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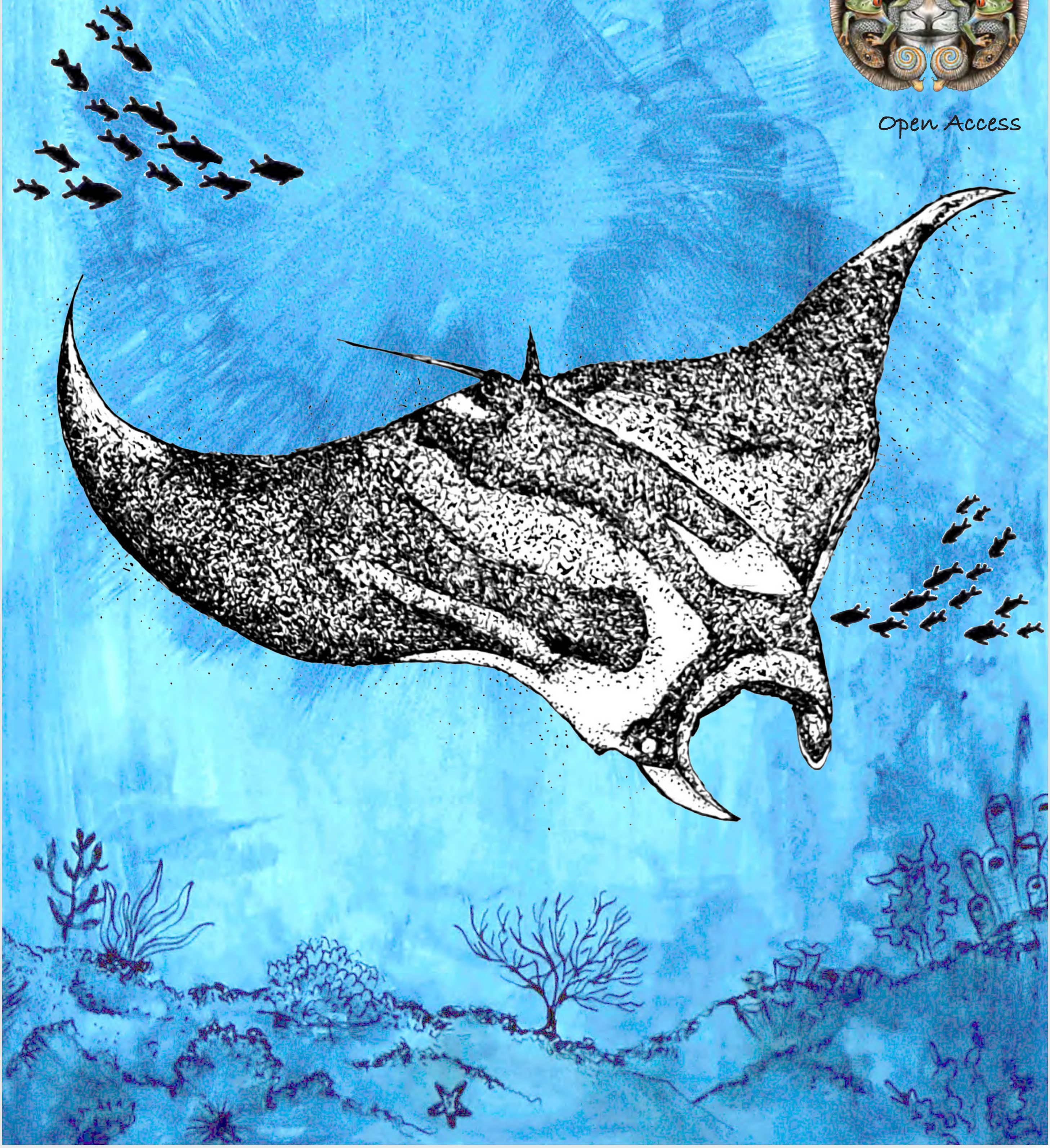

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Cover: Giant Oceanic Manta Ray *Mobula birostris* in ink on acrylic wash by Elakshi Mahika Molur adapted from scientific illustration by Roger Hall.

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COMMUNICATION

Evaluating the influence of environmental variables on fish abundance and distribution in the Singhiya River of Morang District, eastern Nepal

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Abstract: Monitoring the impact of fishing pressure on the Singhiya River is critical for resource development and sustainability, and the present situation is alarming and causing critical concern among the public. This study aimed to identify fish community trends over time and space in the river, and to investigate the impact of environmental variables on fish abundance and dispersion. Monthly fish sampling was performed from October 2020 to September 2021 from the 5th to 10th of each month. We used three cast nets of various mesh sizes (0.5, 2, & 4 cm) and monofilament gill nets with mesh sizes of 6, 8, & 10 cm. A total of 7,593 fish were collected, representing 61 species from seven orders, 20 families, and 37 genera. Similarity percentage (SIMPER) analysis revealed 78.8% similarity among six stations, with the primary contributing species: *Puntius chola* (28.2%), *Puntius sophore* (13.5%), *Pethia ticto* (5.33%), *Chagunius chagunio* (3.76%), *Barbonymus gonionotus* (3.69%), *Puntius terio* (3.46%), *Opsarius shacra* (2.2%), and *Opsarius bendelisis* (2.1%). Analysis of variance (ANOVA) on canonical correspondence analysis revealed that four of the seven selected environmental variables had significant relationship with the fish assemblage such as water parameters velocity, temperature, pH, and hardness. Overfishing and direct discharge of industrial waste into water resources may be the primary causes for the decline in fish diversity in Singhiya River.

Keywords: ANOVA, assemblage structure, cast nets, fish diversity, fish ecology, habitat variable, time-space.

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Author contributions: JHL, DR, AS and RB performed field surveys and collected data. JHL, DR, AS, SS and JT wrote and finalized the manuscript. JHL analyzed the data.

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INTRODUCTION

Freshwater bodies are vulnerable to habitat fragmentation, human encroachment, climate change, pollution, and biological invasions (Radinger et al. 2019). The combined effects of environmental pollution, unprecedented rates of biodiversity change, hydrological alteration, dam construction, and disconnection between the rivers and their lakes are possibly the largest threats to freshwater fish biodiversity (Huang & Li 2016). The diversity of the natural population is partially dependent on the environmental variables which always affect the competing populations (Chowdhury et al. 2011; Hossain et al. 2012). The factors influencing fish assemblages involve the environmental variables which are spatially heterogeneous and temporally variable and biotic interactions such as competition and predation (Harvey & Stewart 1991; Grossman et al. 1998). The environmental variables such as water velocity (Li et al. 2012; Adhikari et al. 2021; Limbu et al. 2021b), water depth (Kadye et al. 2008; Li et al. 2012; Mia et al. 2019; Chaudhary & Limbu 2021), substrate (Vlach et al. 2005; Yan et al. 2010), water temperature (Hossain et al. 2012; Nsor & Obodai 2016), and dissolved oxygen (Guo et al. 2018) all have been found to affect fish abundance and distribution in the rivers and streams.

In Nepal, few studies have looked at fish diversity and its link with environmental factors (Mishra & Baniya 2016; Limbu et al. 2020). Information on the relationship between fish community structure and environmental variables can aid in the preservation and management of aquatic biodiversity in the face of human-caused problems such as pollution and global climate change (Li et al. 2012). The Singhiya River has been altered due to several human encroachments such as settlements, factories, embankments, sand mining, electrofishing, damping and agriculture. To date, the space and time pattern of low-land, Terai region remains relatively unknown. Moreover, the details on fish community structure relating to their anthropogenic activities is also scanty. Facts about the relationship between fish community structure and environmental conditions can help us retain and lead aquatic biodiversity away from human-caused challenges like pollution and global climate change (Li et al. 2012).

The present study aimed to detect fish community patterns in the Singhiya River through time and space, as well as to evaluate the impact of environmental variables on fish abundance and dispersion. The current study expected that during the annual dry season, when water current and volume are reduced, fish abundance in the Singhiya River would be increased. We also hypothesized that the structure of fish assemblages will vary according to seasonal fluctuation defined by environmental variables.

MATERIALS AND METHODS

Study area

Singhiya River is situated in the Morang district of Eastern Nepal (Figure 1). It is a perennial river that originates from the periphery of Hattimuda, Dulary, and Sundar Haraicha and surges through the Budiganga Municipality and Biratnagar Sub-metropolitan, and from the Buddhanagar it crosses the border of India. It lies in the latitude and longitude coordinates of 26.913^{o} N & 87.157^{o} E, respectively. The water of this river is mainly used for irrigation. The vegetation bordering the river is mixed, mostly consisting of bamboo and coniferous forest and the dominant river substrata consist of cobbles, pebbles, gravel, and sand. In total, six sampling stations were set up to gather fish. Residents settled along the entire river in the catchment, and numerous small and large factories were established in stations 1, 2, 3, & 4 whereas, stations 6 & 7 were set up close to the city.

The Singhiya River region experiences mostly sunny weather, with occasional clouds, and the water is muddy due to increased anthropogenic activities near the area of human settlement but crystal clear in its origin parts. The study area for this research includes 22 km of river basin starting from Hattimuda to Buddhanagar of Morang District.

Data collection, Identification, and Preservation

From October 2020 to September 2021, fish samples were taken every month. Sample collection started on the 5th and continued to the 10th of the selected month, i.e., October, November, December (2020), January, February, March, April, May, June, July, August, and September (2021). We made 72 samples at six stations, namely, (S1) Hattimudha, (S2) Puspalal Chowk, (S3) near Hanuman Mandir, (S4) Hatkhola, (S5) Jahda Bridge, and (S6) Buddhanagar, with fish sampling carried out between 0070 h and 0090 h. We employed three cast nets of various sizes, one with a mesh size of 0.5 cm, diameter of 5 m, and a weight of 2 kg, and another with a mesh size of 2 cm, diameter of 5 m, and a weight of 4 kg. A cast net with a diameter of 4 cm, a length of 7 meters, and a weight of 7 kg was also utilized. Cast netting was used to cover 150 m to 200 m across each

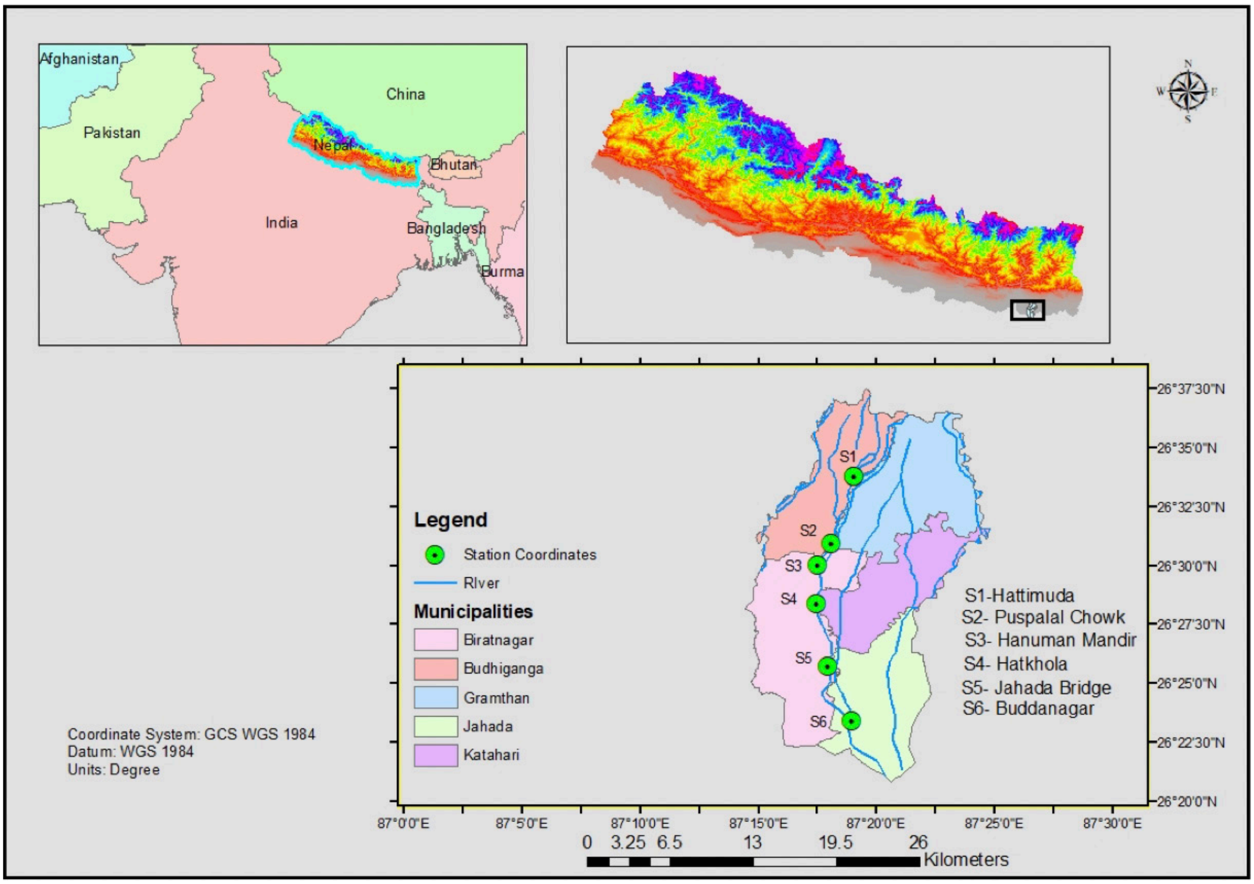


Figure 1. Map of study area showing sampling stations in Singhiya River, Nepal.

station, ensuring that all conceivable habitats were covered (Limbu et al. 2021b). For each cast net, a total of 10 throws were made over one hour. The fish were also caught using monofilament gill nets with mesh sizes of 6, 8, & 10 cm. Nine-gill nets were left late in the evening (1700h–1800 h) and pulled out early in the morning (0600h–0700 h) at a sample distance of 150–200 m at each station

Fish sampled were photographed and identified in the field, and unidentified specimens were preserved in 10% formalin for later identification. The remaining samples were released to their original habitat after the photography. Standard fish taxonomy literatures (Talwar & Jhingran 1991; Jayaram 2010; Shrestha 2019; Fricke et al. 2021) and other available standard literature were used to identify the fish. During field visits environmental variables such as water temperature, dissolved Oxygen (DO), pH, total hardness, water velocity, alkalinity, and free carbon dioxide (CO) were investigated using the American Public Health Association's standard methodology (APHA 2012). A digital thermometer was placed in the water at a depth of 1 foot to measure the water temperature (°C). The Winkler titrimetric method was used to determine DO (mg/l). A pH meter was used to determine the pH (HI 98107, HANNA Instrument). The EDTA titrimetric technique was used to evaluate total hardness (mg/l). With the help of a stopwatch, a small ball and a measuring tape, water velocity (m/s) was determined using the float method. The alkalinity (mg/l) was measured using the titration method. The titrimetric method was used to detect free carbon dioxide (mg/l) using phenolphthalein as an indicator.

Data analysis

To examine potential variation over space and time a one-way analysis of variance (ANOVA) was used for temperature, pH, dissolved oxygen, hardness, and water velocity. To determine which means were significantly different at the 0.05 level of probability, a posthoc Tukey HSD test was used (Spjøtvoll & Stoline 1973). In the first step of data processing, the diversity of the fish assemblage was quantified, and then a statistical comparison was performed. Data on fish abundance were subjected to various diversity indices (Shannon, Simpson dominance, evenness, and species richness). All of the diversity indices were created using data from

12 months (each month, six samples were taken, S1–S6) and the data were used directly in the analysis, according to Magurran (1988) for each fish community sample. The Shannon diversity index (Shannon & Weaver 1963) takes into account both the number of species and the distribution of individuals within species.

The Shannon diversity was calculated using the following formula:

$$H = \sum_{i=1}^{S} Pi * logPi \quad (1)$$

Where S is the total number of species and *Pi* is the relative proportion of i_{th} of species.

The Simpson index (Harper 1999) is a dominance index which gives more weight to common or dominant species.

The Simpson dominance index was calculated by using following formula:

$$D = \sum_i \left(\frac{ni}{n}\right)^2 \quad (2)$$

Where n_i is number of individuals of species *i*.

The Evenness index (Pieleu 1966) measures how evenly or uniformly the relative abundances Pi (i=1..,S) are distributed across the S different species, irrespective of the value of S and the Evenness index was determined by the following equation:

$$E = H'/\log S \quad (3)$$

Where, H' = Shannon diversity index

S = Total number of species in the sample.

In the multivariate analysis, rare species (<1%) were excluded in the analysis as they tend to affect multivariate analyses (Gauch 1982). Samples by species and environmental variables were analyzed through a multivariate analysis tool. Detrended correspondence analysis (DCA) (Hill & Gauch 1983) was performed to determine whether redundancy correspondence analysis (RDA) or canonical correspondence analysis (CCA) would be the most appropriate model to describe the association between species and environmental variables. The value of first axis length (3.14) and eigen value (0.53) obtained from DCA suggested that the uni-model associated with canonical correspondence analysis (CCA) (Ter Braak 1986) was more applicable. Therefore, a direct multivariate ordination method (Legendre & Legendre 1998) based on a linear response of species to environmental gradients was applied. Collected fish abundance and determined environmental variables were used directly in the multivariate analysis (Yan et al. 2010; Hossain et al. 2012; Vieira & Tejerina-Garro 2020; Tumbahangfe et al. 2021).

The one-way permutational multivariate analysis of variance (perMANOVA) (Clarke 1993) was used to determine whether there was a significant difference between the spatial and temporal scales of the collected fish data. A similarity percentage (SIMPER) (Clarke 1993) analysis was used to visualize the major contributing species in both space and time. Furthermore, Individual rarefaction analyses (Colwell et al. 2012), was performed across stations and months. All the statistical analysis were performed in R software (R Core Team 2019), 2.5-6 version.

RESULTS

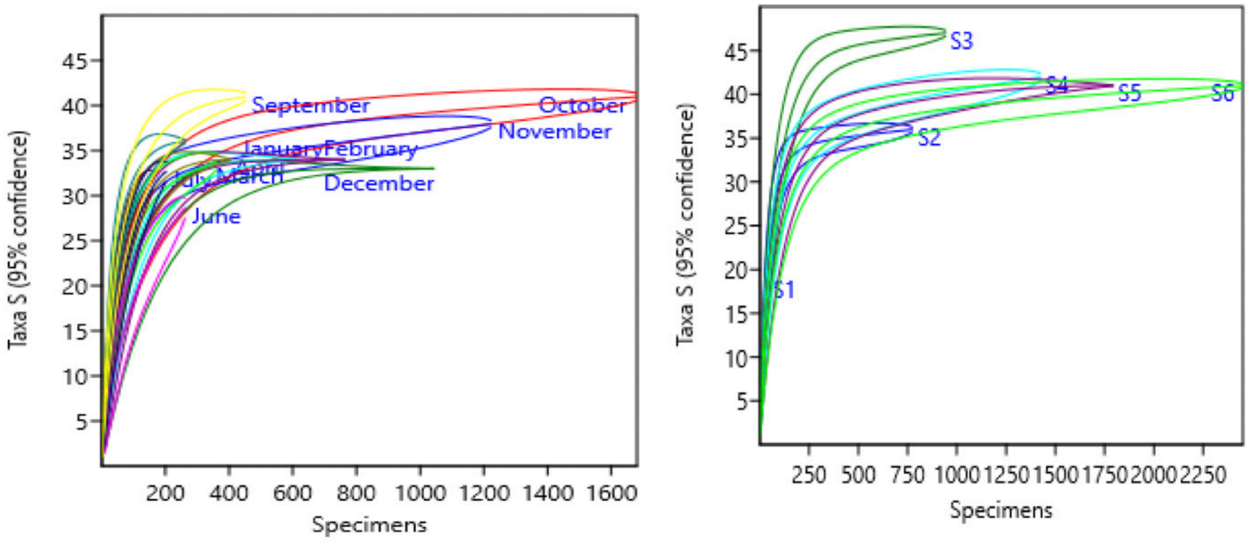
Fish community structure

A total of 7,593 fish were collected, representing 61 species belonging to seven orders, 20 families, and 37 genera (Table 1). The three main orders that represented 84% of the total species count included Cypriniformes (32 species), Siluriformes (11 species), and Anabantiformes (8 species). Synbranchiformes and Perciformes each contained four species and the rest contributed less than 2% to the total species counts. At the family level, the Danionidae family included the most species (16), followed by Cyprinidae (11), Ambassidae (4), Bagridae (4), Channidae (4), Mastacembelidae (3), Cobitidae (2), Siluridae (2), Ailiidae (2), Anabantidae (2), Osphronemidae (2), Psilorhynchidae (1), Nemacheilidae (1), Botiidae (1), Sisoridae (1), Clariidae (1), Heteropneustidae (1), Synbranchidae (1), Mugilidae (1), and Gobiidae (1). The four most abundant species comprised 56% of the total catch, i.e., *Puntius chola* (27%), *Puntius sophore* (18%), *Pethia ticto* (6.3%), and *Barbonymus gonionotus* (5.3%). Considerable differences in fish abundance and diversity were observed among sampling stations and monthly samplings.

The highest number of fish was collected during October (1,707 specimens), followed by the months of November > December > February > January > September > April > March > August > June > July > May (Figure 2a). The highest fish diversity in the study area was calculated during October (42 species), followed by September (41 species), November (38 species), August (36 species), December, February, & April (34 species in each month), March & July (33 species each in each month), May (32 species), January (31 species), and June (29 species). The highest numbers of fish were collected at station (S6), followed by S5>S4>S3>S2>S1 (Figure 2b). According to similarity percentage (SIMPER) analysis (Table 2), 79% similarity was found between the stations, and the primary contributing species were: *Puntius chola* (28%), *Puntius sophore* (14%), *Pethia ticto* (5.3%), *Chagunius chagunio* (3.8%), *Barbonymus gonionotus* (3.7%), *Puntius terio* (3.5%), *Opsarius shacra* (2.2%), and

Table 1. Coding of the Singhiya River, Morang District, Nepal by order, family, and species.

Order / Family	Code	Species	IUCN status
Cypriniformes			
Cyprinidae	C1	*Chagunius chagunio* (Hamilton 1822)	LC
Cyprinidae	C2	*Cirrhinus mrigala* (Hamilton 1822)	LC
Cyprinidae	C3	*Cirrhinus reba* (Hamilton 1822)	LC
Cyprinidae	C4	*Labeo gonius* (Hamilton 1822)	LC
Cyprinidae	C5	*Tariqilabeo latius* (Hamilton 1822)	LC
Cyprinidae	C6	*Puntius chola* (Hamilton 1822)	LC
Cyprinidae	C7	*Puntius sophore* (Hamilton 1822)	LC
Cyprinidae	C8	*Puntius terio* (Hamilton 1822)	LC
Cyprinidae	C9	*Pethia ticto* (Hamilton 1822)	LC
Cyprinidae	C10	*Barbonymus gonionotus* (Bleeker 1849)	LC
Cyprinidae	C11	*Systomus sarana* (Hamilton 1822)	LC
Danionidae	C12	*Barilius barila* Hamilton 1822	LC
Danionidae	C13	*Opsarius bendelisis* Hamilton, 1822	LC
Danionidae	C14	*Opsarius shacra* Hamilton 1822	LC
Danionidae	C15	*Opsarius vagra* Day 1878	LC
Danionidae	C16	*Opsarius barna* Hamilton 1822	LC
Danionidae	C17	*Cabdio morar* (Hamilton 1822)	LC
Danionidae	C18	*Cabdio jaya* (Hamilton 1822)	LC
Danionidae	C19	*Danio rerio* (Hamilton 1822)	LC
Danionidae	C20	*Devario devario* (Hamilton 1822)	LC
Danionidae	C21	*Chela cachius* (Hamilton 1822)	LC
Danionidae	C22	*Esomus danrica* (Hamilton 1822)	LC
Danionidae	C23	*Amblypharyngodon mola* (Hamilton 1822)	LC
Danionidae	C24	*Rasbora daniconius* (Hamilton 1822)	LC
Danionidae	C25	*Bengala elanga* (Hamilton 1822)	LC
Danionidae	C26	*Salmostoma acinaces* (Valenciennes 1844)	LC
Danionidae	C27	*Salmostoma phulo* (Hamilton 1822)	LC
Psilorhynchidae	C28	*Psilorhynchus sucatio* (Hamilton 1822)	LC
Nemacheilidae	C29	*Paracanthocobitis botia* (Hamilton 1822)	LC
Cobitidae	C30	*Canthophrys gongota* (Hamilton 1822)	LC
Cobitidae	C31	*Lepidocephalichthys guntea* (Hamilton 1822)	LC
Botiidae	C32	*Botia lohachata* Chaudhuri 1912	NE
Siluriformes			
Bagridae	C33	*Mystus bleekeri* (Day 1877)	LC
Bagridae	C34	*Mystus cavasius* (Hamilton 1822)	LC
Bagridae	C35	*Mystus tengara* (Hamilton 1822)	LC
Bagridae	C36	*Mystus vittatus* (Bloch 1794)	LC
Siluridae	C37	*Ompok bimaculatus* (Bloch 1794)	NT
Siluridae	C38	*Wallago attu* (Bloch & Schneider 1801)	VU
Ailiidae	C39	*Ailia coila* (Hamilton 1822)	NT
Ailiidae	C40	*Clupisoma montanum* Hora 1937	LC
Sisoridae	C41	*Pseudolaguvia ribeiroi* (Hora 1921)	LC
Clariidae	C42	*Clarius magur* (Hamilton 1822)	EN
Heteropneustidae	C43	*Heteropneustes fossilis* (Bloch 1794)	LC
Synbranchiformes			
Synbranchidae	C44	*Ophichthys cuchia* (Hamilton 1822)	LC
Mastacembelidae	C45	*Macrognathus aral* (Bloch & Schneider 1801)	LC
Mastacembelidae	C46	*Macrognathus pancalus* Hamilton 1822	LC
Mastacembelidae	C47	*Mastacembelus armatus* (Lacepède 1800)	LC
Perciformes			
Ambassidae	C48	*Chanda nama* Hamilton 1822	LC
Ambassidae	C49	*Parambassis baculis* (Hamilton 1822)	LC
Ambassidae	C50	*Parambassis lala* (Hamilton 1822)	NT
Ambassidae	C51	*Parambassis ranga* (Hamilton 1822)	LC
Anabantiformes			
Anabantidae	C52	*Anabas cobojius* (Hamilton 1822)	DD
Anabantidae	C53	*Anabas testudineus* (Bloch 1792)	LC
Osphronemidae	C54	*Trichogaster fasciata* Bloch & Schneider 1801	LC
Osphronemidae	C55	*Trichogaster lalius* (Hamilton 1822)	LC
Channidae	C56	*Channa barca* (Hamilton 1822)	DD
Channidae	C57	*Channa gachua* Bloch & Schneider 1801	VU
Channidae	C58	*Channa striata* (Bloch 1793)	LC
Channidae	C59	*Channa punctata* (Bloch 1793)	LC
Mugiliformes			
Mugilidae	C60	*Minimugil cascasia* (Hamilton 1822)	LC
Gobiformes			
Gobiidae	C61	*Glossogobius giuris* (Hamilton 1822)	LC



a b

Figure 2. a—Individual rarefaction analysis plot based on months | b— Individual rarefaction analysis plot based on stations.

Table 2. Average similarity (%) and discriminating fish species in the Singhiya River, Morang District, Nepal, by month and station using SIMPER analysis.

Code	Species	Station	Code	Species	Months
		Contribution (%)			Contributions (%)
C6	*Puntius chola*	28.2	C6	*Puntius chola*	26.58
C7	*Puntius sophore*	13.51	C7	*Puntius sophore*	13.78
C9	*Pethia ticto*	5.33	C9	*Pethia ticto*	5.7
C1	*Chagunius chagunio*	3.76	C10	*Barbonymus gonionotus*	3.81
C10	*Barbonymus gonionotus*	3.69	C8	*Puntius terio*	3.59
C8	*Puntius terio*	3.46	C1	*Chagunius chagunio*	3.51
C14	*Opsarius shacra*	2.2	C14	*Opsarius shacra*	2.24
C13	*Opsarius bendelisis*	2.1	C13	*Opsarius bendelisis*	2.15

Opsarius bendelisis (2.1%); 77.5% similarity was found between months, and the top contributing species were as listed above.

Diversity status

Tables 3 & 4 show the results of diversity indices. The highest Shannon diversity index (2.79) was found at station 2 (S2) and in the month of August (2.94) whereas the lowest (1.76) was found at station 1 (S1) and in June (1.51). Analysis of variance (ANOVA) testing for both time and space revealed a significant (P <0.05) difference across six stations, but no significant (P >0.05) difference for the Shannon diversity index over twelve months. The highest Simpson dominance index (0.91) was found at station 2 (S2) and in the month of August (0.93) while the lowest Simpson index value (0.67) was found at station 6 (S6) and in the month of June (0.61). There was no significant (P >0.05) difference in the Simpson dominance index across the six sampling points and months. Similarly, the highest Evenness index (0.59) was at stations 1 & 2 and the month of August (0.59) whereas the lowest value (0.44) was found at stations 5 & 6 respectively, and the month of June (0.42). There was also no significant (P >0.05) difference in the Evenness index between the six stations and months. On the other hand, the highest Species richness value was observed at station 6 (S6) and in the month of October (36) while the lowest value was found at station 1 (S1) and the month of June (21). The species richness index differed significantly (P <0.05) between the six sampling

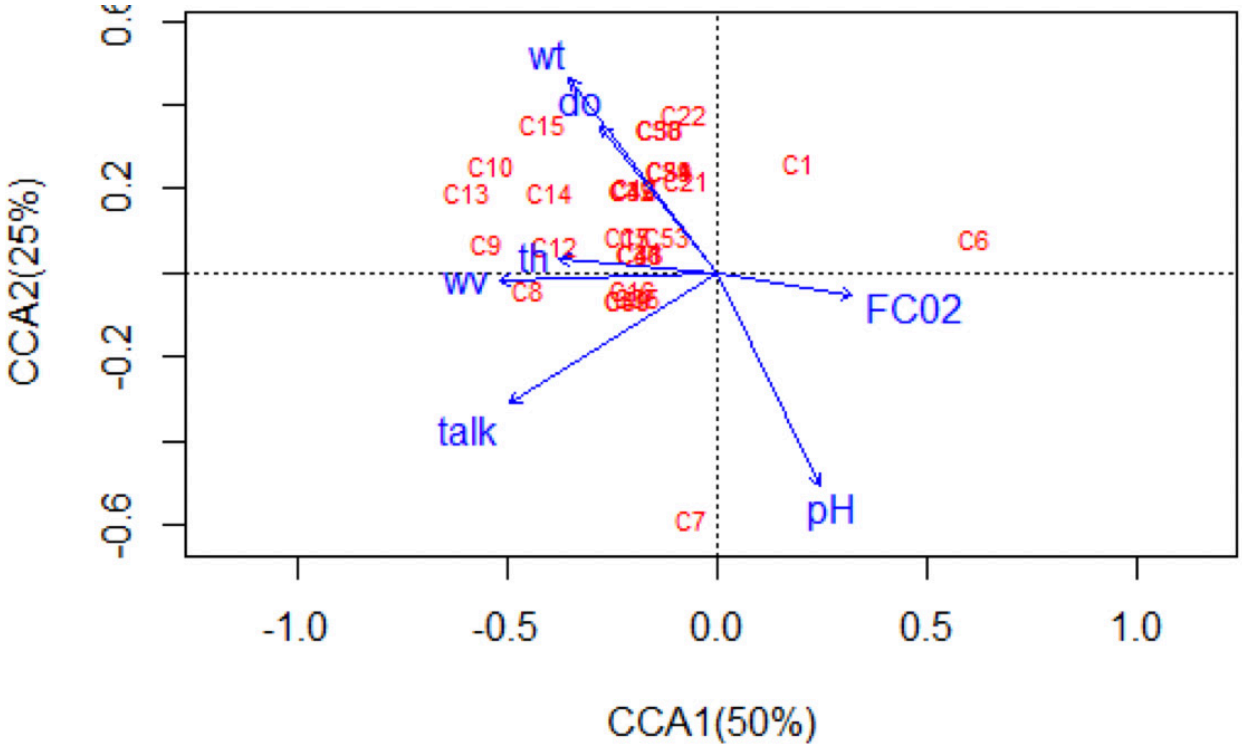


Figure 3. Canonical correspondence analysis ordination between fish community structure and environmental variables (for species code refer to table 1): talk—total alkalinity | do—dissolved oxygen | wv—water velocity | th—total hardness | wt—water temperature | FCO2—free carbon-dioxide.

Table 3. Diversity indices for the Singhiya River, Morang District, Nepal at six stations.

Stations	Shannon index	Simpson dominance index	Evenness index	Species richness
S1	1.76±0.7	0.9±0.14	0.59±0.93	18.44±3.88
S2	2.79±0.5	0.91±0.05	0.59±0.03	27±4.92
S3	2.37±0.64	0.8±0.14	0.51±0.09	31.4±4.77
S4	2.3±0.9	0.77±0.22	0.49±0.13	33.23±4.43
S5	1.87±0.8	0.68±0.2	0.44±0.12	34.51±4.07
S6	1.93±0.74	0.67±0.22	0.44±0.12	35.41±3.91

locations and months.

Environmental factors vs fish community structure

The results obtained after the canonical correspondence analysis are plotted in Figure 3. The first (CCA1) and second (CCA2) axis of the CCA accounted for 50% and 25%, respectively. The CCA biplot indicates the relationship between species and environmental variables. The fish species of *Puntius sophore* (C7), *Puntius terio* (C8), *Opsarius barna* (C16), *Salmostoma acinaces* (C26), and *Mystus tengara* (C35) are positively related to total alkalinity and water velocity but species of *Chagunius chagunio* (C1) and *Puntius chola* (C6) are negatively related to water velocity and total alkalinity. Fish species of *Pethia ticto* (C9), *Barbonymus gonionotus* (C10), *Barilius barila* (C12), *Opsarius bendelisis* (C13), *Opsarius shacra* (C14), *Opsarius vagra* (C15), *Cabdio morar* (C17), *Chela cachius* (C21), *Esomus danrica* (C22), *Mystus bleekeri* (C33), *Wallago attu* (C38), *Heteropneustes fossilis* (C43), and *Chanda nama* (C48) are positively related to water temperature, dissolved oxygen, and total hardness but negatively related to free carbon dioxide and pH. An analysis of variance (ANOVA) on canonical correspondence analysis suggested that water parameters of water velocity, water temperature, total alkalinity, pH and total hardness are the major

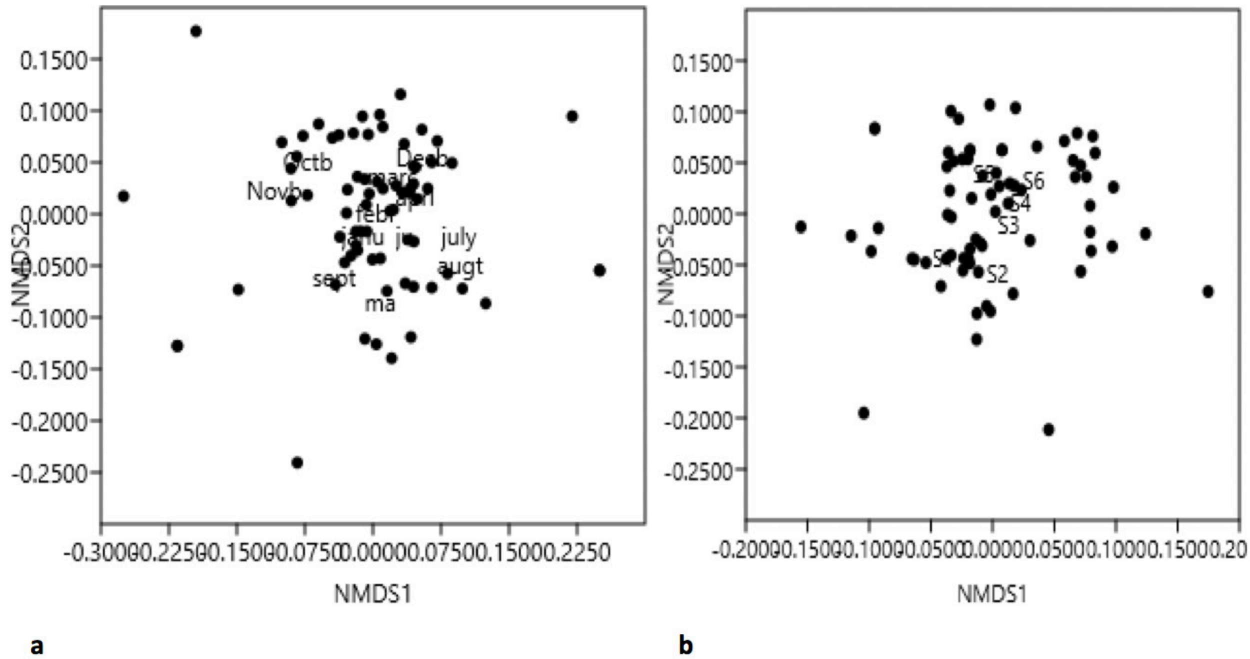


Figure 4. a—NMDS ordination of time variation of fish community structure in the Singhiya River | b—NMDS ordination of space variation fish community structure in the Singhiya River.

influencing factors (*P* <0.05) to determine the fish abundance and distribution.

In addition, one-way permutational multivariate analysis of variance (perMANOVA) on the Non-multidimensional Scaling (NMDS) showed no significant (*P* >0.05) difference between station 3, 4, 5, & 6, but station 1 & 2 showed significant (*P* <0.05) difference (Figure 4b). The fish community structure in October showed a significant (*P* <0.05) difference between January, February, March, April, May, June, July, August, & September but no significant (*P* >0.05) difference was found with November and December (Figure 4a).

DISCUSSION

This is the first study to describe the spatial and temporal fluctuation of fish community structure in a Nepalese low-land river. The outcomes of this study will improve our understanding of the variance in fish communities for the benefit of Nepalese low-land river conservation, which recorded a total of 7,593 individuals, represented by 61 species belonging to seven orders, 20 families, and 37 genera. This suggests that the Singhiya River provides a significant source of livelihood and food to local fisherman and communities. The representation of Cypriniformes, Siluriformes, and Anabantiformes orders found in this study is consistent with the information reported in the other river systems of Nepal such as the Mechi River (Adhikari et al. 2021), Ratuwa River (Rajbanshi et al. 2021), and Phewa Khola (Limbu et al. 2021b).

The present findings revealed that the maximum number and diversity of fish species were collected in October, September, and November. During June and July, water velocity was found to be low and the water temperature was found to be high in the current study. Because of the low water velocity, the fishermen could do the most of the fishes. River discharge and water temperature have a much greater impact on the amount and diversity of fish (Kriauciuniene et al. 2019). Overfishing, industrial discharges, and sand mining may have impacted the amount and diversity of fish in the Singhiya River. Furthermore, essential aquatic ecosystem measurements such as species richness and diversity indices are influenced by changes in abiotic parameters such as river discharge and water temperature (Crane & Kapuscinski 2018; Parker et al. 2018).

According to local fisherman, populations of *Cirrhinus mrigala, Cirrhinus reba, Labeo gonius, Systomus sarana, Danio rerio*, *Devario devario, Amblypharyngodon mola*, *Rasbora daniconius*, *Bengala elanga*, *Salmostoma acinaces*, *Salmostoma phulo*, *Psilorhynchus sucatio*, *Lepidocephalichthys guntea*, *Botia lohachata*, *Heteropneustes fossilis*, *Ophichthys cuchia*, *Macrognathus aral*, *Macrognathus pancalus*,

Table 4. Diversity indices for the Singhiya River, Morang District, Nepal over 12 months.

Months	Shannon index	Simpson dominance index	Evenness index	Species richness
Oct	2.33±0.27	0.84±0.03	0.52±0.01	35.97±7.61
Nov	2.34±0.9	0.82±0.14	0.51±0.09	32.98±5.89
Dec	1.9±0.79	0.75±0.16	0.49±0.11	30.35±5.59
Jan	2.26±0.84	0.83±0.15	0.54±0.1	30.48±6.58
Feb	2.08±1.19	0.71±0.32	0.46±0.21	30.42±5.53
Mar	1.99±0.68	0.74±0.2	0.48±0.13	27.94±6.44
Apr	2.12±0.93	0.72±0.27	0.47±0.17	30.17±5.45
May	2.38±0.53	0.84±0.12	0.56±0.08	26.31±6.04
Jun	1.51±0.91	0.61±0.29	0.42±0.2	20.66±7.31
Jul	1.91±0.48	0.72±0.15	0.47±0.1	26.26±7.13
Aug	2.94±0.59	0.93±0.05	0.59±0.03	33.01±6.65
Sep	2.91±0.2	0.91±0.04	0.56±0.03	34.92±6.98

Mastacembelus armatus, *Trichogaster fasciata*, *Trichogaster lalius*, *Channa barca*, *Channa orientalis*, *Channa striata*, *Minimugil cascasia*, and *Glossogobius giuris* have significantly reduced, with less than five individuals recorded for each over the 12-month study period. Many studies have suggested that ongoing road development, river corridor engineering, dams and water diversion, aquatic habitat loss and fragmentation, deforestation, riparian loss, overfishing, climate change, and direct discharge of industrial waste into water resources are the primary causes of Nepalese fish population reduction (Limbu et al. 2021a,b; Tumbahangfe et al. 2021). River output appears to be influenced by water level variations caused by climate change and water management, as well as fishing pressure (Halls 2015). Monitoring the impact of fishing pressure on the Singhiya River's exploited fish population is critical for resource development and sustainability. The present situation in the Singhiya River is still sounding the alarm and causing critical concern among the public. As the biodiversity of freshwater fish keeps on decreasing mainly due to anthropogenic impacts, it is apparent that there has been a serious lack of scientific basis and truly ecological action for sound river basin management (Li et al. 2012). The populations of *Labeo catla, Bagarius* spp., *Chitala chitala, Sisor* spp., and *Notopterus notopterus* have declined significantly and are not represented in the present study. Only *Cirrhinus* spp., *Channa* spp., *Labeo* spp., *Ophichthys cuchia, Heteropneustes fossilis, Macrognathus* spp., *Mastacembelus armatus, Clarius magur, Opsarius bendelisis, Chagunius chagunio,* and *Salmostoma* spp. are highly preferred fish species by the local community in the Singhiya River basin.

The Shannon diversity index takes into account the richness and proportion of each species, while the evenness and dominance indices reflect the relative number of individuals and the proportion of common species, respectively (Hossain et al. 2012; Yang et al. 2021). The highest Shannon diversity index (2.79) was identified at station 2 and in August (2.94), while the lowest (1.76) was discovered at station 1 and in June (1.51). A high Shannon diversity index is linked to a small number of individuals, whereas a low Simpson's diversity index is linked to a large number of individuals (Hossain et al. 2012; Temesgen et al. 2021). A biodiversity index seeks to categorize a sample's diversity (Magurran 1988) and is easily influenced by the number of specimens, sample size, and environmental factors (Leonard et al. 2006). The highest Simpson dominance index (0.91) was found at station 2 and the month of August (0.93), while the lowest Simpson index value (0.67) was obtained at station 6 and the month of June (0.61). Similarly, the highest evenness index (0.59) was observed at stations 1 and 2 and in August (0.59), while the lowest value (0.44) was recorded at stations 5 and 6 and in June (0.42). The maximum species richness (35.31) value was found at station 6 and the months of October (35.97), while the lowest (18.44) value was recorded at station 1 and the month of June (20.66). The species richness index varies considerably (P <0.05) between the six sampling locations and months. Overall, stations 2, 3, 4, 5, & 6 and the months of October, January, February, May, August, & September were likely to be rich with richness and diversity, because these sections were deeper and larger in terms of water depth and surface cover than station 1 section within the system. The river width and depth may be important for resting and hiding (Li et al. 2012) and for variable habitats for lotic water inhabiting fish such as *Cirrhinus* spp., *Mystus* spp., *Ailia coila*, *Ompok bimaculatus*, & *Wallago attu*

The information on the interaction between environmental variables and fish community structure can assist us in maintaining and managing aquatic biodiversity in the face of human-caused problems such as pollution, global climate change, and so on (Li et al. 2012). The influence of environmental variables on fish abundance, diversity, and distribution was checked by canonical correspondence analysis. In the current study, water velocity, water temperature, total alkalinity, pH, and total hardness are the major influencing factors (P <0.05) to determine the fish diversity, abundance, and distribution of the Singhiya River. Water velocity (Yu & Lee 2002; Yan et al. 2010; Adhikari et al. 2021), water temperature (Kadye et al. 2008; Temesgen et al. 2021), total alkalinity (Edds 1993; Pokharel et al. 2018), pH (Pokharel et al. 2018; Limbu et al. 2021b; Rajbanshi et al. 2021), and total hardness (Rajbanshi et al. 2021; Shrestha et al. 2021) have also been found to be influencing factors to shape the fish assemblage structure.

CONCLUSION

The Singhiya River exhibits a good ichthyofaunal diversity, represented by 61 species of fish belonging to seven orders, 20 families, and 37 genera. Of 61 species, *Puntius chola*, *Puntius sophore*, *Pethia ticto*, and *Barbonymus gonionotus* were the dominant fish species recorded in Singhiya River. However, commercially important species such as *Labeo catla, Bagarius* spp., *Chitala chitala, Sisor* spp., and *Notopterus notopterus* were not recorded during the study period. Thus, conservation of these species has become urgent in Singhiya River. Overfishing and direct discharge of industrial waste into water resources may be the primary causes for the decline in fish diversity in Singhiya River. Therefore, practices like dumping of industrial waste, overfishing, and sand mining should be minimized, monitored, and if required, prohibited to protect the Singhiya River's aquatic flora and fauna and natural ecology. The canonical correspondence analysis suggested that an important environmental variables in structuring the fish community in the Singhiya River were water velocity, temperature, pH, and hardness. Lastly, the current study, in conjunction with the preceding examination, could serve as a baseline scenario for future analysis of the Singhiya River and other connected water bodies in the coming decades.

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