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Correlation of fish assemblages with habitat and environmental variables in the Phewa Khola Stream of Mangsebung Rural Municipality, Ilam, Nepal

Jash Hang Limbu[1*], Suren Subba[2], Jeevan Kumar Gurung[3], Jawan Tumbahangfe[4] and Bharat Raj Subba[5]

[1] *Nature Conservation and Health Care Council, Biratnagar, Nepal*
[2] *Dhankuta Multiple Campus, Tribhuvan University, Dhankuta, Nepal*
[3] *Damak Multiple Campus, Tribhuvan University, Damak, Jhapa, Nepal*
[4] *Central Department of Zoology, Tribhuvan University, Kirtipur, Kathmandu, Nepal*
[5] *Post Graduate Campus, Tribhuvan University, Biratnagar, Nepal*
[*] *Corresponding author* ✉: limbujash@gmail.com

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Abstract

We assessed the correlation of fish assemblages with habitat and environmental variables temporally from July and October, 2019 and January and April, 2020 across 5 study sites in the Phewa Khola stream of Mangsebung Rural Municipality, Ilam, Nepal. We sampled 3571 fish representing 13 species, belonging to 3 orders, 4 families, and 9 genera. An analysis of similarity (ANOSIM) indicated that there is a significant difference between the fish assemblage structure in space (R= 0.833, P= 0.001) but not in time (R= -0.148, P= 0.985). Our habitat study showed that glides, runs, pools and deep pools are the primary habitats contributing to the maximum diversity in the Phewa Khola stream. The canonical correspondence analysis (CCA) affirmed that variables such as pH, water temperature, water velocity, total hardness and dissolved oxygen play an important role in shaping fish species distribution. Results from the similarity percentage analysis (SIMPER) hinted that, 67.08% similarity was found between the months and the major contributing species were *Schistura multifasciata* (20.61%), *Devario aequipinnatus* (16.48%), *Schistura rupecula* (15.65%), *Garra annandalei* (15.36%), *Schistura horai* (7.74%), *Schistura scaturigina* (5.91%), *Schistura savona* (5.74%), *Schizothorax plagiostomus* (4.37%), *Channa punctata* (3.9%), *Puntius terio* (1.9%) and *Neolissochilus hexagonolepis* (1.39%). On the contrary, a 76.23% similarity was found between the sites and the major contributing species were *Schistura multifasciata* (21%), *Devario aequipinnatus* (16.8%), *Garra annandalei* (15.89%), *Schistura rupecula* (15.38%), *Schistura horai* (7.7%), *Schistura scaturigina* (5.66%), *Schistura savona* (4.9%), *Schizothorax plagiostomus* (4.4%), *Channa punctata* (3.97%), *Puntius terio* (2%) and *Neolissochilus hexagonolepis* (1.43%). Ongoing road development, micro-hydropower generation, the use of poisonous herbicides, illegal electro-fishing, deforestation and water diversion are all found to be major threats to the present fish species of the Phewa Khola stream.

Key words: Fish diversity, Falgunanda, habitat, stream, spatio-temporal

Introduction

River habitat generally refers to the physical structure of rivers, including the river-bed, bank and riparian canopy, and it is a key component of stream ecosystems, playing a major role in determining biotic assemblages and stream integrity (Newson and

Newson, 2000; Zeni and Casatti, 2014). Habitat alterations or modification of physical habitat can lead to brief, or long-lasting, changes in the composition of stream fish assemblages depending on the severity of the disturbances (Lammert and Allan, 1999). Riverine fish assemblages are structured by diverse habitats (Schlosser, 1991) and so, most rehabilitation measures attempt to re-establish the structural complexity lost by human activity impacts (Gore et al., 1995), such as stream clearing (Nilsson et al., 2005). Physio-chemical characteristics are crucial determinants of the condition of stream fish assemblages (Li et al., 2012). Habitat variables, such as water temperature (Kadye et al., 2008; Limbu et al., 2019a), depth and distance to source (Vlach et al., 2005), stream width (Gerhard et al., 2004), substrate (Vlach et al., 2005; Kadye et al., 2008; Limbu and Prasad, 2020), altitude (Magalhães et al., 2002), conductivity (Yu and Lee, 2002), climate (Menni et al., 2005) and chlorophyll-a abundance (Blanc et al., 2001) have all been shown to influence fish assemblages.

A review of the literature shows that the study of correlations between fish diversity, environmental variables and fish habitat aspects at different spatial and temporal scales in Nepal are very few (Mishra and Baniya, 2016; Pokharel et al., 2018; Limbu et al., 2018; 2019a; 2020; 2021). However, these studies did not mention which factors (physio-chemical factors, current velocity substrate composition, stream width, water temperature, water volume, etc.) contribute most to fish assemblage variations. Previous published literature on fisheries resources in the rivers of Nepal includes: Edds (1986), Shrestha et al. (2009), Shrestha (2016), Subba et al. (2017), Limbu et al. (2018, 2019a, 2019b, 2020, 2021) and Limbu and Prasad (2020). In Nepal, only a limited number of studies on river habitat and its correlation with biological communities are available. So, many ecological aspects of Nepal's fisheries habitats are yet to be discovered. In recent years, classification and assessment of river habitat have been widely used in, for example, the Liao River Basin, the Naoli River Watershed, the Dong River and the Kaligandaki River (Zheng et al., 2007; Wang et al., 2010; Wang et al., 2011; Pokharel et al., 2018).

The aim of this study is to assess the present status of fish community structure, diversity, and inter-relationships with habitat ecology and environmental variables in the Phewa Khola stream.

Material and Methods

Study area

The Phewa Khola stream is a tributary of the Deumai River and is situated at Mangsebung Rural Municipality in Ilam district, eastern Nepal (Fig. 1). The Dahagau and Khamnuwa are areas where this stream originates and surges eastwards thoroughly in between areas of Gajurmukhi, Dadhagau, Sangrang, Ivang and joins with the Deumai River at Gajurmukhidham. The water of this stream is used for drinking, irrigation, water mills and hydro-power generation. The vegetation bordering the stream is mixed, mostly consisting of coniferous forest and bamboo forest and the dominant stream substrata consist of boulders, cobbles, pebbles, gravel and sand. The Phewa Khola stream region experiences mostly sunny weather, with occasional clouds and the mean annual temperature is 19 °C.

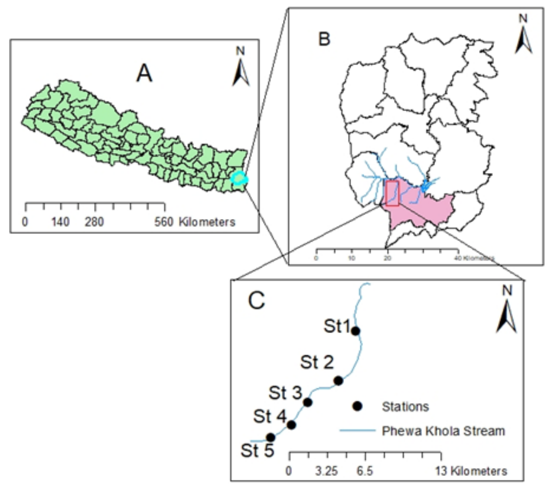

Figure 1: Map of study area of the Phewa Khola stream. A: Nepal including Ilam district, B: Ilam district including Mangsebung Rural Municipality, C: Phewa Khola stream with different sampling stations, St: station.

Sampling method

The study area was divided into five sampling sites (Fig. 1C): Chamlinge (station 1), Kholatar (station 2), Sangreng (station 3), Batta (station 4) and Devghat (station 5) for measuring hydrological parameters and collection of fish. Fish samples were collected based on different habitat representation from July 20–25, October 20–25, 2019 and January 20–25 and April 20–25, 2020. Based on the characteristics of the flow pattern, slope, average water velocity and substrate components in the study area (Han, 2010; Huang et al., 2019), the habitat type was categorized into three groups: lentic habitat (pool and deep pool), slow flow pattern (glide and run) and fast flow habitat (riffle and cascade).

The distance between two adjacent sampling sites was 3 km. Fish were collected either downstream (station 4, 5) or upstream (station 1, 2, 3) of each sampling site. Each sampling site was 200–250 m long with different habitat types present (pool, deep pool, glide, run, riffle and cascade). Mosquito nets, Ghorlang and Bamboo fish traps were used for collection and at the upstream sites (station 1, 2), the hand-picking method was also adopted to catch the fish because water volume was small and the stream bed was strewn with big boulders. Local fishermen were hired to collect all of the fish. Approximately 10% of the sampled fish were preserved in 10% formaldehyde solution in plastic jars as a reference collection. The remaining fish samples were returned to the habitat from where they were captured after photography. The identification was done with the help of standard taxonomic references (Talwar and Jhingran, 1991; Jayaram, 2010; Shrestha, 2019).

During fieldwork, the in situ environmental variables were measured at each sampling site. The water temperature (°C) was measured with a digital thermometer placed in the water to a depth of 1 foot for two to three minutes. Dissolved oxygen (DO) (mg/l) was measured by the Winkler titra-metric method. The pH was measured using a pH meter (HI 98107, HANNA Instrument). Water velocity was measured by the float method with the help of a stopwatch, plastic ball and measuring tape. The total hardness (mg/l) was determined using the EDTA titra-metric method.

Data analysis

One-way analysis of variance (ANOVA) was used for temperature, pH, dissolved oxygen, hardness and water velocity to calculate the existence of any differences between space and time. A post-hoc Tukey HSD test was used to test which means were significantly different at a 0.05 level of probability (Spjøtvoll and Stoline, 1973). One-way analysis of similarities (ANOSIM) (Clarke, 1993) was used to test the significant differences between the spatial and temporal scales.

To visualize the major contributing species both in space and time, a similarity percentage (SIMPER) (Clarke, 1993) analysis was performed. To visualize the differences in the fish assemblage structure between habitat types, a non-metric multidimensional scaling (NMDS) (R Core Team, 2018) was performed. Samples by species, sites and environmental variables were analyzed through multivariate analysis tools. Detrended correspondence analysis (DCA) (Gauch, 1982) 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, sites and environmental variables. The value of axis length and eigen values obtained from the DCA suggested that the linear model associated with CCA was more applicable. Therefore, a direct multivariate ordination method (Legendre and Legendre, 1998), based on a linear response of species to environmental gradients, was applied.

To test the habitat relationships, Principal Component analysis (PCA) was performed.

All the statistical analysis were performed in the R software 2.5-6 version.

Results

Fish assemblage structure

A total of 3571 fish individuals were collected during the study period, belonging to 3 orders, 4 families, 9 genera and 13 species (Tables 1 and 2; Appendix). Among these, the Cypriniformes was the most species rich order accounting for 75%, followed by Siluriformes 15% and Perciformes 10% of the total fish species (Fig. 2). Cyprinidae and Cobitidae were the most abundant families which contributed equally (38.46%) followed by Sisoridae 15.38% and Channidae 7.69% (Fig. 3).

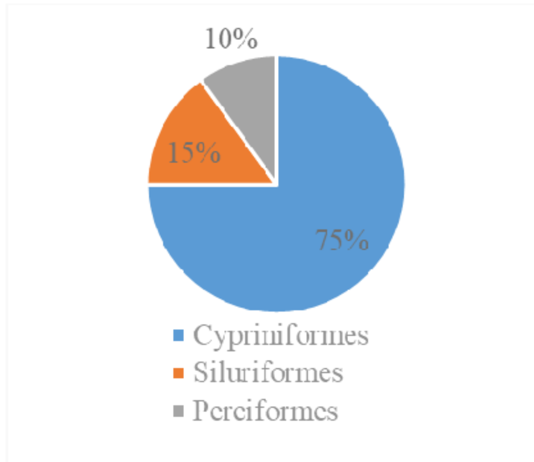

Figure 2: Percentage composition of fish species by order.

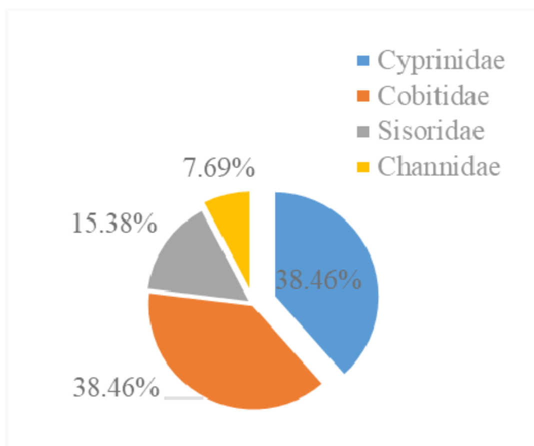

Figure 3: Percentage composition of fish species by family.

According to similarity percentage (SIMPER) analysis, 67.08% similarity was found between the months and the major contributing species were: *Schistura multifasciata* (20.61%), *Devario aequipinnatus* (16.48%), *Schistura rupecula* (15.65%), *Garra annandalei* (15.36%), *Schistura horai* (7.74%), *Schistura scaturigina* (5.91%), *Schistura savona* (5.74%), *Schizothorax plagiostomus* (4.37%), *Channa punctata* (3.9%), *Puntius terio* (1.9%)

and *Neolissocheilus hexagonolepis* (1.39%). On the other hand, 76.23% similarity were found between the sites and the major contributing species were: *S. multifasciata* (21%), *D. aequipinnatus* (16.8%), *G. annandalei* (15.89%), *S. rupecula* (15.38%), *S. horai* (7.7%), *S. scaturigina* (5.66%), *S. savona* (4.9%), *S. plagiostomus* (4.4%), *C. punctata* (3.97%), *P. terio* (2%) and *N. hexagonolepis* (1.43%) (Table 3).

The analysis of ANOSIM indicated that there is a significant difference between the fish assemblage structure in space (R = 0.833, P = 0.001) but not in time (R = -0.148, P = 0.985).

Table 1: List of fish species recorded from the Phewa Khola stream, Ilam district, Nepal.

Order	Family	Code	Species
Cypriniformes	Cyprinidae	dani_aqu	*Devario aequipinnatus* (McClelland, 1839)
		garra_ana	*Garra annandalei* Hora, 1921
		neo_hex	*Neolissochilus hexagonolepis* (McClelland, 1839)
		pun_ter	*Puntis terio* (Hamilton, 1822)
		sciz_plag	*Schizothorax plagiostomus* Heckel, 1838
	Cobitidae	shi_hor	*Schistura horai* (Menon, 1952)
		schi_mul	*Schistura multifasciata* (Day, 1878)
		schi_rup	*Schistura rupecula* McClelland, 1838
		schi_sca	*Schistura scaturigina* McClelland, 1839
		schi_sov	*Schistura savona* (Hamilton, 1822)
Siluriformes	Sisoridae	euc_hod	*Euchiloglanis hodgarti* (Hora, 1923)
		glyp_tri	*Glyptothorax trilineatus* Blyth, 1860
Perciformes	Channidae	chan_punc	*Channa punctata* (Bloch, 1793)

Table 2: List of fish species and their distribution in different habitat types of Phewa Khola stream, Ilam, Nepal.

Family	Species	Pool	Deep pool	Glide	Run	Riffle	Cascade
Cyprinidae	*Devario aequipinnatus*	300	200	71	50	0	0
	Garra annandalei	0	0	9	50	300	200
	Neolissochilus hexagonolepis	0	0	10	12	25	2
	Puntius terio	10	26	18	0	0	0
	Schizothorax plagiostomus	0	10	20	13	60	7
Cobitidae	*Schistura horai*	170	81	51	0	0	0
	Schistura multifasciata	400	200	73	67	0	0
	Schistura rupecula	190	150	100	88	66	0
	Schistura scaturigina	160	20	11	10	0	0
	Schistura savona	100	55	57	0	0	0
Sisoridae	*Euchiloglanis hodgarti*	0	0	0	0	7	3
	Glyptothorax trilineatus	0	0	0	0	4	10
Channidae	*Channa punctata*	80	25	0	0	0	0
Total		1410	767	420	290	462	222

Table 3: Average similarity (%) and the discriminating fish species in each month and site using SIMPER analysis of the Phewa Khola stream, Ilam district, Nepal.

Month (67.08%)		**Site (76.23%)**	
Contributory species	**%**	**Contributory species**	**%**
Schistura multifasciata	20.61	*Schistura multifasciata*	21
Devario aequipinnatus	16.48	*Devario aequipinnatus*	16.8
Schistura rupecula	15.65	*Garra annandalei*	15.89
Garra annandalei	15.36	*Schistura rupecula*	15.38
Schistura horai	7.74	*Schistura horai*	7.70
Schistura scaturigina	5.91	*Schistura scaturigina*	5.66
Schistura sovana	5.74	*Schistura savona*	4.93
Schizothorax plagiostomus	4.37	*Schizothorax plagiostomus*	4.40
Channa punctata	3.9	*Channa punctata*	3.97
Puntius terio	1.95	*Puntius terio*	2
Neolissochilus hexagonolepis	1.39	*Neolissochilus hexagonolepis*	1.43

With regard to habitats, 39.48% (1410) of the individuals, belonging to 2 orders, 3 families, 4 genera and 8 species, were collected in the pools. The most abundant species were *Schistura multifasciata* (28.36%), *Devario aequipinnatus* (21.27%) and *Schistura rupecula* (13.47%). In the deep pools, 21.53% (767) were collected, belonging to 2 orders, 3 families, 5 genera and 9 species. The most abundant species were *S. multifasciata* (26.07%), *D. aequipinnatus* (26.07%) and *S. rupecula* (19.53%).

In the glide areas, 11.76% (420) specimens were collected, belonging to 1 order, 2 families, 6 genera and 10 species. The most abundant species were *S. rupecula* (23.8%) and *S. multifasciata* (17.38%). In the runs, 8.12% (290) were collected, belonging to one order, 2 families, 5 genera and 7 species. The most abundant species were *S. rupecula* (30.34%) and *S. multifasciata* (23.1%). In the riffles, 12.93% (462) were collected, belonging to 2 orders, 3 families, 6 genera and 5 species. The most abundant species was *Garra annandalei* (64.93%). In the cascades, 6.21% (222) specimens were collected, belonging to 2 orders, 2 families, 5 genera and 5 species. The most abundant species was *G. annandalei* (90.09%).

Results of NMDS showed that fish assemblage structure differed between pool and cascade habitats (ANOSIM, $P < 0.001$) but not all assessed habitats (Fig. 4).

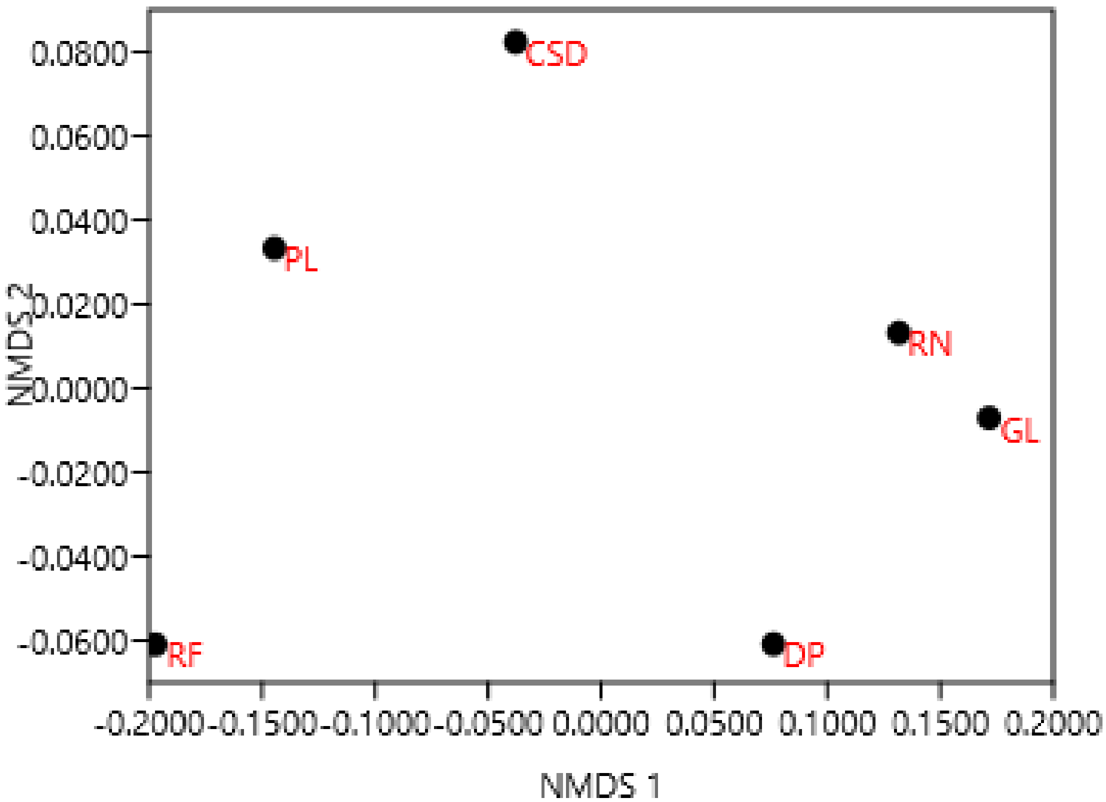

Figure 4: Non-metric Multidimensional Scaling (NMDS) ordination of fish assemblages in different habitat types of the Phewa Khola stream, Nepal (RN = run, RF = riffle, CSD = cascade, PL = pool, DP= deep pool, GL = glide).

Results of PCA affirmed that *G. annandalei* is positively related to riffle and cascade but *Channa punctata*, *Schistura horai* and *S. scaturigina* are negatively related to these habitats. The occurrence of *Devario aequipinnatus*, *S. multifasciata* and *S. rupecula*, are all highly associated with run, glide, deep pool and pool habitats. *Schizothorax plagiostomus*, *Schistura rupecula*, *Neolissochilus hexagonolepis*, *Euchiloglansis hodgarti*, *Glyptothorax trilineatus*, *S. scaturigina*, *S. horai* and *Channa puctata* are all negatively related to the select habitat types (Fig. 5).

Correlations between fish species and environmental variables

The fish species, *Schistura rupecula* and *Channa punctata* are positively related to water temperature and water velocity but negatively related to pH (Fig. 6). Occurrence of *Schizothorax plagiostomus*, *Schistura scaturigina*, *Schistura savona* and *Euchiloglansis hodgarti* are highly associated with total hardness and dissolved oxygen (Fig. 6). *Glyptothorax trilineatus*, *Schistura horai* and *Schistura multifaciata* are positively related to pH but negatively related to water temperature and water velocity and *Puntius terio*, *Garra annandalei*, *Neolissochilus hexagonolepis* and *Danio aequipinnatus* are not positively related to any variables but are negatively related to total hardness and dissolved oxygen (Fig. 6).

The RDA affirmed that water quality parameters of pH, water temperature, water velocity, total hardness and dissolved oxygen play an important role in shaping the fish assemblage structure of Phewa Khola stream (Fig. 6).

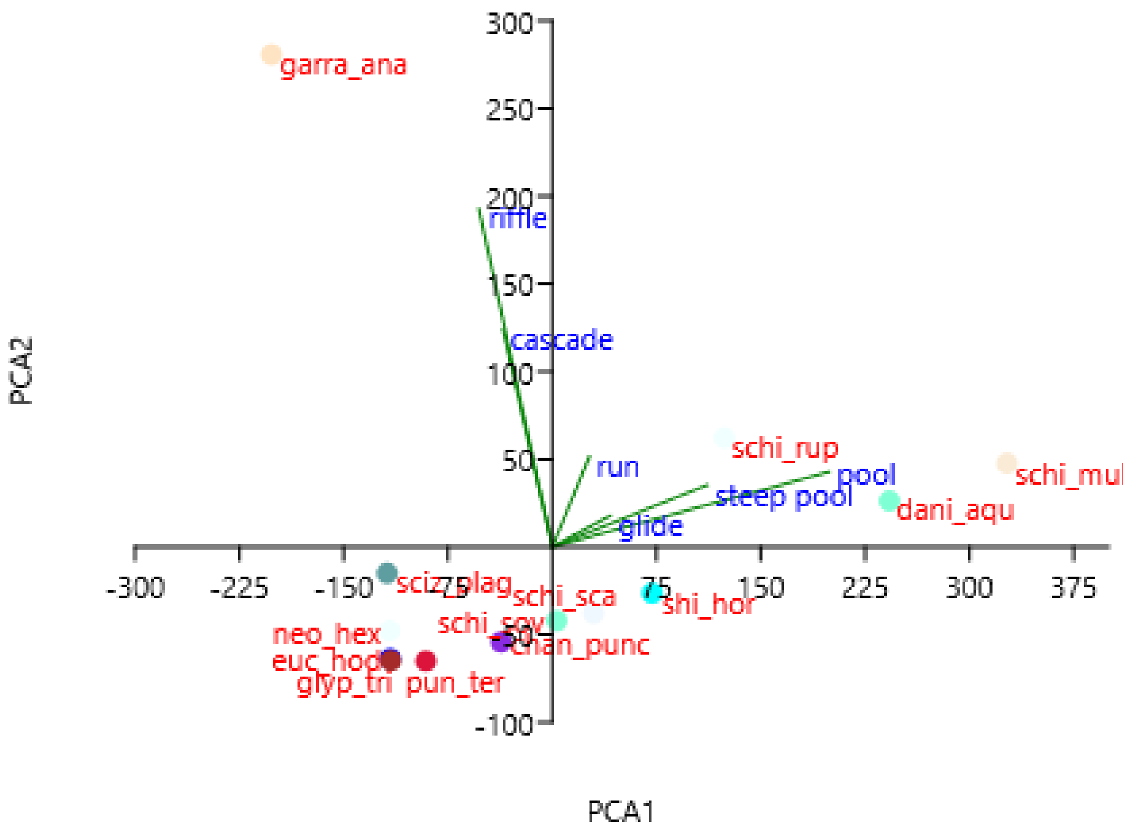

Figure 5: Principal Component analysis (PCA) of the habitats evaluated in the Phewa Khola stream (for species code see Table 1).

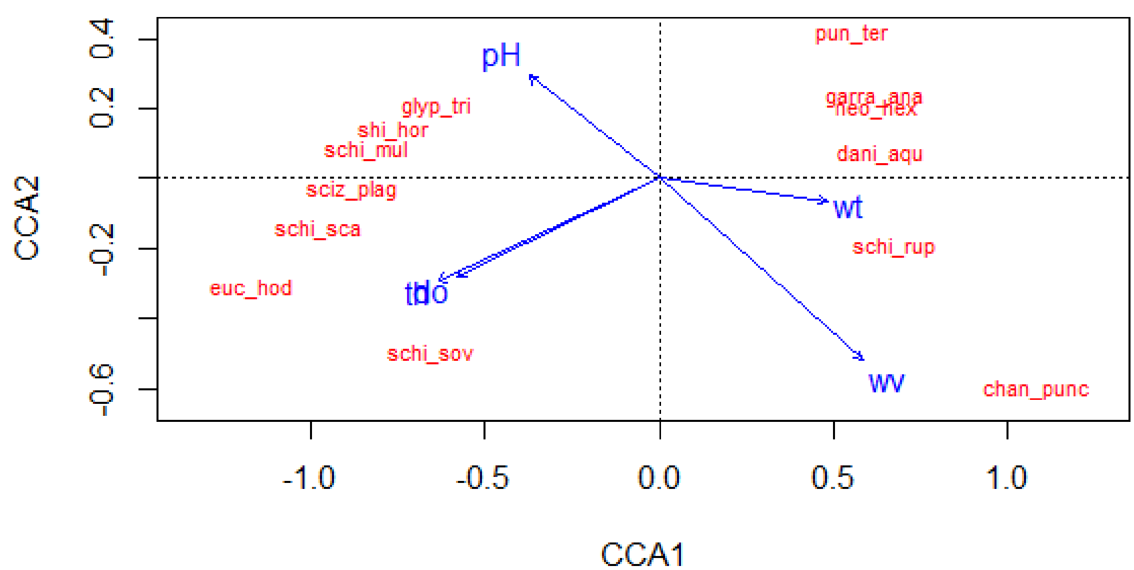

Figure 6: Canonical correspondence analysis (CCA) ordination between fish assemblage and environmental variables (for species code see Table 1) (th = total hardness, do = dissolved oxygen, wt = water temperature and wv = water velocity).

Discussion

A total of 13 fish species were recorded during the study period. Of these, on the basis of the similarity percentage analysis (SIMPER), the species *Schistura multifasciata*, *Devario aequipinnatus*, *S. rupecula*, *Garra annandalei*, *S. horai*, *S. scaturigina*, *S. sovana*, *Schizothorax plagiostomus*, *Channa punctata*, *Puntius terio* and *Neolissochilus hexagonolepis* were the major contributing species. The diversity is 4 species greater than that reported by Prasad and Limbu (2017) from the same stream. This might be due to the limited study areas covered in this earlier study and the fact that the fish were collected simply by throwing a cast net into the stream without sampling the different habitat types. The most diverse order and family sampled were the Cypriniformes and Cyprinidae, respectively. This result is consistent with the findings of several previous studies (Shrestha et al., 2009; Mishra and Baniya, 2016; Shrestha, 2016; Subba et al., 2017; Yadav, 2017; Limbu et al., 2018, 2019a, b, c; Limbu and Gupta, 2019; Punam and Limbu, 2019; Chaudhary et al., 2020; Limbu et al., 2020; Limbu and Prasad, 2020; Prasad et al., 2020) and the fact that the majority of freshwater fish are in the order Cypriniformes and family Cyprinidae (Nelson, 2006).

The analysis of similarity (ANOSIM) showed significant differences in the species assemblage in space (*R*= 0.833, *P*= 0.001) but not in time (*R*= -0.148, *P*= 0.985) which is similar to findings of Yan et al. (2010) and Punam and Limbu (2020).

Different habitats are essential for diverse fish assemblages (Huang et al., 2019). The highest number of individual fish were collected from the pool and deep pool habitat types. The total species richness in pool and deep pool habitats play a pivotal role in the breeding and growth for the fish community (Espírito-Santo and Zuanon, 2017; Favrot et al., 2018). Many species preferred those types of habitat, such as the multiple *Schistura* species, *Devario aequipinnatus*, *Channa punctata*, *Schizothorax plagiostomus* and *Puntius terio*.

In this study, we have reported the highest number of fish species (diversity) from the slow-flow habitats (glides and runs) (Table 2), but Huang et al. (2019) reported the highest number of species from the riffle habitat type. Fish species such as *Schistura* species, *Devario aequipinnatus*, *Neolissochilus hexaqgonolepis*, *Garra annandalei*, *Schizothorax plagiostomus* and *Puntius terio* preferred the glide and run habitat types. The fast-flow habitats (riffles and cascades), with highest water velocity are the most prevalent in the stream and are preferred by many fish species of Gobiidae, Cobitidae and Balitoridae (Huang et al., 2019). The fish species *Garra annandalei*, *Glyptothoras trilineatus*, *Euchiloglansis hodgarti* and *Schizothorax plagiostomus* were all collected from the riffles and cascades in our study.

The present habitat study, shows that glide, run, pool and deep pool habitats are the primary ones contributing to the maximum diversity, therefore, protection of these particular habitats is recommended for conservation and management of the fish biodiversity (Sarkar et al., 2010). In the present study area instances of habitat fragmentation are increasing due to ongoing road development and hydro-power generation. In addition, deforestation, illegal electro-fishing, use of agricultural poisons, canalization for irrigation and fishing by water diversion have serious consequences in the present study area and are responsible for declining fish populations (Limbu et al., 2018). For instance, according to local fishermen the fish species *Catla catla*, *Psylorhynchus* spp. were commonly found a decade ago but, we could not find these species in our collection. They fear that the population of these above-mentioned species might have been severely depleted.

Physical and chemical characteristics of freshwater are crucial determinants of the condition of fish assemblages (Li et al., 2012). Canonical correspondence analysis (CCA) indicated that the environmental variables, such as water velocity, pH, total hardness, dissolved oxygen (DO) and water temperature were shown to shape the fish assemblages in the Phewa Khola stream. Previous studies, such as Yu and Lee (2002), Kadye et al. (2008), Mishra and Baniya (2016), Limbu et al. (2019a) and Limbu and Prasad, 2020 have also documented that these environmental variables play a crucial role in shaping fish assemblages.

To conclude, all the selected environmental variables are shown to influence the fish community structure of the Phewa Khola stream. Similarly, of the selected habitat types, the glides, runs, pools and deep pools are the primary habitats contributing to the maximum diversity, therefore, protection of these particular habitats is recommended for conservation and management of the fish biodiversity. Moreover, habitat alterations such as electrofishing, irrigation and hydro-electric dams have accelerated the vulnerability of fishes in Nepal's rivers and streams.

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Conflict of interest

All the authors declare that there are no conflicting issues related to this research article.

References

Blanc, L., Aliaume, A., Zerbi, L. and Lasserre, G. (2001). Spatial and temporal co-structure analyses between ichthyofauna and environment: an example in the tropics. *Comptes Rendus de l'Académie des Sciences - Series III - Sciences de la Vie*, 324 (7): 635–646. https://doi.org/10.1016/S0764-4469(01)01338-5

Chaudhary, S., Limbu, J. H., Subba, S., Gurung, J. K., Pandey, N. and Singh, D. K. (2020). Fish assemblage structure and environmental correlates in Nepal's West Rapti River. *Our Nature*, 18 (1): 28–37.

Clarke, K. R. (1993). Non-parametric multivariate analyses of changes in community structure. *Australian Journal of Ecology*, 18 (1): 117–143. https://doi.org/10.1111/j.1442-9993.1993.tb00438.x

Edds, D. R. (1986). Fishes of the Kali Gandaki/Narayani River, Nepal. *Journal of the Nepal Natural History Museum*, 10: 13–22.

Espírito-Santo, H. M. V. and Zuanon, J. (2017). Temporary pools provide stability to fish assemblages in Amazon headwater streams. *Ecology of Freshwater Fish*, 26 (3): 475–483. https://doi.org/10.1111/eff.12292

Favrot, S. D., Jonasson, B. C. and Peterson, J. T. (2018). Fall and winter microhabitat use and suitability for Spring Chinook Salmon Parr in a U.S. Pacific Northwest River. *Transactions of the American Fisheries Society*, 147 (1): 151–171. https://doi.org/10.1002/tafs.10011

Gauch, H. G. (1982). *Multivariate analysis in community ecology*. Cambridge University Press, Cambridge, UK. 312 pp.

Gerhard, P., Maraes, R. and Molander, S. (2004). Stream fish communities and their associations to habitat variables in a rain forest reserve in southeastern Brazil. *Environmental Biology of Fishes*, 71 (4): 321–340. https://doi.org/10.1007/s10641-004-1260-y

Gore, J. A., Bryant, F. L. and Crawford, D. J. (1995). River and stream restoration, *In*: Cairns, J. (Ed.), *Rehabilitating damaged ecosystems*. 2nd Edition. Lewis Publishers, Boca Raton, USA. pp. 245–275.

Han, Y. Q. (2010). Studies on fish species diversity and evolution trend in Lijiang River. *Journal of Hydroecology*, 3: 132–135.

Huang, J., Huang, L., Wu, Z., Mo, Y., Zou, Q., Wu, N. and Chen, Z. (2019). Correlation of fish assemblages with habitat and environmental variables in a Headwater Stream Section of Lijiang River, China. *Sustainability*, 11 (4): 1–14. https://doi.org/10.3390/su11041135

Jayaram, K. C. (2010). *The freshwater fishes of Indian region*. Narendra Publishing House, Delhi, India.

Kadye, W. T., Magadza, C. H. D., Moyo, N. A. G. and Kativu, S. (2008). Stream fish assemblages in relation to environmental factors on a montane plateau. *Environmental Biology of Fishes*, 83: 417–428. https://doi.org/10.1007/s10641-008-9364-4

Lammert, M. and Allan, J. D. (1999). Assessing biotic integrity of streams: effects of scale in measuring the influence of land use/cover and habitat structure on fish and macroinvertebrates. *Environmental Management*, 23: 257−270. http://dx.doi.org/10.1007/s002679900184

Legendre, P. and Legendre, L. (1998). *Numerical Ecology*. Second Edition. Elsevier, Amsterdam, Netherlands. 853 pp.

Li, J., Huang, L., Zou, L., Kano, Y., Sato, T. and Yahara, T. (2012). Spatial and temporal variation of fish assemblages and their associations to habitat variables in a mountain stream of north Tiaoxi River, China. *Environmental Biology of Fishes*, 93: 403–417. https://doi.org/10.1007/s10641-011-9928-6

Limbu, J. H. and Gupta, S. K. (2019). Fish diversity of Damak and lower Terai region of Ratuwa River of Jhapa district, Nepal. *International Journal of Fauna and Biological studies*, 6 (1) 01–04.

Limbu, J. H. and Prasad, A. (2020). Environmental variables and fisheries diversity of the Nuwa River, Panchthar, Nepal. *Scientific World*, 13 (13): 69–74.

Limbu, J. H. and Punam, G. C. (2019c). Spatio-temporal variation of fish assemblages in Babai River of Danag district, Province No. 5, Nepal, *Our Nature*, 17 (1): 14–25.

Limbu, J. H., Acharya, G. S. and Shrestha, O. H. (2018). A brief report on ichthyofaunal diversity of Dewmai Khola of Ilam district, Nepal. *Journal of Natural History Museum*, 30: 312–317.

Limbu, J. H., Archana, P. and Baniya, C. B. (2019a). Spatio-Temporal Variation of Fish Assemblages in Ratuwa River, Ilam, Nepal. *Journal of Ecology and Natural Resources*, 3 (3): 1–11.

Limbu, J. H., Bhurtel, B., Adhikari, A., Punam, G. C., Maharjan, M. and Sunuwar, S. (2020). Fish community structure and environmental correlates in Nepal's Andhi Khola. *Borneo Journal of Resource Science and Technology*, 10 (2): 85–92. https://doi.org/10.33736/bjrst.2510.2020

Limbu, J. H., Chapagain, N, Gupta, S. K. and Sunuwar, S. (2019b). Review on fish diversity of eastern Nepal. *International Journal of Fisheries and Aquatic Studies*, 7 (3): 177–181.

Limbu, J. H., Gurung. J. K., Subba. S., Khadka. N., Adhikari, A. and Baniya, C. B. (2021). An Impact Assessment of Betani Irrigation Dam on Fish Diversity of Damak Municipality, Jhapa, Nepal. *Egyptian Journal of Aquatic Biology and Fisheries*, 25 (2): 163–175. https://dx.doi.org/10.21608/ejabf.2021.161363

Limbu, J. H., Shrestha, O. H. and Prasad, A. (2018). Ichthyofaunal diversity of Bakraha River of Morang district, Nepal. *International Journal of Fisheries and Aquatic Studies*, 6 (5): 267–271.

Magalhães, M. F., Batalha, D. C. and Collares-Pereira, M. J. (2002). Gradients in stream fish assemblages across a Mediterranean landscape: contributions of environmental factors and spatial structure. *Freshwater Biology*, 47 (5): 1015–1031. https://doi.org/10.1046/j.1365-2427.2002.00830.x

Menni, R. C., Miquelarena, A. M. and Volpedo, A. V. (2005). Fishes and environment in northwestern Argentina: from lowland to Puna. *Hydrobiologia*, 544: 33–49. https://doi.org/10.1007/s10750-004-8299-9

Mishra, A. R. and Baniya, C. B. (2016). Ichthyofaunal diversity and physico-chemical factors of Melamchi River, Sindhupalchok, Nepal. *Journal of Institute of Science and Technology*, 21 (1): 10–18.

Nelson, J. S. (2006). *Fishes of the World*. Fourth Edition. John Wiley and Sons, New York. 601 pp.

Newson, M. D. and Newson, C. L. (2000). Geomorphology, ecology and river channel habitat: Mesoscale approaches to basin-scale challenges. *Progress in Physical Geography: Earth and Environment*, 24 (2): 195–217. https://doi.org/10.1177/030913330002400203

Nilsson, C., Reidy, C. A., Dynesius, M. and Revenga, C. (2005). Fragmentation and flow regulation of the world's large river systems. *Science*, 308 (5720): 405–408. https://doi.org/10.1126/science.1107887

Pokharel, K. K., Basnet, K. B., Majupuria, T. C. and Baniya, C. B. (2018). Correlations between fish assemblage structure and environmental variables of the Seti Gandaki River Basin, Nepal. *Journal of Freshwater Ecology*, 33 (1): 31–43. https://doi.org/10.1080/02705060.2017.1399170

Prasad, A. and Limbu, J. H. (2017). Ichthyofaunal diversity of PhewaKhola of Ilam district, Nepal. *Recent Life Science Mirror*, 6 (1–2): 1–8.

Prasad, A., Shrestha, A., Limbu, J. H. and Swar, D. (2020). Spatial and Temporal Variation Of Fish Assemblages In Seti Gandaki River, Tanahu, Nepal. *Borneo Journal of Resource Science and Technology*, 10 (2): 93–104. https://doi.org/10.33736/bjrst.2048.2020

Punam, G. C. and Limbu, J. H. (2019). Spatio-temporal variation of fish assemblages in Babai River of Dang district, Province No. 5, Nepal. *Our Nature*, 17 (1): 19–30.

R Core Team (2018). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available online at https://www.R-project.org.

Sarkar, U. K., Gupta, B. K. and Lakra, W. S. (2010). Biodiversity, ecohydrology, threat status and conservation priority of freshwater fishes of River Gomti, a tributary of River Ganga (India). *Environmentalist*, 30: 3–17. https://doi.org/10.1007/s10669-009-9237-1

Schlosser, J. I. (1991). Stream fish ecology: a landscape perspective: land use, which influences the terrestrial-aquatic interface, can affect fish populations and their community dynamics. *BioScience*, 41 (10): 704–712. https://doi.org/10.2307/1311765

Shrestha, J. N. (2016). Fish diversity of Triyuga River, Udayapur District, Nepal. *Our Nature*, 14 (1): 124–134.

Shrestha, J., Singh, D. M. and Saund, T. B. (2009). Fish diversity of Tamor River and its major tributaries of eastern Himalayan region of Nepal. *Nepal Journal of Science and Technology*, 10: 219–223.

Shrestha, T. K. (2019). *Ichthyology of Nepal: A study of fishes of the Himalayan waters*. Second Edition. Prism Color Scanning and Press Supportive Pvt. Ltd, Kathmandu. 388 pp.

Spjøtvoll, E. and Stoline, M. R. (1973). An extension of the T-Method of multiple comparison to include the cases with unequal sample sizes. *Journal of the American Statistical Association*, 68 (344): 975–978. https://doi.org/10.1080/01621459.1973.10481458

Subba, B. R., Pokharel, N. and Pandey, M. R. (2017). Ichthyo-faunal diversity of Morang district, Nepal. *Our Nature*, 15 (1–2): 55–67.

Talwar, P. K. and Jhingram, A. G. (1991). *Inland fishes of India and adjacent countries*. Oxford and IBH Publishing Company, Ltd., New Delhi, India. 2 Volumes. 1158 pp.

Vlach, P., Dušek, J., Švátora, M. and Moravec, P. (2005). Fish assemblage structure, habitat and microhabitat preference of five fish species in a small stream. *Folia Zoologica*, 54 (4): 421–431.

Wang, J. H., Tian, J. H. and Lü, X. G. (2010). Assessment of stream habitat quality in Naoli River Watershed, China. *Acta Ecologica Sinica*, 30 (2), 481–486.

Wang, Q., Yuan, X.-Z., Liu, H. and Zhang, Y.-W. (2011). Rapid assessment model for mountain stream habitat and its application. *Journal of Hydraulic Engineering*, 42 (8): 928–933.

Yadav, S. N. (2017). Studies on fish diversity and need for their conservation of Singhiya River, Morang district, Eastern Nepal. *Agriculture, Forestry and Fisheries*, 6 (3): 78–81. https://doi.org/10.11648/j.aff.20170603.12

Yan, Y., Shan, H., Chu, L., Xiang, X., Jia, Y., Tao, J. and Chen, Y. (2010). Spatial and temporal variation of fish assemblages in a subtropical small stream of the Huangshan Mountain. *Current Zoology*, 56 (6): 670–677. https://doi.org/10.1093/czoolo/56.6.670

Yu, S.-L. and Lee, T.-W. (2002). Habitat preference of the stream fish, *Sinogastromyzon puliensis* (Homalopteridae). *Zoological Studies*, 41 (2): 183–187.

Zeni, J. O. and Casatti, L. (2014). The influence of habitat homogenization on the trophic structure of fish fauna in tropical streams. *Hydrobiologia*, 726: 259–270

Zheng, B. H., Zhang, Y. and Li, Y. B. (2007). Study of indicators and methods for river habitat assessment of Liao River Basin. *Acta Scientiae Circumstantiae*, 27: 928–936.

Appendix: Common fish species of the Phewa Khola stream, Ilam district, Nepal.

Devario aequipinnatus

Garra annandalei

Neolissochilus hexagonolepis

Puntius terio

Schizothorax plagiostomus

Schistura horai

Schistura multifasciata

Schistura rupecula

Schistura scaturigina

Schistura savona

Euchiloglansis hodgarti

Glyptothorax trilineatus

Channa punctata