Characterization of the complete mitochondrial genome of the paddy frog *Fejervarya multistriata* (Anura: Dicroglossidae) and its phylogeny

Lichun Jiang\(^a,b\), Guihua Lv\(^c\), Lei Liu\(^d\), Bingxiu Wu\(^a\), Zhongwen Xu\(^a\) and Yu Li\(^a\)

\(^a\)Key Laboratory for Molecular Biology and Biopharmaceutics, School of Life Science and Technology, Mianyang Normal University, Mianyang, Sichuan, P.R. China; \(^b\)Ecological Security and Protection Key Laboratory of Sichuan Province, Mianyang Normal University, Mianyang, Sichuan, P.R. China; \(^c\)Dongyang Institute of Maize Research, Zhejiang Academy of Agricultural Sciences, Dongyang, Zhejiang, P.R. China

**ABSTRACT**

The mitochondrial genome has proven to be a highly successful resource for the investigation of evolution and population genetics. Here, we present the complete mitochondrial genome of *Fejervarya multistriata*. The circle genome was found to be 17,759 bp in length, containing 13 protein-coding genes, 23 transfer RNA genes, 2 ribosomal RNA genes, and 1 noncoding control region that are conserved in most Dicroglossidae mitogenomes. The total base composition of the *F. multistriata* mitogenome is 28.04% A, 29.82% T, 26.99% C, and 15.15% G, which is typical for Amphibious animals’ mitochondrial genomes. Eight tRNAs are encoded on the light strand (tRNA-Gln, tRNA-Ala, tRNA-Asn, tRNA-Cys, tRNA-Tyr, tRNA-Ser, tRNA-Glu and tRNA-Pro). Only one PCG is encoded on the light strand (ND6), whereas the other genes are located on the heavy strand. Phylogenetic analyses were performed on the concatenated dataset of 13 PCGs at nucleotide levels with maximum likelihood (ML) and Bayesian analysis (BI) methods. The results showed that *F. multistriata* and *F. limnocharis* integrated into a big branch and they have a close genetic relationship. This study could provide important molecular data for species identification and the phylogenetic relationship of *Fejervarya* and related species.

The paddy frog (*Fejervarya multistriata*) is a species widely distributed in the area from temperate to tropical Asia, belonging to the family Dicroglossidae (Frost 2018). There are many disputes about the classification of *F. kawamurai* and the closely related *F. limnocharis* and *F. multistriata* (Huang and Tu 2016). For example, *F. kawamurai* was a member of the *F. limnocharis* from western Honshu, Japan, and it is described as a new species of dicroglossid frog (Djong et al. 2011). The complete mitochondrial genome has proven to be a highly effective resource for the investigation of the genus *Fejervarya* species evolution and population genetics (Cheng et al. 2018). Thus, we sequenced the mitochondrial genome of *F. multistriata* to discuss the relationship within the genus *Fejervarya* phylogenetic analyses.

Specimens of *F. multistriata* were collected from Santai County, Mianyang City, Sichuan province, China, in August 2019 (105°7′49.89″E, 31°10′41.55″N), and immediately preserved in 95% ethanol at −80 °C until use. The clipped toe of the frog leg was taken and lysed by SDS/protease K and extracted by the phenol-chloride method (Ren et al. 2004) and deposited in the Key Laboratory for Molecular Biology and Biopharmaceutics, Mianyang Normal University (LC2019092206). We employed Long-and-Accurate PCR methods to amplify the whole mitogenomic region of *F. multistriata* with the self-designed and partial universal PCR primers for the mtDNAs of modern frogs (Liu et al. 2005; Kurabayashi and Sumida 2009). The sequence was submitted to the GenBank with the accession number MN733918.

The total length of the mitochondrial genome of *F. multistriata* is 17,759 bp with a base composition of 28.04% A, 29.82% T, 26.99% C, and 15.15% G. The complete mitochondrial genome consists of 13 protein-coding genes, 2 tRNA genes, 23 tRNA genes, and 1 control region. The mitogenome of *F. multistriata* shows the typical gene content observed in vertebrate mitogenomes (Maurino and Viscusi 2009; Zhou et al. 2009; Li et al. 2014). Eight tRNAs are encoded on the light strand (tRNA-Gln, tRNA-Ala, tRNA-Asn, tRNA-Cys, tRNA-Tyr, tRNA-Ser, tRNA-Glu and tRNA-Pro). Only one PCG is encoded on the light strand (ND6), whereas the other genes are located on the heavy strand. Most protein-coding genes begin with ATG as the start codon, except for ND2 gene with ATC, COI gene with ATA, ND3 gene with GTG, and ND3 gene with GTG. The ND6 gene is terminated with AGG as the stop codon, whereas ND1, ND2, ND5, COI,
COII, ATP6, and COIII genes end with an incomplete stop codon (T––) and the other protein-coding genes end with TAA.

To elucidate phylogenetic relationships of *F. multistriata* with the other related species, phylogenetic trees were reconstructed using BI and ML methods based on the nucleotide dataset (Alam et al. 2010; Yu et al. 2012). The best-fit GTR + I + G model was selected in jModelTest 0.1 (Darriba et al. 2012), and yielded identical phylogenetic trees by high node-supporting values, including that 22 reported dicroglossids frogs (Figure 1). In the phylogenetic tree, *F. multistriata* clustered with *F. limnocharis* into a branch and they have a close genetic relationship. The monophyly of *Fejervarya*, *Quasipaa*, *Limnonectes* and *Nanorana* was well supported as also reported in other recent studies (Zhang et al. 2009; Cai et al. 2018). In this study, we present the complete mitochondrial genome sequence of *F. multistriata*, which would contribute to further phylogenetic analysis of this species. More mitochondrial genomic data of undetermined taxa and further analysis are required to reveal the phylogeny and evolution of Dicroglossidae.

**Disclosure statement**

No potential conflict of interest was reported by the author(s).

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