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MITOGENOME ANNOUNCEMENT

The complete mitochondrial genome of sea slug *Phyllidiopsis krempfi* Pruvot-Fol, 1957 (Nudibranchia, Phyllidiidae) from Pacific Ocean

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ABSTRACT

The mitogenome of *Phyllidiopsis krempfi* has been determined for the first time. The assembled mitogenome was 14,970 bp in length, including 13 protein-coding genes, 22 transfer RNA, and 2 ribosomal RNA genes. The gene content and order were identical with those of the other Phyllidiid species. The molecular taxonomic position of *P. krempfi* was clustered with the Phyllidiid species. The genus *Phyllodiopsis* clade is closely related with the genus *Phyllidia*. The mitogenome of *P. krempfi* provides significant DNA molecular data for further identification and phylogenetic analysis within the Phyllidiid.

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Nudibranchs are called ‘ocean cleaners’ likened to hyenas on land because they feed on sponges, hydoids (Folino 1997), bryozoans (Dominguez et al. 2008), etc. The genus *Phyllidiopsis* Bergh, 1876 including the Phyllidiidae are distributed mostly throughout Indo-Pacific tropical waters, which comprises 37 valid species (Valdés 2001; MolluscaBase 2020). Its members are characterized by having an elongate foregut and fused oral tentacles (Brunckhorst 1993). The phylogenetic relationships within the Phyllidiidae based on molecular and morphology analyses has been conducted, but position of the genera within the family remains unclear (Brunckhorst 1993; Valdés 2002; Valdés 2003; Stoffels 2016). The mitochondrial genome is a useful tool for inferring phylogenetic relationships (Matsudaira and Ishida 2010). In this study, we analyzed the full-length mitochondrial genome (mitogenome) of the *P. krempfi* for the first time and it is valuable information for further study on molecular systematics and phylogenetic relationships within the Phyllidiidae.

Specimens of *P. krempfi* were collected from the Pacific Ocean (7°16′20.02′N, 134°31′21.68′E). The voucher specimens are deposited in the National Marine Biodiversity Institute of Korea (MABIK Lot no. 0018233). The genomic DNA was extracted from the muscle tissue and the mitogenome sequences were analyzed in two ways: first, after the COI gene was amplified through the universal primers, the primer sets were designed from the partial sequences of the COI gene of *P. krempfi* and highly preservative gene regions for the *Phyllidiopsis* species. Then, we conducted the long-range PCR (LR-PCR) to amplify the targeted genomic intervals and sequenced by the Sanger method. The sequences were assembled and annotated in comparison with the previously reported mitogenome sequences of the Phyllidiid species (Yu et al. 2018; Dinh Do et al. 2019) using the Geneious v9.1.2 (Kearse et al. 2012). Additionally, the mitochondrial genome annotation (MITOS) server (Bernt et al. 2013), and the tRNAscan-SE server (Lowe and Chan 2016) were used for annotation.

The assembled *P. krempfi* mitogenome (GenBank accession number MT726194) was a 14,970 bp long circular DNA with 2 ribosomal RNA (rRNA), 22 transfer RNA (tRNA), and 13 protein-coding genes (PCGs). The gene content and order were identical to those of the species belonging to the order Nudibranchia (Xiang et al. 2016; Yu et al. 2018; Dinh Do et al. 2019). All PCGs get off by the typical ATG as start codon. Eight (*COI, nad6, nad1, nad4l, CO2, atp6, nad3, nad4*) of 13 PCGs use TAA for the stop codon, and three genes (*nad5, atp8, CO3*) ends with TAG while *nad2* and *cytb* genes have an incomplete stop codon, T.

A maximum-likelihood (ML) tree was constructed to investigate the molecular taxonomic position of these species using RAxML 8.2 (Stamatakis 2014), and the dataset used the nucleotide sequences of the 12 PCGs excluding *atp8* of the other species belonging to the Nudibranchia. *Phyllidiopsis krempfi* was clustered with the Phyllidiid species previously announced from the GenBank, with high bootstrap values of 100% (Figure 1). The genus *Phyllodiopsis* clade is closely related with the genus *Phyllidia*, as well as support for the previously published 16s gene tree (Valdés 2003).

Disclosure statement

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

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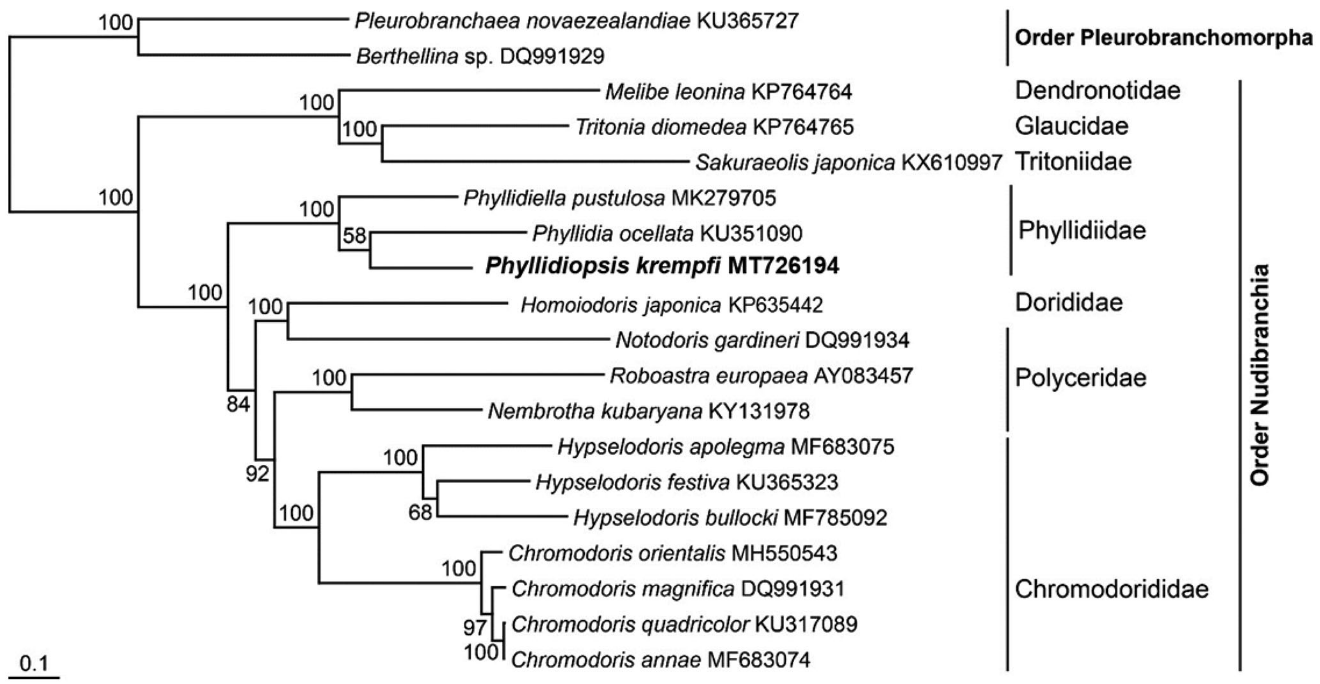

Figure 1. Maximum-likelihood (ML) phylogeny based on the full-length mitochondrial genomes from the marine invertebrate species belonging to the order Nudibranchia. The nucleotide sequence matrix included the first, second and third codon positions of the 12 protein-coding genes. A bootstrap value above 50% in the ML analysis is indicated at each node. *Phyllidiopsis krempfi* analyzed in this study is shown in bold.

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Data availability statement

The data that support the findings of this study are openly available in GenBank of NCBI at https://www.ncbi.nlm.nih.gov, reference number MT726194.

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