The complete mitochondrial genome of *Sardinella zunasi* (Clupeiformes: Clupeidae)

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**ABSTRACT**

The complete mitogenome of *Sardinella zunasi* was determined by next-generation sequencing. The *S. zunasi* mitogenome was a circular 16,307 bp molecule that contained 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes, and one control region (D-loop). The gene arrangement was consistent with other *Sardinella* mitogenomes. The phylogenetic relationships of 29 Clupeoidei species based on 13 protein-coding genes from the available mitogenomes were analyzed. *Sardinella zunasi* clustered with *Sardinella* among Clupeidae, suggesting a closer relationship with this genus. These results will be useful for understanding the phylogenetic relationships, taxonomic classification, and phylogeography of the genus *Sardinella* relative to other genera of Clupeoidei.

*Sardinella zunasi* (Japanese sardinella) is a ray-finned fish in the family Clupeidae, which includes herrings and sardines. It is native to the northwestern Pacific, where it occurs near shore along the Asian coastline from southwestern Korea and southern Japan to Taiwan. It is commercially important in Korea, Japan, and China (Wang et al. 2008) and has been heavily fished and is considered overexploited. Numerous ecological and morphological studies have examined the species; however, the complete mitochondrial DNA (mtDNA) sequence of this species has not been reported (Kim et al. 2001).

A specimen was bought at a Korean fish market and stored at Soonchunhyang University, Republic of Korea, under accession no. SUC-30000. The species identification of the specimen was confirmed morphologically.

Total genomic DNA was extracted from muscle tissue using a NucleoSpin Tissue Kit (Macherey-Nagel, Düren, Germany) and a genomic library for next-generation sequencing (NGS) was constructed using the MGIEasy DNA Library Prep Kit (MGI Tech, Shenzhen, China). After obtaining raw NGS data on an MGISEQ-2000 (MGI Tech), the mtDNA sequence of a circular 16,307 bp molecule was assembled using CLC Assembly Cell 5.1.1 (Qiagen, Hilden, Germany) and a genomic library for next-generation sequencing (NGS) was constructed using the MGISEQ-2000 (MGI Tech). The complete mtDNA sequence was a circular 16,307 bp molecule containing 13 protein-coding genes, 22 tRNA genes, 2 rRNA genes, and one non-coding region (D-loop). The gene arrangement was identical to those of its relatives: *S. lemuru* (Jiang et al. 2017), *Sardinella melanura* (Andriyono et al. 2019), *Sardinella fijiensis* (Wang et al. 2019), *Sardinella longiceps*, and *Sardinella gibbosa* (Sebastian et al. 2017). Twelve PCGs were on the heavy strand, while ND6 was on the light strand. All of the tRNAs were predicted to form typical stem-loop secondary structures, as in *S. lemuru*. The noncoding D-loop region was located between tRNA-Pro and tRNA-Phe. All PCGs had the typical ATG start codon, except for COX1 (ATC), COX2 (GTG), and ND6 (GTG). Incomplete stop codons were identified in ND2, COX2, COX3, ND3, ND4, and cytB, as in *S. melanura*.

A maximum-likelihood phylogenetic tree of *S. zunasi* and 28 other Clupeoidei species was constructed based on the 13 protein-coding genes using MEGA-X (Kumar et al. 2018). Interestingly, this showed that *S. zunasi* clustered with...
S. lemuru, suggesting a very close relationship, confirming that the current sequence belongs to Sardinella (Figure 1). To confirm that the species identification of S. lemuru NC_039553 was correct, a p-distance analysis using the complete mitogenome sequences of Sardinella species gave values of 0.196–0.214 with other species in Sardinella, while the distance between S. zunasi and S. lemuru was 0.008. In a more detailed analysis, the p-distances were determined for the other cox1 genes of S. zunasi and S. lemuru in GenBank and ranged from 0.0031 to 0.0053 between the cox1 genes of S. zunasi; NC_039553 S. lemuru had a value of 0.0044, while other S. lemuru sequences had values of 0.1799–0.1928 and Sardinella maderensis had a value of 0.1920. Therefore, NC_039553 was verified to be the S. zunasi mitochondrial sequence. The complete mtDNA sequence of S. zunasi should improve the taxonomic classification of Sardinella species with similar mtDNA sequences.

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**Data availability statement**

The genome sequence data that support the findings of this study are openly available in GenBank of NCBI at (https://www.ncbi.nlm.nih.gov/) under the accession no. MW118114. The associated BioProject, SRA, and Bio-Sample numbers are PRJNA695411, SRX9967975 and SAMN17613384, respectively.

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