

A Study on Phylogenetic Analysis of Fishes in the Sone River of Sidhi District, Madhya Pradesh (India)

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ABSTRACT

The Sone River is a significant river in India, located primarily in the states of Madhya Pradesh, Uttar Pradesh, and Bihar. It is one of the major tributaries of the Ganges River and plays a crucial role in the region's ecology, economy, and cultural heritage. In the present study 1000 individuals of fish specimens were collected from four different geographical locations of Sone River of Sidhi district region. Morphologically 19 species were identified and Phylogenetic trees were constructed based on CO1 gene of mitochondrial DNA using Maximum Parimony approaches using MEGA 11 software. The Phylogenetic tree revealed that species clustered differently under diverse nodes. It revealed that fish species clustered together because they were in the same order and family. These results showed that fish species share identical genera but with diverse genetic variations due to diverse habitats involving a common ancestor.

KEYWORDS: *Phylogenetic analyses, fish biodiversity, Sone River, Sidhi district, Madhya Pradesh*

INTRODUCTION

The Sone River basin is known for its rich biodiversity. The river and its tributaries support diverse ecosystems, including wetlands, marshes, and floodplains, which provide habitats for a wide range of flora and fauna (Bunkar and Tiwari, 2017). The river is home to numerous fish species, including the Common carp, labeo species and Mahseer, as well as other aquatic organisms (Sahu and Tiwari, 2023; Abhishek, 2020). With their diverse morphology and coloration, fish are among the most beautiful creations on earth (Sheikh and Goswami, 2014).

India is one of the 17 mega biodiversity hotspot and occupies the ninth position on account of freshwater mega biodiversity (Sharma et al., 2021; Shinde et al., 2009). A better understanding of the taxonomy of freshwater fish species has become crucial today because of the serious conservation concerns as well as the importance of fish as a major source of protein diet available to society (Lakra et al., 2016; Afridi et al., 2019).

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Recently, molecular techniques are being used as a reliable and precise approach for systematics (Ward et al., 2009). Due to the advancement in computational techniques, DNA sequencing becomes the major source to understand genetics and evolutionary relationships (Hajibabaei et al., 2006). For the discrimination between closely resembled species across dissimilar animal phyla mitochondrial Cytochrome c oxidase subunit I is best for molecular analysis of marine and freshwater fishes (Hebert et al., 2003). COI is a bio-identical marker because it has good discrimination power for almost all animal phyla due to the following important characteristics: it is easy to isolate, high copy number (Kamran, 2020; Cywinska et al., 2006)

Material and Methods

Study site and sample

The present study was focused to assess fish diversity in different localities of Sone River of Sidhi district.

Sone river originates near Amarkantak hills of Madhya Pradesh and flows for approximately 780 kilometers before joining the Ganges. Total 1000 individuals of fish specimens were collected and identified using morphometric or geometric analyses.

DNA extraction and amplification

Total genomic DNA was extracted from fish muscles using standard phenol chloroform isoamyl (24:23:1) method will be followed (Sambrook and Russel, 2001). Fish Species will be further analysed by PCR

Result and Discussion

In the present the freshwater fish biodiversity was estimated in Sone River of Sidhi district, in which 19 fish species belong to 9 families including Cyprinidae (66.66%), Barbinae (33.33%), Bagridae (22.2%), Siluridae (22.2%), Gobiidae (22.2%), Channidae (11.1%), Pangasiidae (11.1%), Mastacembelidae (11.1%), Clupeidae (11.1%), were identified (Table 1).

Table 1: List of fish species obtained from Sone river of Sidhi district, Madhya Pradesh

Species	Family/Subfamily	Order
<i>Catla catla</i>	Cyprinidae	Cypriniformes
<i>Cyprinus carpio</i>		
<i>Labeo rohita</i>		
<i>Labeo bata</i>		
<i>Garra mullya</i>		
<i>Tor</i>		
<i>Puntius sophore</i>	Barbinae	
<i>Esomus danricus</i>		
<i>Salmophasia</i>		
<i>Rita rita</i>	Bagridae	Siluriformes
<i>Mystus tengara</i>		
<i>Pangasius</i>	Pangasiidae	
<i>Wallago attu</i>	Siluridae	
<i>Ompok</i>		
<i>Channa striatus</i>	Channidae	Perciformes
<i>Glossogobius giuris</i>	Gobiidae	
<i>Macrognathus</i>	Mastacembelidae	Synbranchiformes
<i>Gudusia chapra</i>	Clupeidae	Clupeiformes

The Phylogenetic tree based on nucleotide sequence of CO1 genes revealed that species clustered differently under diverse nodes. The pairwise genetic distance among 19 species from 5 orders i.e., Cypriniformes, Siluriformes, Perciformes, Synbranchiformes, and Clupeiformes showed less divergence between a minimum of 0.000% and a maximum of 0.14% (figure 1). The nodes in K2P distance-based NJ trees were supported by high bootstrap values ranges from 50– 100% (Karim et al, 2018).

The Percentage base composition of sequenced samples was calculated and overall AT content was found higher in all sequences as compared to GC content (Figure 2). Observations showed that fish species were clustered together because they were in the same order and family. Similar results were obtained by Sajjad et al., 2023, a higher AT content (53.7%) and low GC content (46.2%). The alignment of cytochrome gene sequences indicated that all five species belong to the same subfamily based on the shared conserved region (Barat et al 2012).

and DNA sequencing analysis with help of universal primers for COI gene.

Phylogenetic analyses

The short mitochondria DNA gene sequence of COI was amplified, sequenced, and analyzed by using different bioinformatics tools. Phylogenetic tree was constructed by MEGA11 using the ClustalW alignment tool using Maximum Likelihood (ML) method with 1000 replicates of bootstrapping.

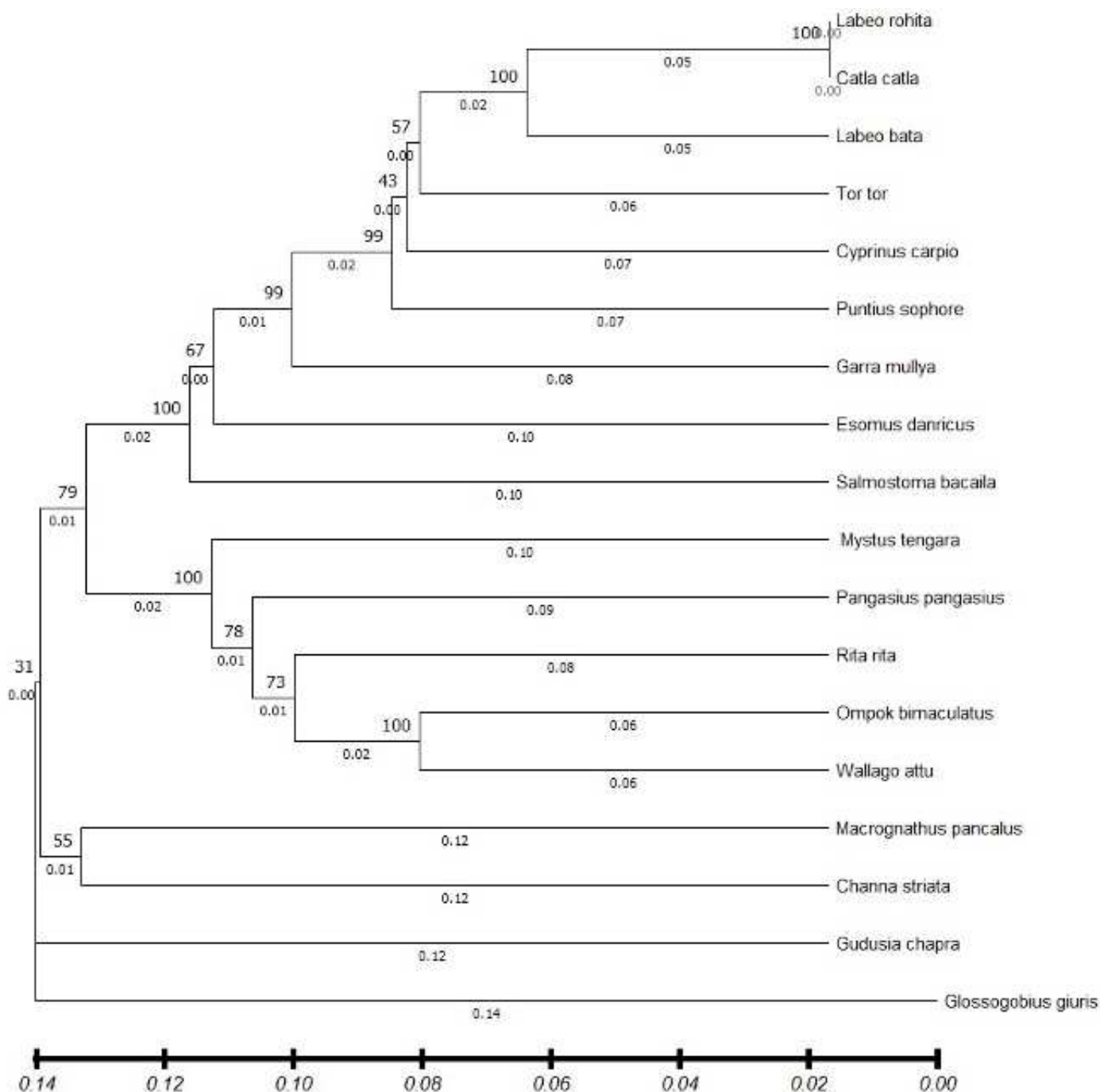


Figure 1: Phylogenetic analysis of different fish species based on the mitochondrial COI gene

The mitochondrial cytochrome gene has been widely used to study genetic variation (McVeigh and Davidson, 1991), phylogenetic relationships (Bajpai and Tewari, 2010; Kumar *et al.*, 2011), biogeographical patterns (Gilles *et al.*, 2001; Durand *et al.*, 2002) and taxonomy (Xiao *et al.*, 2001) in many fishes and higher invertebrates. A recent study demonstrated that species-specific molecular tags derived from COI genes could be used to identify species at the species level in an efficient manner (Lakra *et al.*, 2009; Ward *et al.*, 2005).

<i>Gudusia chapra</i>	GGCAATCACGCTCTCTACTACCAGCTGTACTTACTTGTATGGTCTTGACCGAATGGGAACCGATTAAGCCT
<i>Macragnathus</i>	GGCAATCACATATTTCTCGACCATCAGCATGCACCTATTAGATCGTGTCTTACCGGAATGGGAAGCCCTAAG
<i>Glossogobius</i>	GATATTAATTAATCTCTCAACCACCTAGCACGTACCATTAGATCGCGCCTGCCGGAATGGGAATGCTTTAAG
<i>Channa striatus</i>	GGCAATCACACATTTCTCGACTATCAGCATGCACCTATCCGATCGTGCCTACCGGAATGGGAAGCCCTAAG
<i>Ompok</i>	GACAATCACACATCTCTCAACCACCAGCATGCACCTACCAGATTGTGCCTACCGGAATTGGTAAGCCCTCAG
<i>Wallago attu</i>	GACAATCGCACATCTCTCAACCACCAGCATGCACCTACCAGATTGTGCCTTACCGGAATTGGTAAGCCCTCAG
<i>Pangasius</i>	CACGATCACGCATTTCTCAACTACTAGCATGTACCATCAGGTTGTGCCTACCGGAATTGGCAAGCCCTTAG
<i>Rita rita</i>	GACAATCACGCATCTCTCAACTACCAGCATGCACCCATCAGATTGTGCTTGTGGGGTTGGTAAGCCCTCAG
<i>Mystus tengara</i>	CACGATCACACATGTCTCAACTACCAGCATGCACCTACCAGATCGTGCCTACCGGAATTGGCAAGCCCTGAG
<i>Salmophasia</i>	GGCAATCACATATTTCTCAACAACCTAGCATGCACCCATTAGATTGTGCCTACCGGAGTGGGCATGCTCTGAG
<i>Esomus danricus</i>	GGCAACTACTTATCTCTCCACCACCAGCATGCACCAATTAGATTGTGCTTACCGGAATAGGAACGCCCTCAG
<i>Puntius sophore</i>	AGCAATCACATATCTTTCTACCATCAGCATGCACCTATTAGATTGTGCCTACTTGGAAATGGGAACGCCCTAAG
<i>Tor</i>	GGCAATCACGTATCTCTCTACCACCAGCATGTACCTATCCGATTGTGCCTACCGGAATAGGAACGCCCTAAG
<i>Garra mullya</i>	GGCAATCACGCATCTCTCTACCACCAGCATGCACCTATCTGATTGTGCCTACCGGAATAGGAACGCCCTAAG
<i>Labeo bata</i>	GACAATTACACATTTCTCTACCACCTAGCATGCACCTATCTGATTGTGCCTACTGGGATAGGAACGCCCTAAG
<i>Labeo rohita</i>	GGCAATCACGCATCTCTCTACTACCAGCATGTACCTATCCGATTGTGCCTACCGGAATAGGAACGCCCTAAG
<i>Cyprinus carpio</i>	GGCAATCACGCATCTCTCTACTACCAGCATGTACCTATCCGATTGTGCCTACCGGAACAGGAACGCCCTAAG
<i>Catla catla</i>	GGCAATCACGCATCTCTCTACTACCAGCATGTACCTATCCGATTGTGCCTACCGGAACAGGAACGCCCTAAG

Figure 2: Alignment of gene sequences of various fish species using ClustalW alignment tool

Conclusion

The diverse fish species inhabits Sone River in India, contributing to the ecological balance and supporting local livelihoods. The undertaken study is useful for taxonomic characterization and effectiveness of COI gene region of mtDNA in molecular identification of the diverse fishes required to improve their conservation measures and to preserve the fish biodiversity of the Sone River for future generations.

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