1. Introduction

Freshwater Gobionellinae fish Rhinogobius filamentosus (Teleostei: Gobiidae) is a small benthic fish (60–80 mm) of great ornamental development value (Zheng et al. 2016; Liu et al. 2021). *Ctenogobius filamentosus* was treated as the synonym of *R. filamentosus* in the past. It is an endemic species from China mainly distributed in the Lijiang River, Xijiang River and Beijiang River, inhabiting the tributaries of rivers and streams. *R. filamentosus* can be distinguished from other *Rhinogobius* fish by dorsal fin rays VI, I-8-9; anal fin rays I-8; pectoral fins 16-17; ventral fin rays I-5; caudal fins 2+18+2; vertical scales 30–33; horizontal scales series 8–10; predorsal scales 8–11; gill rakes 5–8; body with 5–6 dark transverse stripes; reticular fine lines on the back of the head; fine stripes extending to the ventral surface on the cheeks; a large spot on the fin membrane between the spines of the first and third fins of the first dorsal fin for male (Wu and Zhong 2008) (Figure 1). Although some studies on the complete mitochondrial genome in *Rhinogobius* fishes have been reported (Zhong et al. 2018; Chen et al. 2019; Zhang and Shen 2019), there were still no relevant reports on the genetic characteristics and phylogenetic position of *R. filamentosus*. This research aimed to sequence the complete mitogenome of *R. filamentosus*, provide fundamental molecular data for evolution, and determine its phylogenetic placement in the genus *Rhinogobius*. Therefore, this study is of great significance.
quality control, subsequently, the DNA library was constructed and amplified by PCR, followed by size selection and library quality check (The concentration of DNA sample detected by NanoDrop 2000 (Thermo Fisher Scientific, USA) was not less than 20 ng/μL, the total amount was not less than 100 ng, and OD260/OD280 = 1.8–2.2. The main band of genomic DNA detected by agarose gel electrophoresis was clearly visible, and there was no obvious degradation dispersion). Finally, library pooling and sequencing were carried out on Illumina Hiseq platform 2500 (Genesky Biotechnologies Inc. Shanghai, China). The next-generation sequencing raw data (2.39 GB) were assembled using MetaSPAdes 3.13.0 (Nurk et al. 2017) with Rhinogobius duaspilus MH127918 as

Figure 1. Rhinogobius filamentosus. The specimen is from the Lijiang River, Guilin city, Guangxi province, China. Photographs by Xiao Jiang CHEN on Jun 10, 2021.

Figure 2. The complete mitochondrial genome map of Rhinogobius filamentosus (GenBank accession no.OM678440), consists of 13 PCGs, 22 tRNAs, two rRNAs, the origin of L-strand replication (OL) and the control region (D-loop). The arrows represent the direction of transcription, H-strand is located in the outer ring and the L-strand is located in the inner ring.
reference (Tan et al. 2020), and then the assembled mitochondrial genome sequences were annotated by MitoMaker 1.14 (Bernt et al. 2013) with default parameters. OGDRAW (https://chlorobox.mpimp-golm.mpg.de/OGDraw.html) was used to draw the genome maps (Greiner et al. 2019).

2.3. Phylogenetic analysis

To confirm the phylogeny of *R. filamentosus*, all 17 mitochondrial genomes from genera *Rhinogobius* and *Gymnogobius* were obtained from Genbank. The phylogenetic tree was reconstructed using the Maximum-likelihood (ML) method based on the concatenated amino acids sequences of 13 PCGs, with a bootstrap of 1000 replicates. *Gymnogobius heptacanthus* and *Gymnogobius laevis* were used as outgroups to root the tree. Accession numbers were given with species names, and the numbers at the nodes represented bootstrap values.

![Phylogenetic tree](image)

**Figure 3.** A phylogenetic tree was constructed for the genera *Rhinogobius* and *Gymnogobius*, using the Maximum-likelihood (ML) method based on the protein-coding regions of their mitogenomes, with a bootstrap of 1000 replicates. *Gymnogobius heptacanthus* and *Gymnogobius laevis* were used as outgroups to root the tree. Accession numbers were given with species names, and the numbers at the nodes represented bootstrap values.

Among the 37 genes, 28 genes were encoded on the H-strand while tRNA-Asn, tRNA-Ala, tRNA-Gln, tRNA-Pro, tRNA-Glu, ND6, tRNA-Ser(UGA), tRNA-Tyr, and tRNA-Cys were encoded on L-strand (Figure 2). The overall base composition of the genome was estimated to be T (25.4%); C (30.2%); A (27.6%); and G (16.7%), with an AT bias (53%). The length of the 13 protein-coding genes ranged from 165 bp (ATP8) to 1,821 bp (ND5). All protein-coding genes started with ATG as an initiation codon except for COX1, which started with GTG. As for stop codons, seven PCGs (ND1, ND2, COX1, ATP6, ATP7, ND4L, and ND5) used TAA, two genes (ND3 and ND6) used TAG. Four PCGs (COX2, COX3, ND4, and CYTB) ended with an incomplete stop codon (T or TA). The length of 22 tRNAs was in the range of 66–76 bp. As for rRNA, the 12S rRNA was 957 bp in length and the 16S rRNA was 1,658 bp.

3. Results and discussion

3.1. The mitochondrial genome of *Rhinogobius filamentosus*

The complete mitochondrial genome of *R. filamentosus* was 16,510 bp in length, consisting of 13 protein-coding genes (PCGs, 11,414 bp), 22 tRNAs (1,555 bp), two rRNAs (2,615 bp), and two non-coding regions (D-loop: control region displacement loop, 478 bp; OL: origin of L-strand replication, 30 bp).

Figure 3. A phylogenetic tree was constructed for the genera *Rhinogobius* and *Gymnogobius*, using the Maximum-likelihood (ML) method based on the protein-coding regions of their mitogenomes, with a bootstrap of 1000 replicates. *Gymnogobius heptacanthus* and *Gymnogobius laevis* were used as outgroups to root the tree. Accession numbers were given with species names, and the numbers at the nodes represented bootstrap values.
structure of Rhinogobius in this paper was similar to that of Song et al (Song et al. 2022). This study provided information on the complete mitochondrial genome of R. filamentosus for the first time, and confirmed its phylogenetic position in genus Rhinogobius, which would be beneficial for further studies on population genetics and biodiversity.

4. Conclusions

The complete mitochondrial genome of Rhinogobius filamentosus had been firstly sequenced and annotated on the Illumina HiSeq platform using high-throughput sequencing technology. The assembly circular mitogenome was 16,510 bp long. The genetic data has been submitted to NCBI with the accession number OM678440, and the phylogenetic analysis results strongly supported that R. filamentosus was clustered with R. duospilus. The mitochondrial genomic data of R. filamentosus would be essential for further genetic studies such as phylogenetic relationship investigations, genetic taxonomy, DNA barcode development, and so on. In the future, we will supplement more mitochondrial genomes of other goby fishes.

Ethical approval

Experiments were approved by the Ethical Committee for Animal Experiments of Jiangsu Agri-animal Husbandry Vocational College and conducted following the Chinese Association for the Laboratory Animal Sciences and the Institutional Animal Care and Use Committee (IACUC) protocols.

Author contributions

Conception and design, Chen XJ and Song L; Data curation, Song L, Wang Q, Liu WZ, and Chen XJ; Analysis and interpretation of the data, Song L, Wang Q, and Chen XJ; Funding acquisition, Chen XJ and Wang Q; Writing – original draft, Chen XJ, Song L, Wang Q, and Liu WZ; Writing – review & editing, Chen XJ, Song L, Liu WZ, and Wang Q. All authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Disclosure statement

The authors declare no potential conflict of interest.

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Data availability statement

The genome sequence data that support the findings of this study are openly available in NCBI at (https://www.ncbi.nlm.nih.gov/) under the accession no.OM678440. The associated “BioProject”, “Bio-Sample” and “SRA” numbers are PRJNA808168, SAMN26030913, and SRR18131290, respectively. https://www.ncbi.nlm.nih.gov/bioproject/PRJNA808168.

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