

## The complete mitochondrial genome of the *Anas platyrhynchos* Linnaeus, 1758 breed Longshengcui and its phylogenetic analyses

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### ABSTRACT

Duck breed Longshengcui (*Anas platyrhynchos* Linnaeus, 1758 breed Longshengcui, LSC) is one of the famous native breed of the Guangxi Zhuang Nationality Autonomous Region in China. In this study, we report the complete mitochondrial genome of LSC. The mitogenome (GenBank accession no. MZ895120) has 16,602 bp in length and consisted of the well-known 13 protein-coding genes, 22 tRNA genes, two rRNA genes, and the control region. The phylogenetic analysis showed that LSC and Zhijiang duck have highly similar genetic relationship. These results are helpful for the conservation of genetic resources and phylogeny of this species.

### ARTICLE HISTORY

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### KEYWORDS

*Anas platyrhynchos*; duck breed Longshengcui; mitochondrial; phylogenetic analyses

Duck breed Longshengcui (*Anas platyrhynchos* Linnaeus, 1758 breed Longshengcui, LSC) is one of the famous native breeds of the Guangxi Zhuang Nationality Autonomous Region in China (Figure 1). LSC is a rare genetic resource of duck species with its high ornamental quality and tender meat. It is important to preserve the genetic resources of native species for urgent conservation of the species. A complete mitochondrial genome is one of the unique genetic characters suitable for the purpose and plays an important role in phylogenetic relationships discovery (Zhai et al. 2021). In this study, we report LSC's complete mitochondrial genome sequence.

The adult individuals of LSC were collected at its originally breeding farm in Longsheng County (25°29'N and 109°43'28"E), Guilin City, Guangxi Zhuang Nationality Autonomous Region. And the specimens were stored at –80°C in our laboratory (School of life sciences Guangxi Normal University, Guilin, <http://www.bio.gxnu.edu.cn/>, Xin Wang is the contact person 2950631569@qq.com) under the voucher number GXNU-LSC-202105066. Total genomic DNA was extracted from the thorax muscle of a single individual using the EasyPure Kit of Genomic DNA (Transgen Biotech, Beijing, China). Polymerase chain reaction (PCR) was carried out to amplify the complete mitochondrial genome with 13 pairs of primers. PCR products of the gel electrophoresis


were purified by Gel Advanced™ Gel Extraction (Rich Biotech, Taiwan, China) and sequenced by BioSune Biotech (Shanghai, China) and using an ABI 3730 automatic sequencer (Sanger sequencing). The complete mitochondrial genome sequence was assembled manually using DNASTAR v7.1



Figure 1. Appearance of Longshengcui duck. The photo was taken by the authors.

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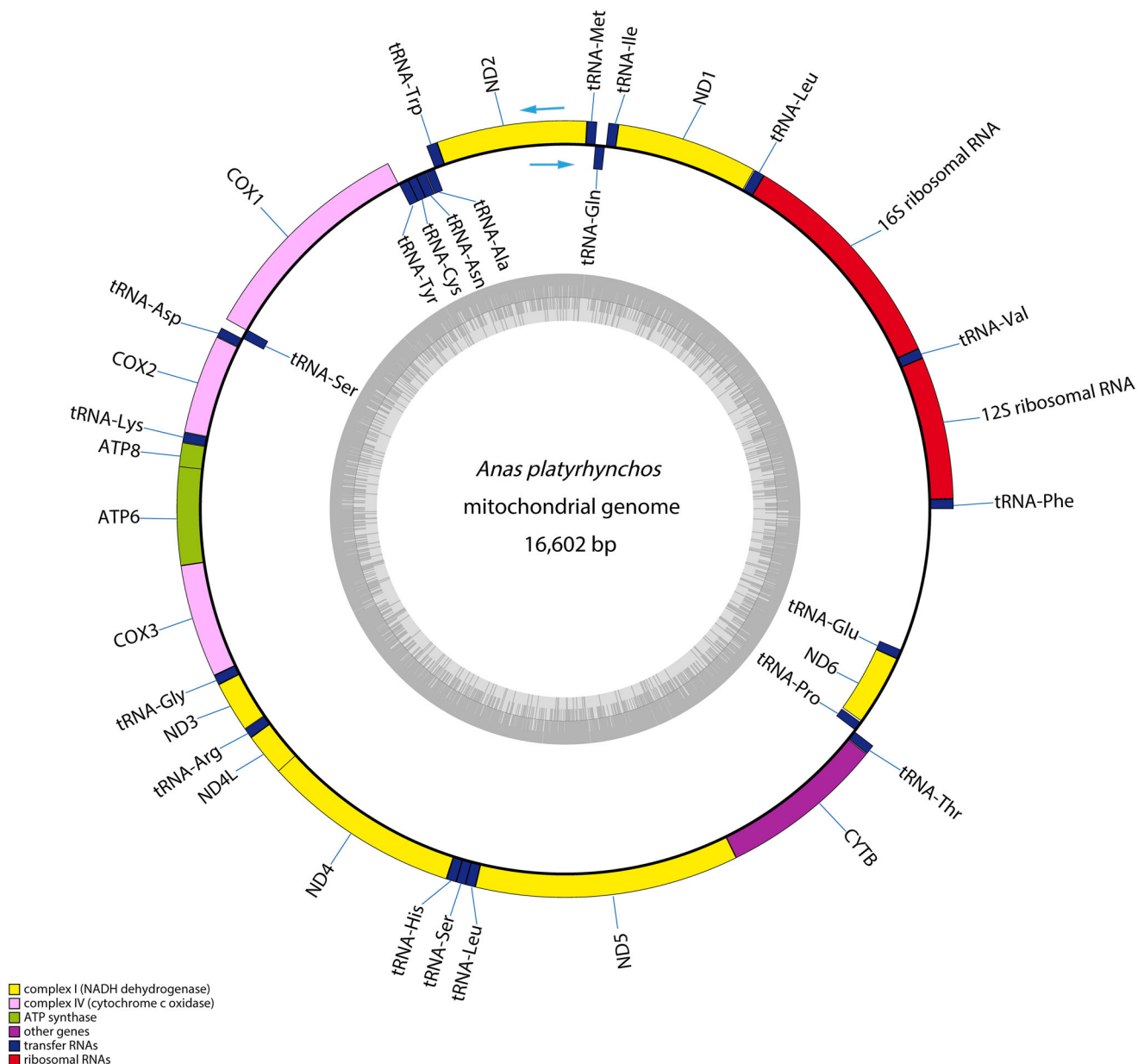
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software. The LSC duck mitogenome was annotated using DOGMA (Wyman et al. 2004).

The complete circular mitogenome of LSC presented 16,602 bp in size (GenBank accession no. MZ895120). Its mitogenome contains 13 protein-coding genes which were identified by an ORF finder analysis at NCBI (<https://www.ncbi.nlm.nih.gov/orffinder/>), and two ribosomal RNA genes were identified by sequence alignments and compared to several related species. Moreover, 22 transfer RNA genes were identified with tRNAscan-SE (Chan and Lowe 2019); and a control region (D-loop) of 1048 bp was also identified. The 12S rRNA and 16S rRNA lengths were 985 and 1602 bp. The OGDRAW version 1.3.1 was used to draw the physical map of the complete genome (Figure 2) (Greiner et al. 2019). Most of the genes were located on the heavy chain, except for ND6 and

eight tRNAs genes (tRNA<sup>Gln</sup>, tRNA<sup>Ala</sup>, tRNA<sup>Asn</sup>, tRNA<sup>Cys</sup>, tRNA<sup>Tyr</sup>, tRNA<sup>Ser</sup>, tRNA<sup>Pro</sup>, tRNA<sup>Glu</sup>), which were similar to most birds mtDNAs (Lin et al. 2016, Lin et al. 2016, Liu et al. 2021). For 13 PCGs in the LSC mtDNA, besides the COX1, COX2, ND5 initiation codon is GTG, ND6 is CTA, and the rest of the PCGs are ATG. LSC mt DNA had five types of termination codons, including AGG (ND1 and COX1), TAG (ND2), TAA (COX2, ATPase8, ATPase6, ND3, ND4L, ND5 and Cytb), CAT (ND6) and an incomplete termination codon "T-" (COX3 and ND4). LSC duck mtDNA possess TAA or TAG as their termination codon and this difference in termination codon is also frequently found in other mitogenomes (Table 1) (Jia et al. 2023).

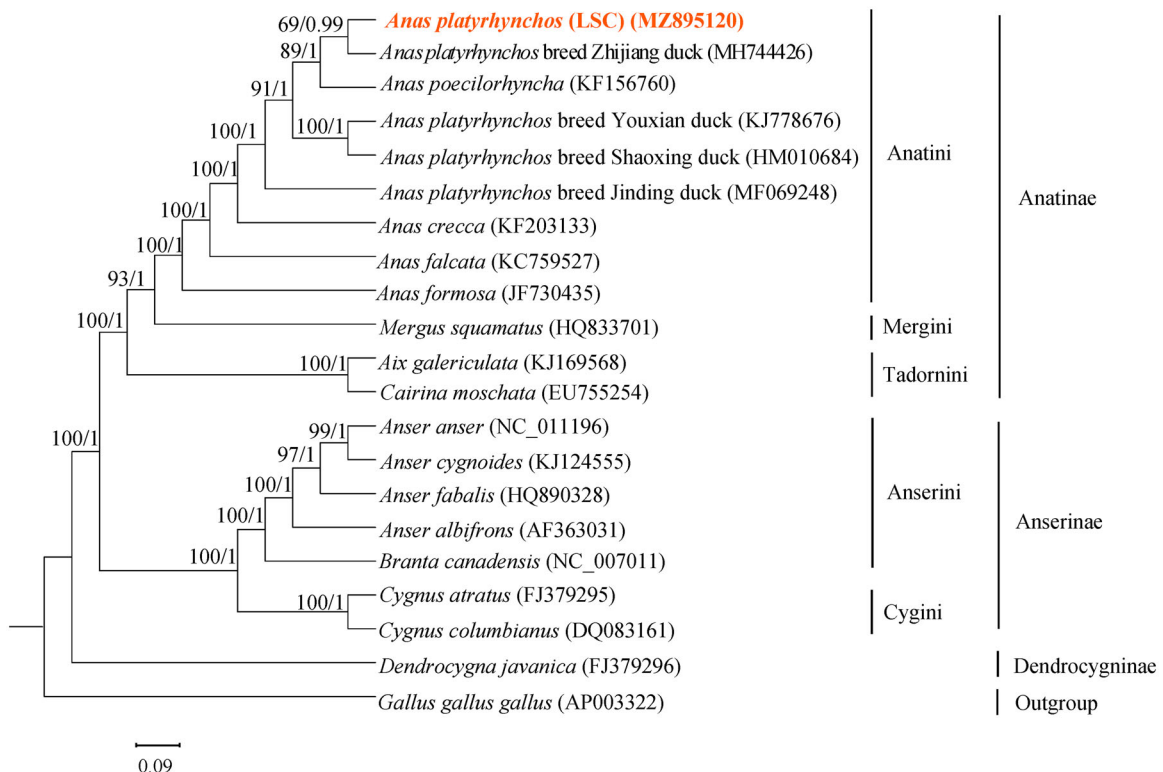
Phylogenetic analysis was performed using the complete mitochondrial DNA sequences of 20 Anseriforms and



**Figure 2.** Mitochondrial genome map of Longshengcui duck. The inner circle indicates the GC content, and the external circle indicates the genes having different colors based on their functions. The arrows represent direction of transcription, genes encoded on the heavy and light strand are shown outside and inside the circle, respectively.

**Table 1.** Organization of the mitochondrial genome of Longshengcui duck.

	Gene	Position		Size	Codon		Anti-codon	Strand	Space /overlap
		Start	End		Start	Stop			
1	tRNA-Phe	1	70	70			GAA	H	
2	12S	71	1055	985				H	
3	tRNA-Val	1056	1126	71			TAC	H	
4	16S	1127	2728	1602				H	
5	tRNA-Leu	2729	2802	74			TAA	H	
6	nad1	2807	3784	978	ATG	AGG		H	4
7	tRNA-Ile	3783	3854	72			GAT	H	-2
8	tRNA-Gln	3862	3932	71			TTG	L	7
9	tRNA-Met	3932	4000	69			CAT	H	-1
10	nad2	4001	5041	1041	ATG	TAG		H	
11	tRNA-Trp	5040	5115	76			TCA	H	-2
12	tRNA-Ala	5119	5187	69			TGC	L	3
13	tRNA-Asn	5190	5262	73			GTT	L	2
14	tRNA-Cys	5263	5328	66			GCA	L	
15	tRNA-Tyr	5328	5399	72			GTA	L	-1
16	cox1	5401	6951	1551	GTG	AGG		H	1
17	tRNA-Ser	6943	7015	73			TGA	L	-9
18	tRNA-Asp	7018	7086	69			GTC	H	2
19	cox2	7088	7774	687	GTG	TAA		H	1
20	tRNA-Lys	7776	7844	69			TTT	H	1
21	atp8	7846	8013	168	ATG	TAA		H	1
22	atp6	8004	8687	684	ATG	TAA		H	-10
23	cox3	8687	9470	784	ATG	T		H	-1
24	tRNA-Gly	9471	9539	69			TCC	H	
25	nad3	9540	9891	351	ATG	TAA		H	
26	tRNA-Arg	9893	9962	70			TCG	H	1
27	nad4L	9963	10259	297	ATG	TAA		H	
28	nad4	10253	11630	1378	ATG	T		H	-7
29	tRNA-His	11631	11699	69			GTG	H	
30	tRNA-Ser	11700	11765	66			GCT	H	
31	tRNA-Leu	11765	11834	70			TAG	H	-1
32	nad5	11835	13658	1824	GTG	TAA		H	
33	cytb	13658	14800	1143	ATG	TAA		H	-1
34	tRNA-Thr	14803	14871	69			TGT	H	2
35	tRNA-Pro	14882	14951	70			TGG	L	10
36	nad6	14962	15483	522	CTA	CAT		L	10
37	tRNA-Glu	15484	15554	71			TTC	L	
38	D-loop	15555	16602	1048					

**Figure 3.** Phylogenetic analysis based on complete mitochondrial genome sequences. An ML/BI tree was built based on the phylogenetic analysis of 20 Anseriform species' complete mitochondrial genomes. The mitochondrial genome sequences of the Anseriform species were obtained from the GenBank databases (accession numbers have marked on the figure). Abbreviation of species indicates: LSC, Longshengcui duck.

outgroup mitogenomes. Each of the sequences datasets was aligned by ClustalX (Thompson et al. 1997). Phylogenetic tree was inferred by Maximum Likelihood (ML) and Bayesian Inference (BI) models and constructed *via* IQtree (Nguyen et al. 2015) and MrBayes 3.2.7 (Huelsenbeck and Ronquist 2001), respectively. The ML/BI tree showed that the analyzed species are divided into three major clades: Anatinae, Anserinae and Dendrocygninae (Figure 3). Anatinae is the first lineage, which is sister to the second group, Anserinae; Dendrocygninae forms the third group and is sister to Anatinae and Anserinae. The outgroup, *Gallus gallus gallus*, is located at the base of the tree. The first lineage Anatinae includes tribes Anatini (*Anas platyrhynchos* breed LSC, *Anas platyrhynchos* breed Zhijiang duck, *Anas poecilorhyncha*, *Anas platyrhynchos* breed Youxian duck, *Anas platyrhynchos* breed Shaoxing duck, *Anas platyrhynchos* breed Jinding duck, *Anas crecca*, *Anas falcata*, *Anas formosa*), Mergini (*Mergus squamatus*), Tadornini (*Aix galericulata*, *Cairina moschata*). The second lineage includes tribes Anserini (*Anser anser*, *Anser cygnoides*, *Anser fabalis*, *Anser albifrons* and *Branta canadensis*) and Cygnini (*Cygnus atratus* and *Cygnus columbianus*), forming the subfamily Anserinae. The third lineage Dendrocygninae groups together with Anatinae and Anserinae. Meanwhile, we also found that LSC and *Anas platyrhynchos* breed Zhijiang duck having a highly similar genetic composition.

### Ethics statement

All the experiment procedures were approved by the Animal Care Committee of School of life sciences Guangxi Normal University, Guilin, China.

### Authors' contributions

The research was designed and conducted by QL, CJL, SYB. The animal experiment was conducted by JMZ, XW and QXW. The detection and analysis works were conducted by JMZ, XW, QXW, LPL, FMC and YZW. The manuscript draft was prepared by QL, CJL, SYB, JMZ, XW and QXW. All authors participated in the discussion and editing of the manuscript.

### Disclosure statement

No potential conflict of interest was reported by the authors.

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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/>) under the accession no. MZ895120.

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