

The complete mitochondrial genome of the Asian stinging catfish, *Heteropneustes fossilis* (Siluriformes, Heteropneustidae) and its comparison with other related fish species

Bijay Kumar Behera^a, Vishwamitra Singh Baisvar^a, Kavita Kumari^a, Ajaya Kumar Rout^a, Sudip Pakrashi^a, Prasenjet Paria^a, Abhishek Das^a, A. R. Rao^b and Anil Rai^b

^aFish Biotechnology Laboratory, ICAR-Central Inland Fisheries Research Institute, Barrackpore, Kolkata, West Bengal, India; ^bICAR-Indian Agricultural Statistics Research Institute, New Delhi, India

ABSTRACT

The complete mitogenome of *Heteropneustes fossilis* is described using Ion Torrent (PGM sequencer), which showed it was 16,489 bp in size comprising 13 mRNAs, 22 tRNAs, 2 rRNA genes, and 858 bp as D-Loop control region, along with gene order and organization, being similar to most of the other related Siluriformes fish mitogenome of NCBI databases. The 20 RNAs were packed into a typical cloverleaf structure. The mitogenome in the present study has 99% similarity to the complete mitogenome sequence of *H. fossilis* mitogenome reported earlier and also would be helpful in understanding the population genetics, phylogenetics, and evolution of catfishes.

ARTICLE HISTORY

Received 2 July 2016

Revised 22 July 2016

Accepted 29 July 2016

KEYWORDS

Next-generation sequencing; mitogenome; *H. fossilis*; phylogenetic tree

The complete mitochondrial genome sequences of Indian Catfish, *Heteropneustes fossilis* under Heteropneustidae family, have been identified as an important fish species due to its medicinal value (Froese & Pauly 2011). This species come under Red List status as Least Concern (LC) category (IUCN 2015). Here, we presented the complete mitochondrial genome of *H. fossilis* (GenBank Accession Number KT001154.1) collected from river Ganga at Barrackpore, Kolkata, West Bengal (22.76° N; 88.35° E) during the period of March 2014 and the total genomic DNA was extracted from 40 mg of muscle tissues using Mitochondrial DNA Isolation Kit ABCAM (Life technologies, Abcam, Carlsbad, CA) following the manufacturer's instructions with slight modification. The total G-DNA was prepared in paired-end libraries, tagged, and subjected to PGM sequencer (Ion Torrent) using the Ion PGM 200 sequencing kit v2 (Life technologies, Carlsbad, CA) according to the manufacturer's instructions. The Ion Torrent was used to parse bar-coded reads and to generate run metrics, including chip loading efficiency and total read counts and quality, in order to confirm the sequence of specific regions with low coverage. The 858bp putative control region was amplified using the primers F5'-GCCTAAGAGCATCGGTCTTGTAA-3' and R 5'-GTCAGGACCATGCC TTTGTG-3' as illustrated by Sivasundar et al. (2001). The complete assembled mitochondrial genome was annotated by comparing with recently published complete mitogenome of *H. fossilis* (Nakatani et al. 2011) along with complete mitogenomes of related catfish (Zhao et al. 2013).

The 16,489 bp, complete mitogenome of Asian stinging catfish comprised 13 protein-coding genes, 22 tRNA genes,

and 2 rRNA genes and D-loop. The DNA organization of complete mitogenome of *H. fossilis* is in accordance with other related catfish species present at NCBI database. The major number of genes was encoded on the H-strand except total nine genes in which eight tRNAs (tRNA^{Gln}, tRNA^{Ala}, tRNA^{Asn}, tRNA^{Cys}, tRNA^{Tyr}, tRNA^{Ser}, tRNA^{Glu}, and tRNA^{Pro}) and ND6 gene (519bp) were encoded on L-strand. The remaining tRNAs were encoded on H-strand of mitochondrial genome. The overall base composition of *H. fossilis* mitogenome was A (32.17%), T (26.07%), C (26.87%), and G (14.88%) and was A + T (58.24%) rich similar to the other related mitogenomes. The study showed the encoding of all the mRNAs, starting with ATG codon except COI that used GTG. All the 20 tRNAs of *H. fossilis* could fold into a typical cloverleaf structure and tRNA^{Ser} is closely packed in stem-loop structure whereas the acceptor arm of tRNA^{Gly} was formed a hair loop-like structure, identified by tRNAscan SE 1.21 (Lowe & Eddy 1997). The lengths of tRNAs varied from 66 bp (tRNA^{Cys}) to 75 bp (tRNA^{Leu}) and genes of protein-coding region varied from 955 bp (12S rRNA) to 1827 bp (ND5) in size.

The evolutionary history was derived using the Minimum Evolution method (Rzhetsky & Nei 1992) and the optimize tree with sum of total branch length (1.23004838) and the replicate trees in percentage, where associated Taxa clusters with bootstrap value (1000 replicates) were present above the branches (Felsenstein 1985). The phylogenetic tree was drawn by MEGA 5.05 (McAllister Ave, USA). (Tamura et al. 2011), using 18 related fish mitogenome from NCBI database.

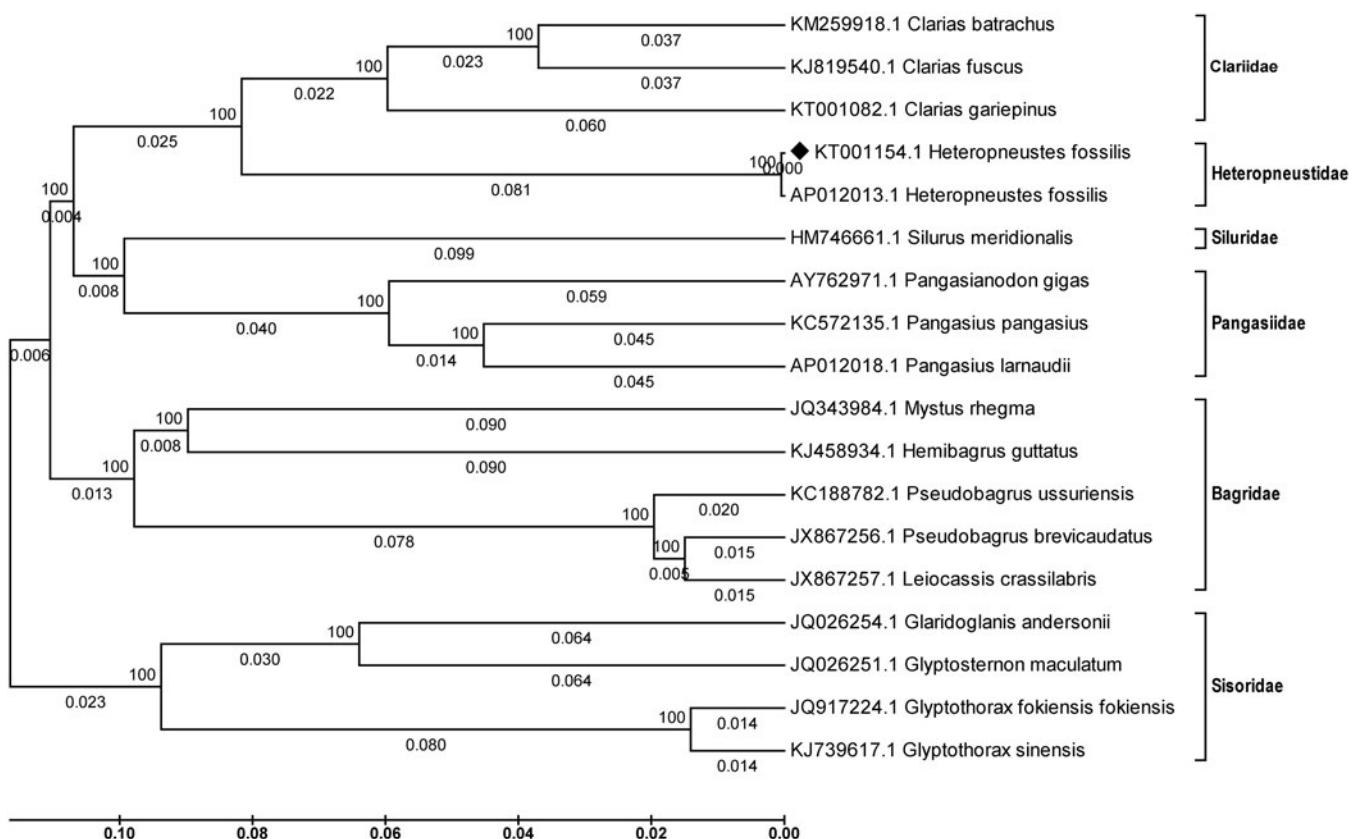


Figure 1. Minimum evolutionary Phylogenetic tree of Siluriformes by taking 18 related fish mitogenome sequences(all parameters were used as default with gap opening penalty 15, gap extension penalty 6.66, and multiple alignment parameters set as gap opening penalty of 15, gap extension 6.66, DNA weight matrix IUB and transition weight 0.5 for alignment. Minimum evolutionary Phylogenetic tree were bootstrap constructed by using complete deletion of gaps with implementations of 3000 value due to computational power).

The *H. fossilis* is very close to Clariidae and other related Siluriformes fish species (Figure 1).

Acknowledgements

The authors are thankful to the Director ICAR-Central Inland Fisheries Research Institute for providing good facilities and Mr. A. K. Jana for fish sample collection and assists Technically.

Disclosure statement

The authors report no conflicts of interest.

Funding

Indian Agricultural Research Institute, 10.13039/501100005384

References

Felsenstein J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution*. 39:783–791.

- Froese R, Pauly D, editors. [Internet] 2011. *Heteropneustes fossilis*. FishBase; [cited 2014 Dec 8]. <http://www.fishbase.org>

IUCN [Internet]. 2015. IUCN Red list of threatened species (ver. 2015.3); [cited 2015 Oct]. Available from: <http://www.iucnredlist.org>

Lowe TM, Eddy SR. 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. *Nucleic Acids Res.* 25:955–964.

Nakatani M, Miya M, Mabuchi K, Saitoh K, Nishida M. 2011. Evolutionary history of Otophysi (Teleostei), a major clade of the modern freshwater fishes: Pangaeian origin and mesozoic radiation. *BMC Evol Biol.* 11:177.

Rzhetsky A, Nei M. 1992. A simple method for estimating and testing minimum evolution trees. *Mol Biol Evol.* 9:945–967.

Sivasundar A, Bermingham E, Ortí G. 2001. Population structure and biogeography of migratory freshwater fishes (Prochilodus: Characiformes) in major South American rivers. *Mol Ecol.* 10:407–417.

Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S. 2011. MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. *Mol Biol Evol.* 28:2731–2739.

Zhao H, Kong X, Zhou C. 2013. The mitogenome of *Pangasiussuttkii* (Teleostei, Siluriformes: Pangasiidae). Mitochondrial DNA. 25:342–344.