Zhang et al. *Zool. Res. Divers. Conserv.* 2024, 1(1): 82−85
https://doi.org/10.24272/j.issn.2097-3772.2023.050

ZRDC Zoological Research: Diversity and Conservation

Letter to the editor Open Access

A new species of *Bostrychus* (Gobiiformes: Eleotridae) from the East China Sea

DEAR EDITOR,

Bostrychus, a genus in the family Eleotridae of the order Gobiiformes, was first established by Lacepède in 1801 (Buffon, 1801). *Bostrychus* currently contains seven recognized species, including two recent additions (*B. microphthalmos* and *B. scalaris*) described in 2005 (Hoese & Kottelat, 2005) and 2008 (Larson, 2008), respectively. The natural range of *Bostrychus* species extends from East Asia to Australia, with the exception of *B. africanus*, Steindachner, 1879, which is distributed in West Africa (Herre, 1946). Among the recognized species, *B. sinensis, B. zonatus,* and *B. africanus* are relatively widespread, inhabiting diverse areas from estuaries to freshwater streams, while *B. scalaris* is only found at a single mangrove site in the Selangor State of Malaysia (Larson, 2008). The remaining three species exhibit a high degree of habitat specialization and are highly localized (Hoese & Kottelat, 2005): *B. microphthalmos* inhabits a cave stream in the Maros karst of southern Sulawesi, *B. aruensis* is confined to freshwater environments in the Aru Islands of Indonesia, and *B. strigogenys* is found only in freshwater areas in southern Papua New Guinea and Irian Jaya.

Bostrychus sinensis Lacepède, 1801, a species with broad salt tolerance, is known to inhabit intertidal mudflats or mangroves. Despite exhibiting limited mobility and a tendency for cave-dwelling, it is widely distributed across the tropical Indo-Pacific, extending westward to India, northward to China and Japan, southward to Queensland, Australia, and eastward to Samoa. In East Asia, *B. sinensis* is predominantly observed in the Ryukyu Archipelago of Japan and the coastal waters of Southeast China (Miyake et al., 2019). Qiu et al. (2016) conducted a population genetic analysis on *B. sinensis* from different geographical groups in the coastal waters of China based on a single gene, revealing the existence of two geographical lineages in the South China Sea (SCS) and East China Sea (ECS), which also exhibited morphological differentiation, notably the presence or absence of rib-like stripes. Furthermore, Wu et al. (2021) later reported distinct variances in the reproductive cycles and egg-laying quantities of these two lineages.

Bostrychus sinensis was initially identified and described by Lacepède in 1801, with the description based solely on a Chinese landscape painting (Buffon, 1801). The depiction of *Bostrychus sinensis* in Lacepède, 1800 (pl. 14 fig. 2, Buffon, 1801), was titled “*Bostryche chinois*”, a vernacular term, showing the species without rib-like stripes on the body side. In this study, we distinguished individuals exhibiting rib-like stripes from *B. sinensis* through both morphological analysis (see detailed comparison below) and molecular data. Thus, these specimens are identified as a new species and are herein described as *Bostrychus donghaiensis* **sp. nov.**

Between 2021 and 2022, we collected 40 specimens of *Bostrychus* from the coastal waters of China (Figure 1A), including 17 specimens identified as the new species, *Bostrychus donghaiensis* **sp. nov.** All specimens were preserved in ethanol and deposited in the Marine Organisms Resources Group (MORG) of Xiamen University, China.

All 40 specimens were measured and counted following Hoese & Kottelat (2005). All measurements were made with a digital vernier caliper to the nearest 0.01 mm. Genomic DNA was extracted from muscle tissue using a standard phenol-chloroform protocol. Sample resequencing libraries were constructed based on standard procedures using the DNBSEQ platform with high sequencing depth. After mapping all data to the chromosome level of *B. sinensis*, a nuclear neighbor-joining (NJ) tree was constructed based on single nucleotide polymorphism (SNP) loci using VCF2Dis. The mitogenomes of the samples were assembled using MitoZ v.2.3 with default parameters. The assembled mitogenome sequence was annotated using MITOS (http://mitos2.bioinf.uni-leipzig.de/index.py). Mitochondrial sequences were obtained from GenBank (Supplementary Table S1). The phylogenetic tree based on 13 protein-coding mitochondrial genes was constructed using MrBayes v.3.2.7. The average intraspecific and interspecific genetic distances were calculated using the Kimura two-parameter (K2P) distance model with MEGA v.7.0. Two fossils were used to constrain the MCMC tree estimations, and log-normal distribution settings followed Li et al. (2018). Molecular dating based on the 13 mitochondrial genes was performed using BEAST v.2.7.0. The analysis was run twice with 10 000 000 MCMC generations and parameter sampling every 1 000 generations.

Taxonomy

Bostrychus donghaiensis **sp. nov.** (Figure 1C–E; Supplementary Tables S2, S3)

Holotype: *Bostrychus donghaiensis* **sp. nov.** MORG20220001, 118.26 mm standard length (SL), collected from the coastal waters of Zhoushan City, Zhejiang Province, China, September 2021 (Figure 1A).

Received: 11 April 2023; Accepted: 28 December 2023; Online: 05 January 2024

Foundation items: This work was supported by the National Natural Science Foundation of China (41776143)

Paratypes: *Bostrychus donghaiensis* **sp. nov.** MORG20220002–MORG20220017, 94.04–111.40 mm SL, collected from the coastal waters of Zhoushan, Yuhuan, Wenzhou, Ningde in China, September 2021–May 2022.

Etymology: The species name *donghaiensis* is derived from its current distribution in the East China Sea. We suggest the Chinese common name "东海乌塘鳢".

Suggested English name: Donghai gudgeon

Diagnosis: *Bostrychus donghaiensis* **sp. nov.** can be distinguished from its congeners based on a combination of the following characteristics: (1) Neatly arranged rib-like stripes visible on side of body (vs. no stripe on body side in *B. sinensis*, ladder-like bars in *B. scalaris* (Larson, 2008), distinct transverse bands in *B. zonatus*, conspicuous dark stripes on white lower part of face behind eye in *B. strigogenys* (Nichols et al., 1937), and no distinct pigmentation on body in *B. microphthalmus* (Hoese & Kottelat, 2005)). (2) Row of equally spaced, bright yellow spots arranged on body side (vs. many small dots scattered on side of body in *B. sinensis*) (Figure 1C–H). (3) Standard body length range of 94.04–118.26 mm (vs. 89.69–165.92 mm in *B. sinensis*). (4) Six first dorsal fin spines and 27 vertebrae (vs. 7–10 first dorsal fin spines and 36 or 38 vertebrae in *B. zonatus*, 8–9 first dorsal fin spines and 39–40 vertebrae in *B. aruensis*, 34–36 vertebrae in *B. strigogenys*, and 29–30 vertebrae in *B. microphthalmus* (Supplementary Table S3). (5) Second dorsal fin with 10–11 rays and anal fin with 9–10 rays (vs. 9 and 8 in *B. scalaris*, respectively, and 8–9 and 7 in *B. africanus*, respectively (Supplementary Table S3).

Descriptions: Body elongated, anterior subcylindrical, compressed posterior. Row of equally spaced, bright yellow spots arranged on side of body, with rib-like stripe also along body side. Measurements based on 118.26 mm SL in holotype and 94.04–111.40 mm SL in paratypes. Medium large, slightly flat head, head width greater than head height. Head length of holotype 27.71% (18.37%–27.07% in paratypes) of SL. Head width at posterior preopercular margin 18.17% (15.71%–18.16%) of SL. Mouth wide, anterior, upper and lower jaw of similar length, jaw teeth thin and sharp, with many rows. Vomer with 4–5 rows of conical short teeth arranged in a semi-oval shape. Eyes small, with two nostrils on each side. Eye length 2.20% (1.48%–2.22%) of SL. Distance from end of head to anal-fin origin (36.40% of SL in holotype, 34.38%–38.62% of SL in paratypes), nearly subequal to distance from anal-fin origin to base of caudal fin (36.17% of SL in holotype, 34.12%–37.99% of SL in paratypes).

Body covered with small cycloid scales extending onto head. Pre-dorsal scales 60–70, longitudinal scales 120–140, and transverse scales 35–50. First dorsal fin with six hard spines and second dorsal fin with one hard spine, 10–11 rays. Anal fin with I–9~10 rays, opposite to and shorter than second dorsal fin. Pectoral fins with 17–18 translucent branching rays, broadly rounded. Left and right ventral fins contain I–5 rays. Caudal fin contains 18–20 segmented rays.

Color in life: Back black, sometimes turning dune brown. Ventral body white, with reddish-white round (female) or triangular (male) reproductive papillae under anus. First dorsal fin with one bright yellow stripe, second dorsal fin with 3–4 bright yellow stripes (Figure 1J). Large eye-shaped dot with gold edges above base of caudal fin.

Color in preservation: Body surface covered by white film when preserved in alcohol, possibly formed by mucus on body surface. Rib-like stripes and row of yellow spots on body faintly visible, not as bright as in living state (Figure 1K).

Distribution: The new species is currently found in the East China Sea, at sampling points in Zhoushan, Yuhuan, Wenzhou, and Ningde (Figure 1A).

Morphological comparison: *Bostrychus donghaiensis* **sp. nov.** and *B. sinensis* can be distinguished based on the following body surface differences: (1) Presence of pronounced rib-like stripes on sides of body, extending to ventral margin (vs. absent in *B. sinensis*); (2) Row of large yellow spots arranged almost equidistant on sides of body (vs. many small dots scattered on sides of body in *B. sinensis*); (3) Overall smaller body size (94.04–118.26 mm SL) (vs. 89.69–165.92 mm SL in *B. sinensis*). A detailed summary comparing morphological characteristics between *Bostrychus donghaiensis* **sp. nov.** and its congeners is provided in Supplementary Table S3.

Phylogenetic and genetic comparisons: The observed mitochondrial phylogenetic relationships within the genus *Bostrychus* strongly suggest that *Bostrychus donghaiensis* **sp. nov.** is an independent branch, forming a sister clade with *B. sinensis* (China), while *B.* **cf.** *sinensis* (Japan) is placed outside the sister clade (Supplementary Figure S1). The nuclear NJ tree showed consistent results with the mitochondrial gene topology regarding the relationship between *Bostrychus donghaiensis* **sp. nov.** and *B. sinensis* (China) (Supplementary Figure S2). In addition, the average genetic distance between *Bostrychus donghaiensis* **sp. nov.** and *B. sinensis* was 0.11, approximately 5.5 times the estimated intraspecific genetic distance. The average genetic distance between *B.* **cf.** *sinensis* (Japan) and *B. sinensis* (China) was 0.54–0.55, almost equivalent to the average genetic distance (0.58) between the outgroup *Oxyeleotris marmorata* and *B.* **cf.** *sinensis* (Japan) (Supplementary Table S4). Mitogenomic phylogeny recovered *B. sinensis* and *Bostrychus donghaiensis* **sp. nov.** as sister species within *Bostrychus*, both of which diverged from their common ancestor approximately 4 million years ago (Ma). Furthermore, *B.* **cf.** *sinensis* (Japan) diverged from *B. sinensis* (China) as early as around 19 Ma and was recovered outside the sister branch of *Bostrychus donghaiensis* **sp. nov.** and *B. sinensis* (Figure 1B).

In this study, we identified a new species within the genus *Bostrychus*, named *Bostrychus donghaiensis* **sp. nov.**, through a combination of morphological and phylogenetic analyses. Our phylogenetic results revealed that *B. sinensis* constitutes a cryptic species complex, containing three well-supported clades representing three separate species. With the inclusion of *Bostrychus donghaiensis* **sp. nov.**, the genus now contains eight known species. Additionally, the morphological characteristics of the unnamed species, *B.* **cf.** *sinensis* (Japan), require further description.

NOMENCLATURAL ACTS REGISTRATION

The electronic version of this article in portable document format represents a published work according to the International Commission on Zoological Nomenclature (ICZN), and hence the new names contained in the electronic version are effectively published under that Code from the electronic edition alone (see Articles 8.5–8.6 of the Code). This published work and the nomenclatural acts it contains have been registered in ZooBank, the online registration system for

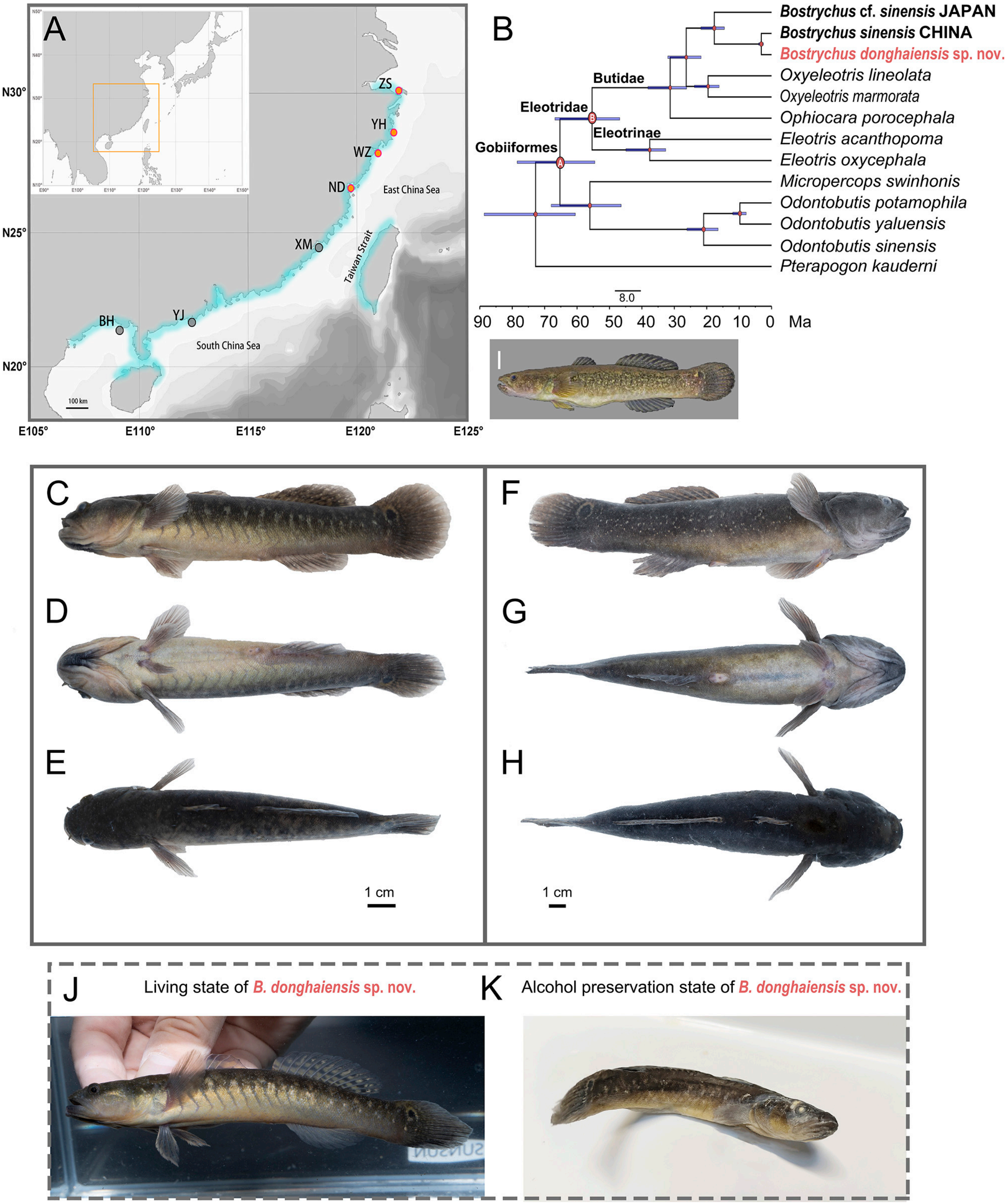

Figure 1 Collection sites, divergence time estimations, and photographs of *Bostrychus donghaiensis* sp. nov. (MORG20220001)

Photo of *Bostrychus sinensis* was collected in China (ZS, Zhoushan; YH, Yuhuan; WZ, Wenzhou; ND, Ningde; XM, Xiamen; YJ, Yangjiang; BH, Beihai) and *Bostrychus* **cf.** *sinensis* was collected in Japan. A: Collection sites of *Bostrychus donghaiensis* **sp. nov.** (orange-red circle) and *B. sinensis* (circle) in coastal waters of China. Blue-green area is the known range of *B. sinensis* in coastal waters of China (https://obis.org/taxon/279944). B: Divergence time estimation of the genus *Bostrychus* based on 13 mitochondrial genes. Shaded bars on nodes represent 95% highest posterior credible age intervals for each node. Red dots with letters (A and B) represent fossil calibration points. C–E: Lateral, ventral and dorsal views of *Bostrychus donghaiensis* **sp. nov.** Photos by Rong-Rong Zhang. F–H: Lateral, ventral, and dorsal views of *B. sinensis*. Photos by Rong-Rong Zhang. I: Photo of *B.* **cf.** *sinensis* collected in Japan (Miyake et al., 2019). J, K: Photos of *Bostrychus donghaiensis* **sp. nov.** in living state and alcohol preservation, respectively. Photos by Rong-Rong Zhang.

the ICZN. The ZooBank LSIDs (Life Science Identifiers) can be resolved and the associated information can be viewed through any standard web browser by appending the LSID to the prefix http://zoobank.org/.

Publication LSID:

LSIDurn:lsid:zoobank.org:pub:88ECCFB6-D616-4218-8279-44BC3B8445FA

Bostrychus donghaiensis **sp. nov.** LSID:

LSIDurn: lsid:zoobank.org:act:F80F773E-4B8D-4508-A357-A3B49090AEB1

SCIENTIFIC FIELD SURVEY PERMISSION INFORMATION

Field collections followed all rules of the Fisheries Law of the People's Republic of China. All activities conformed to the Laboratory Animal Guidelines for the Ethical Review of Animal Welfare (GB/T 35892-2018).

PERMISSIONS

Figure 1I is a faithful reproduction of the original figure presented in Miyake et al. (2019), with its use licensed under the Creative Commons Attribution 4.0 International License. To view this license, visit https://creativecommons.org/licenses/by/4.0/ (Permissions were obtained).

DATA AVAILABILITY

All assembled mitochondrial genome sequences were deposited in the National Center for Biotechnology Information (NCBI) database (https://www.ncbi.nlm.nih.gov) under GenBank accession Nos. OP759468–OP759478, OP759482–OP759509, and OP747050.

SUPPLEMENTARY DATA

Supplementary data to this article can be found online.

COMPETING INTERESTS

The authors declare that they have no competing interests.

AUTHORS' CONTRIBUTIONS

S.X.D. designed the study. R.R.Z. and K.Y. collected specimens in the field. R.R.Z. performed the experiments and analyzed the data. R.R.Z., K.Y., and D.L. conducted data measurements. R.R.Z. wrote the manuscript. All authors read and approved the final version of the manuscript.

ACKNOWLEDGMENTS

We thank Hong-Du Lin for valuable suggestions.

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