japan	DRR398485–DRR398487
KUZ Z4234	Lake Biwa, Yokoehama, Takashima City, Shiga Prefecture, Japan	DRR398630–DRR398634
KUZ Z4235, Z4290	Lake Biwa, Satsuma-cho, Hikone City, Shiga Prefecture, Japan	DRR398590–DRR398594
KUZ Z4236, Z4291	Lake Biwa, Horikiri Port, Okishima-cho, Omihachiman City, Shiga Prefecture, Japan	DRR398465–DRR398469
KUZ Z4237, Z4292	Lake Biwa, Wani Beach, Wani-nakahama, Otsu City, Shiga Prefecture, Japan	DRR398615–DRR398619
KUZ Z4238, Z4239, Z4293	Lake Biwa, Mano, Otsu City, Shiga Prefecture, Japan	DRR398511–DRR398522
KUZ Z4240, Z4294	Lake Biwa, Otsu Port, Hamaotsu, Otsu City, Shiga Prefecture, Japan	DRR398574–DRR398577
Semisulcospira nakasekoae		
KUZ Z4241, Z4295	Seta River, around Araizeki, Nango, Otsu City, Shiga Prefecture, Japan	DRR398434, DRR398435
KUZ Z4242, Z4296	Seta River, Nango, Otsu City, Shiga Prefecture, Japan	DRR398539–DRR398544
KUZ Z4243, Z4297	Uji River, Ujiotokata, Uji City, Kyoto Prefecture, Japan	DRR398610–DRR398614
KUZ Z4244, Z4298	Horikawa River flows into Uji River, Yoshijimakanaido-cho, Fushimi-ku, Kyoto City, Kyoto Prefecture, Japan	DRR398455–DRR398458
KUZ Z4245, Z4299	Lake Biwa Canal, Horiike-cho, Higashiyama-ku, Kyoto City, Kyoto Prefecture, Japan	DRR398464
KUZ Z4246, Z4300	Uji River, Yawata-zaiohji, Yawata City, Kyoto Prefecture, Japan	DRR398625–DRR398629
KUZ Z4247, Z4301	Yodo River, Shimeno, Neyagawa City, Osaka Prefecture, Japan	DRR398545–DRR398554
Semisulcospira fluvialis		
LBM 13-16 (holotype)	Seta River, Nango, Otsu City, Shiga Prefecture, Japan	
Semisulcospira ourensis		
LBM 13-7 (holotype)	Lake Biwa, Oura, Nagahama City, Shiga Prefecture, Japan	
KUZ Z4248, Z4249, Z4302	Lake Biwa, Oura, Nagahama City, Shiga Prefecture, Japan	DRR398583–DRR398587
KUZ Z4250, Z4303	Lake Biwa, Okudeenchi, Sugaura, Nagahama City, Shiga Prefecture, Japan	DRR398561, DRR398562
KUZ Z4251, Z4304	Lake Biwa, Sugaura, Nagahama City, Shiga Prefecture, Japan	DRR398595–DRR398599
KUZ Z4252	Lake Biwa, Chikubu-shima Island, Nagahama City, Shiga Prefecture, Japan	DRR398438, DRR398439
Semisulcospira elongata sp. nov.		
KUZ Z4305 (holotype)	Lake Biwa, Kitafunaki, Takashima City, Shiga Prefecture, Japan	DRR398489
KUZ Z4306–Z4308 (paratypes)	Lake Biwa, Kitafunaki, Takashima City, Shiga Prefecture, Japan	DRR398492, DRR398493, DRR398495
KUZ Z4309	Lake Biwa, Kitafunaki, Takashima City, Shiga Prefecture, Japan	DRR398491, DRR398494
KUZ Z4310	Lake Biwa, Okudeenchi, Sugaura, Nagahama City, Shiga Prefecture, Japan	DRR398558–DRR398560
KUZ Z4311, Z4312	Lake Biwa, Imazu Beach, Hamabun, Takashima City, Shiga Prefecture, Japan	DRR398470–DRR398475
KUZ Z4313–Z4315	Lake Biwa, Mano, Otsu City, Shiga Prefecture, Japan	DRR398525–DRR398529

Semisulcospira cryptica sp. nov.		
KUZ Z4316 (holotype)	Lake Biwa, Chikubu-shima Island, Nagahama City, Shiga Prefecture, Japan	DRR398440
KUZ Z4317–Z4319 (paratypes)	Lake Biwa, Chikubu-shima Island, Nagahama City, Shiga Prefecture, Japan	DRR398441, DRR398442
KUZ Z4320	Lake Biwa, Chikubu-shima Island, Nagahama City, Shiga Prefecture, Japan	DRR398443, DRR398444
KUZ Z4321, Z4322	Lake Biwa, Okudeenchi, Sugaura, Nagahama City, Shiga Prefecture, Japan	DRR398563–DRR398573
A putative hybrid between *S. arenicola* and *S. nakasekoae*		
KUZ Z4253, Z4323	Seta River, around Araizeki, Nango, Otsu City, Shiga Prefecture, Japan	DRR398431–DRR398433
KUZ Z4254	Seta River, Nango, Otsu City, Shiga Prefecture, Japan	DRR398538

Table 2. Morphometric characters of the nine *Semisulcospira* species examined in the present study. Measurements and counts: minimum–maximum value (mean ± SD).

Species	*S. decipiens*	*S. habei*	*S. rugosa*	*S. reticulata*	*S. arenicola*	*S. nakasekoae*	*S. ourensis*	*S. elongata* sp. nov.	*S. cryptica* sp. nov.
specimen number of shells (mature female teleoconch / protoconch)	117 / 72	40 / 23	24 / 18	4 / 3	106 / 70	86 / 63	31 / 26	29 / 24	30 / 21
specimen number of radulae	35	9	5	3	27	22	11	11	6
Morphological characters of mature female teleoconchs									
Aperture height (AH) (mm)	5.8–15.9 (11.2 ± 1.9)	9.0–12.2 (10.4 ± 0.8)	9.8–13.0 (11.1 ± 0.9)	8.0–12.0 (10.3 ± 1.7)	6.8–11.1 (8.4 ± 0.9)	5.5–11.4 (8.8 ± 1.3)	7.3–10.0 (8.4 ± 0.6)	7.8–11.6 (9.6 ± 1.1)	9.1–11.9 (10.2 ± 0.6)
Aperture length (AL) (mm)	5.7–15.8 (11.4 ± 1.8)	9.1–12.4 (10.5 ± 0.9)	10.1–13.5 (11.4 ± 0.9)	8.7–12.1 (10.7 ± 1.4)	6.8–10.9 (8.2 ± 0.8)	5.8–12.8 (9.1 ± 1.5)	7.2–10.4 (8.4 ± 0.7)	7.5–11.6 (9.3 ± 1.1)	9.0–12.2 (10.4 ± 0.7)
Aperture slenderness ratio (ASR)	1.49–1.88 (1.7 ± 0.1)	1.58–1.92 (1.7 ± 0.1)	1.59–2.10 (1.7 ± 0.1)	1.30–1.50 (1.4 ± 0.1)	1.43–1.80 (1.6 ± 0.1)	1.50–1.86 (1.7 ± 0.1)	1.49–1.92 (1.6 ± 0.1)	1.34–1.76 (1.6 ± 0.1)	1.43–1.86 (1.6 ± 0.1)
Aperture width (AW) (mm)	3.5–9.8 (6.8 ± 1.1)	5.2–6.9 (6.0 ± 0.5)	5.6–7.8 (6.6 ± 0.6)	6.1–8.4 (7.6 ± 1.0)	4.3–6.7 (5.2 ± 0.5)	3.4–7.3 (5.4 ± 0.9)	4.5–6.1 (5.1 ± 0.4)	5.0–7.4 (6.0 ± 0.6)	5.8–7.1 (6.3 ± 0.4)
Basal cord number (BCN)	2–6 (4.0 ± 1.0)	3–6 (3.8 ± 0.8)	2–5 (3.8 ± 0.8)	3–4 (3.5 ± 0.6)	1–4 (2.3 ± 0.6)	2–6 (3.8 ± 1.0)	2–4 (2.6 ± 0.7)	2–4 (2.8 ± 0.7)	1–5 (2.9 ± 0.9)
Body whorl length (BWL) (mm)	9.2–26.0 (18.6 ± 3.0)	15.1–20.0 (17.2 ± 1.2)	16.5–20.9 (18.5 ± 1.4)	13.7–19.9 (17.6 ± 2.8)	12.0–18.9 (14.6 ± 1.5)	9.4–19.2 (15.0 ± 2.4)	12.7–18.0 (14.4 ± 1.2)	13.5–20.2 (16.5 ± 1.8)	16.1–20.2 (17.7 ± 0.9)
Fourth whorl length (FWL) (mm)	2.0–5.7 (4.0 ± 0.6)	2.9–4.5 (3.6 ± 0.3)	3.2–4.8 (3.8 ± 0.4)	3.4–4.4 (4.1 ± 0.5)	2.9–4.8 (3.6 ± 0.4)	1.2–4.0 (2.8 ± 0.6)	3.0–4.9 (3.6 ± 0.5)	3.1–5.3 (4.1 ± 0.5)	4.0–5.5 (4.6 ± 0.4)
Penultimate whorl length (PWL) (mm)	3.1–9.7 (6.7 ± 1.1)	5.4–7.2 (6.2 ± 0.5)	5.6–7.9 (6.8 ± 0.7)	5.1–7.4 (6.7 ± 1.1)	4.4–7.5 (5.8 ± 0.6)	3.4–7.5 (5.5 ± 0.9)	4.9–7.5 (5.7 ± 0.6)	4.9–7.9 (6.5 ± 0.7)	6.2–8.1 (7.0 ± 0.5)
Longitudinal rib number of penultimate whorl (RN)	12–25 (16.7 ± 2.3)	15–23 (19.7 ± 1.8)	10–22 (15.5 ± 2.9)	25–34 (29.3 ± 4.4)	10–22 (16.0 ± 2.8)	12–26 (18.2 ± 3.4)	11–20 (13.7 ± 2.3)	13–29 (20.0 ± 3.8)	9–28 (15.2 ± 5.4)
Spire angle (SA) (degrees)	14.3–24.8 (19.4 ± 2.4)	13.7–23.6 (18.4 ± 2.5)	15.1–25.6 (21.2 ± 2.8)	18.5–20.3 (19.5 ± 0.8)	10.2–18.6 (13.5 ± 1.7)	12.3–40.3 (22.0 ± 5.5)	10.0–16.8 (13.4 ± 1.8)	7.7–18.3 (13.6 ± 2.7)	8.1–17.3 (14.0 ± 2.0)
Spiral cord number of penultimate whorl (SCN)	4–8 (6.0 ± 0.9)	5–8 (6.5 ± 0.8)	4–8 (5.6 ± 1.1)	5–6 (5.5 ± 0.6)	6–9 (7.4 ± 0.8)	7–13 (9.2 ± 1.1)	6–8 (6.9 ± 0.7)	5–10 (7.9 ± 1.2)	7–8 (7.5 ± 0.5)

Shell height (SH) (mm)	17.1–47.7 (32.9 ± 5.1)	25.5–36.3 (29.5 ± 2.6)	25.1–33.7 (30.3 ± 2.1)	28.2–41.2 (36.6 ± 5.7)	21.9–39.8 (27.0 ± 3.3)	11.8–28.6 (21.7 ± 4.0)	20.9–37.3 (25.7 ± 3.3)	25.4–43.2 (32.3 ± 4.3)	28.7–40.5 (33.7 ± 3.3)
Shell width (SW) (mm)	6.1–16.9 (11.8 ± 1.8)	9.1–12.7 (10.6 ± 0.9)	10.4–13.4 (11.7 ± 0.9)	10.8–14.6 (13.3 ± 1.7)	7.1–11.5 (8.8 ± 0.9)	6.0–12.9 (9.4 ± 1.7)	7.6–11.1 (9.2 ± 0.8)	9.2–13.6 (10.4 ± 1.1)	10.6–12.8 (11.6 ± 0.6)
Third whorl length (TWL) (mm)	2.7–7.9 (5.2 ± 0.8)	4.0–5.6 (4.7 ± 0.4)	4.1–6.2 (4.9 ± 0.4)	4.2–5.9 (5.4 ± 0.8)	3.6–6.6 (4.6 ± 0.5)	1.8–5.3 (4.0 ± 0.6)	3.6–5.8 (4.6 ± 0.5)	4.1–7.1 (5.2 ± 0.7)	5.1–6.5 (5.7 ± 0.4)
Whorl elongation ratio (WER)	2.25–3.48 (2.8 ± 0.2)	2.44–3.58 (2.9 ± 0.3)	2.38–3.24 (2.9 ± 0.2)	2.37–2.73 (2.5 ± 0.2)	1.93–2.90 (2.3 ± 0.2)	2.50–8.05 (3.6 ± 1.3)	2.02–2.70 (2.4 ± 0.2)	2.02–2.74 (2.4 ± 0.2)	1.82–2.57 (2.2 ± 0.2)
Whorl number (WN)	3.25–7.00 (4.9 ± 0.8)	3.50–7.00 (5.0 ± 0.8)	3.50–5.00 (4.3 ± 0.4)	6.25–7.00 (6.7 ± 0.4)	3.25–8.25 (4.6 ± 0.9)	1.75–5.75 (3.4 ± 0.8)	3.00–5.50 (4.2 ± 0.7)	3.25–8.00 (5.3 ± 1.3)	3.50–7.00 (4.9 ± 1.1)
Sculpture Type (node / granulated rib / smooth rib / spiral cord / smooth) (%)	26 / 65 / 2 / 3 / 5	85 / 15 / 0 / 0 / 0	4 / 38 / 17 / 4 / 38	100 / 0 / 0 / 0 / 0	10 / 55 / 6 / 13 / 16	17 / 29 / 5 / 38 / 10	0 / 35 / 61 / 3 / 0	0 / 48 / 31 / 14 / 7	0 / 53 / 33 / 7 / 7
Morphological characters of protoconchs									
Number of protoconchs (PN)	8–166 (58.3 ± 39.4)	8–82 (31.2 ± 14.9)	10–160 (70.7 ± 45.3)	2–4 (3.0 ± 1.0)	4–59 (19.6 ± 11.3)	1–50 (14.5 ± 9.1)	1–42 (17.3 ± 10.9)	6–63 (31.9 ± 16.1)	9–82 (36.7 ± 20.1)
Longitudinal rib number on body whorl of the largest protoconch (RNP)	8–16 (12.1 ± 1.3)	11–15 (12.4 ± 1.1)	9–12 (10.9 ± 1.0)	10–15 (12.7 ± 2.5)	8–13 (10.5 ± 1.1)	9–17 (11.4 ± 1.7)	8–14 (10.0 ± 1.5)	10–15 (12.1 ± 1.6)	10–16 (12.3 ± 1.7)
Shell height of the largest protoconch (SHP) (mm)	1.3–3.3 (2.4 ± 0.4)	1.9–3.6 (2.7 ± 0.4)	1.6–2.9 (2.2 ± 0.3)	2.7–4.5 (3.9 ± 1.0)	1.8–3.4 (2.6 ± 0.4)	1.6–4.0 (3.0 ± 0.5)	1.1–3.4 (2.3 ± 0.6)	1.2–3.4 (2.7 ± 0.4)	1.9–3.6 (2.8 ± 0.4)
Shell width of the largest protoconch (SWP) (mm)	1.2–2.3 (1.8 ± 0.2)	1.6–2.6 (2.1 ± 0.2)	1.4–2.2 (1.8 ± 0.2)	2.8–3.7 (3.3 ± 0.5)	1.3–2.2 (1.7 ± 0.2)	1.4–3.0 (2.2 ± 0.3)	1.0–2.0 (1.6 ± 0.3)	1.4–2.0 (1.7 ± 0.2)	1.4–2.1 (1.8 ± 0.2)
Whorl number of the largest protoconch (WNP)	2.00–3.75 (3.0 ± 0.4)	2.50–4.00 (3.2 ± 0.4)	2.25–3.25 (2.9 ± 0.3)	2.50–3.50 (3.1 ± 0.5)	3.00–4.25 (3.4 ± 0.3)	2.25–4.00 (3.3 ± 0.4)	2.25–3.75 (3.2 ± 0.5)	3.00–4.00 (3.5 ± 0.3)	2.75–4.00 (3.4 ± 0.4)
Number of nodes on body whorl of the largest protoconch (Node Number) (3 / 2 / 1 / 0) (%)	1 / 3 / 93 / 3	21 / 67 / 13 / 0	0 / 6 / 94 / 0	0 / 67 / 33 / 0	0 / 7 / 41 / 51	0 / 0 / 10 / 90	0 / 4 / 65 / 31	0 / 0 / 48 / 52	0 / 0 / 57 / 43
Spiral cord type on body whorl of the largest protoconch (Spiral Cord Type) (prominent / weak / absent) (%)	96 / 0 / 4	100 / 0 / 0	33 / 67 / 0	0 / 0 / 100	46 / 50 / 4	16 / 63 / 21	50 / 38 / 12	57 / 43 / 0	90 / 10 / 0
Morphological characters of radulae									
Cusp number of rachidian	5–7	5–7	5–7	5–8	5–7	5–9	5–7	5–7	5–7
Cusp number of laterial teeth	4–7	5–7	3–5	6–7	3–7	3–6	3–7	4–7	5–7
Cusp number of interior marginal teeth	4–6	4–6	4–5	3–5	4–7	3–7	4–6	4–7	4–6
Cusp number of exterior marginal teeth	4–6	4–6	4–5	3–6	4–7	3–7	4–6	4–7	4–6
Shape of the central cusp of rachidian (pointed / rounded / flat) (%)	34 / 53 / 13	78 / 11 / 11	40 / 0 / 60	100 / 0 / 0	88 / 13 / 0	77 / 18 / 5	73 / 18 / 9	100 / 0 / 0	100 / 0 / 0
Shape of the central cusp of laterial teeth (pointed / rounded / flat) (%)	0 / 25 / 75	22 / 33 / 44	0 / 0 / 100	100 / 0 / 0	42 / 58 / 0	0 / 77 / 23	0 / 55 / 45	91 / 9 / 0	0 / 100 / 0

2016

2017

Table 3. Results of the Random Forest analyses for the four *Semisulcospira niponica*-group species with specimen numbers and the contribution of each character and the mean Gini coefficients to each species.

Species	*S. decipiens*	*S. habei*	*S. rugosa*	*S. reticulata*	Mean Gini coefficient
Specimen number	117	40	24	4	
Aperture slenderness ratio (ASR)	0.0072	0.0252	0.0096	0.1180	6.8875
Basal cord number (BCN)	0.0020	0.0003	0.0003	-0.0001	1.1322
Body whorl length (BWL)	0.0200	0.0619	0.0081	-0.0066	6.0493
Longitudinal rib number of penultimate whorl (RN)	0.0155	0.0951	0.0621	0.1350	10.5619
Spire angle (SA)	0.0027	0.0085	0.0067	0.0066	4.1103
Spiral cord number of penultimate whorl (SCN)	0.0019	0.0025	0.0034	0.0007	1.8010
Whorl elongation ratio (WER)	0.0075	0.0066	0.0227	0.0275	4.5596
Whorl number (WN)	0.0031	-0.0005	0.0287	0.0470	3.4210
Sculpture Type	0.0084	0.0427	0.0591	0.0595	6.7765
Number of protoconchs (PN)	0.0220	0.0530	0.0280	0.1237	7.1688
Longitudinal rib number on body whorl of the largest protoconch (RNP)	0.0041	0.0092	0.0636	-0.0056	4.5267
Shell height of the largest protoconch (SHP)	0.0141	0.0245	0.0254	0.0525	6.1618
Whorl number of the largest protoconch (WNP)	0.0082	0.0021	0.0031	-0.0040	2.5485
Number of nodes on body whorl of the largest protoconch (Node Number)	0.0872	0.3326	0.0263	0.0122	20.8279
Spiral cord type on body whorl of the largest protoconch (Spiral Cord Type)	0.0393	0.0379	0.2354	0.1791	12.0522

Table 4. Results of the Random Forest analyses for the five *Semisulcospira nakasekoae*-group species with specimen numbers and the contribution of each character and the mean Gini coefficients to each species.

Species	*S. arenicola*	*S. nakasekoae*	*S. ourensis*	*S. elongata* sp. nov.	*S. cryptica* sp. nov.	Mean Gini coefficient
Specimen number	95	86	31	29	30	
Aperture slenderness ratio (ASR)	0.0047	0.0107	0.0065	0.0218	0.0145	8.8103
Basal cord number (BCN)	0.0397	0.0104	0.0020	0.0034	-0.0042	6.6482
Body whorl length (BWL)	0.0559	0.0063	0.0442	0.0457	0.1952	16.8534
Longitudinal rib number of penultimate whorl (RN)	0.0320	0.0073	0.0889	0.0524	0.0759	14.7342
Spire angle (SA)	0.0528	0.1597	0.0396	0.0118	0.0388	24.6305
Spiral cord number of penultimate whorl (SCN)	0.0119	0.0727	0.0853	0.0380	0.0737	17.5765
Whorl elongation ratio (WER)	0.0810	0.2831	0.0553	0.0550	0.1040	33.3922
Whorl number (WN)	0.0401	0.0349	0.0062	0.0329	0.0386	13.5530
Sculpture Type	0.0085	0.0011	0.0222	0.0113	-0.0003	4.2137
Number of protoconchs (PN)	0.0306	0.0168	0.0051	0.0509	0.0947	13.7278
Longitudinal rib number on body whorl of the largest protoconch (RNP)	0.0430	0.0028	0.0476	0.0907	0.0524	13.6698
Shell height of the largest protoconch (SHP)	0.0256	0.0156	0.0314	0.0103	0.0097	11.2620
Whorl number of the largest protoconch (WNP)	0.0172	0.0042	0.0170	0.0080	0.0021	5.9489
Number of nodes on body whorl of the largest protoconch (Node Number)	0.0135	0.0331	0.0462	0.0115	0.0192	8.0142
Spiral cord type on body whorl of the largest protoconch (Spiral Cord Type)	0.0122	0.0165	0.0206	0.0038	0.0839	6.6146

Table 5. Morphometric characters of the holotype of *Semisulcospira decipiens* and juveniles of the four *Semisulcospira* species newly collected from Mano. Measurements and counts: minimum–maximum value (mean ± SD).

Specimen	Holotype of *S. decipiens*	Newly collected *S. decipiens*	Newly collected *S. reticulata*	Newly collected *S. arenicola*	Newly collected *S. elongata* sp. nov.
Specimen number	1	4	2	6	3
Aperture height (AH) (mm)	6.6	6.0–7.4 (6.8 ± 0.7)	5.8–6.5 (6.2 ± 0.5)	5.5–6.3 (5.8 ± 0.3)	6.1–7.0 (6.7 ± 0.5)
Aperture length (AL) (mm)	6.6	6.3–7.7 (7.1 ± 0.7)	6.0–6.8 (6.4 ± 0.6)	5.5–6.1 (5.8 ± 0.2)	6.5–7.3 (6.9 ± 0.4)
Aperture slenderness ratio (ASR)		1.56–1.85 (1.8 ± 0.1)	1.47–1.56 (1.5 ± 0.1)	1.48–1.91 (1.6 ± 0.1)	1.52–1.70 (1.6 ± 0.1)
Aperture width (AW) (mm)		3.7–4.4 (4.1 ± 0.3)	4.1–4.3 (4.2 ± 0.2)	3.0–3.8 (3.5 ± 0.3)	3.8–4.8 (4.2 ± 0.5)
Basal cord number (BCN)	4	2–3 (2.75 ± 0.5)	3	2–3 (2.5 ± 0.6)	2–3 (2.7 ± 0.6)
Body whorl length (BWL) (mm)	10.6	10.0–12.1 (11.1 ± 0.9)	9.6–11.0 (10.3 ± 1.0)	8.9–10.1 (9.4 ± 0.4)	10.5–11.9 (11.4 ± 0.8)
Fourth whorl length (FWL) (mm)	2.2	2.5–2.7 (2.6 ± 0.1)	2.1–2.6 (2.3 ± 0.3)	2.2–2.5 (2.4 ± 0.1)	2.6–3.5 (3.1 ± 0.4)
Penultimate whorl length (PWL) (mm)	3.5	3.7–4.2 (3.9 ± 0.2)	3.5–4.0 (3.8 ± 0.3)	3.0–3.7 (3.5 ± 0.3)	3.9–4.6 (4.3 ± 0.5)
Longitudinal rib number of penultimate whorl (RN)	15	12–15 (13.8 ± 1.5)	15–18 (16.5 ± 2.1)	10–14 (11.7 ± 1.4)	14–21 (17.0 ± 3.6)
Spire angle (SA) (degrees)	22	18.3–22.3 (19.5 ± 1.9)	21.1	14.5–16.4 (15.5 ± 0.8)	15.6–18.6 (17.0 ± 1.5)
Spiral cord number of penultimate whorl (SCN)		5	5–6 (5.5 ± 0.7)	7	6
Shell height (SH) (mm)	20.2	20.0–23.6 (21.8 ± 1.5)	16.3–20.2 (18.2 ± 2.8)	17.9–20.4 (18.9 ± 0.9)	21.8–27.4 (24.5 ± 2.8)
Shell width (SW) (mm)	7.1	7.1–7.9 (7.5 ± 0.4)	7.2–7.8 (7.5 ± 0.4)	5.2–6.4 (6.0 ± 0.4)	7.1–8.1 (7.6 ± 0.5)
Third whorl length (TWL) (mm)	2.8	3.2–3.4 (3.2 ± 0.1)	2.7–3.0 (2.8 ± 0.3)	2.8–3.2 (3.0 ± 0.2)	3.2–3.8 (3.5 ± 0.4)
Whorl elongation ratio (WER)	3.05	2.32–2.86 (2.6 ± 0.3)	2.54–2.76 (2.7 ± 0.2)	2.34–2.55 (2.5 ± 0.1)	2.00–2.33 (2.2 ± 0.2)
Whorl number (WN)	7.50	6.00–7.00 (6.5 ± 0.6)	4.25–5.00 (4.6 ± 0.5)	5.25–6.50 (5.8 ± 0.5)	4.50–7.00 (6.0 ± 1.3)
Sculpture Type (node / granulated rib / smooth rib / spiral cord / smooth) (%)	0 / 0 / 100 / 0 / 0	0 / 100 / 0 / 0 / 0	50 / 50 / 0 / 0 / 0	0 / 100 / 0 / 0 / 0	0 / 67 / 33 / 0 / 0

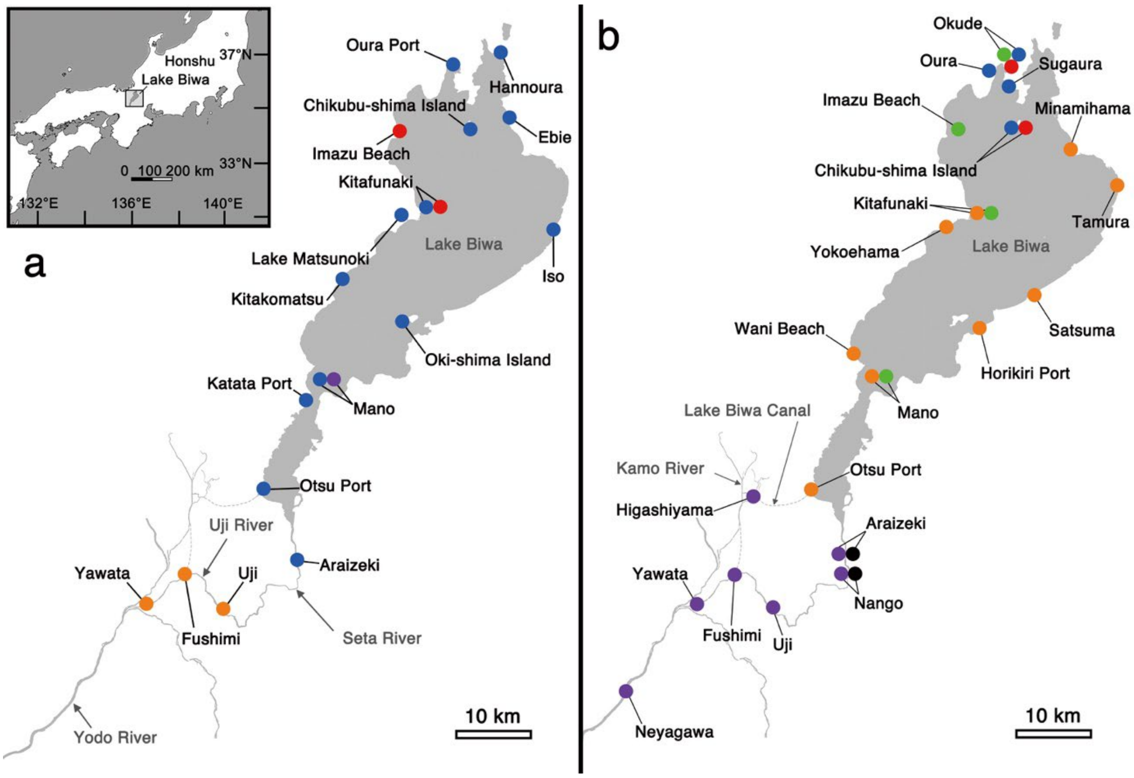

a
Honshu
37°N
Lake Biwa
0 100 200 km
33°N
132°E
136°E
140°E
Oura Port
Hannoura
Chikubu-shima Island
Ebie
Imazu Beach
Kitafunaki
Lake Biwa
Iso
Lake Matsunoki
Kitakomatsu
Oki-shima Island
Katata Port
Mano
Otsu Port
Uji River
Yawata
Uji
Araizeki
Fushimi
Seta River
Yodo River
10 km
b
Okude
Oura
Sugaura
Imazu Beach
Minamihama
Chikubu-shima Island
Kitafunaki
Tamura
Yokoehama
Lake Biwa
Wani Beach
Satsuma
Horikiri Port
Mano
Lake Biwa Canal
Kamo River
Otsu Port
Higashiyama
Araizeki
Yawata
Nango
Fushimi
Uji
Neyagawa
10 km

Fig. 1

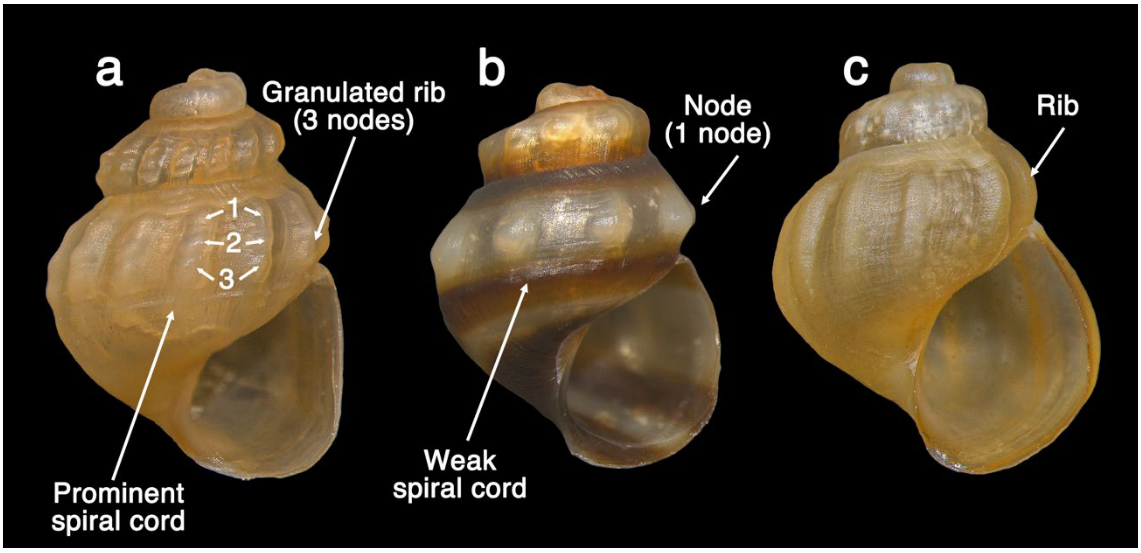

a
Granulated rib
(3 nodes)
1
2
3
Prominent
spiral cord
b
Node
(1 node)
Weak
spiral cord
c
Rib

Fig. 2

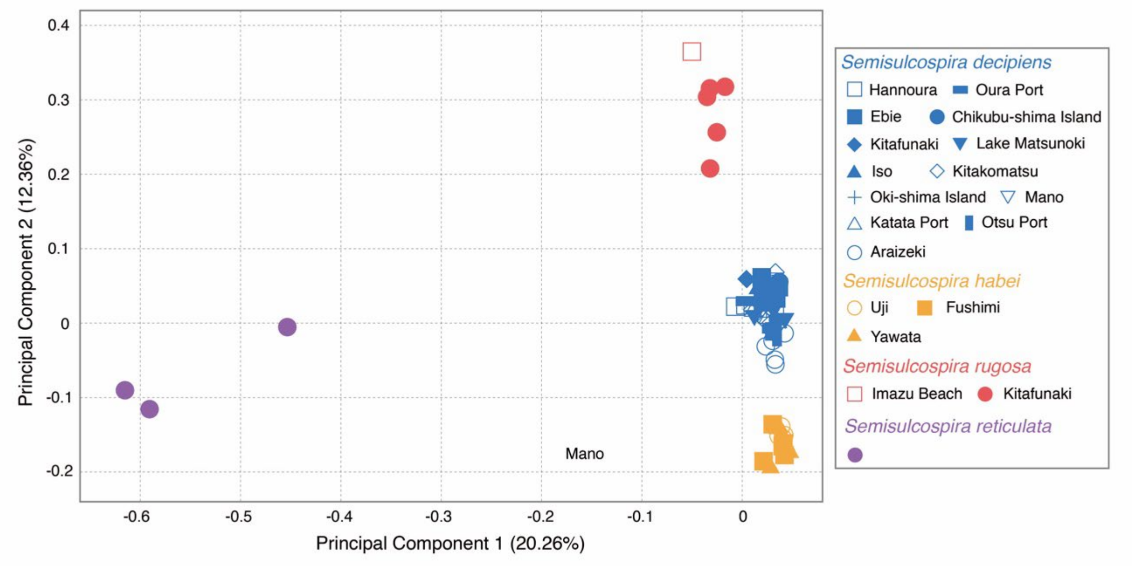

0.4
0.3
0.2
0.1
0
-0.1
-0.2
Principal Component 2 (12.36%)
-0.6
-0.5
-0.4
-0.3
-0.2
-0.1
0
Principal Component 1 (20.26%)
Mano
Semisulcospira decipiens
Hannoura
Oura Port
Ebie
Chikubu-shima Island
Kitafunaki
Lake Matsunoki
Iso
Kitakomatsu
Oki-shima Island
Mano
Katata Port
Otsu Port
Araizeki
Semisulcospira habei
Uji
Fushimi
Yawata
Semisulcospira rugosa
Imazu Beach
Kitafunaki
Semisulcospira reticulata

Fig. 3

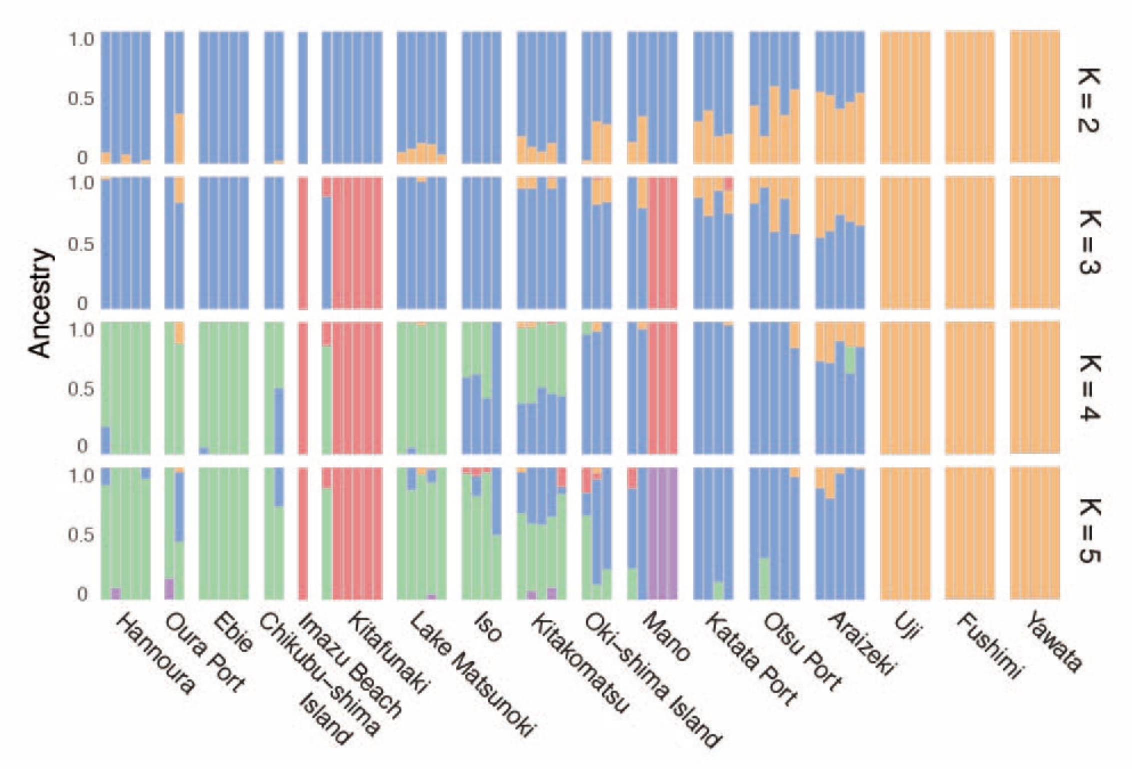

Ancestry
1.0
0.5
0
K = 2
K = 3
K = 4
K = 5
Hannoura
Oura Port
Ebie
Chikubu–shima Island
Imazu Beach
Kitafunaki
Lake Matsunoki
Iso
Kitakomatsu
Oki–shima Island
Mano
Katata Port
Otsu Port
Araizeki
Uji
Fushimi
Yawata

Fig. 4

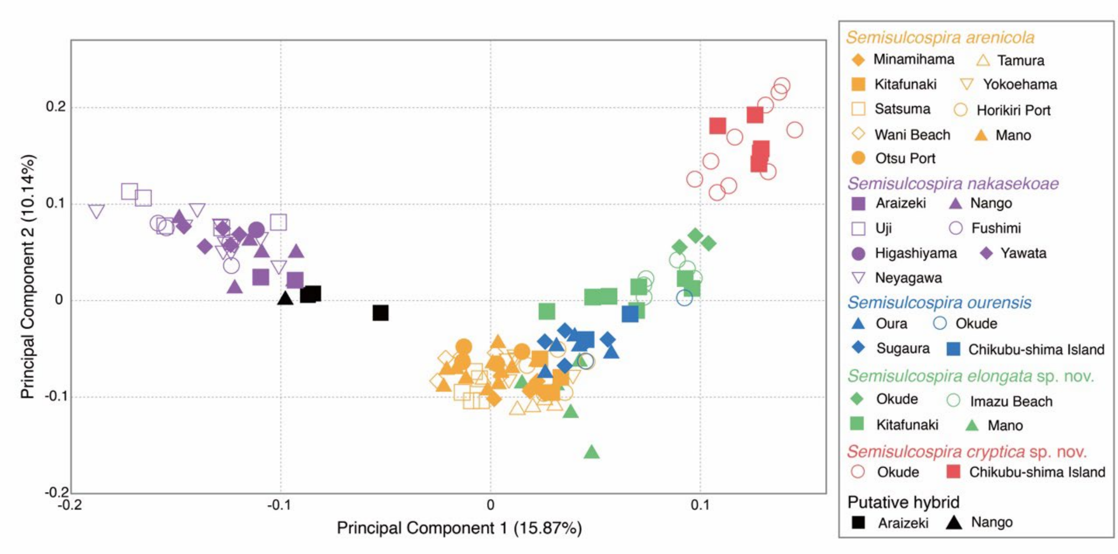

Semisulcospira arenicola
Minamihama
Tamura
Kitafunaki
Yokoehama
Satsuma
Horikiri Port
Wani Beach
Mano
Otsu Port
Semisulcospira nakasekoae
Araizeki
Nango
Uji
Fushimi
Higashiyama
Yawata
Neyagawa
Semisulcospira ourensis
Oura
Okude
Sugaura
Chikubu-shima Island
Semisulcospira elongata sp. nov.
Okude
Imazu Beach
Kitafunaki
Mano
Semisulcospira cryptica sp. nov.
Okude
Chikubu-shima Island
Putative hybrid
Araizeki
Nango
Principal Component 1 (15.87%)
Principal Component 2 (10.14%)
0.2
0.1
0
-0.1
-0.2
-0.2
-0.1
0
0.1

Fig. 5

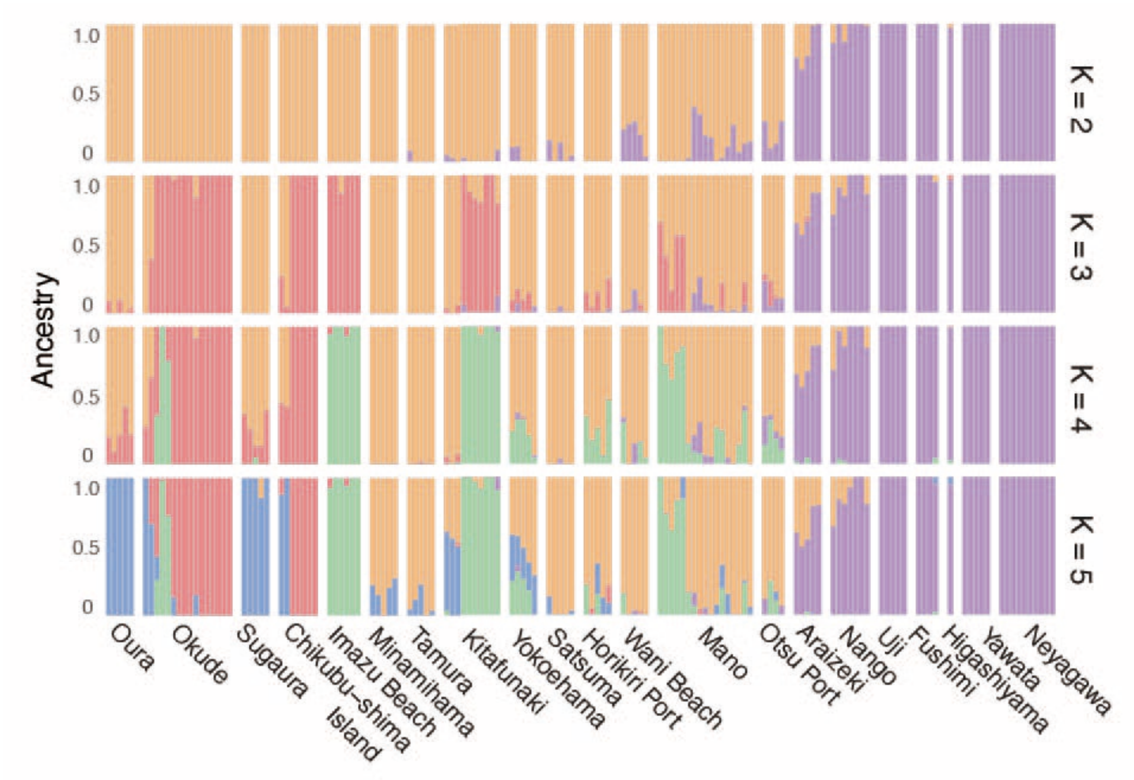

Ancestry
1.0
0.5
0
K = 2
K = 3
K = 4
K = 5
Oura
Okude
Sugaura
Chikubu-shima
Island
Imazu Beach
Minamihama
Tamura
Kitafunaki
Yokoehama
Satsuma
Horikiri Port
Wani Beach
Mano
Otsu Port
Araizeki
Nango
Uji
Fushimi
Higashiyama
Yawata
Neyagawa

Fig. 6

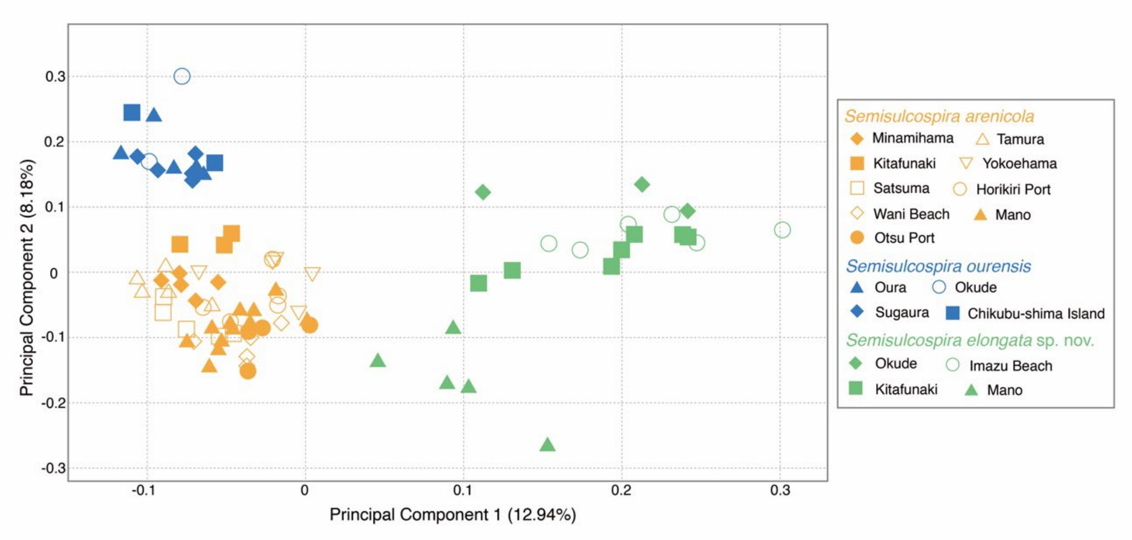

Principal Component 1 (12.94%)
Principal Component 2 (8.18%)
Semisulcospira arenicola
Minamihama
Tamura
Kitafunaki
Yokoehama
Satsuma
Horikiri Port
Wani Beach
Mano
Otsu Port
Semisulcospira ourensis
Oura
Okude
Sugaura
Chikubu-shima Island
Semisulcospira elongata sp. nov.
Okude
Imazu Beach
Kitafunaki
Mano

Fig. 7

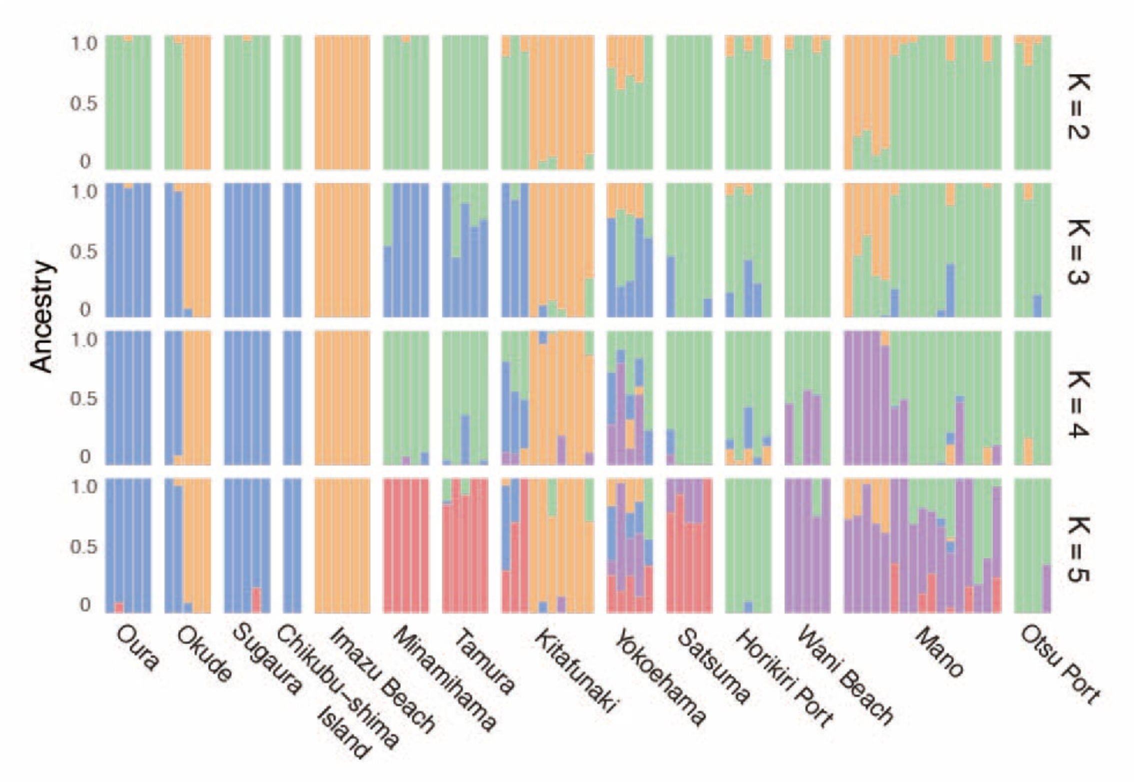

Ancestry
1.0
0.5
0
K = 2
K = 3
K = 4
K = 5
Oura
Okude
Sugaura
Chikubu-shima Island
Imazu Beach
Minamihama
Tamura
Kitafunaki
Yokoehama
Satsuma
Horikiri Port
Wani Beach
Mano
Otsu Port

Fig. 8

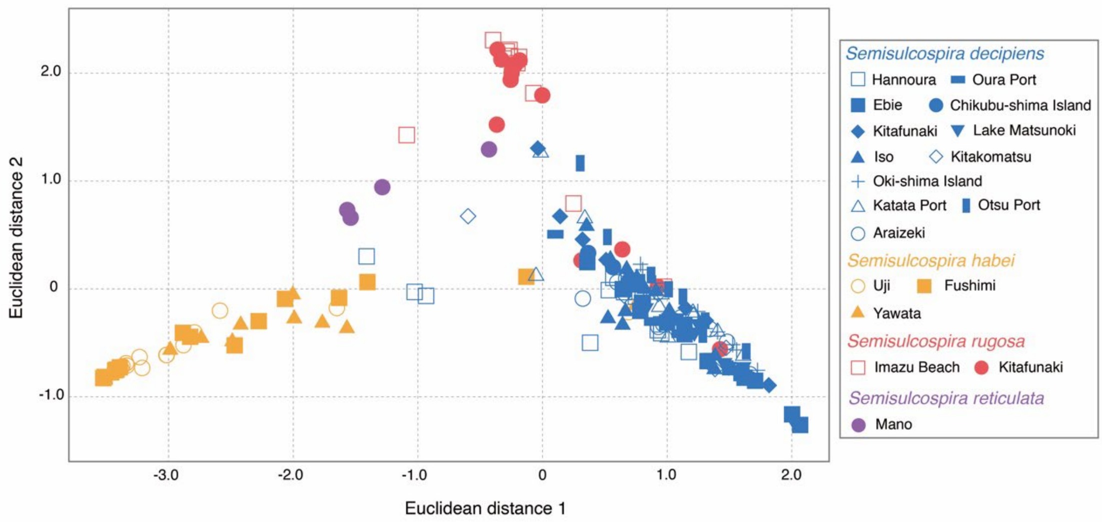

Euclidean distance 2
Euclidean distance 1
2.0
1.0
0
-1.0
-3.0
-2.0
-1.0
0
1.0
2.0
Semisulcospira decipiens
Hannoura
Oura Port
Ebie
Chikubu-shima Island
Kitafunaki
Lake Matsunoki
Iso
Kitakomatsu
Oki-shima Island
Katata Port
Otsu Port
Araizeki
Semisulcospira habei
Uji
Fushimi
Yawata
Semisulcospira rugosa
Imazu Beach
Kitafunaki
Semisulcospira reticulata
Mano

Fig. 9

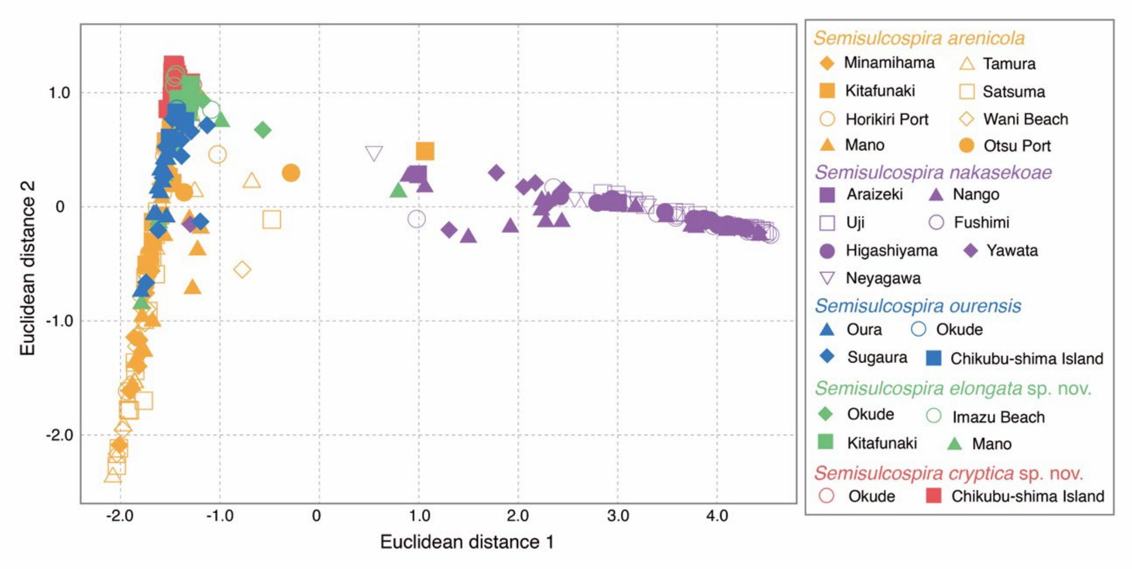

Euclidean distance 2
1.0
0
-1.0
-2.0
-2.0
-1.0
0
1.0
2.0
3.0
4.0
Euclidean distance 1
Semisulcospira arenicola
Minamihama
Tamura
Kitafunaki
Satsuma
Horikiri Port
Wani Beach
Mano
Otsu Port
Semisulcospira nakasekoae
Araizeki
Nango
Uji
Fushimi
Higashiyama
Yawata
Neyagawa
Semisulcospira ourensis
Oura
Okude
Sugaura
Chikubu-shima Island
Semisulcospira elongata sp. nov.
Okude
Imazu Beach
Kitafunaki
Mano
Semisulcospira cryptica sp. nov.
Okude
Chikubu-shima Island

Fig. 10

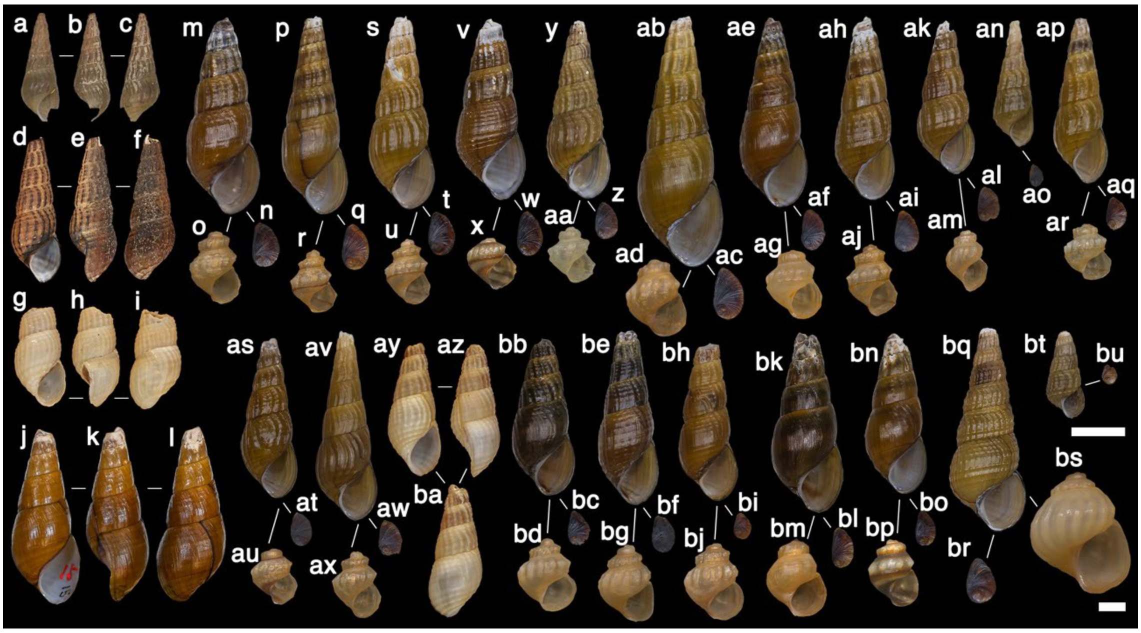

a b c d e f g h i j k l
m n o p q r s t u v w x y z aa ab ac ad ae af ag ah ai aj ak al am an ao ap aq ar
as at au av aw ax ay az ba bb bc bd be bf bg bh bi bj bk bl bm bn bo bp bq br bs bt bu

Fig. 11

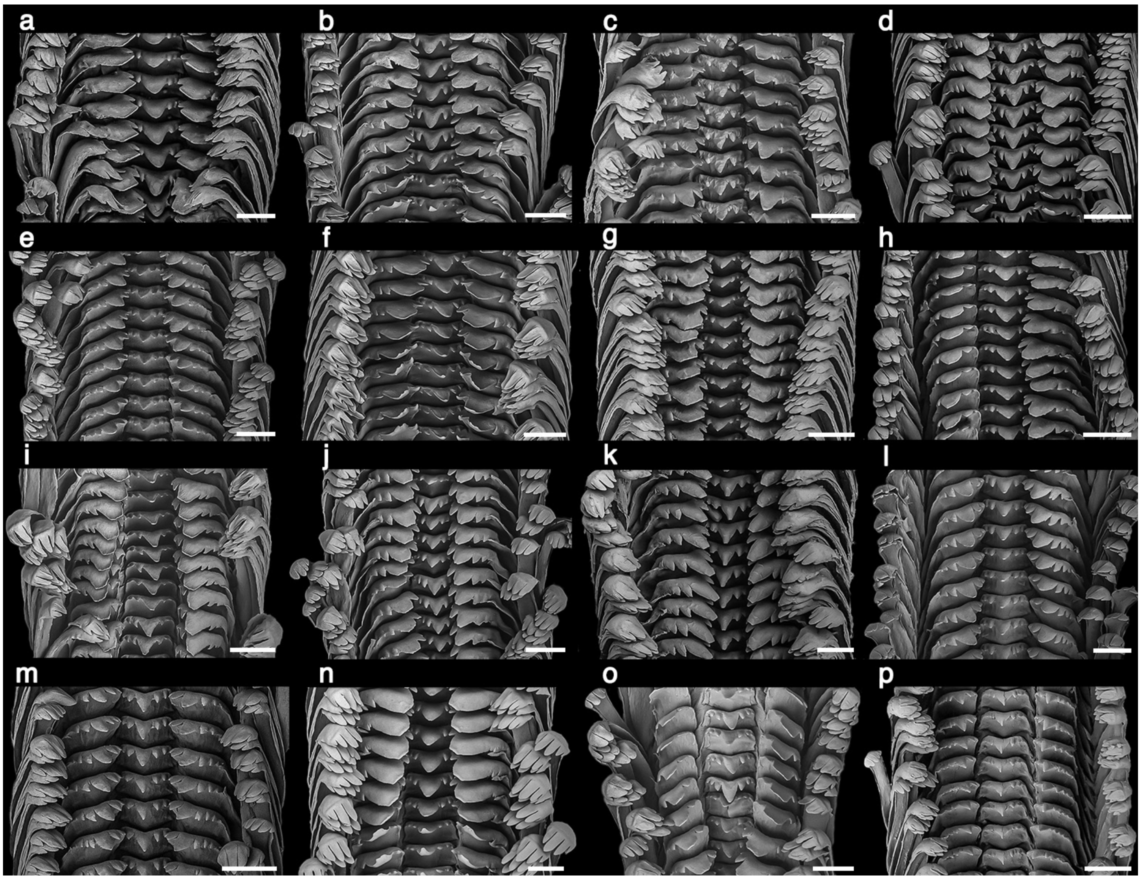


Fig. 12

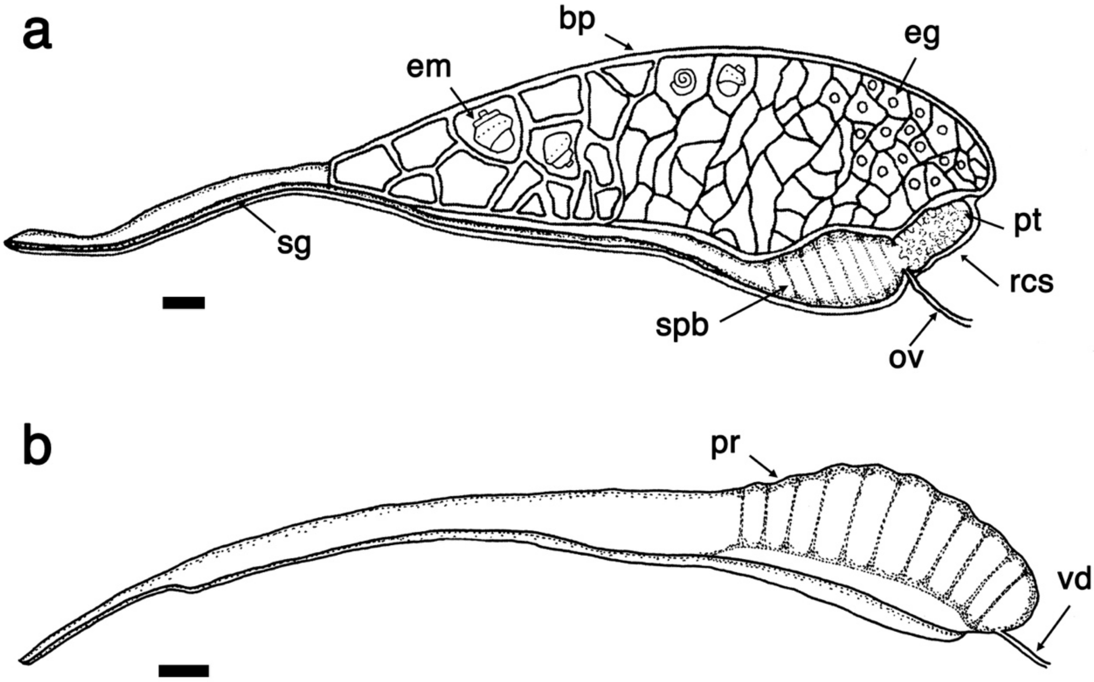


Fig. 13

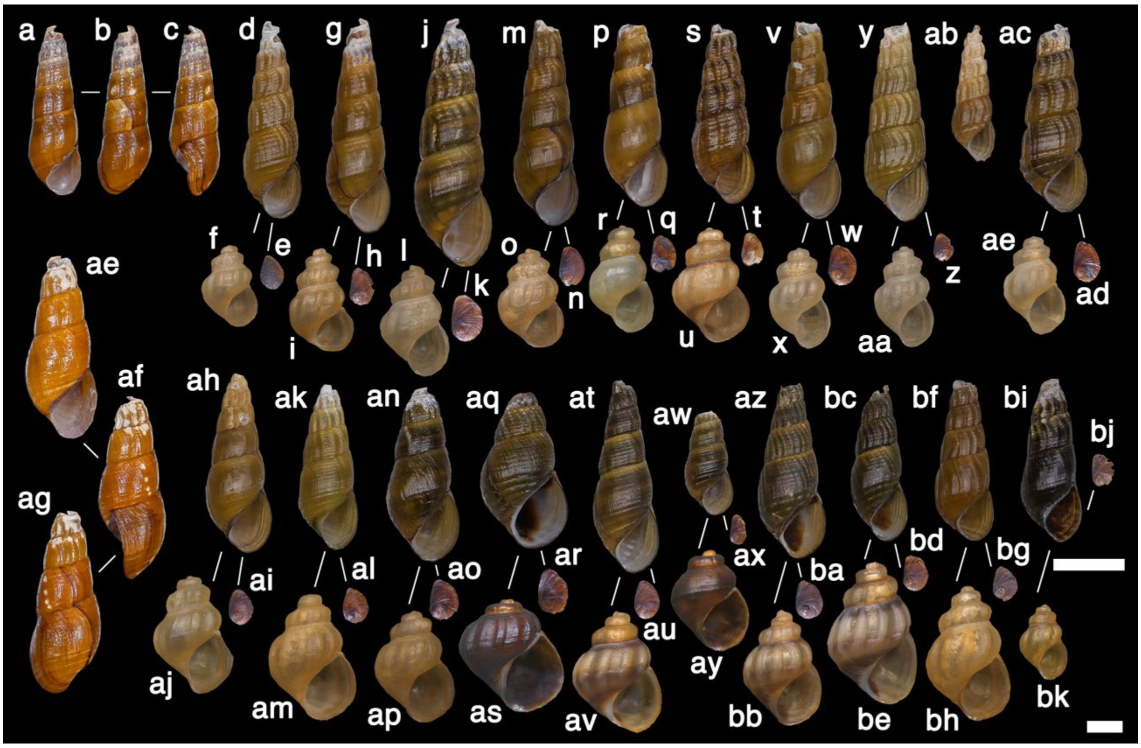

a b c d g j m p s v y ab ac
f e i h l k o n r q u t x w aa z ae ad
ae af ag
ah ak an aq at aw az bc bf bi bj
aj ai am al ap ao as ar av au ay ax bb ba be bd bh bg bk

Fig. 14

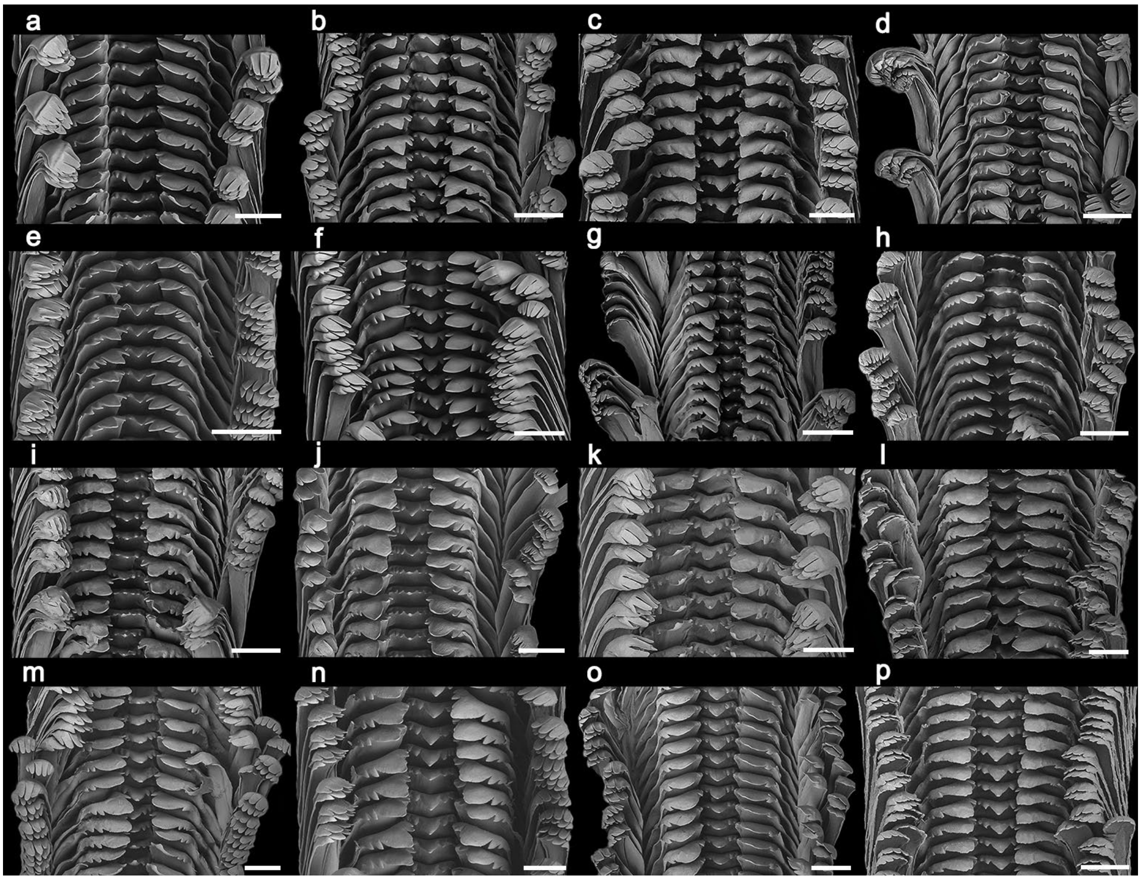


Fig. 15

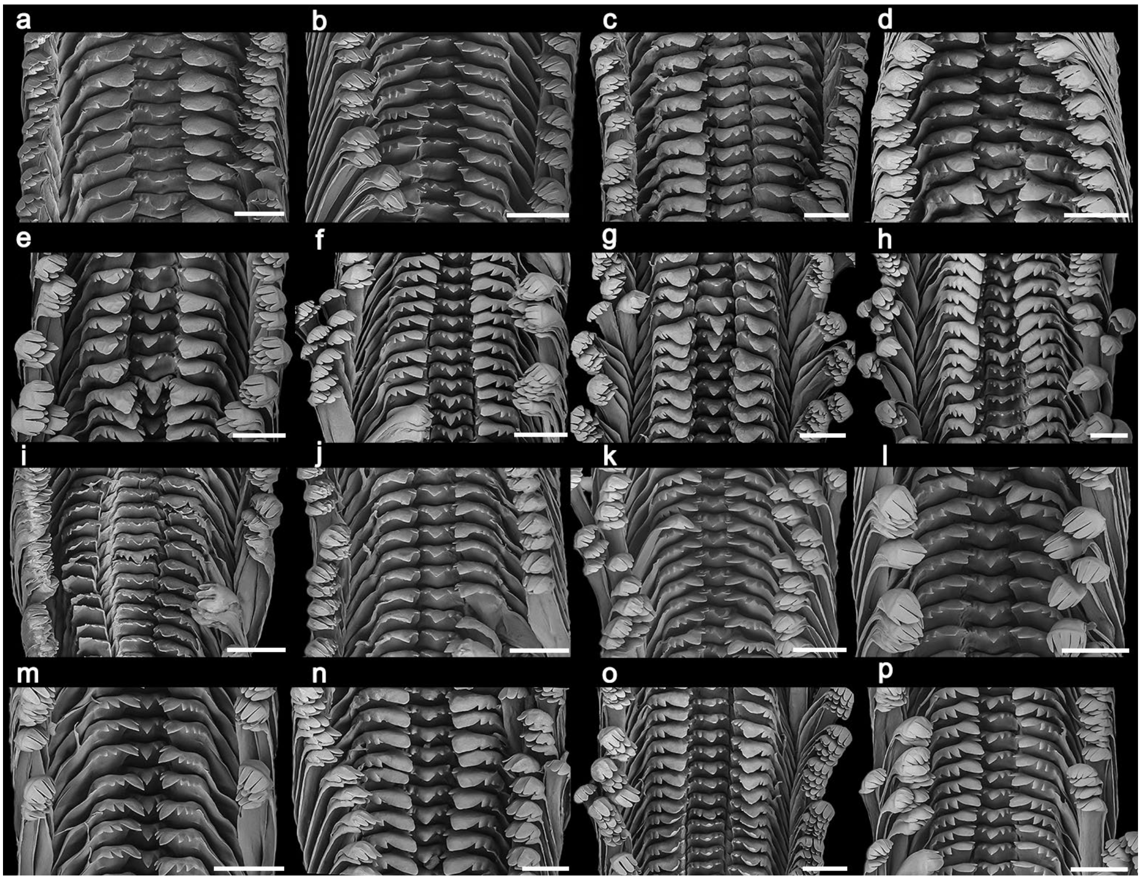
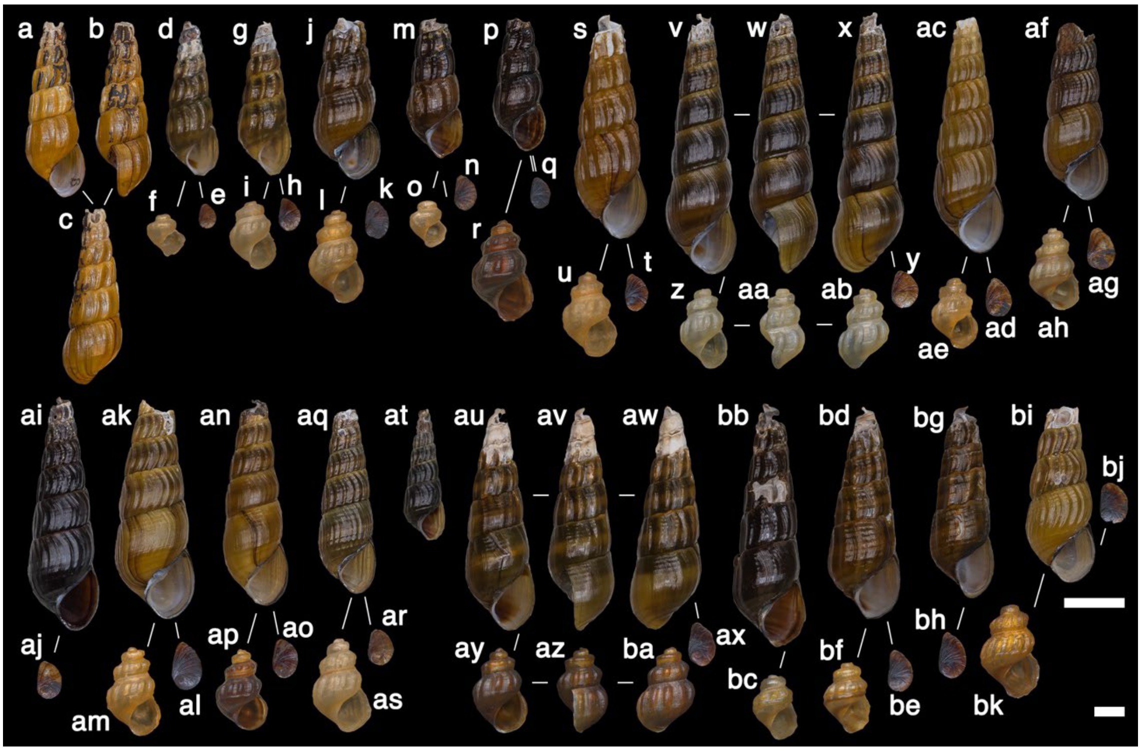


Fig. 16


a
b
c
d
e
f
g
h
i
j
k
l
m
n
o
p
q
r
s
t
u
v
w
x
y
z
aa
ab
ac
ad
ae
af
ag
ah
ai
aj
ak
al
am
an
ao
ap
aq
ar
as
at
au
av
aw
ax
ay
az
ba
bb
bc
bd
be
bf
bg
bh
bi
bj
bk
Fig. 17