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MITOGENOME ANNOUNCEMENT

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Complete mitochondrial genome of the calanoid copepod *Eurytemora affinis* (Calanoida, Temoridae)

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ABSTRACT

The complete mitochondrial genome was sequenced from the calanoid copepod *Eurytemora affinis*. The sequenced total genome size was 18,553 bp. The mitochondrial genome of *E. affinis* has 13 protein-coding genes (PCGs), two rRNAs, and 22 tRNAs. Of 13 PCGs, *ND1*, *ND5*, and *ATP6* genes had incomplete stop codons TA–, T—, and TA–, respectively. Furthermore, the stop codons of the remaining eleven PCGs were TAG or TAA while the start codon of 13 PCGs was ATG (*Cytb*, *ATP8*, *ATP6*, and *CO3* genes), ATT (*CO1*, *ND2*, *ND3*, *ND4L*, *ND5*, and *ND6* genes), and ATA (*ND1*, *ND4*, and *CO2* genes), respectively. The ratio of A + T and G + C nucleotides of 13 PCGs of *E. affinis* mitogenome showed 63.9% and 36.1%, respectively while those ratio of the entire sequences were 65.5% and 34.5%, respectively.

To date, 12 species have been retrieved in the genus *Eurytemora* (http://v3.boldsystems.org/index.php/Taxbrowser_Taxonpage?taxid=5798); however, not a single complete mitochondrial genome has been reported from those species. Despite the limited mitochondrial genome information of these species, the population genetic analyses were examined to reveal how they invade freshwater ecosystem, using *16S rRNA* and cytochrome oxidase 1 (*CO1*) gene (Lee, 1999, 2000, 2016; Winkler et al. 2008; Sukhikh et al. 2016). Also, environmental ecotoxicology using the RNA-seq (Legrand et al. 2016; Lee et al. 2018) of *E. affinis* have been widely reported (Lee et al. 2017; Legrand et al. 2017; Zidour et al. 2019), in addition to usefulness of *E. affinis* in studying the environmental adaptation (e.g. temperature) (Karlsson et al. 2018), but the taxonomical consideration of the copepod *E. affinis* based on complete mitogenome is still unavailable. Based on its wide distribution of the genus *Eurytemora* with ecological importance, complete mitochondrial genome will be useful for a better understanding on the phylogenetic divergence of *E. affinis* populations in the genus *Eurytemora*.

The calanoid copepod *E. affinis* was obtained from the laboratory cultures that originated from the sample copepods on September 2014 from the oligohaline zone of the Seine Estuary (49°45′34.38″N, 0°17′33.68″E) by Prof. Sami Souissi (Michalec et al. 2017) and maintained in Nagasaki University in Japan. The specimen was deposited in the copepod collection

of the Fisheries Science Museum of Nagasaki University, Nagasaki University under the accession no. FFNU-Cr-00393.

We sequenced 300 bp paired-end library of *E. affinis* from the whole body genomic DNA using the Illumina HiSeq 2500 platform (GenomeAnalyzer, Illumina, San Diego, CA). *De novo* assembly was conducted using spades v3.13.0 (http://cab.spbu.ru/software/spades/) with K-mer auto. Of the assembled *E. affinis* 729,332 contigs (N50 = 2051 bp) with Newbler (version 2.9; identity 100) (http://www.454.com), one supercontig was obtained. After a manual curation of one supercontig with Consed (version 19.0) (http://www.phrap.org/consed/consed.html) with a gap closing, a single supercontig was mapped to the mitochondrial DNA of *E. affinis*. The total length of the complete mitochondrial genome of *E. affinis* was 18,553 bp (GenBank accession no. MN043905). The mitochondrial genome of *E. affinis* contained 13 protein-coding genes (PCGs), two rRNAs, and 22 tRNAs. The direction of 13 PCGs of *E. affinis* was mostly different to those of other copepods but the directions of the mitochondrial genome of the harpacticoid copepods *Tigriopus japonicus* and *Tigriopus californicus* were identical (Figure 1). The ratio of A + T and G + C nucleotides of 13 PCGs of *E. affinis* mitogenome showed 63.9% and 36.1%, respectively, while those ratio of all the sequences were 65.5% and 34.5%, respectively.

The phylogenetic placement of *E. affinis* was obtained using previously reported complete copepod mitogenomes
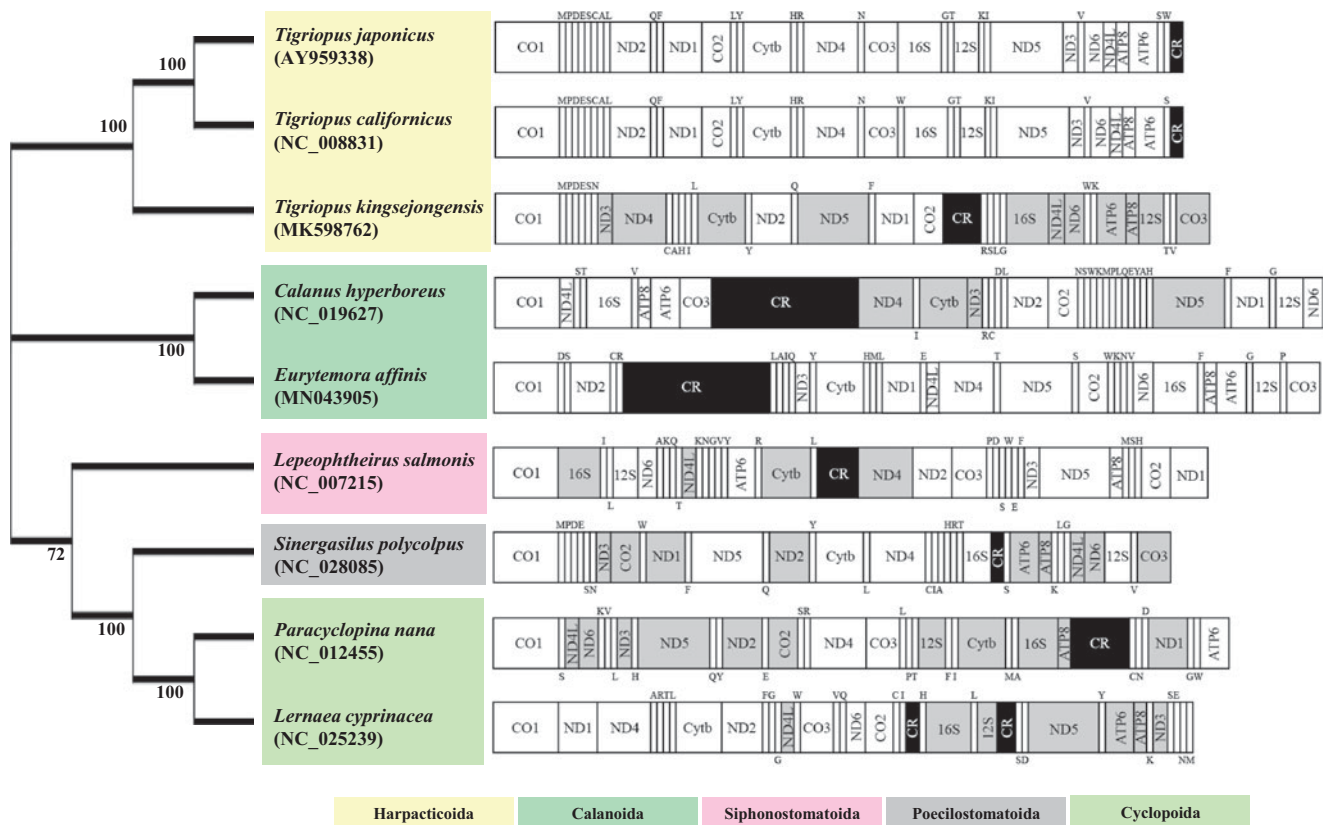

Figure 1. Phylogenetic analysis. We conducted comparison of the mitochondrial genomes of nine copepods. Amino acids of 13 PCGs gene from nine copepods were aligned using MEGA software (ver. 10.0.1) with the ClustalW alignment algorithm. To establish the best-fit substitution model for phylogenetic analysis, the model with the lowest Bayesian Information Criterion (BIC) and Akaike Information Criterion (AIC) scores were estimated using a maximum-likelihood (ML) analysis. According to the results of model test, maximum likelihood phylogenetic analyses were performed with the LG + G + I model. Gray color represents the opposite direction of PCGs and the Black color represents the control regions; Upper and lower single letters indicate the direction of tRNA clockwise and anticlockwise, respectively. Modified from Hwang et al. (2019).

(Figure 1). The phylogenetic placement of the two calanoid copepods *Calanus hyperboreus* and *E. affinis* were in the same clade, but their orientations of 13 PGCs and tRNAs were different, indicating rearrangements of those particular components of mitochondrial DNA in the calanoid copepods over evolution. This information will be helpful for a better understanding of mitogenome evolution in the genus *Eurytemora*.

Disclosure statement

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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