The Status Of Fish Diversity Of Dudhkoshi River Of Eastern Nepal

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Abstract
Conservation of freshwater biodiversity is a major challenge, globally. It is obvious that for management and policy making, taxonomic data and their current status become inevitable. Dudhkoshi River is one of the major tributaries of the Koshi basin, and the least explored river regarding fish diversity. In this study, we explored the fish diversity status of this river incorporating species composition, diversity indices, and (relative) abundance, and also endeavored to reveal the conservation status of its ichthyofauna for the first time. Fish sampling was carried out using combinations of fishing gears and with the assistance of local fishermen. A total of 22 species belonging to 13 genera, eight families, and three orders, making a total of 1,265 individuals were collected. Cypriniformes and Cyprinidae were recorded as the dominant order and family which accounted for 77.27% and 36.36%, respectively. Species like *Schizothorax richardsonii*, *S. progastus*, *Labeo dyocheilus*, *Barilius bendelisis*, *B. barila*, *Glyptothorax pectinopterus* and *Psilorhynchus pseudecheneis* were found as the most abundant species, from highest to lowest, and *Botia dario*, *Channa barca*, *Eutropiichthys murius*, *Garra annandalei*, *Puntius sophore* and *Schistura multifasciata* were recorded as the occasional (rare) species. The Shannon-Wiener's diversity index shows that the overall fish diversity status of Dudhkoshi River was medium. While the majority of species belong to the Least Concerned (LC) category accounting for 67%, the four species, *Neolissochilus hexagonolepis*, *Psilorhynchus pseudecheneis*, *Schizothorax progastus* and *S. richardsonii* belong to NRDB status; those species had been proposed for legal protection in Nepal. To appreciate more generalized inferences about the fish diversity status of Dudhkoshi River, a comprehensive study integrating more seasons (e.g. winter and rainy) is highly recommended for future study.

Keywords: Fish diversity, Diversity index, Conservation, Dudhkoshi River

Introduction

Protection of freshwater biodiversity is a major conservation challenge at the present time (Dudgeon *et al.*, 2006). Rivers and lakes represent <1% of the total earth's surface (Leveque *et al.*, 2008; Su *et al.*, 2021), but support higher biodiversity. Studies show that temperate rivers are highly threatened in terms of biodiversity (Su *et al.*, 2021). Likewise, major threats to the Himalayan rivers include pollution (Edds, 2002), climate change and associated range shifting of species (Heino *et al.*, 2009; Conti *et al.*, 2015), Glacial Lakes Outburst Floods (GLOF), making huge disastrous situations, also to humans (Bajracharya and Mool, 2009), and hydropower dams, causing chief impacts in migratory routes, destruction of feeding and breeding grounds, alteration of flow regimes and water quality parameters (Allen *et al.*, 2010; Kano *et al.*, 2016; ADB, 2018; Kumar *et al.*, 2020). It is claimed that about 76% of migratory freshwater fishes have declined on a global scale; however, migratory status (data) of freshwater fishes of the Asian continent is largely lacking (Deinet *et al.*, 2020). In Nepal, 203 hydropower dams have been reported, of which 83 are operational and 120 are under construction (NEA, 2019), so the country's Himalayan rivers are very likely to be fragmented more and more in the days to come.

Fishes are the most numerous and diverse vertebrate groups. About 200 to 393 new species are discovered each year globally (Moyle and Cech, 2011; Fricke *et al*., 2023). Fricke *et al.* (2023) reported a total of 36,584 valid fish species, of which 18,574 are being freshwater. South Asian country, Nepal is home to 252 documented freshwater fishes including 18 endemic and 16 exotic species (Shrestha, 2019). Their systematic revision is highly necessary, however (Conway *et al.*, 2011; Shrestha, 2019). Although Nepal contributes about 0.1% of the total earth's surface, it accounts for 1.38% of the global freshwater fish (Fricke *et al.*, 2023). A total of 176, 193, and 152 fish species have been documented from the Koshi, Gandaki, and Karnali, respectively (Rajbanshi, 2012). They are three major river basins of the eastern, central, and western regions of Nepal.

Species diversity in communities can be achieved in several ways (see Whittaker, 1972; Pielou, 1966) and it represents the condition of organisms in a particular environment (Heda, 2009; Suvernaraksha *et al.*, 2012) such that the higher the diversity the more stable the community is in the ecosystem. Fish communities and their distribution in the lotic system are governed mainly by habitat variables, river geomorphology, and interacting biotic and abiotic variables

(Suvernaraksha *et al.*, 2012). Likewise, the availability of food and shelter regulates their occupancy at particular river sections (Tamonova *et al.*, 2007).

Twenty-five global biodiversity hotspots have been described which represent only 1.4% of the earth's surface (Myers *et al.*, 2000). Such hotspots are characterized by not only having outstanding endemism, but also are suffering from significant habitat loss (Myers *et al.*, 2000). The Koshi basin (river system) (see Shrestha *et al.*, 2023 for details) of Nepal lies within the biodiversity hotspot of the eastern Himalayan region (Shrestha *et al.*, 2009), so biodiversity research becomes crucial in this basin. However, previous studies have explored only a few major tributaries for fish diversity in this basin [e.g. Bajracharya, 2001; Shrestha *et al.*, 2009; Singh, 2017 (unpubl.); Jha *et al.* (2018); Tumbahangfe *et al.*, 2021; Shrestha *et al.*, 2023]. Dudhkoshi River (of Dudhkoshi sub-basin) is one of the major tributaries of the Koshi basin. Dudhkoshi sub-basin is threatened by 12 glacial lakes for possible outbursts due especially to climate change (Bajracharya and Mool, 2009). However, very little is known about fish diversity status of this river (see Shrestha *et al.*, 2023). The main aim of this study was to investigate fish diversity status by incorporating species richness, diversity indices, and conservation concerns. As taxonomic data is essential for the conservation of fish (Bhatt *et al.*, 2012; Suryaningsih *et al.*, 2018), we hope that the results of this study can contribute to effective conservation plans for fishes of Dudhkoshi River.

Materials and Methods

Study area

The present study was conducted in the Dudhkoshi River of eastern Nepal. It is one of the important tributaries of the Koshi River system, and as the latter has seven major tributaries, this river system is also called Sapta Koshi (sapta = seven) which eventually merges with the Ganges River system of India. The Dudhkoshi River flows through high altitudinal gradients (8848 to 465 masl) (Nepal, 2016) during its course from north to south. The upper stretches of this river have rocks and boulders as dominant substrates while the lower reaches have cobbles and pebbles. Some islands of sand bars could also be seen when water flow is comparatively lower (e.g. pre-monsoon), especially at downstream reaches. Habitat heterogeneity includes pools, riffles, runs, and cascades. The banks of the river are characterized by tall trees, shrubs, agricultural lands, bare lands, or sandy areas. Finally, the Dudhkoshi River joins another major tributary, the Sunkoshi River a few kilometers downstream of Jayramtar (s10).

Data collection

Fish sampling was conducted in two seasons (making four collections, hereafter four study seasons) for a period of two years (Post-monsoon 2020 to Pre-monsoon 2022). For data collection, 10 sampling sites were fixed: Jubhing (s1), Dibli (s2), Barkhughat (s3), Maikupul (s4), Tuintar (s5), Rabuwa bazar (s6), Rawa confluence (s7), Ghopatar (s8), Karki bensi (s9), and Jayramtar (s10) (Table 1). A combination of fishing gears was used (e.g. Jang *et al.*, 2003; Hossain *et al.*, 2012; Negi and Mamgain, 2013; Singh *et al.*, 2021) and details of fish sampling (and measurement of water quality parameters) are available (Shrestha *et al.*, 2023).

Table 1 Ten sampling sites of Dudkoshi River with their elevation, latitude, and longitude

Sites (code)	Elevation (masl)	Latitude (Deg. min.)	Longitude (Deg. min.)
Jubhing (s1)	1486	27° 35.68446'	86° 40.71786'
Dibli (s2)	876	27° 28.05042'	86° 43.41462'
Barkhughat (s3)	757	27° 26.07558'	86° 42.06204'
Maikupul (s4)	632	27° 23.19648'	86° 40.83978'
Tuintar (s5)	591	27° 21.87631'	86° 40.74408'
Rabuwa bazar (s6)	475	27° 16.27698'	86° 40.21488'
Rawa confluence (s7)	448	27° 16.11042'	86° 39.64422'
Ghopatar (s8)	388	27° 12.83011'	86° 32.66544'
Karki bensi (s9)	371	27° 12.26496'	86° 32.18611'
Jayramtar (s10)	358	27° 10.65426'	86° 28.47222'

Fish Identification and preservation

The collected fish were sorted out, counted, and identified in the field (as far as possible). The unidentified fishes were brought to the laboratory of the Central Department of Zoology, Kirtipur, Kathmandu. While some representative specimens were preserved in preservatives (e.g. 10% formalin), the rest were released back to the sites of their capture. For identification, morphometric, meristic, and other characters were followed (Day, 1878; Shrestha, 1981; Talwar & Jhingran, 1991; Jayaram, 2010; Shrestha, 2019). Online portals like FishBase (Froese and Pauly, 2023) and Catalogue of Fishes (Fricke *et al.*, 2023) were also followed where applicable.

Data analysis

Fish diversity status was assessed through Shannon-Wiener's diversity index (H′) (Shannon, 1949), Margalef's richness index (d) (Margalef, 1968), Pielou's evenness index (J) (Pielou, 1966) and Simpson's index of diversity (1-D)

(Simpson, 1949). Relative abundance (RA) was also calculated to assess the dominancy of the collected fish species. The species accumulation curves were created to inspect whether the sampling efforts were sufficient. We used PAlentological Statistics (PAST, version 4.03; Hammer *et al.*, 2001) and Microsoft Excel 2010 for data analysis.

Results and Discussion

Species diversity

Twenty-two species belonging to 13 genera, eight families, and three orders, making a total of 1,265 individuals have been recorded in this study (Table 2, 3). Nineteen species under eight families were recorded in Pre-monsoon 2021, followed by 17 species under six families (Pre-monsoon 2022) and 16 species under six families (Post-monsoon 2021). The minimum species, 14 under five families were recorded in Post-monsoon 2020 (Table 3, Fig. 2). In overall, the number of species (species richness) was higher in Pre-monsoons than in Post-monsoons which may be associated with several environmental parameters.

Table 2 Fishes of the Dudhkoshi River (* = NRDB status)

S.N.	Order	Family	Species
1.	Anabantiformes	Channidae	*Channa barca* (Hamilton, 1822)
2.	Cypriniformes	Botiidae	*Botia dario* (Hamilton, 1822)
			Botia histrionica Blyth, 1860
			Botia lohachata Chaudhuri, 1912
		Cyprinidae	*Garra annandalei* Hora, 1921
			Garra gotyla (Gray, 1830)
			Garra lamta (Hamilton, 1822)
			Labeo dyocheilus (McClelland, 1839)
			**Neolissochilus hexagonolepis* (McClelland, 1839)
			Puntius sophore (Hamilton, 1822)
			**Schizothorax progastus* (McClelland, 1839)
			**Schizothorax richardsonii* (Gray, 1832)
		Danionidae	*Barilius barila* (Hamilton, 1822)
			Barilius bendelisis (Hamilton, 1807)
			Barilius shacra (Hamilton, 1822)
		Nemacheilidae	*Schistura multifasciata* (Day, 1878)
		Psilorhynchidae	*Psilorhynchus homaloptera* Hora and Mukerji, 1935
			**Psilorhynchus pseudecheneis* Menon and Datta, 1964
3.	Siluriformes	Schilbidae	*Eutropiichthys murius* (Hamilton, 1822)
		Sisoridae	*Glyptothorax cavia* (Hamilton, 1822)
			Glyptothorax pectinopterus (McClelland, 1842)
			Pseudecheneis eddsi Ng, 2006

Table 3 Season-wise composition of fish abundance of Dudhkoshi River

Species	Seasons (No. of species)			
	Post-monsoon 2020 (14)	Pre-monsoon 2021 (19)	Post-monsoon 2021 (16)	Pre-monsoon 2022 (17)
Barilius barila	17	31	31	6
Barilius bendelisis	20	21	50	21
Barilius shacra	--	28	16	11
Botia dario	--	8	--	--
Botia histrionica	4	7	16	7
Botia lohachata	4	6	9	6
Channa barca	--	6	--	--
Eutropiichthys murius	--	1	--	4
Garra annandalei	--	24	--	--
Garra gotyla	--	14	14	12
Garra lamta	8	--	40	13
Glyptothorax cavia	--	21	--	5
Glyptothorax pectinopterus	11	28	29	14
Labeo dyocheilus	26	43	34	25
Neolissochilus hexagonolepis	10	14	21	14
Pseudecheneis eddsi	7	21	15	11
Psilorhynchus homaloptera	6	12	7	19
Psilorhynchus pseudecheneis	18	21	20	18
Puntius sophore	1	--	--	--
Schistura multifasciata	--	--	1	--
Schizothorax progastus	39	42	42	18
Schizothorax richardsonii	64	59	49	25
Total	**235**	**407**	**394**	**229**

Table 4 Total and relative abundances of fish species of Dudhkoshi River

Species	Total Abundance	Relative abundance (%)
Barilius barila	85	6.72
Barilius bendelisis	112	8.85
Barilius shacra	55	4.35
Botia dario	8	0.63
Botia histrionica	34	2.69
Botia lohachata	25	1.98
Channa barca	6	0.47
Eutropiichthys murius	5	0.40
Garra annandalei	24	1.90
Garra gotyla	40	3.16
Garra lamta	61	4.82
Glyptothorax cavia	26	2.06
Glyptothorax pectinopterus	82	6.48
Labeo dyocheilus	128	10.12
Neolissochilus hexagonolepis	59	4.66
Pseudecheneis eddsi	54	4.27
Psilorhynchus homaloptera	44	3.48
Psilorhynchus pseudecheneis	77	6.09
Puntius sophore	1	0.08
Schistura multifasciata	1	0.08
Schizothorax progastus	141	11.15
Schizothorax richardsonii	197	15.57
Total	**1265**	**100**

Figure 1 Map of the study area

Species like *Barilius barila*, *B. bendelisis*, *Botia histrionica*, *B. lohachata*, *Glyptothorax pectinopterus*, *Labeo dyocheilus*, *Neolissochilus hexagonolepis*, *Pseudecheneis eddsi*, *Psilorhynchus homaloptera*, *P. pseudecheneis*, *Schizothorax progastus* and *S. richardsonii* were recorded from all study seasons (Table 3). They represented about 55% of the total species recorded during the study. Three species, *Barilius shacra*, *Garra gotyla*, and *G. lamta* were captured in three study seasons. Similarly, *Eutropiichthys murius* and *Glyptothorax cavia* were recorded from only two study seasons. The rare species were *Botia dario*, *Channa barca*, *Garra annandalei*, *Puntius sophore*, and *Schistura multifasciata* which were captured in only one study season (Table 3). Gear choice, mesh size of a net, and time of sampling may be directly associated with fish capture (Ahmed and Hambrey, 2005). We used both active (e.g. cast net) and passive (e.g. gill net) fishing gears during the study.

Exotic species reported in the Koshi basin include *Hypophthalmichthys nobilis* (big-head carp), *Clarias gariepinnus* (African catfish), *Ctenopharyngodon idella* (grass carp), *Cyprinus carpio* (common carp), *Hypophthalmichthys molitrix* (silver carp), *Oreochromis niloticus* (Tilapia), and *Pangasius* sp. (Pangas) (e.g. Doody *et al.*, 2016), but we could not find any of them during this study, which may be an indication of pristine nature of riverine habitat (Vishnu *et al.*, 2023). Previous studies reported five endemic species in this basin: *Batasio macronotus*, *Erethistoides ascita*, *Pseudecheneis crassicauda*, *Psilorhynchus pseudecheneis*, and *Turcinoemacheilus himalaya* (Rajbanshi, 2012). We collected *Psilorhynchus pseudecheneis* from all study seasons whose relative abundance was recorded as 6.09%. The same species was also reported by Shrestha *et al.* (2009) as a dominant species in the Tamor River.

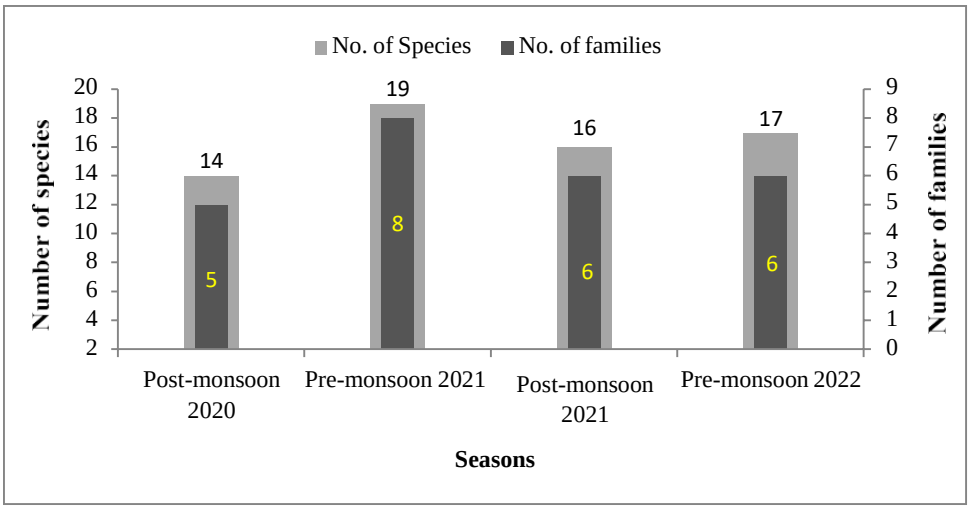
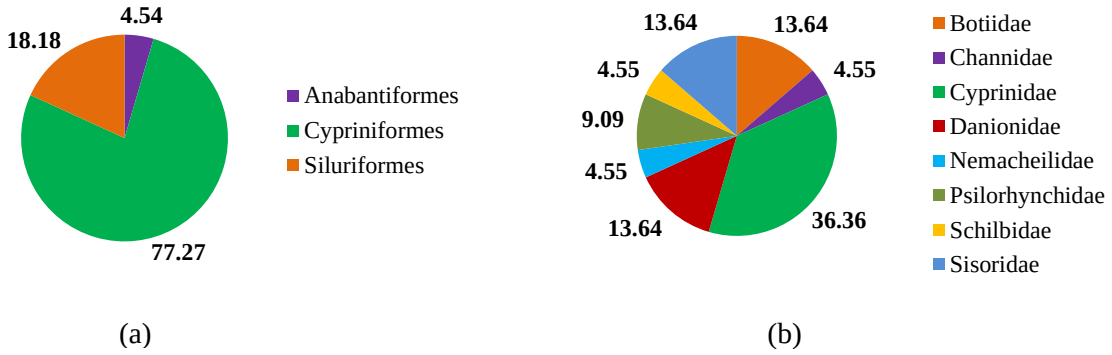


Figure 2 Number of species and families recorded in two seasons in Dudhkoshi River

Like Dudhkoshi, Tamor is also a snow-fed major tributary of the Koshi basin. Some previous fish diversity research conducted in Tamor in recent past includes Shrestha *et al.* (2009), Jha *et al.* (2018), and Tumbahangfe *et al.* (2021). They reported a total of 30, 13, and 28 species, respectively. Variations of species richness may be associated with sampling procedures, time of sampling, and duration of the study. Cypriniformes and Cyprinidae were recorded as the dominant order and family in the present study, and accounted for 77.27% and 36.36%, respectively (Fig. 3a, b). The overall composition in terms of species richness (primary y-axis) and families (secondary y-axis) in Post-monsoons and Pre-monsoons is shown in Fig 2. Our results are in line with Ward-Campbell *et al.* (2005), Shrestha *et al.* (2009), Shahnawaz *et al.* (2010), Mishra and Baniya (2016), Shrestha (2016), Jha *et al.* (2018), Shrestha *et al.* (2023) and Vishnu *et al.* (2023). While Shrestha *et al.* (2009) reported the dominancy of Cypriniformes and Cyprinidae as 87% and 61%, Vishnu *et al.* (2023) reported the dominancy of Cypriniformes and Cyprinidae as 60% and 53%, respectively.

The species we collected like *B. barila*, *B. bendelisis*, *G. gotyla*, *N. hexagonolepis*, *P. pseudecheneis* and *S. richardsonii* were also reported by Shrestha *et al.* (2009) and Jha *et al.* (2018), but species in our collections like *B. dario*, *B. histrionica*, *B. lohachata*, *G. annandalei*, *S. multifasciata*, and *S. progastus* were common only with Shrestha *et al.* (2009). While *S. richardsonii* was the most dominant species in our study, *P. pseudecheneis* and *Schistura beavani* were the dominant species in Shrestha *et al.* (2009) and Jha *et al.* (2018), respectively. It is widely accepted that because of possessing diverse mouth structures and the presence of adaptive body morphology, Cyprinids are widely distributed in all possible habitats in rivers and streams (Ward-Campbell *et al.*, 2005). For example, the authors reported that about 40% of the species composition belongs to Cyprinidae in the Southeast Asian watershed.

Besides Cyprinidae, each of the Botiidae, Danionidae, and Sisoridae accounted for (13.64%), Psilorhynchidae (9.09%), and each of the Schilbidae, Channidae, and Nemacheilidae accounted for only 4.55% (Fig. 3b). Species under five families (Botiidae, Cyprinidae, Danionidae, Psilorhynchidae and Sisoridae) were found in all study seasons, but species under Schilbidae was recorded in two study seasons (Pre-monsoon 2021 and Pre-monsoon 2022). Likewise, species under Nemacheilidae were found in Post-monsoon 2021 and Channidae in Pre-monsoon 2021. Fishes of Nepal are either long-distance migratory, short-distance migratory, or resident species (Gubhaju, 2011). In this study, short-distance migratory species (e.g. *Schizothorax* spp., *Barilius* spp., *L. dyocheilus*, *N. hexagonolepis*) and resident species (e.g. *Garra* spp.) were captured, but could not collect any long-distance migratory species.



(a) (b)

Figure 3 Fish composition (%) of Dudhkoshi River: (a) Order-wise and (b) Family-wise

Relative abundance

The abundance here reflects the number of individuals captured based on the length or areas of the river sampled, and the relative abundance (RA) of a species indicates its relative proportion. To assess the dominance of fishes in the Dudhkoshi River, we calculated the relative abundance of each species. Although the relative abundance of species greatly varied in four study seasons (Table 4), in overall, the highest relative abundance was observed in *S. richardsonii* which was 15.57%. Because snow trout prefer wide range of habitats including riffle, run and pool (Singh and Agrawal, 2013), their capture is very likely in comparison to other species which prefer specific habitat types. Among study seasons, its highest RA was respectively, 27.23% (Post-monsoon 2020), 14.50% (Pre-monsoon 2021), and 12.44% (Pre- monsoon 2022). But RA of *B. bendelisis* was highest in Post-monsoon 2021. The next species with higher RA (in overall) was *S. progastus* (11.15%), followed by *L. dyocheilus* (10.12%), *B. bendelisis* (8.85%), *B. barila* (6.72%), *G. pectinopterus* (6.48%) and *P. pseudecheneis* (6.09%). While nine species have RA less than 5%, five species have even less than 1% (see Table 4). Habitat preferences are highly specific in fish (Singh and Agrawal, 2013). So, variability of catch rate was also influenced by fishing schedule, distance, and month (Murillo-Posada *et al.*, 2019). They also highlighted the combinations of environmental, spatial, and temporal factors are mainly associated with variation in the relative abundance of species.

Diversity indices

Several measures are used to determine ecological diversity, or community diversity or species diversity (Izsák and Popp, 2000). For example, Dominance, Evenness, Margalef, and Shannon-Wiener are commonly used indices to examine the status of aquatic animals (Kindong *et al.*, 2020), of which H′ and 1/D are the most widely used diversity indices (Ricotta, 2005). Previous studies claimed that food resources, environment, and seasons can influence diversity indices (Hossain *et al.*, 2012).We used captured data to compare species richness and diversity indices. The diversity indices of fishes of the Dudhkoshi River are shown in Fig. 4. The highest average value of the Shannon diversity index (H′) was found in the Pre-monsoon of 2022 (1.74±0.199), followed by the Pre-monsoon of 2021 (1.65±0.212) and Post-monsoon of 2021 (1.52±0.231). The lowest value was observed in the Post-monsoon of 2020 (1.33±0.164). Values of H′ are associated with abundance and relative abundance of homogeneity of species (Kallianiotis *et al*., 2000), here we found higher H′ in Pre-monsoons than Post-monsoons, comparatively. However, H′ was not statistically significant in four study seasons (F=0.71, p=0.548). Our results are not in accordance with Jha *et al.* (2018), Tumbahangfe *et al.* (2021), andVishnu *et al.* (2023) who reported comparatively higher values of H′ including 2.21, 2.88, and 3.12, respectively.

The average Simpson's index of diversity (1-D) was also highest in Pre-monsoon 2022 (0.77±0.221), followed by Pre-monsoon 2021 (0.76±0.047), and Post-monsoon 2021 (0.71±0.07). The lowest value (0.69±0.044) was found in Post-monsoon 2020. Similar to the Shannon diversity index, Simpson's index of diversity was also not statistically significant (F=0.502, p=0.682). Our results are not in line with Tumbahangfe *et al.* (2021) and Vishnu *et al.* (2023). The former reported higher (0.93) and the latter reported lower (0.31) values than ours. The highest Margalef's richness index (d) was observed in Pre-monsoon 2022 (1.93±0.342), followed by Pre-monsoon 2021(1.5±0.288), and Post-monsoon 2021 (1.31±0.25). The lowest value of 'd' was found in Post-monsoon 2020 with 1.13±0.215. The value of d was also not statistically significant (F=1.561, p=0.215). Our results are not in line with Vishnu *et al.* (2023), who reported comparatively higher values (4.13) than ours (1.93).

The highest Pielou's evenness index (J) was found to be 0.95±0.009 and 0.95±0.253 in each of the Pre-monsoon 2021 and Pre-monsoon 2022, followed by Post-monsoon 2021 (0.94±0.022). The lowest value of 'J' was observed in Post-monsoon 2020 (0.93±0.016). The value of J however was not statistically significant in four study seasons (F=0.258, p=0.854). The results are not in accordance with Jha *et al.* (2018) and Tumbahangfe *et al.* (2021), who reported comparatively lower values, i.e. 0.74 and 0.64, respectively than ours (0.95). But, our results were very close to the value of Vishnu *et al.* (2023), which was 0.96. Previous studies revealed that flooding events have been associated with oscillations in evenness values (Vishnu *et al.*, 2023). All diversity indices were lowest in the Post-monsoon of 2020, comparatively.

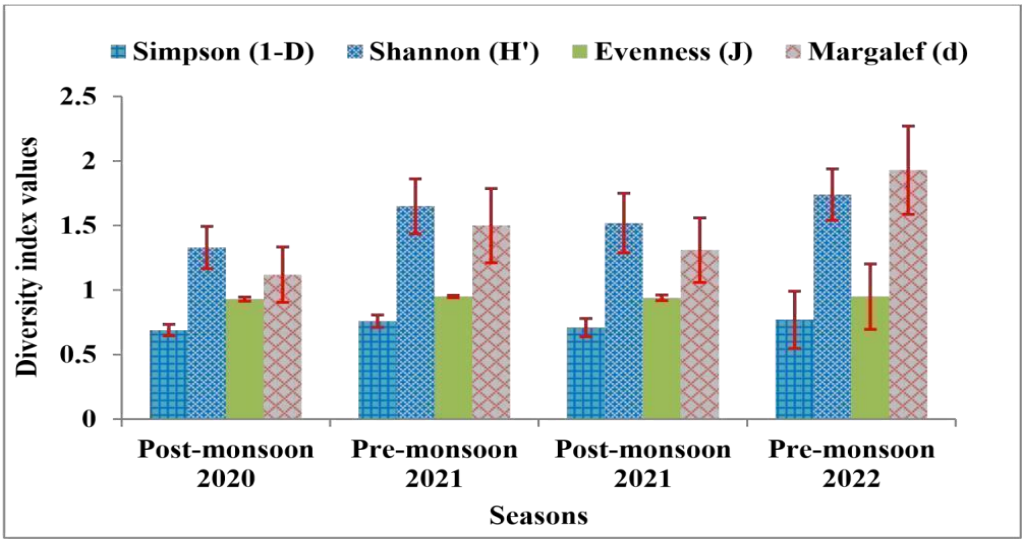


Figure 4 Fish diversity indices (Simpson, Shannon, Evenness, and Margalef) of the Dudhkoshi River

Species accumulation curve

The species accumulation curve represents a graph showing species numbers on the y-axis and sampling efforts on the x- axis (Colwell *et al.*, 2004). This curve tells whether the sampling efforts are sufficient or not in diversity research. In other words, such curves indicate the relationship between observed species and sampling efforts, the latter may be a number of samples, traps, or trap-days (Moreno and Halffter, 2000).

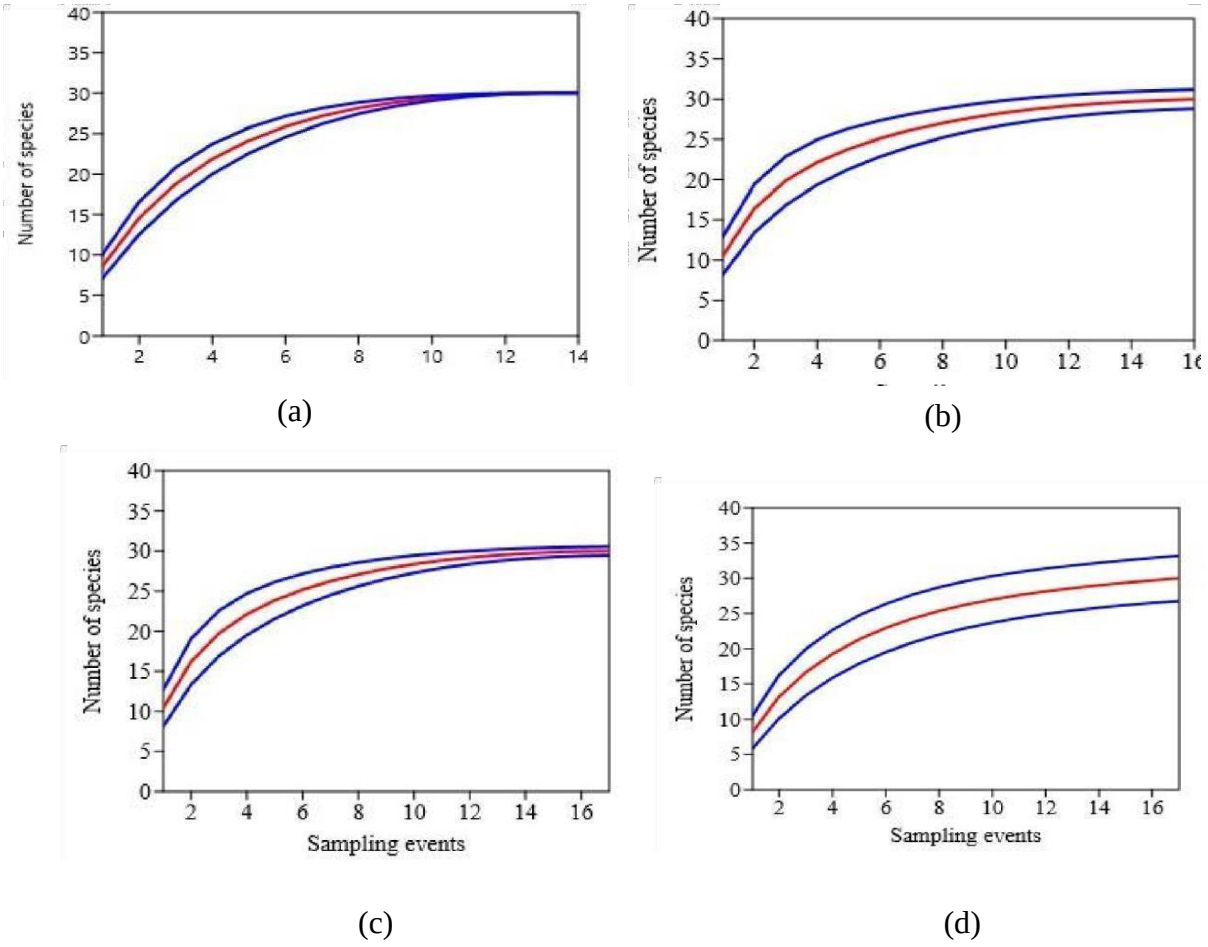


Figure 5 Species accumulation curves of two seasons (four collections) with mean and 95% confidence interval of the cumulative number of species (y-axis) for given number of samples (x-axis): (a) Post-monsoon 2020, (b) Post- monsoon 2021, (c) Pre-monsoon 2021 and (d) Pre-monsoon 2022

The species accumulation curves generated from the data collected in two seasons (in four collections: Post-monsoon 2020, Pre-monsoon 2021, Post-monsoon 2021, and Pre-monsoon 2022) are shown in Fig. 5. The red and blue lines indicate the accumulation curves and confidence intervals, respectively. The curves in all collections showed asymptotic conditions with slight variations. When the curve reaches to an asymptote, there is less possibility of adding more new species even if sampling efforts are increased (Moreno and Halffter, 2000). In this study, except Pre-monsoon of 2022, curves in other seasons are somehow well flattened towards the right side, which is an indication of enough sampling efforts during data collection. Additionally, it also informs that if more sample collections are made in the Pre-monsoon of 2022, results will be different in terms of the number of species turnover than the current situation. In that case, the accumulation curve of that season also becomes more flattened. The confidence interval of the Pre-monsoon of 2022 was comparatively wider than others, however (Fig.5).

Conservation status

Conservation of fish diversity plays an important role in ecological, nutritional, and socio-economic balance (Shams *et al.*, 2013). Unlike other vertebrates, the DNPWC (Dept. of National Parks and Wildlife Conservation) of Nepal has not enlisted any single fish species seeking protection, and no single species of the country has gone extinct, to date. However, ten species, *Anguilla bengalensis*, *Chagunius chagunio*, *Danio rerio*, *N. hexagonolepis*, *P. pseudecheneis*, *Schizothorax plagiostomus*, *S. progastus*, *S. richardsonii*, *Tor putitora* and *T. tor* have been proposed for protection in National Red Data Book (NRDB) status (Shrestha, 1995). Moreover, 35 fish species are included in NRDB status under different categories, and was based on the Biodiversity Profile Project (BPP), which endeavored to enumerate the fishes of Nepal (see Shrestha, 1995). While the majority of collected species in Dudhkoshi River belong to the Least concern (LC) (IUCN, 2022), accounting for 67% (see Shrestha *et al.*, 2023 for other categories), four species however belong to NRDB status (Table 2). Previous studies have also assigned different conservation statuses for the same fish, *Psilorhynchus pseudecheneis*, as evident in Jha *et al.* (2018) and Shrestha (2019). Such issues need to be resolved through systematic and meticulous investigation. As study about fish conservation status is scanty (but see Jha *et al*., 2018), there is a huge scope for future research in Nepal.

Conclusion

More fish species were recorded in Pre-monsoons than in Post-monsoons, comparatively. Based on Shannon-Wiener's diversity index (H′), the overall diversity status of Dudhkoshi River was categorized as medium. Cypriniformes and Cyprinidae were found as the dominant order and family in this study exhibiting the typical trend of high occurrence of Cyprinids in rivers and streams. Species such as *S. richardsonii*, *S. progastus*, *L. dyocheilus*, *B. bendelisis*, *B. barila*, *G. pectinopterus* and *P. pseudecheneis* were found as the most abundant species as per their relative abundances, from highest to lowest. On the other hand, *B. dario*, *C. barca*, *G. annandalei*, *P. sophore* and *S. multifasciata* were occasional (rare) species recorded. A large percentage of species collected in this study belongs to the LC category and only four species collected belongs to NRDB status. We hope that the results of this study can contribute to i) future researchers and ichthyologists, as baseline taxonomic data and ii) aquatic ecologists and conservationists, as reference data for proper management and conservation of fish species in Dudhkoshi River. Future research incorporating other seasons (e.g. winter and rainy) is highly recommended to further generalize the diversity status of the fishes of this river.

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