METHODS AND RESOURCES ARTICLE



The complete mitochondrial genome of the hybrid of *Acipenser* dabryanus $(\bigcirc) \times A$. schrenckii (\bigcirc) and its phylogeny

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Abstract The complete mitochondrial genome sequence of the hybrid of Acipenser dabryanus $(\bigcirc) \times A$. schrenckii (\bigcirc) was first determined by using the next-generation sequencing in this study. The circular mitochondrial genome was 16,439 bp in length, which contained 13 protein-coding genes, 2 ribosomal RNA genes, 22 transfer RNA genes, one displacement loop locus and an origin of replication on the light-strand, showed a typical vertebrate pattern. All of the genes were encoded on the heavy strain except for ND6 and eight tRNA genes. The overall nucleotide composition was 30.19% A, 23.74% T, 29.65% C, 16.42% G, with 53.93% AT, respectively. Compared with the complete mitogenome of the parents, the results indicated that the mitochondria of hybrid sturgeon could be consistent with a maternal inheritance. Phylogenetic analyses using concatenated nucleotide sequences of the 13 protein-coding genes with two different methods (maximum likelihood and neighbor-joining analysis) both highly supported that A. dabryanus $(\bigcirc) \times A$. schrenckii (\mathcal{O}) showed a close relationship with A. dabryanus and A. sinensis. These data provide useful information for a better understanding of the mitogenomic diversities and evolution in fish as well as novel genetic markers for studying population genetics and species identification.

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² School of Life Sciences, Southwest University, Chongqing 400700, China **Keywords** Acipenser dabryanus · A. schrenckii · Hybrid sturgeon · Mitochondrial genome · Phylogeny

Acipenser dabryanus and A. schrenckii are two species of Acipenseriformes, which belong to the genus of Acipenser. A. dabryanus, a critically endangered and endemic species, is also called "the Yangtze River sturgeon", which has been distributed in the upper and middle sections of the Yangtze River (Liu et al. 2017a). The natural production of A. dabryanus have declined sharply in the past decades due to dam construction, overfishing, pollution, etc., in the Yangtze River (Liu et al. 2017a). A. schrenckii of the Amur River basin and its tributaries is one of the most important economically valuable fish in Russia and in China (Liu et al. 2017b; Li et al. 2016). However, growth depression, late sexual maturity, and disease susceptibility restrict the culture of population of A. schrenckii (Dong et al. 2014; Wei et al. 2011). Because of heterosis, hybrid sturgeon has been an important species in sturgeon aquaculture in China (Gao et al. 2017; Shen et al. 2014).

In the present study, we first determined the complete mitogenome sequence of the hybrid sturgeon $(A \times A)$. The specimen of $A \times A$ used in this study were obtained by artificial hybridization from the Conservation and Utilization of Fishes resources in the Upper Reaches of the Yangtze River Key Laboratory of Sichuan Province, Neijiang Normal University. A 30–40 mg fin clip was collected and preserved in 95% ethanol at 4 °C. Total genomic DNA was extracted from these caudal fins by a Tissue DNA Kit (OMEGA E.Z.N.A.) following the manufacturer's protocol. Subsequently, the genomic DNA was sequenced using the next-generation sequencing, and then the mitogenome was assembled using *A. dabryanus* as reference.

The complete mitochondrial genome of the A × A was determined to be 16,439 bp in length, including 13 protein-coding genes, 2 ribosomal RNA (rRNA) genes, 22 transfer RNA (tRNA) genes, a displacement loop (D-loop) locus and an origin of replication on the light-strand (OL) (Table 1; Fig. 1). The overall nucleotide composition was 30.19% A, 23.74% T, 29.65% C, 16.42% G, with 53.93%AT, respectively. Most of coding genes were encoded on the heavy strain (H-strand) except for *ND6* and eight tRNA genes, which were encoded on the light strain. The common initiation codon was ATG in the 13 protein-coding genes, except *COX1*, which used GTG. 9 protein-coding genes stop with the complete termination codon TAG (*ND1*, *COXI*, *ND3*, *ND6*) or TAA (*ATP8*, *ATP6*, *COXIII*, *ND4L*, *ND5*), while the rest ends with an incomplete termination codon T(*ND2*, *COXII*, *ND4*, *Cyt b*). Moreover, all the 22 tRNA genes, length ranging from 67 to 75 bp, and the *tRNA^{Cys}* was the shortest while *tRNA^{Leu}* was the longest. The *12S rRNA* and *16S rRNA* were 960 and 1701 bp, respectively. Additionally, an 32 bp origin of replication on the light-strand

Table 1 Completemitochondrial genomecharacteristics of the hybrid ofAcipenser dabryanus $(\mathcal{Q}) \times A$.	Gene	Position		Size		Codon		Strandy	Intergenic
		From	То	Nucleotide (bp)	Amino acid	Start	Stop*		nucleotide (bp)
schrenckii (ð)	tRNA ^{Phe}	1	68	68				Н	0
	12S rRNA	69	1028	960				Н	0
	tRNA ^{Val}	1029	1099	71				Н	0
	16S rRNA	1100	2800	1701				Н	0
	tRNA ^{Leu}	2801	2875	75				Н	0
	ND1	2876	3850	975	324	ATG	TAG	Н	9
	tRNA ^{Ile}	3860	3930	71				Н	-1
	tRNA ^{Gln}	3930	4000	71				L	-1
	tRNA ^{Met}	4000	4069	70				Н	0
	ND2	4070	5114	1045	348	ATG	T	Н	0
	tRNA ^{Trp}	5115	5187	73				Н	1
	tRNA ^{Ala}	5189	5257	69				L	1
	tRNA ^{Asn}	5259	5331	73				L	32
	tRNA ^{Cys}	5364	5430	67				L	0
	tRNA ^{Tyr}	5431	5501	71				L	1
	COXI	5503	7056	1554	517	GTG	TAG	Н	7
	tRNA ^{Ser}	7064	7134	71				L	8
	tRNA ^{Asp}	7143	7214	72				Н	14
	COXII	7229	7919	691	230	ATG	T	Н	0
	tRNA ^{Lys}	7920	7993	74				Н	1
	ATP8	7995	8162	168	55	ATG	TAA	Н	-10
	ATP6	8153	8836	684	227	ATG	TAA	Η	-1
	COXIII	8836	9621	786	261	ATG	TAA	Н	-1
	tRNA ^{Gly}	9621	9693	73				Η	0
	ND3	9694	10,044	351	116	ATG	TAG	Н	-2
	tRNA ^{Arg}	10,043	10,112	70				Н	0
	ND4L	10,113	10,409	297	98	ATG	TAA	Н	-7
	ND4	10,403	11,783	1381	460	ATG	T	Н	0
	tRNA ^{His}	11,784	11,852	69				Н	0
	tRNA ^{Ser2}	11,853	11,920	68				Н	0
	tRNA ^{Leu2}	11,921	11,993	73				Н	0
	ND5	11,994	13,835	1842	613	ATG	TAA	Н	-4
	ND6	13,832	14,353	522	173	ATG	TAG	L	0
	tRNA ^{Glu}	14,354	14,423	70				L	2
	Cyt b	14,426	15,566	1141	380	ATG	T	Н	0
	tRNA ^{Thr}	15,567	15,639	73				Н	3
	tRNA ^{Pro}	15,643	15,712	70				L	0
	D-loop	15,713	16,439	727				Н	

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Fig. 1 The gene map of the complete mitochondrial genome of the hybrid of *Acipenser dab-ryanus* (\mathcal{Q})×*A. schrenckii* (\mathcal{S}). Arrows indicate the direction of transcription. tRNA genes are named using single-letter amino acid abbreviations. Genes encoded on the H-strand and L-strand are shown outside and inside the circular map of the mitogenome, respectively. The innermost circle indicates the GC content graph



(OL) was found in the A × A mitogenome between $tRNA^{Asn}$ and $tRNA^{Cys}$. Furthermore, the displacement loop (D-loop) was located between $tRNA^{Pro}$ and $tRNA^{Phe}$, with 727 bp in length (Fig. 1; Table 1). The genomic sequence has been deposited in GenBank with an accession number MF958972.

To confirm the phylogenetic relationships between A × A and other Acipenserinae subfamily fishes, phylogenetic analyses were performed on the concatenated dataset of 13 PCGs at nucleotide level with neighbor-joining (NJ) and maximum likelihood (ML) methods (Zou et al. 2017). The tree topologies produced by NJ and ML analyses were equivalent (Fig. 2). The 15 taxa all belong to Acipenseridae, Acipenserinae except Culter erythropterus, which belongs to Cyprinidae, Culterinae. So C. erythropterus was used as an outgroup. The other 14 species from 2 genus were divided into two clades. Species of Huso huso, A. stellatus, A. ruthenus, A. baerii and, A. gueldenstaedtii were clustered into clade B, and the rest of species were clustered into clade A. The $A \times A$ and its female parent (A. dabryanus) aggregated into a clade at the first. The situation also showed in *H. dauricus* × *A. schrenckii* (Liu et al. 2017b). This phenomenon is inseparable from the maternal inheritance characteristics of the mitochondria. The A \times A has a closer relationship with A. sinensis than its male parent (A. schrenckii), due to its female (A. dabryanus) has a close relationship with A. sinensis (Li et al. 2016). Although H. dauricus and H. huso both pertain to Huso but located in different branches, which is slightly different from conventional morphology-based classification of Acipenserinae species (Li et al. 2016; Birstein et al. 1999). The difference may be owing to the limited availability of mitogenomes from Acipenserinae species (Wei et al. 2011).

In summary, the present study first reported the complete mitochondrial genome of the hybrid sturgeon of $A \times A$, The circular molecule was 16,439 bp long and showed a typical vertebrate mitogenome structure. Phylogenetic analyses showed that mitochondrial genomes of $A \times A$ remain maternally inherited, which was consistent with the mitochondrial inheritance mechanism. The complete mitochondrial genome sequence of the $A \times A$ provided an important dataset for a better understanding of the mitogenomic diversities and evolution in fish as well as novel genetic markers for studying population genetics and species identification.



Fig. 2 The phylogenetic analyses investigated using neighbor-joining (NJ) and maximum likelihood (ML) analysis indicated evolutionary relationships among 15 taxa based on nucleotide sequences of 13 concatenated protein-coding genes. NJ posterior probability (blue

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number) and ML bootstrap support values (black number) are shown on the nodes. *Culter erythropterus* (GenBank: NC 024749) was used as an outgroup. (Color figure online)

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