Complete mitochondrial genome of the *Yunnanilus Sichuanensis* and phylogenetic analysis

Yuanchao Zou, Junying Zhang, Tian Wu and Zhengyong Wen

Conservation and Utilization of Fishes resources in the Upper Reaches of the Yangtze River Key Laboratory of Sichuan Province, College of Life sciences, Neijiang Normal University, Neijiang, Sichuan, 641100, China

**ABSTRACT**

The complete mitochondrial genome of the *Yunnanilus sichuanensis* (Cypriniformes: Nemacheilinae) was first sequenced using the next-generation sequencing in this study. The mitogenome was 16,571 bp in length and contained 13 protein-coding genes, 2 ribosomal RNA genes, 22 transfer RNA genes, 1 displacement loop (D-loop) locus, and an origin of replication on the light-strand (OL) region. Most of the genes were encoded on the heavy strand except ND6 and 8 tRNA genes encoded on the light strand. The mitogenome organization and gene order were similar to a typical vertebrate mitogenome. The overall base composition was 26.96% A, 28.62% T, 25.75% C, and 18.67% G, with 55.58% AT. Phylogenetic analysis based on the 13 protein-coding genes nucleotide sequences with two different methods (neighbour-joining and maximum likelihood analysis) showed that *Y. zichuanensis* was nearest to *Y. jinxiensis*, coming together into a branch, meanwhile together with *Triplophysa venusta* formed a sister group. This study enriched the data of the mitogenome of the genus *Yunnanilus*, and provide an important information for morphological classification of Nemacheilinae species and the study of biological origin.

**Yunnanilus sichuanensis** (Cypriniformes: Nemacheilinae) is endemic to the Upper Yangtze River in China, which is mainly distributed in the slow water area of river trunk and branch flow, with an altitude of 1500–2300 mm (Ding 1995). Currently, only the mitogenome of *Y. jinxiensis* (GenBank Accession number MG 818720) in the same genus has been reported. Recent research has indicated that the natural population number of this fish strongly decreased due to the construction of hydroelectric dams, habitat loss, overfishing and environmental pollution (Ding 1995).

The complete mitogenome of *Y. sichuanensis* was first sequenced using the next-generation sequencing (NGS) in this study. The specimens were collected from Guangan, Sichuan province of China (30° 52' 51.41" N, 104° 14' 55.77" E) in September 2018 and were stored in Zoological Specimen Museum of Neijiang Normal University (accession number: 20180925BB102). A 30–40 mg fin clip was collected and preserved in 95% ethanol at 4 °C. Total genomic DNA was extracted from these caudal fins by a Tissue DNA Kit (OMEGA E.Z.N.A., Norcross, GA, USA) following the manufacturer’s protocol. Subsequently, the genomic DNA was sequenced using the NGS, and then the mitogenome was assembled using *Y. jinxiensis* as a reference.

The complete mitogenome of *Y. sichuanensis* was a circular molecule of 16,571 bp in length (GenBank Accession number MK181571), including 13 protein-coding genes, 2 ribosomal RNA genes, 22 transfer RNA genes, 1 displacement loop (D-loop) locus, and an origin of replication on the light-strand (OL) region, which was in conformity with typical vertebrate mitogenome (Sun et al. 2014; Murienne et al. 2016). Most of the genes were encoded on the heavy strand except ND6 and 8 tRNA genes (tRNA^{Gl}

**Y. jinxiensis** is endemic to the Upper Yangtze River in China, which is mainly distributed in the slow water area of river trunk and branch flow, with an altitude of 1500–2300 mm (Ding 1995). Currently, only the mitogenome of *Y. jinxiensis* (GenBank Accession number MG 818720) in the same genus has been reported. Recent research has indicated that the natural population number of this fish strongly decreased due to the construction of hydroelectric dams, habitat loss, overfishing and environmental pollution (Ding 1995).

**CONTACT** Yuanchao Zou zou3891@163.com College of Life Sciences, Neijiang Normal University, Neijiang, Sichuan 641100, China

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Phylogenetic tree has been known as a model of biological evolution, and its establishment is almost all based on amino acid sequences or DNA sequences (Chor and Tuller, 2005). Neighbor-joining (NJ) and maximum likelihood (ML) methods are two common methods to construct the phylogenetic tree (Dong et al., 2008; Zou et al., 2018). Additionally, ML is an increasingly popular optimality criterion for selecting evolutionary trees (Chor and Tuller, 2005). To confirm the phylogenetic relationships between *Y. sichuanensis* and other Cobitidae family fishes, phylogenetic analysis were performed on the concatenated dataset of 13 protein-coding genes (PCGs) at nucleotide level with NJ and ML (Li et al., 2016; Zou et al., 2017). There were 14 species in the phylogenetic tree, including *Channa argus* (Channoidei, Channidae) as an outgroup. The tree topologies produced by NJ had nearly the same topology as that of ML tree (Figure 1). Both trees showed that *Y. sichuanensis* was nearest to *Y. jinxiensis*, coming together into a branch, meanwhile together with *Triplophysa venusta* formed Clade B, and the rest of species were clustered into clade A. Additionally, results suggested that *Yunnanilus* has a closer relationship with *Triplophysa* than *Schistura* and *Barbatula*. These results may provide an important information for morphological classification of Nemacheilinae species and the study of biological origin.

**Disclosure statement**

No potential conflict of interest was reported by the authors.

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**Figure 1.** The phylogenetic analyses investigated using neighbor-joining (NJ) and maximum likelihood (ML) methods analysis indicated evolutionary relationships among 14 taxa based on the nucleotide sequences of 13 concatenated protein-coding genes. The yielded NJ tree had a same topology as that of ML tree. NJ posterior probability and ML bootstrap support values are shown on the nodes. *Channa argus* (GenBank: KC823605) was used as an outgroup.