



Identification of Functional Nursery Habitats of *Opsarius bendelisis* in an Important Tropical River in the Eastern Himalayan Biodiversity Hotspot

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Abstract

The present study deals with the documentation of *Opsarius bendelisis* larvae habitats in Siang River, Arunachal Pradesh. Larvae of *Opsarius bendelisis* were identified with the help of molecular tools. The study also highlights the threats facing these functional nursery habitats of fishes in the river. The findings of the study are of paramount significance as identification of larval grounds can help in formulating habitat-based conservation approaches.

Keywords: Biodiversity conservation, Functional nursery habitats, *Opsarius bendelisis*, Siang River

Introduction

Freshwater ecosystems within biodiversity hotspots support high fish diversity, including numerous endemics and recently described species. Despite their ecological significance, freshwater systems, particularly rivers are severely threatened by habitat degradation, resource extraction and hydrological alterations, channel modification, pollution and sand mining. Hence, there is an urgent need to formulate and implement effective conservation measures for protecting fish diversity in rivers. Identification of active spawning and functional larval rearing or nursery habitats is of paramount significance in this context. It is to be noted that conservation strategies that do not explicitly protect larval breeding and nursery habitats are unlikely to ensure long-term population persistence. However, persistent challenges in larval identification hinder the recognition of breeding and nursery habitats and the implementation of habitat-based conservation measures. Recent advances in

molecular biology have accurate, cost-effective and efficient species-level identification of fish larvae, greatly improving the study of early life stages. DNA barcoding using the mitochondrial cytochrome c oxidase subunit I (COI) gene is now a widely accepted and reliable approach for linking larval specimens to adult reference sequences, overcoming limitations of morphological similarity (Antil *et al.*, 2023).

Siang River is an important transboundary river in the Eastern Himalayan biodiversity hotspot. This headstream of River Brahmaputra flows through the North-East Indian state of Arunachal Pradesh and is a rich repository of indigenous fish diversity with 78 fish species being reported from it (Borah *et al.*, 2026). ICAR-Central Inland Fisheries Research Institute, Barrackpore carried out an exploratory survey to document the fish diversity of Siang River from Yingkiong in Arunachal Pradesh to Oiramghat in Assam and it was observed that Barillinae group of fishes, particularly *Opsarius bendelisis* was one of the most dominant species.

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Molecular Identification as a Tool for Resolving Larval Fish Identity

Larval specimens of fish (Figure 1a and 1b) with sizes ranging from 20.01 to 22.50 mm total length were collected from shallow marginal habitats (Figure 1c) at the confluence of Siang and Simang rivers (N 28°18'53.42" and E 94°57'47.90"; Figure 2) near Boleng, Arunachal Pradesh. The larval specimens could not be identified using conventional morphological keys; however, COI-based phylogenetic analysis of 81 sequences of 81 different Barillinae fishes unambiguously confirmed their identity as *O. bendelisis* (Figure 3) as they formed internal clade within *O. bendelisis*,

with <0.01 intraspecific K2P divergence (Table 1). The NCBI GenBankAccession No. of *O. bendelisis* larvae generated in the present study are: PX905470; PX905471; PX905472; PX905473. This represents the first direct molecular evidence of existence as well as identification of active spawning and functional nursery habitat of *O. bendelisis* in River Siang at its confluence with Simang RiveGenBank. These findings also underscore the importance of molecular identification in freshwater biodiversity research, particularly in poorly studied regions where newly described species and limited baseline ecological data constrain effective conservation planning.

Table 1: Pairwise Genetic Divergence reveals clear species delimitation among *Opsarius* and allied Barillinae fishes

Species	[1]	[2]	[3]	[4]	[5]	[6]	[7]	[8]
<i>O. bendelisis</i> [1]	0.010							
<i>B. barila</i> [2]	0.192	0.010						
<i>B. vagra</i> [3]	0.192	0.044	0.000					
<i>O. bakeri</i> [4]	0.190	0.177	0.187	0.000				
<i>O. barna</i> [5]	0.223	0.208	0.203	0.199	0.010			
<i>O. canarensis</i> [6]	0.182	0.168	0.173	0.060	0.189	0.010		
<i>O. caudicellatus</i> [7]	0.189	0.164	0.162	0.172	0.215	0.147	0.000	
<i>O. dogarsinghi</i> [8]	0.197	0.195	0.192	0.188	0.245	0.166	0.117	0.000
<i>O. gatensis</i> [9]	0.212	0.184	0.167	0.140	0.192	0.124	0.143	0.179
<i>O. koratensis</i> [10]	0.276	0.259	0.239	0.243	0.264	0.240	0.234	0.257
<i>O. malabaricus</i> [11]	0.209	0.186	0.191	0.058	0.191	0.060	0.163	0.180
<i>O. ngawa</i> [12]	0.195	0.189	0.175	0.197	0.215	0.152	0.060	0.117
<i>O. pulchelus</i> [13]	0.213	0.242	0.237	0.153	0.246	0.158	0.160	0.179
<i>O. shacra</i> [14]	0.237	0.247	0.237	0.258	0.247	0.242	0.190	0.203
<i>O. signicaudus</i> [15]	0.194	0.176	0.167	0.206	0.222	0.166	0.127	0.114
<i>O. tileo</i> [16]	0.224	0.199	0.195	0.208	0.184	0.186	0.188	0.187
<i>O. siangi</i> [17]	0.247	0.256	0.223	0.232	0.239	0.212	0.208	0.199

Table 1: Continue...

Species	[9]	[10]	[11]	[12]	[13]	[14]	[15]	[16]	[17]
<i>O. gatensis</i> [9]	0.000								
<i>O. koratensis</i> [10]	0.230	0.000							
<i>O. malabaricus</i> [11]	0.149	0.223	0.000						
<i>O. ngawa</i> [12]	0.155	0.229	0.176	0.000					
<i>O. pulchelus</i> [13]	0.194	0.240	0.176	0.177	0.010				
<i>O. shacra</i> [14]	0.262	0.250	0.259	0.203	0.236	0.040			
<i>O. signicaudus</i> [15]	0.176	0.241	0.203	0.139	0.180	0.205	n/c		
<i>O. tileo</i> [16]	0.197	0.281	0.212	0.190	0.225	0.240	0.221	0.000	
<i>O. siangi</i> [17]	0.240	0.293	0.245	0.196	0.209	0.126	0.204	0.215	0.000

Threats to Larval Habitats in Biodiversity Hotspots

Freshwater biodiversity hotspots are increasingly threatened by anthropogenic pressures and Siang River is no exception. As observed in our study, sand mining and destructive fishing practices are the major threats facing the river. Further, the ecology and hydrology of Siang River will likely undergo

significant changes in the near future as a result of the large hydropower projects which are reportedly being constructed in its headwaters in Tibet. Thus, channel modification, water abstraction, habitat fragmentation and alteration will put tremendous additional pressure on ichthyofaunal diversity of the river.

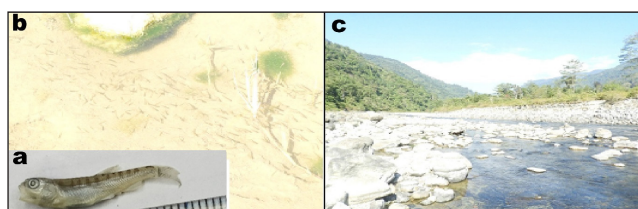


Figure 1: (a-b) Larvae of *Opsarius bendelisis*; (c) *Opsarius bendelisis* larvae habitat in Siang River, Arunachal Pradesh



Figure 2: Collection locality of *Opsarius bendelisis* larvae from Siang River, Arunachal Pradesh

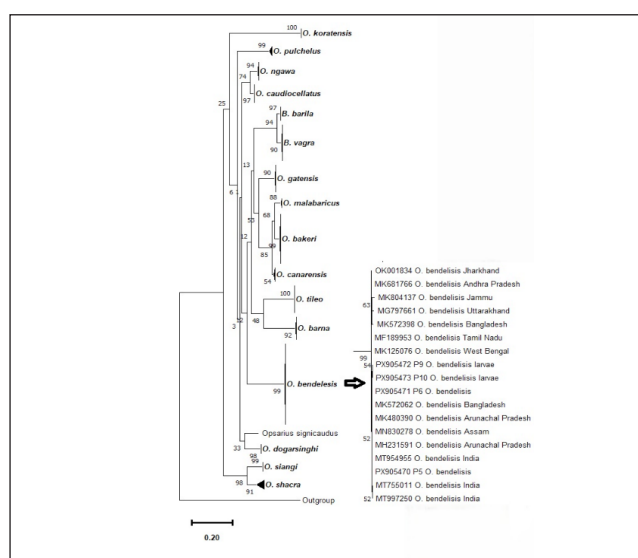


Figure 3: COI-based phylogenetic analysis of 81 sequences of 81 different Barillinae fishes

Conclusion

Considering these aspects, the present findings are of paramount significance as identification of larval grounds can help in formulating habitat-based conservation approaches (Mangel *et al.*, 2006) and can facilitate establishment of riverine protected areas or fish breeding conservation zones within freshwater biodiversity hotspots (Acreman *et al.*, 2020). These areas can be specifically designed to safeguard larval breeding and nursery habitats, which are often overlooked in conventional conservation planning. By conserving habitats critical to early life stages, these protected areas act as source areas, enhancing recruitment and resilience throughout the river system (Piczak *et al.*, 2023). Protecting larval habitats not only benefits individual species but also conserves entire aquatic communities and ecosystem processes (Piczak *et al.*, 2023). Habitat-based protection informed by molecular ecology aligns with modern conservation principles that emphasize ecosystem integrity, connectivity and long-term sustainability (Behrens-Chapuis *et al.*, 2021). This finding underscores the conservation significance of identification of breeding and nursery grounds of fishes and highlights the urgent need for habitat-based protection and the establishment of protected areas focused on larval breeding grounds. Integrating molecular tools with habitat-centric management offers a robust and scientifically defensible pathway to conserve freshwater biodiversity in the face of increasing anthropogenic pressure.

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