ASED http://e-ASED.org
Animal Systematics, Evolution and Diversity

Anim. Syst. Evol. Divers. Vol. 37, No. 4: 354-357, October 2021
https://doi.org/10.5635/ASED.2021.37.4.032

Short communication

DNA Barcoding of *Eurydice longiantennata* (Isopoda, Cymothooidea, Cirolanidae) from South Korea

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ABSTRACT

In Korean waters, the cirolanid isopod, *Eurydice longiantennata* Nunomura and Ikehara, 1985 has been reported only from the subtidal zone of Jeju island. We obtained the mitochondrial cytochrome *c* oxidase subunit I (*COI*) sequences of this species and determined the DNA barcoding data of *E. longiantennata* based on a genetic comparison of *E. longiantennata* and its congeners. The intra-specific genetic distance between the three *COI* sequences of *E. longiantennata* ranged from 0 to 0.6%. The inter-specific distances between *E. longiantennata* and other cirolanid isopods ranged from 24 to 33.2%. In this study, we provided the DNA information of *E. longiantennata* with a morphological diagnosis and images of the species.

Keywords: cirolanid, *COI*, DNA barcode, isopods, Korean waters

INTRODUCTION

The genus *Eurydice* Leach, 1815, which includes 58 species, can be distinguished from the other cirolanid genera by the following characteristic features: (1) the antennule has geniculate peduncular articles between articles 1 and 2, (2) the antenna is composed of a 4-articled peduncle and a multi-articled flagellum, (3) the maxilliped has a reduced endite lacking coupling hooks, and (4) pleonite 5 is not surrounded by pleonite 4 (Bruce, 1986; Brusca et al., 1995; Jones and Nithyanandan, 2012). To date, five *Eurydice* species, *E. akiyamai* Nunomura, 1981; *E. longiantennata* Nunomura and Ikehara, 1985; *E. nipponica* Bruce and Jones, 1981; *E. nunomurai* Saito, 2012, and *E. saikaiesis* Nunommura, 2008, have been reported from the Far East (Bruce and Jones, 1981; Nunomura, 1981, 2008; Nunomura and Ikehara, 1985; Saito, 2012; Kim and Yoon, 2019). In Korean waters, Kim and Yoon (2019) reported the finding of *E. longiantennata*, originally described from Japanese waters, from the subtidal zone of Jeju Island.

DNA barcoding has been widely used to recognize species in taxonomic studies of variable animals (Hebert and Gregory, 2005; Raupach et al., 2015; Song, 2020; Lee and Shin, 2021). Even though this molecular method can accelerate species identification and the discovery of new taxa, DNA barcode libraries for isopods are very poor (PrasannaKumar et al., 2020). Therefore, we herein presented the first cytochrome *c* oxidase subunit I (*COI*) sequences of *E. longiantennata* as a barcoding marker for molecular identification.

Eurydice longiantennata materials were collected from the subtidal zone of Jeju Island (33°09′56″N, 126°14′50″E) using a Smith-McIntyre grab. The sediment samples were sieved with a mesh size of 1 mm and then fixed in 95% ethyl alcohol. The morphological observations were conducted under a dissecting microscope (SMZ 1500; Nikon, Japan) and a compound microscope (BX 50; Olympus, Japan). Photographs were captured using iSolution Lite software (IMT i-solution, Bernardy, Canada). The voucher specimens were deposited at the National Institute of Biological Resources (NIBRIV0000860026). Genomic DNA was extracted using a DNeasy Blood and Tissue Kit (Qiagen, Hilden, Germany) according to the manufacturer's protocol. The *COI* sequences of *E. longiantennata* were obtained using LCO 1490 and HCO 2198 primers (Folmer et al., 1994). These *COI* sequences were deposited in GenBank under the accession number OK081997–OK081999 and aligned with one species of *Eury-*

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eISSN 2234-8190

dice, *Eurydice pulchra* Leach, 1815, and three species of *Excirolana* Richardson, 1912, *Excirolana armata* (Dana, 1853), *Excirolana latipes* (Barnard, 1914), and *Excirolana natalensis* (Vanhöffen, 1914), using Geneious Prime v2021. 2.2 (Biomatters, Auckland, New Zealand). The genetic distances were acquired using the Kimura-2-parameter (K2P) model in MEGA v.6.06 (Tamura et al., 2013).

RESULTS AND DISCUSSION

We newly obtained three partial *COI* sequences each 658 bp from three *Eurydice longiantennata* individuals collected from South Korea. We conducted the genetic analysis of this species and four other cirolanid species, *Eurydice pulchra*, *Excirolana armata*, *Excirolana latipes*, and *Excirolana natalensis*, available from GenBank (Raupach et al., 2015; von der Heyden et al., 2020; Tourinho et al., 2021). The alignment length of five species in the genetic comparison was 474 bp. The intra-specific variations were 0 to 0.6% and the inter-specific genetic distance between *Eurydice longiantennata* and four cirolanid species measured by the K2P model ranged from 24 to 33.2% (Table 1). Consequently, *COI* identification of the cirolanid isopods was provided based on this study, although further studies with additional DNA information on more *Eurydice* species as well as other cirolanid isopods are needed for detailed identification. In this study, we reported the DNA barcoding data of *Eurydice longiantennata* with the morphological diagnosis and photographs. These features would be helpful for distinguishing it from other congeners.

Order Isopoda Latreille, 1817
Superfamily Cymothooidea Leach, 1814
Family Cirolanidae Menzies and Glynn, 1968
Genus *Eurydice* Leach, 1815

Eurydice longiantennata **Nunomura and Ikehara, 1985 (Fig. 1)**

Eurydice longiantennata Nunomura and Ikehara, 1985: 52, figs. 1, 2; Kim and Yoon, 2019: 169, figs. 1–5.

Diagnosis. Body elongated oval, about 3 times longer than the greatest width, smooth dorsally; lateral margins subparallel in males, whereas more ovoid in females. Cephalon with minute rostral process anteriorly; eyes located dorsoventrally; frontal lamina lanceolate, tapering proximally; clypeus acute distally, projecting ventrally. Pereonite 1 subequal to pereonite 2 in length. Pleonites similar to each other in length; pleonite 5 not surrounded by pleonite 4. Pleotelson with truncated posterior margin bearing 11 teeth and 12 plumose setae.

Table 1. Genetic distance (K2P) based on 658 bp-size *COI* sequences in *Eurydice longiantennata* and three cirolanid isopods

No.	Species	Accession No.	1	2	3	4	5	6	7	8	9	Data source
1	*Eurydice longiantennata*	OK081997										Present study
2	*Eurydice longiantennata*	OK081998	0.006									Present study
3	*Eurydice longiantennata*	OK081999	0.006	0.000								Present study
4	*Eurydice pulchra*	KT208670	0.254	0.261	0.261							Raupach et al. (2015)
5	*Eurydice pulchra*	KT209053	0.254	0.261	0.261	0.000						Raupach et al. (2015)
6	*Eurydice pulchra*	KT209446	0.254	0.261	0.261	0.000	0.000					Raupach et al. (2015)
7	*Excirolana natalensis*	MN593876	0.270	0.277	0.277	0.242	0.242	0.242				von der Heyden et al. (2020)
8	*Excirolana latipes*	MN594025	0.328	0.332	0.332	0.287	0.287	0.287	0.267			von der Heyden et al. (2020)
9	*Excirolana armata*	MT239963	0.287	0.294	0.294	0.273	0.273	0.273	0.240	0.264		Tourinho et al. (2021)

K2P, Kimura-2-parameter; *COI*, cytochrome *c* oxidase subunit I.

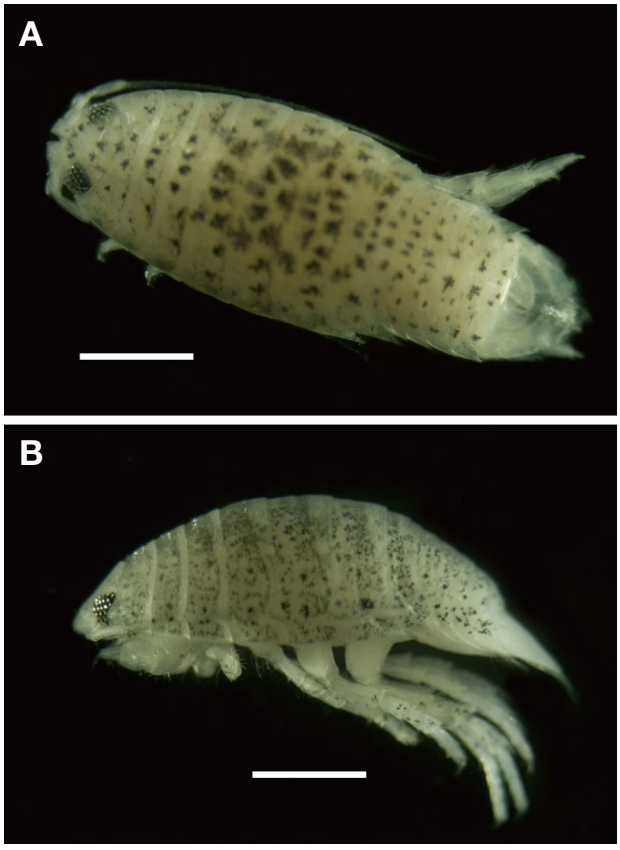

Fig. 1. Photographs of *Eurydice longiantennata* from Jeju Island of South Korea. Dorsal view (A) and lateral view (B). Scale bars: A, B = 1 mm.

Antennule nearly reaching the posterior margin of the cephalon, geniculated between peduncular articles 1 and 2. Antenna reaching the proximal region of the pleotelson, consisting of 31 flagellar articles. Maxillipedal palp 5-articled; endite reduced. Appendix masculina longer than rami, inserted medially, serrated on the subdistal region; apex with a subacute process. Penes separated, rounded distally.

Distribution. Korea, Japan.

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CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

ACKNOWLEDGMENTS

This study was supported by research funds from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR 202131101).

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Received September 23, 2021
Revised October 17, 2021
Accepted October 17, 2021