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# Complete mitochondrial genomes of the tooth of a poached Bornean banteng (*Bos javanicus lowi*; Cetartiodactyla, Bovidae)

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## MITOGENOME ANNOUNCEMENT

Complete mitochondrial genomes of the tooth of a poached Bornean banteng (*Bos javanicus lowi*; Cetartiodactyla, Bovidae)Taichiro Ishige<sup>1</sup>, Takashi Gakuhari<sup>2</sup>, Kei Hanzawa<sup>3</sup>, Tomohiro Kono<sup>1</sup>, Indra Sunjoto<sup>4</sup>, Jum Rafiah Abdul Sukor<sup>5</sup>, Abdul Hamid Ahmad<sup>6</sup>, and Hisashi Matsubayashi<sup>3</sup><sup>1</sup>NODAI Genome Research Center, Tokyo University of Agriculture, Setagaya, Tokyo, Japan, <sup>2</sup>Faculty of Medicine, Kitasato University, Sagami-hara, Kanagawa, Japan, <sup>3</sup>Faculty of Agriculture, Tokyo University of Agriculture, Atsugi, Kanagawa, Japan, <sup>4</sup>Sabah Forestry Department, Sandakan, Sabah, Malaysia, <sup>5</sup>Sabah Wildlife Department, Kota Kinabalu, Sabah, Malaysia, and <sup>6</sup>Institute for Tropical Biology and Conservation, Universiti Malaysia Sabah, Kota Kinabalu, Sabah, Malaysia

## Abstract

Here we report the complete mitochondrial genome of the Bornean banteng *Bos javanicus lowi* (Cetartiodactyla, Bovidae), which was determined using next-generation sequencing. The mitochondrial genome is 16,344 bp in length containing 13 protein-coding genes, 21 tRNAs and 2 rRNAs. It shows the typical pattern of bovine mitochondrial arrangement. Phylogenetic tree analysis of complete mtDNA sequences showed that Bornean banteng is more closely related to gaur than to other banteng subspecies. Divergence dating indicated that Bornean banteng and gaur diverged from their common ancestor approximately 5.03 million years ago. These results suggest that Bornean banteng might be a distinct species in need of conservation.

## Keywords

Banteng, *de novo* assembly, mtDNA, next-generation sequencing

## History

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The banteng (*Bos javanicus* d'Alton, 1923) is a type of wild cattle that is found in the open forests of Southeast Asia. The animal is categorized as an endangered species on the IUCN Red List of Threatened Species (Timmins et al., 2008) due to excessive hunting for bush meat and horns, as well as habitat loss and degradation. Banteng is classified into three different subspecies occurring in three different regions: *Bos javanicus javanicus* in Java, *Bos javanicus birmanicus* in the mainland of Southeast Asia, and *Bos javanicus lowi* in Borneo. Genetic analysis of Bornean banteng was first reported in our previous study (Matsubayashi et al., 2014). The results suggested that Bornean banteng is more closely related to the gaur (*Bos gaurus*) than to other banteng subspecies. However, these results were based on only partial cytochrome *b* and D-loop sequences.

Here, the complete mitochondrial genome of Bornean banteng was sequenced from a poached male skull collected in 2010 in Malua (N05°16'24.0", E117°37'33.3", 171 m a.s.l); the tooth was used for DNA extraction. Genomic DNA was used to generate a sequences library that was then sequenced as paired-end, 100-bp reads on an Illumina HiSeq 2000 sequencer (Illumina, San Diego, CA). The paired-end reads were assembled *de novo* using the CLC Genomics workbench. Protein-coding genes, ribosomal and transfer RNA genes were identified by comparing with published mitochondrial genomes of gaur (JN632604). Phylogenetic trees were constructed by the maximum likelihood method using the MEGA 5 software (Tamura et al., 2011). To find out the divergence dates, Bayesian phylogenetic analysis was conducted in BEAST v2 (Bouckaert et al., 2014) and fossil calibration was

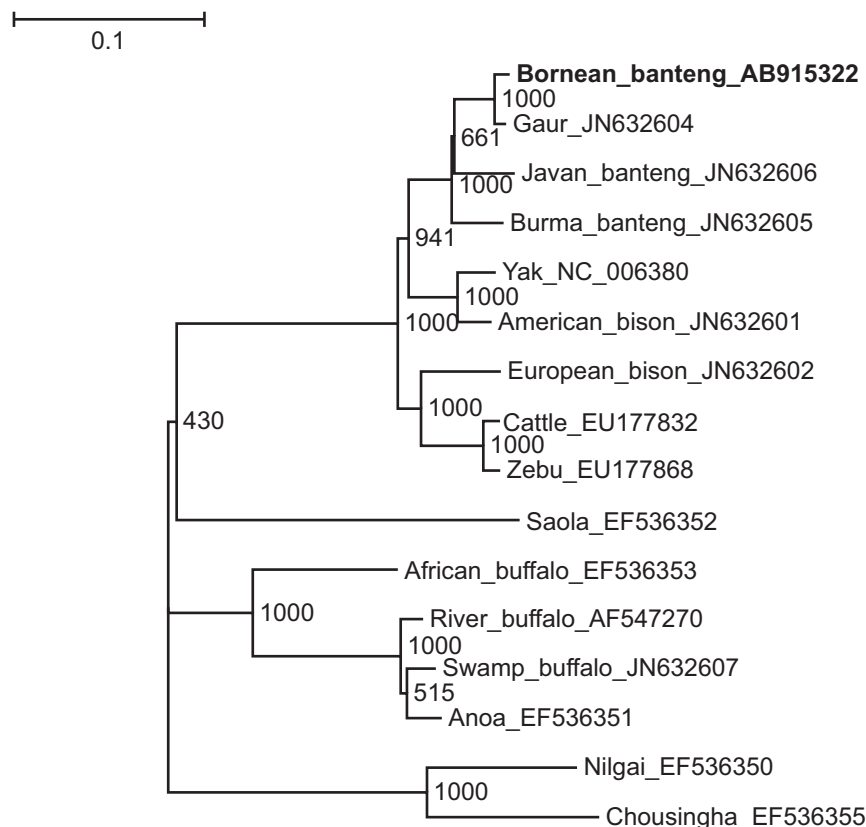
used based on the first occurrence of the Bovidae 20 ± 2 Mya (Hassanin et al., 2012).

This whole-genome draft shotgun sequence has been deposited at the DDBJ under the accession number DRA001728. The annotated sequence was deposited in GenBank under the accession number AB915322. The teeth is now catalogued and deposited in the BORNEENSIS (BORNEENSIS MAL 05550), the reference collection museum at the Institute for Tropical Biology and Conservation, Universiti Malaysia Sabah.

Whole-genome sequencing was carried out using the 100 bp paired-end library, which generated 275,795,798 reads totaling 27,579,579,800 bases of DNA. There was a total of 75,815 contigs assembled *de novo* based on the paired-end reads, which were blasted against the gaur mtDNA sequence. The BLAST result revealed that the Bornean banteng mtDNA is covered by one non-ambiguous contig, with an average sequence depth of about 61.6 times and 10,445 mapping reads.

The assembled Bornean banteng mtDNA sequence was similar to that of gaur, containing 13 protein-coding genes, 21 tRNAs, 2 rRNAs and a D-loop. This contig was consistent with the partial Malua Bornean banteng cytochrome *b* and D-loop sequences (AB703046 and AB703048). There were no internal stop codons found in the protein-coding genes. The gene arrangement was identical to that of gaur mtDNA and other members of Bovini. The phylogenetic tree showed that the three subspecies of banteng did not form sister groups, whereas the Bornean banteng and the gaur emerged as sister groups (Figure 1). The divergence date showed that the Bornean banteng and the gaur diverged from a common ancestor approximately 5.03 Mya, before the occurrence of the Sundaland. These results indicated that the Bornean banteng might be a distinct species with high conservation value.

Figure 1. Phylogenetic trees of the complete mitochondria DNA among Bovini. The numbers at the branches stand for bootstrap values, based on 1000 replicates.



## Declaration of interest

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