



# Complete Mitochondrial Genome and Phylogenetic Analysis for the Korean Field Mouse *Apodemus peninsulae* Found on Baengnyeong Island in South Korea

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## ABSTRACT

The Korean field mouse, *Apodemus peninsulae* mitochondrial genome has previously been reported for mice obtained from mainland Korea and China. In this investigation the complete mitochondrial genome sequence for a mouse obtained from Baengnyeong Island (BI) in South Korea was determined using high-throughput whole-genome sequencing for the first time. The circular genome was determined to be 16,268 bp in length. It was found to be composed of a typical complement gene that encodes 13 protein subunits of enzymes involved in oxidative phosphorylation, two ribosomal RNAs, 22 transfer RNAs, and one control region. Phylogenetic analysis involved 13 amino acid sequences and demonstrated that the *A. peninsulae* genome from BI was more closely grouped with two Korean samples (HQ660074 and JN546584) than the Chinese (KP671850) sample. This study verified the evolutionary status of *A. peninsulae* inhabiting the BI at the molecular level, and could be a significant supplement to the genetic background.

**Keywords:** *Apodemus peninsulae*, Baengnyeong island, Korean field mouse, Mitochondrial genome

## Introduction


The genus *Apodemus* contains at least 20 rodent species (Musser & Carleton, 2005), of which only three have been recorded in South Korea, according to the National Institute of Biological Resources, and they are as follows: *Apodemus agrarius*, *A. chejuensis*, and *A. peninsulae* (Won & Smith, 1999; Yoon *et al.*, 2004). The Korean

field mouse, *A. peninsulae* is distributed from the Russian Far East and Japan (Hokkaido) to China and the Korean mainland (Batsaikhan *et al.*, 2016; Jo *et al.*, 2018). The habitats of these mice include fallow fields, grasslands, shrubs, and mixed forests (Lu & Zhang, 2005), but understory coverage and an abundance of fallen trees are significantly more important for forest-dwelling mice when compared to the striped field mouse (Lee *et al.*, 2008; 2012). To date, the mitochondrial genome data for *A. peninsulae* has been reported for two inland samples from Korea and one from China (Jeon *et al.*, 2016; Kim & Park, 2011; Oh *et al.*, 2011).

Baengnyeong Island (BI) is located in the northernmost part of the West Sea on the Korean Peninsula, approximately 228 km from Incheon (Fig. 1). It has an area of

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45.8 km<sup>2</sup>, a coastline road of 56.8 km, and it was connected to the Ongjin Peninsula in Hwanghae-do during the Quaternary Ice Age (Park & Lee, 2007). Despite its small area, BI is home to a variety of vertebrates such as birds, mammals, and amphibians, some of which are rare species that are not found inland (Kang *et al.*, 2012; Park *et al.*, 2019). The geographical location, topography, and climatic factors of BI and its complex biogeographic history indicate that it is an area with a high possibility for speciation (Cho *et al.*, 2013).

In this study, the complete mitochondrial genome sequence from an *A. Peninsulae* specimen from BI in Korea was obtained for the first time. Furthermore the phylogenetic relationships between the genus *Apodumus* and the Korean and Chinese *A. peninsulae* were investigated. This study verified the evolutionary status of *A. peninsulae* at the molecular level and provided important complementary data for its genetic background.



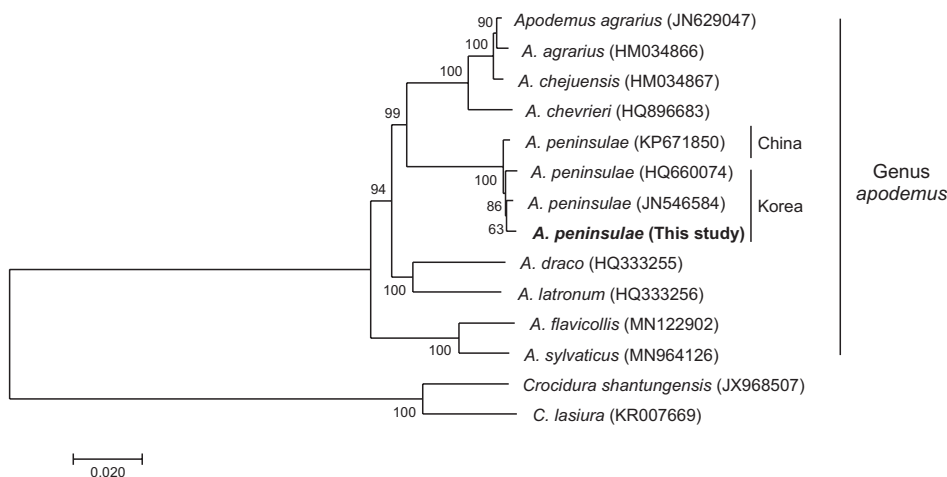
**Fig. 1.** Sampling location of *Apodumus peninsulae* species used in this study.

## Materials and Methods

Tissue (muscle) samples from an *A. peninsulae* specimen (obtained at 37°58'25.85", 124°41'57.49"; Ongjin-gun, Incheon, Korea; voucher number NIBRMM0000106403) were collected and stored in the National Institute of Biological Resources (NIBR, Incheon, Korea). Sample collection was approved by the Institutional Animal Care and Use Committee (approval number: NIBR IACUC 20220001). Genomic DNA was extracted from the tissues using a DNeasy Blood and Tissue Kit (Qiagen) in accordance with the manufacturer's protocol and deposited in the NIBR. DNA Link (Seoul, Korea) was used to perform whole-genome resequencing on an Illumina HiSeq 2500 platform. The mitochondrial genome was assembled using MitoZ (v.2.3) (Meng *et al.*, 2019) and small gaps in the assembly were re-mapped using MITOBim (v.1.9.1) (Hahn *et al.*, 2013). DOGMA (Wyman *et al.*, 2004) and ARWEN (Laslett & Canback, 2008) were used for gene prediction. The neighbor-joining method was used to construct a tree with the software MEGA 6.0 (Arizona State University) using the Kimura 2-parameter model with 1,000 bootstrap replicates.

## Results and Discussion

The total length of the *A. peninsulae* (GenBank accession: OL978460) mitochondrial genome was determined to be 16,268 bp with a GC base composition of 34.7%. The two mitochondrial genomes previously identified from inland South Korea samples were determined to be 16,269 bp (JN546584; Kim & Park, 2011) and 16,266 bp (HQ660074; Oh *et al.*, 2011), while the sample from China was 16,457 bp (Jeon *et al.*, 2016). There were one or two base pair differences with the Korean samples and a 189 bp difference with the Chinese sample. The mitochondrial genome was comprised of 13 protein-coding genes



**Fig. 2.** Phylogenetic relationships among 12 *Apodemus* sp. mitochondrial genomes and two soricomorpha out group. The numbers present on the nodes indicate percentages of 1000 bootstrap values, estimated for concatenated sequences of 13 protein-coding genes.

(PCGs), 22 transfer RNAs, two ribosomal RNAs, and a control region. The most common start codon, ATG, was found in nine of the PCGs, but not in the other four [*ND1*, *ND3*, *ND5* (ATA), and *ND2* (ATC)].

A neighbor-joining tree was constructed with the 13 PCGs sequenced here, along with 11 *Apodemus* mitochondrial genomes downloaded from GenBank (Fig. 2). Phylogenetic analysis confirmed that the clade of *A. peninsulae* was a sister to *A. agrarius* and *A. chejuensis*, which corresponded to the three species of the *Apodemus* genus recorded in South Korea. The three domestic *A. peninsulae* species, including individuals from BI, were grouped closer to each other than the Chinese *A. peninsulae*. In conclusion, this study enriches our evolutionary and ecological understanding of *A. peninsulae* through the contribution of mitochondrial genome data from an isolated region.

### Conflict of Interest

The authors declare that they have no competing interests.

### Acknowledgments

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