

Complete mitochondrial genome of *Trematomus newnesi* (Perciformes, Nototheniidae)

Phuong Thi Nguyen^{a,b*}, Seungyeon Lee^{a,b*}, Jihye Jeong^a, Jihun Kim^{a,b}, Dong-Won Han^a, Il-Chan Kim^a, Jun Hyuck Lee^{b,c}, Jisoo Park^{b,d} and Jin-Hyoung Kim^{a,b}

^aDivision of Life Sciences, Korea Polar Research Institute, Incheon, Korea; ^bPolar Science, University of Science and Technology, Daejeon, Korea; ^cResearch Unit of Cryogenic Novel Material, Korea Polar Research Institute, Incheon, Korea; ^dDivision of Ocean Sciences, Korea Polar Research Institute, Incheon, Korea

ABSTRACT

The complete mitochondrial genome of *Trematomus newnesi* was sequenced using an Illumina platform. The 18,602 bp mitogenome contains 13 protein-coding genes, two rRNAs, and 23 tRNAs (tRNA^{Met} is duplicated). The eight stop codons are TAA, TAG, CTT, GTA, AAT, ACT, AGG, and TTA. Two start codons ATG and GTG are present. The GC content is 44.4% and AT content is 55.6%. A phylogenetic tree was generated using 13 species from three families. The results showed that *T. newnesi* is closely related to *Pagothenia borchgrevinki* in Nototheniidae. This study provides fundamental data for further genetic evolutionary studies on *T. newnesi*.

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Introduction

The suborder Notothenioidei is the dominant fish fauna distributed in the Antarctic Ocean. This suborder accounts for almost all (approximately 95%) of the total fish biomass in this polar region (Eastman 2005). Notothenioids are characterized by the presence of antifreeze glycoproteins (AFGPs), lack of hemoglobin, and/or heat-shock response capacity in some species (DeVries and Wohlschlag 1969; Hofmann et al. 2000; Buckley et al. 2004; Beers and Jayasundara 2015; Kim et al. 2019). Dusky rockcod, *Trematomus newnesi* (Boulenger 1902) is a demersal fish that is distributed in depths ranging from 0 to 400 m, often found in shallow near shore areas and intertidal zones according to the fish base (<https://www.fish-base.se/summary/7058>) (Miller 1993) (Figure 1). The genus *Trematomus* has a total of 11 species. The whole mitochondrial genome sequence has been reported from only five species (Song et al. 2016b; Alam et al. 2019; Choi et al. 2021; Papetti et al. 2021). In this study, we provide a complete mitochondrial genome of *T. newnesi* with the phylogenetic analysis to increase the understanding of its evolution compared to other Antarctic fishes.



Materials

The fish were collected near the Jang Bogo Station (Korean Antarctic Research Base) in the north of Victoria Land (74°37.8792'S, 164°14.8883'E) from December 2020 to


February 2021, stored at –20 °C immediately after death, and transferred to the Korea Polar Research Institute (KOPRI), Incheon, Korea. A specimen was deposited at KOPRI (<https://www.kopri.re.kr/eng/>, Jin-Hyoung Kim, kimjh@kopri.re.kr) under voucher number (Antarctic fish_012).

Methods

The total genomic DNA was extracted from the muscle tissue using the conventional phenol-chloroform method. An Illumina paired-end library was prepared from the extracted DNA following the manufacturer's instructions. Sequencing was performed on an Illumina HiSeq XTM Ten by a commercial company (Phyzen, Seong-Nam, South Korea). Adapter sequences and low-quality reads were eliminated using quality_trim option under Phred score 20; the pre-processing completed data was performed de novo assembly using QIAGEN CLC Assembly Cell package (Version 4.21, CLC Inc., Aarhus, Denmark). Subsequently, contigs derived from the mitochondrial genome were selected, and the final mitochondrial genome was assembled using Illumina data-based polishing. Next, the complete mitochondrial genome sequence was annotated using the GeSeq program (<https://chlorobox.mpimp-golm.mpg.de/geseq-app.html>) based on related species. Manual curation was performed using the Artemis annotation tool (Liang et al. 2018) with NCBI BLASTN search.

CONTACT Jin-Hyoung Kim  kimjh@kopri.re.kr  Division of Life Sciences, Korea Polar Research Institute, Incheon, Korea

*Phuong Thi Nguyen and Seungyeon Lee contributed equally on this manuscript.

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Figure 1. A photo of the *Trematomus newnesi*. The photo has been taken by KJH.

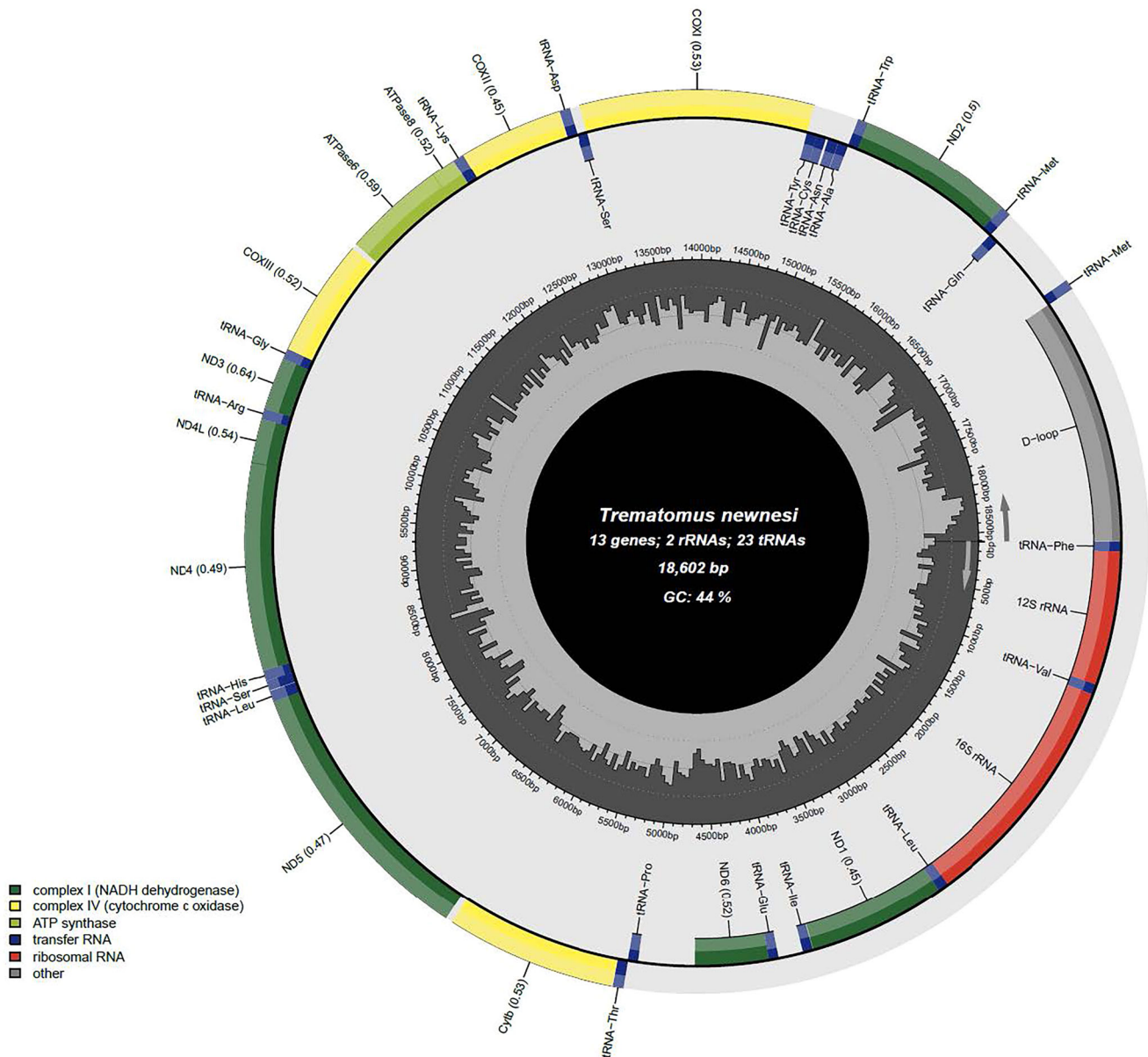


Figure 2. Circular gene map of the *Trematomus newnesi* mitochondrial genome using MitoFish (<http://mitofish.aori.u-tokyo.ac.jp/annotation/input/>).

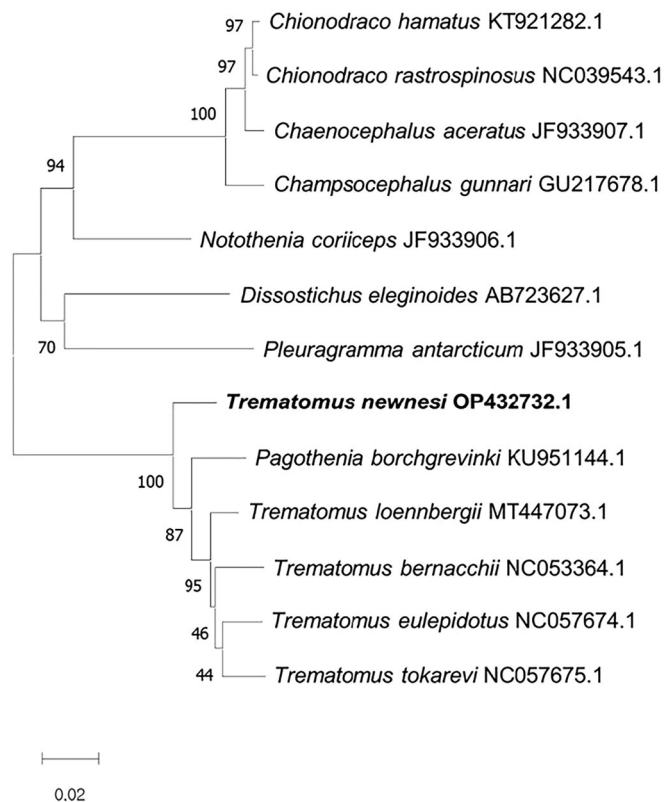


Figure 3. Phylogenetic tree for the genus *Trematomus*, with outgroup species, with MEGA (version 11.0.13) by the Likelihood method and JTT model based on 13 protein-coding genes. The scientific name and GenBank number were included for each species.

The phylogenetic analysis was performed by MEGA11 (Lin et al. 2012) involved 13 protein-coding genes. The following sequences were used: KT921282.1 (Song et al. 2016a), NC039543.1 (Liang et al. 2018), JF933907.1, GU217678.1 (Lin et al. 2012), JF933906.1, AB723627.1, JF933905.1, KU951144.1 (Liu et al. 2016), MT447073.1 (Choi et al. 2021), NC053364.1 (Song et al. 2016b), NC057674.1 (Papetti et al. 2021), NC057675.1 (Papetti et al. 2021). The Maximum Likelihood (ML) tree was constructed with 1000 bootstrap replicates and JTT matrix-based model (Felsenstein 1981).

Results and discussions

The complete mitochondrial genome of *T. newnesi* (GenBank Number: OP432732.1) is 18,602 bp in length and consists of 13 protein-coding genes (ND1, ND2, ND3, ND4, ND4L, ND5, ND6, CYTB, COX1, COX2, COX3, ATP6, ATP8), two rRNA genes (*rrnL* and *rrnS*), and 23 tRNA genes (*tRNA^{Met}* was duplicated) (Figure 2). In the 23 tRNA genes, the duplication of *tRNA^{Met}* gene might because *T. newnesi* have a high frequency of gene duplications and rearrangement of mitochondrial genomes (Lin et al. 2012; Song et al. 2016b; Papetti et al. 2021; Minhas et al. 2022; Patel et al. 2022). In 13 protein-coding genes, GTG was the start codon for COX1, and ATG was the start codon for the rest. There were eight stop codons, TAA for COX1, ATP6, CYTB, ND4L, and ND1 genes, and the stop codon TAA of five genes (COX1, ATP6, CYTB, ND4L, and ND1) was predicted to be completed by the addition of 3' A residues to the mRNA, TAG for ATP8 and ND5 genes, CTT for

COX2 gene, GTA for COX3 gene, AAT for ND3 gene, ACT for ND4 gene, AGG for ND6 gene, and TTA for ND2 gene. The GC contents were 44.4% and AT contents were 55.6%. These results are corresponded with a previous result regarding of the inversion of the control region and mechanisms of rearrangement in notothenioid mitochondrial genomes (Papetti et al. 2021). In particular, it is confirmed that the mitochondrial genome of *T. newnesi* also has an extremely rare inversion of a large genomic segment as shown in other fishes of Trematominae such as *Trematomus eulepidotus*, *Trematomus tokarevi*, *Trematomus borchgrevinki*, *Lindbergichthys nudifrons* (Papetti et al. 2021).

The phylogenetic tree (Figure 3) showed that *T. newnesi* was close to *Pagothenia borchgrevinki* in Nototheniidae. The phylogenetic analysis would provide an intuitive insight into the evolutionary relationship of *T. newnesi* with other fish species living in the Antarctic Ocean.

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Author contributions

Conceptualization, JHK, PTN, SL, and JK; methodology, PTN, SL, JJ, JK, and DWH; resources, DWH, ICK, and JP; writing original draft preparation, PTN, SL, and JJ; writing review and editing JHK, ICK, JHL, and JP; interpretation of data, JHK, PTN, SL, ICK, JP, and JHL; project administration, JHK; funding acquisition, JHL and JHK. All the authors have read and agreed to the published version of the manuscript.

Ethics statement

All handling and experimental procedures followed the ethical guidelines regulated by the Animal Experimental Ethics Committee established by the Korea Polar Research Institute (KACC2201-007).

Disclosure statement

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

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Data availability statement

The genome sequence data that support the findings of this study are available in GenBank at <https://www.ncbi.nlm.nih.gov/under> accession no. OP432732.1. The associated BioProject, SRA, and Bio-Sample numbers were PRJNA881762, SRR21669782, and SAMN30864146, respectively.

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