Short Communication

The Complete Mitochondrial Genome of Leopard Cat, *Prionailurus bengalensis euptilurus* (Carnivora:Felidae)

Feng Gao[1]* and Jinping Zhang[2]

[1]Beijing Wildlife Rescue Center, China.

[2]Jiangsu Key Laboratory for Biodiversity and Biotechnology, College of Life Sciences, Nanjing Normal University, Nanjing, 210023, China

ABSTRACT

The full-length of the mitochondrial (mt) genome of leopard cat (*Prionailurus bengalensis euptilurus*) was first determined in this study and consisted of a 17,246bp fragment , including 13 protein-coding genes, two rRNA genes, 22 tRNA genes, a control region (CR), and an origin of L-strand replication (OLR). The total base composition of the heavy strand was A, 33.05%; G, 13.52%; C,25.94%; and T, 27.49%, with a slight AT bias of 60.54%. The complete mitochondrial data of *P. b.euptilurus* may provide an important for further phylogenetic and taxonomic analyses of Genus *Prionailurus* species.

The Asian leopard cat (Felidae: *Prionailurus bengalensis* Kerr, 1792) occurs in forests from South Asia through East Asia to the Russian Far East, and from Southeast Asia to western Indonesia and the Philippines (Nowell and Jackson 1996). Leopard cat (*Prionailurus bengalensis*) is a small wild cat and widely distributed in Asia. The name of leopard cat is derived from the leopard-like spots prevalent in all subspecies, but its relationship with the leopard seems very distant. There are 12 leopard cat subspecies, which differ largely in appearance (Wilson and Mittermeier, 2009). It is widely distributed in China, living in the mountain broad-leaved forest, mixed forest, copse and so on. It is mainly active at night, and can occasionally see activities during the day. It is good at climbing. Its diet includes grass rabbits, rats, squirrels, birds, frogs, lizards, insects and a few fruits (Chen *et al.*, 2002).

* Corresponding author: radio6182000@163.com

Since 2002, it has been listed as least concern by IUCN as it is widely distributed but threatened by habitat loss and hunting in parts of its range (Sanderson *et al.*, 2008; Shuai-Tan *et al.*, 2015).

In order to better protect the leopard cat China classified the leopard cat as a National Grade II protected wild animal in 2021.

However, the molecular data of this species mainly focus on the samples collected from Japan, Korean, and some islands of Southeast Asian (Park, 2011; Tamada *et al.*, 2006), as well as molecular data for this species are mainly concentrated in samples collected from Japan, Korea, some islands in Southeast Asia, and Sichuan, China (Park, 2011; Tamada *et al.*, 2006; Shuai-Tan *et al.*, 2015), and the ocelot genome in Beijing and North China has not yet been sequenced. In this study, the complete sequence of the euptilurus was determined to obtain additional molecular information available for phylogenetic and taxonomic analyses in the future. In this study, combined with data from GenBank, we sequenced the mitotic whole genome of *P. bengalensis* from northern China and constructed its relatives to other ocelots.

Materials and methods

This sample was obtained from Cuihu Wetland Park (116°11′26.52''E,40°6′25.08''N), Haidian District, Beijing,

F. Gao

China. On the night of December 13, 2021, Blood samples are stored in the Animal Hospital of Beijing Wildlife Rescue Center. Sample number is 2021121301 and the email address of the collection director is bwrrc@126. com.the leopard cat was hit by a car in Cuihu Wetland Park and was saved by Beijing Wildlife Rescue Center. We employed polymerase chain reaction (PCR) methods to amplify the complete of the *P. b. chinensis* with the PCR primers designed by ourselves. A previous published sequence (NC_016189) of Amur leopard cat was used to design these primers for PCR-amplification and a template for gene annotation.

The genome sequence data that support the findings of this study are openly available in GenBank of NCBI at (https://www.ncbi.nlm.nih.gov; https://www.ncbi.nlm.nih. gov/) under the accession no. OR282482.

Results

The full-length of *P.b euptilurus* mitogenome consists of a 17,246 bp fragment and the gene content of the mitogenome is identical to most of other vertebrates (Table I). The base composition of mitochondrial genome, protein-coding gene and rRNA gene were calculated respectively. The creation of circos was used to map the whole mitochondrial genome, and the results were shown in (Fig. 1).
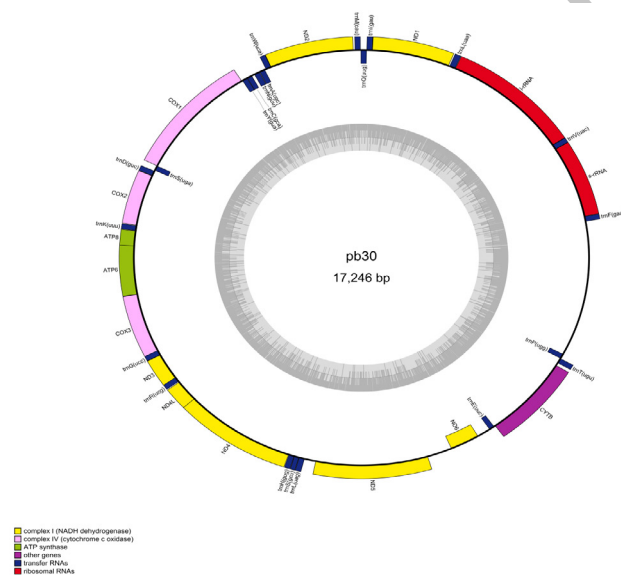

Fig. 1. Mitochondrial complete genome circle.
Note: From outside to inside are: genome structure chart, coverage of reads on genome, GC content.

It contains 13 protein-coding genes, 22 tRNAs, two rRNAs, one control region, and an origin of L-strand

Table I. Characteristics of the mitochondrial genome of *Prionailurus bengalensis euptilurus.*

Gene	Position		Length (bp)	Codon		Intergenic nucleotides[b]	Strand
	From	To		Start	Stop[a]		
tRNAPhe	1	70	70			0	H
12S rRNA	71	1030	960			0	H
tRNAVal	1028	1095	68				H
16S rRNA	1094	2666	1573				H
tRNALeu (UUR)	3148	3222	75			+2	H
nd1	2754	3701	948	ATG	TA-	0	H
tRNAIle	3701	3769	69			-3	H
tRNAGln	3767	3840	74			+1	L
tRNAMet	4322	4390	69			0	H
nd2	3929	4954	1026	ATG	T--		H
tRNATrp	5433	5500	68			14+	H
tRNAAla	5035	5103	69			+1	L
tRNAAsn	5105	5177	73			0	L
OriL	5178	5210	33			0	L
tRNACys	5691	5755	65				L
tRNATyr	5756	5342	67			+1	L
cox1	5350	6888	1539	ATG	TAA	-3	H
tRNASer (UCN)	7366	7434	69			+6	L
tRNAAsp	6961	7029	69			0	H
cox2	7030	7713	684	ATG	TAA	+3	H
tRNALys	7717	7785	69			+1	H
atp8	7787	7990	204	ATG	TAA	-43	H
atp6	7948	8628	681	ATG	TAA	-1	H
cox3	8628	9462	835	ATG	T--	0	H
tRNAGly	9412	9480	69			0	H
nd3	9478	9837	360	ATA	TA-	0	H
tRNAArg	9828	9896	69			0	H
nd4l	9903	10193	291	ATG	TAA	-7	H
nd4	10187	11562	1376	ATG	A--	0	H
tRNAHis	11565	11633	69			0	H
tRNASer (AGY)	11634	11692	59			0	H
tRNALeu (CUN)	11693	11762	70			0	H
nd5	12378	13746	1369	ATA	TAA	+4	H
nd6	13567	13932	366	ATG	TAA	0	L
tRNAGlu	14095	14163	69			+3	L
cytb	14176	15271	1096	ATG	AGA	0	H
tRNAThr	15307	15375	69			0	H
tRNAPro	15376	15442	67			0	L
Control region	15443	17246	1804				H

[a]T– – and TA– represent incomplete stop codons. [b]Numbers correspond to the nucleotides separating adjacent genes. Negative numbers indicate overlapping nucleotides.

replication (OLR). The total base composition of the heavy strand was A, 33.05%; G, 13.52%; C,25.94%; and T, 27.49%, with a slight AT bias of 60.54%. All 13 protein-coding genes share the start codon ATG, except for ND4 and ND5, which start with ATA. Twelve out of 13 protein-coding genes are encoded on the heavy strand, while ND6 is encoded on the light strand. As in other mammals, most protein-coding genes share the typical termination codon TAA (COXI, COXII, ATPase8, ATPase 6, ND4L, ND5, and ND6), while Cytb terminates with AGA. In particular, incomplete stop codons are found in ND1 (TA–) and ND2, ND3, ND4, COX3 (T—). The length of 22 tRNAs ranges from 59 to 75 bp. The two ribosomal RNAs, 12S rRNA (960 bp) and 16S rRNA (1573 bp), are located between tRNAPhe and tRNALeu(UUR) and separated by tRNAVal. Additionally, it is also important to note that the L-strand replication origin (OL) (33 bp in length) is located within the WANCY region containing five tRNA genes (tRNATrp, tRNAAla, tRNAAsn, tRNACys, and tRNATyr) as in the most vertebrates (Seutin *et al*., 1994).

The other non-coding region, the control region (1804 bp in length), is bound by tRNAPro and tRNAPhe. It is expected that the complete mitogenomic data of *P. b. euptilurus* could provide an important data set for further phylogenetic and taxonomic analyses of Genus *Prionailurus* species.

Discussion

We downloaded the sequences of another 11 ocelots from NCBI to construct the phylogenetic tree, the phylogenetic tree reveals that Prionailurus viverrinus nests in the *P. bengalensis* ontogenetic population, close to *Prionailurus bengalensis euptilurus,* its Acession ID are JN392459 and NC016189, the similarity is 48 percent (Fig. 2). It is necessary to further examine the monophyly of *P. bengalensis* and taxonomy of the species *P. viverrinus* using more samples and/or morphological and molecular data.
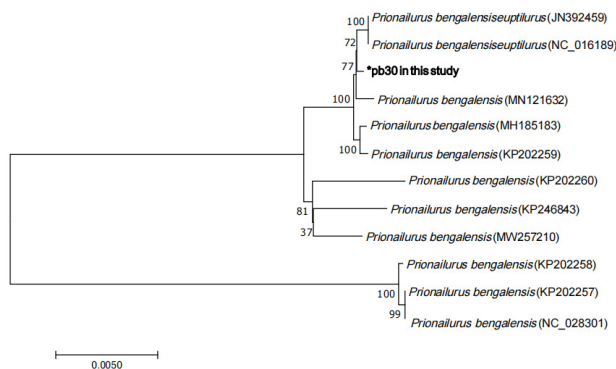

Fig. 2. Phylogenetic tree of *Prionailurus bengalensis* based on 13 protein-coding genes.

References

Andrew, T. and Smith, Y.X., 2009. *A guide to the mammals of China*. Hunan Education Press.

Chen, W., Gao, W. and Fu, B.Q., 2002. *Zoology of Beijing*. China Forestry Publishing Press, Beijing.

Nowell, K. and Jackson, P., 1996. *Wild cats: Status survey and conservation action plan*. IUCN, Switzerland.

Park, Y.C., 2011. *Mitochond. DNA,* **22**: 89–90. https://doi.org/10.3109/19401736.2011.624607

Seutin, G., Lang, B.F., Mindell, D.P. and Morais, R., 1994. *Mol. Biol. Evol*., **11**: 329–341.

Shuai-Tan, Jie-Tian, X., Fang-Dong, Z., Que-Kun, P. and Rui, P., 2015. *Mitochond. DNA early online*, **1–2**: 1940-1736.

Tamada, T., Siriaroonrat, B., Subramaniam, V., Hamachi, M., Lin, L.K., Oshida, T., Rerkamnuaychoke, W. and Masuda, R., 2006. *Zool. Sci*., **25**: 154–163. https://doi.org/10.2108/zsj.25.154

Wilson, D.E. and Mittermeier, R.A., 2009. *Handbook of the mammals of the world*. Vol 1: Carnivores. Lynx Edicions, Barcelona, Spain.

Zhang, J.J., Liang, Y.L. and Ren, Z.M., 2019. *Mitochond. DNA B*, **4**: 3072-3074. https://doi.org/10.1080/23802359.2019.1666678