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RESEARCH ARTICLE

Impacts of Hydropower Dams of the Himalayan Rivers (Trisuli and Marsyangdi) on Macroinvertebrates

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ABSTRACT

This study examines the impact of hydropower dams on benthic macroinvertebrates in two Himalayan rivers, Marsyangdi and Trisuli. River waters were collected from 5 sites (six replicates) in each river representing the reference, disturbed and recovery sites during three different seasons(pre-monsoon, post-monsoon, and base-flow) during 2021-2022. The water sampling was accompanied by macroinvertebrates sampling using themultihabitat sampling approach(20 sub-samples in each site). There weresignificant differences in macroinvertebrate compositions amongseasons and sampling stations, although no significant differences were found in the two rivers. Nevertheless,there was a less abundance of macroinvertebrates in Marsyangdi indicating that the PRoR hydropower scheme has more impact compared to the RoR hydropower scheme.The hydropower dams had higher impactson macroinvertebrates just aboveand below the dams.Moreover, dissolved oxygen(among the water quality variables), post-monsoon(among the seasons), and dam and recovery sites(among the spatial locations) were the major factors for variability in benthic macroinvertebrate compositions.Identification ofsuch environmental variables affecting the overall distribution patterns of benthic macroinvertebrates in hydropower-affected rivers providesuseful information for sustainable hydropower development in future.

INTRODUCTION

Rivers provide biological, ecological, and hydrological benefits to local human communities (Smakhtin et al., 2006) besides providing habitats for aquatic species which play a crucial role in global biodiversity protection (Adapa et al., 2016). Moreover, freshwater ecosystems are biodiversity hotspots (Strayer and Dudgeon, 2010) as they occupy less than 1% of the total surface of the earth with providing appropriate habitat for approximately 10% of the known global biodiversity, of which half are insects (Balian et al., 2008). River systems are unique amongst aquatic ecosystems due to their unidirectional flow, so change in catchment influences river ecosystems resulting in an impact on downstream sections (Malmqvist and Rundle, 2002). However, these habitats and biodiversity hotspots are damaged by an estimated 1 million dams above 15 m high in the rivers across the globe, particularly for hydropower generation, transportation and irrigation resulting to fragmented habitats (Nilsson et al., 2005). For example, a study on larger rivers suggests that only 37 percent of rivers longer than 1,000 kilometers are in free-flowing conditions with only 23 percent unblocked to the ocean (Grill et al., 2019).

Natural flow regime modification causes a range of ecological and hydromorphological changes (Poff and Zimmerman, 2010; Schneider and Petrin, 2017). Such modifications in the natural flow regime due to the water diversion projects, such as hydropower dams, may trigger alterations in the abundance and composition of the in-stream biotic community (Bunn and Arthington, 2002; Wright et al., 2002). Dam construction is more responsible for altering the wetland ecosystem than other anthropogenic activities (Lees et al., 2016), altering the natural flow regime as well as water quality, sediment transport, and aquatic habitat (Botelho et al., 2017). Studies have shown a remarkable change in the faunal characteristics downstream of a dam (Bunn and Arthington, 2002; OgbeibuandOribhabor, 2002). Populations of freshwater vertebrates have declined by 83% between 1970 and 2014, the major cause being the dams (WWF, 2020). Therefore, dam construction has a substantive global impact on biodiversity (Grumbine and Pandit, 2013; WWF, 2020). Besides, dam building has significant impacts on the macroinvertebrate compositions just above the dam within a small reservoir (Sharma et al., 2005).

Impacts on community structures of benthic macroinvertebrates were dealt inwith reference to physicochemical parameters(Makumbe et al.,2022) as well as pollutants (Adams et al.,2020).Moreover, dams affect the ecosystem by modifying the physicochemical characteristics of riverine water and fragmenting the continuity of rivers (Malik and Richardson, 2009; Zdankus et al., 2008).

Benthic macroinvertebrates are regarded as one of the best bio-indicators for assessing the changes in river ecosystems such as river flow alteration, and water pollution as they are less mobile than fishes, have long life cycles, and respond to small environmental variations (Poff and Zimmerman, 2010; Wu et al., 2019). Macroinvertebrates showed changes in community composition below dams with increased temperature (Lessard and Hayes, 2003), mixed responses to changes in abundance and diversity in response to flow change owing to dam construction (Poff and Zimmerman, 2010; Vallaniaand Corigliano Mdel, 2007; Vinson, 2001;Wu et al., 2019).

In this study, we assessed the impacts of hydropower dams on indicative aquatic species in the Trisuli and Marsyangdi rivers in response to river flow fluctuations. The study aimed to assess the abundance, richness, diversity, and density of the macroinvertebrate community with respect to changes in river flow. We hypothesized that environmental variables lead to a change in macroinvertebrate communities in hydropower’s dewater area (abstraction area) and compared them to the reference (natural) sites.

2. MATERIALS AND METHODS

2.1 Study area

The present study area was carried out in the Trisuli and Marsyangdi rivers of the Gandaki River basin (Fig. 1).Bigger hydro projects (more than 50 MW) and operational stages were the criteria to select the hydro-projects under study.

Trisuli sub-basin is one of the eight sub-basins of the Gandaki River basin, covering an area of 32,000 square kilometers (km^2), approximately 13 percent of the total Gandaki basin. Trisuli is located on the Eastern corner of the Gandaki basin within the physiographic Highland and Midland zones and is characterized by average altitudes of 2,000 meters and high valley landscapes. It originates in Tibet, where it is known as Bhote Koshi. The catchment area of Bhote Koshi in Tibet is about 3,170 km^2 for a river length of 120 kilometers. The Trishuli River extends approximately 106 kilometers within Nepal, with high gradients in the initial 40 kilometers and rapids along its entire length. As of 2022, the Trisuli sub-basin has already been altered by anthropogenic activities with six hydro-projects that are currently in operation.

Marsyangdi sub-basin has a total area of 4,787 sq. km. of which about 2,150 sq. km. (45 %) lies above the elevation of 4,000 masl. The elevation of the basin varies between 200 masl to 7,800 masl. Physiographically, the basin extends from the High Himalayas in the north to the Lesser Himalayan region in the south (Shrestha and Aryal, 2011). The Marsyangdi basin is an important river basin in Nepal from the hydropower perspective. At present, two PRoR types of hydropower projects namely the Marsyangdi Hydropower Project (69 MW) and the Middle Marsyangdi Hydroelectric Project (70

MW) are operating in the basin. Further, Upper Marsyangdi Hydroelectric Project (600 MW), Lower Manang Marsyangdi Hydroelectric Project (100 MW), and Nyadi Hydropower Project (30 MW) are under different stages of development.

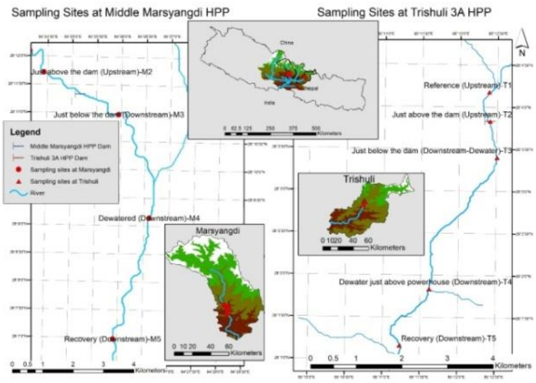

Figure 1. Location of sampling sites in Marsyangdi (red circle) and Trisuli (red triangles) rivers of Gandaki River basin inside Nepal map.

2.2 Sampling sites

A total of 10 sampling sites, 5 in each selected river (Fig. 1), were sampled covering post-monsoon (November), based-flow (late February), and pre-monsoon (April) seasons during 2021 and 2022 to collect biological, physicochemical, and hydrological data. Five sampling stations were selected in Upper Trisuli 3A HPP (60 MW) covering an upstream (starting from 876 m.), dewater, and downstream (639 m). The distance of the selected reach of the river from the reference zone to the recovery zone is about 6 km.The first station was marked with T1 (1.7 km upstream; reference), the second with T2 (just above the dam), the third with T3 (just below the dam), the fourth with T4 (just above the powerhouse outlet) and the fifth was marked with T5 (2.7 km downstream of powerhouse), respectively (Fig. 1).

Similarly,five sampling stations were selected covering upstream (622 m), dewater, and downstream (after the powerhouse releases the diverted water from the tunnel into the natural stretch; 486 m). The distance of the selected reach of the River is about 7 Kilometers (from the reference zone to the recovery zone). Below the dam and before the confluence of Dordi Khola, the stretch of 4.5 km is more important from the aquatic diversity impact point of view. The first station was marked with M1, the second with M2, the third with M3, the fourth with M4 (after major Tributary Dordi Khola joined the dewater zone), and the fifth were marked with M5, respectively (Fig. 1).

Physicochemical and hydraulic parameters

Six replicate samples were collected in 1-liter HDPE bottles at each station from both Trisuli and Marsyangdi rivers, making a total of 60 water samples. The bottles were rinsed with river water before the collection of water samples. Physicochemical parameters such as Temperature, Turbidity, pH, Electrical Conductivity (EC), and Total Dissolved Solids (TDS) were measured *insitu*. pH and temperature were measured by pH meter (Milwaukee pH55, UK), EC and TDS by EC meter (Milwaukee EC59, Europe), and dissolved oxygen (DO) was measured by using OXY 70 Vio (Chromserviss.r.o, Czech). Similarly, ammonia, nitrate, and phosphate were measured on site by using Photometer MD 600. Turbidity was measured on-site by turbidity meter (Wagtech, Micro 950). The remaining physical parameters such asalkalinity, chloride, total hardness, Ca hardness, potassium, free ammonia, and free CO_2were brought to the laboratory for further analysis. Other chemicals like chloride (Cl^-), total hardness (TH), alkalinity (HCO_3^-), and free CO_2 by argentometric titration, complexometric (EDTA) titration, acid-base titration, and phenolphthalein titration, respectively.

Velocity was measured at 0.6 times of total water depth from the water surface by using a current meter at 1 m intervals covering the wetted river section. The discharge (m^3/s) value was obtained by multiplying velocity with the cross-sectional area of the wetted river channel.

Macroinvertebrates sampling

Benthic macroinvertebrates were sampled following the multihabitat sampling procedure which reflects the proportion of microhabitat types equal to or greater than 5% in a 100 m river stretch (Moog, 2007). Before microhabitat sampling, surface flow types such as % rifle, % run, % rapid, and % pool were estimated. 100 m river stretch in each sample site was stratified into 20 subsampling units according to habitat types and percentage coverage and assigned the microhabitats (the mineral and organic). Adopting the multihabitat sampling, microhabitat with less than 5% coverage was discarded for the sampling of aquatic macroinvertebrates. Macroinvertebrates were collected from 20 subsampling units against the flow of water by using a hand net of 500-micrometer mesh and a frame of 25 cm x 25 cm. The benthic samples covered an area of 0.625 m^2 in each sample station.

All macroinvertebrates were identified at the genus, family, and subfamily levels. Mainly Ephemeroptera, Plecoptera, Trichoptera, Mollusca, and Oligochaeta were identified at the genus level, Odonata, Diptera, Megaloptera, Coleoptera, and Heteroptera at the family level, and Chironomidae and Psephenidae at the subfamily level with employing available keys (Assess-HKH, 2006).

2.5 Statistical Analyses

The non-metric multidimensional scaling (NMDS) ordination technique was applied through a *vegan* package (Oksanen et al., 2022) to determine the macroinvertebrate composition structures. Macroinvertebrates were transformed to log (x+1) before NMDS analysis. Differences in community composition among sites and seasons were tested by using a permutational multivariate analysis of variance (PERMANOVA) with 9999 permutations and the Bray-Curtis distance measure. A pairwise comparison was undertaken to visualize the difference within the site in the NMDS.

One-way analysis of variance (ANOVA) was used for macroinvertebrates diversity to find outthe existence of a statistically significant difference between the sites and seasons. A post-hoc Tukey LSD test was used to test whether the means were significantly different at a 0.05 level of probability (SpjøtvollandStoline, 1973).

Principal Component Analysis (PCA) was performed to determine the correlation among the macroinvertebrates, sites, seasons, and environmental variables. Detrended Correspondence Analysis (DCA, Hill and Gauch, 1980) was used to select the most appropriate model either redundancy analysis (RDA) or canonical correspondence analysis (CCA) to describe the association among species, sites, and environmental variables based on linearity or unimodal datasheet (ter Braak andSmilauer, 2002). As the length of the first axis, and eigenvalue did not exceed 2.5 SD and 0.5, respectively in the macroinvertebrate dataset (2.2 SD and 0.3), redundancy analysis (RDA) was chosen to what extent the variance in the distribution of the macroinvertebrates could be explained by the environmental variables. Before conducting RDA, multicollinearity in these environments was testedby using *vif.cca* function (Oksanen et al., 2022). All these statistical analyses were carried out by R Software version 4.2.1(R Core Team, 2018).

RESULTS

Overview of water quality variables

The descriptive statistical results of fourteen physicochemical parameters of Trisuli and Marsyangdi river waters and their comparison with the WHO guidelines are presented in Table 1. Most of the measured hydrochemical variables except pH, DO and Total hardness in Marsyangdi river was found within the limits of the WHO Guideline (WHO 2017).

The inferential statistics from both rivers combined showed statistically significant differencesin most of the water quality parameters between rivers, among sampling sites, and seasons. There were statistical significant differences in pH ($F_{(1,179)}$ = 7.803, p < 0.006), TDS ($F_{(1,179)}$=63.948, p<0.000), EC ($F_{(1,179)}$=312.38, p<0.000), DO ($F_{(1,179)}$=32.282, p<0.000), chloride ($F_{(1,179)}$=185.072, p<0.000), free CO_2 ($F_{(1,179)}$= 24.114, p<0.000), $CaCO_3$ $F_{(1,179)}$=32.506, p<0.000), and TH ($F_{(1,179)}$=41.265, p<0.000) between two rivers. However, no significant differences were found for temperature, turbidity, alkalinity, nitrate, and ammonia.

Similarly, most of the physicochemical parameters were significantly different among sampling stations in both rivers, such as pH ($F_{(1,179)}$=4.037, p<0.004), Temperature, ($F_{(1,179)}$=3.308, p<0.012), TDS ($F_{(1,179)}$=4.703, p<0.001), EC ($F_{(1,179)}$=6.857, p<0.000), DO ($F_{(1,179)}$=11.153, p<0.000), Turbidity ($F_{(1,179)}$=8.107), p<0.006), Chloride $F_{(1,179)}$=6.114, p<0.000), and Ammonia ($F_{(1,179)}$=4.290, p<0.000). No significant differences were found between the alkalinity, free CO_2, $CaCO_3$, TH, nitrate, and phosphate.

The season-wise differences were significant for pH ($F_{(1,179)}$=124.692, p<0.000), Temperature ($F_{(1,179)}$=167.528), p<0.000), TDS ($F_{(1,179)}$=12.852, p<0.000), DO ($F_{(1,179)}$=21.099, p<0.000), Turbidity ($F_{(1,179)}$=20.898, p<0.000), Alkalinity ($F_{(1,179)}$=342.751, p<0.000), $CaCO_3$ ($F_{(1,179)}$=131.005, p<0.000), Total hardness ($F_{(1,179)}$=116.864, p<0.000), Nitrate ($F_{(1,179)}$=67.435, p<0.000) and Ammonia ($F_{(1,179)}$=28.584, p<0.000). However, no significant differences were observed for EC and chloride only.

Table 1 here

Table 1. Descriptive statistics of water quality variables of rivers and their comparison with WHO guidelines (WHO, 2017).

Physicochemical variables	Trisuli River			Marsyangdi River			WHO guideline (WHO, 2017)
	Mean (SD)	Median	Range	Mean (SD)	Median	Range	
pH	8.86(0.82)	8.8	7.2-10.7	9.18(0.72)	9.30	8-10.7	6.5-8.5
Temperature (∘C)	15.9(5.11)	16.0	9.7-27.5	17.17(4.88)	17.15	10.8-28.4	--
TDS (mg/L)	69.33(6.03)	70.0	62-85	110.05(47.34)	131.5	6.75.-158	< 600
EC (μS/cm)	139.67(12.21)	141	122-170	245.89(55.69)	263.5	27.0-316	--
DO (mg/L)	8.61(0.52)	8.69	7.49-9.69	9.17(0.77)	9.19	7-11.17	--
Turbidity (NTU)	10.10(4.43)	8.64	3.51-21	9.98(3.75)	10.66	2.06-22	0.2
Chloride (mg/L)	9.10(3.12)	8.52	5.68-18.46	17.81(5.21)	18.46	4.26-25.56	200-300
Alkalinity (HCO_3^-)(mg/L)	39.06(38.86)	15.0	5.0-115	43.28(46.94)	15.0	5-140	--
Free CO_2(mg/L)	4.89(1.54)	4.40	2.20-8.80	6.31(2.27)	6.60	2.20-13.20	--
Calcium hardness $CaCO_3$(mg/L)	19.03(8.55)	19.64	6.41-40.08	30.35(16.78)	34.87	8.02-65.73	--
TH(mg/L)	67.33 (44.910)	52.00	20-150	133.11(86.13)	120.0	32-300	100-300
Nitrate (NO_3^-)(mg/L)	0.16(0.08)	0.14	0.03-0.36	0.14(0.08)	0.09	0.05-0.31	50

Phosphate (PO_4^{3-}) (mg/L)	0.20(0.16)	0.18	0.01-0.9	0.14(0.14)	0.08	0.01-0.58	--
Ammonia (NH_4^+) (mg/L)	0.22(0.27)	0.09	0.01-1.09	0.20(0.53)	0.02	0.01-2.96	1.5

Overview of benthic macroinvertebrates community composition according to sampling sites, and seasons.

A total of 8 Orders representing 21 Families of macroinvertebrates were recorded in all sampling sites. In terms of seasons, a total of 8, 7, and 8 orders representing 21, 18, and 19 families were recorded for post-monsoon, base-flow, and pre-monsoon, respectively. Most of the taxa belonged to aquatic insects (95%) including Ephemeroptera, Plecoptera, Trichoptera (EPT) taxa, Odonata, Coleoptera, and Diptera. Among other groups of aquatic organismsoutsideInsecta, Mollusca in the other category (5%) demonstrated the greatest taxa richness and individual abundance.

The taxonomic abundance was found significantly different among the sampling stations. The study found that Ephemeroptera was the dominant order followed by Diptera and Trichoptera (Fig. 2) in both rivers. The mean value for Ephemeroptera, Diptera, and Trichoptera was found 106.40, 48.87, and 33.27, respectively in the Trisuli River. The same pattern was followed in Marsyangdi river where the mean value for Ephemeroptera, Diptera, and Trichoptera found 77.67,42.27 and 12.67 implying a higher abundance of macroinvertebrates assemblages in Trisuli River.

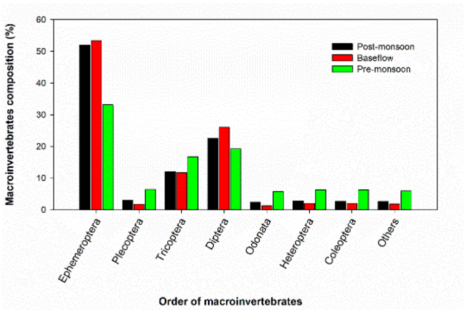

Figure 2. Composition of benthic macroinvertebrates by seasons. Others include Megaloptera and Lepidoptera.

The mean value for Ephemeroptera was found highest in the reference site (244), followed by the recovery site (116) followed by dewater site (111), and lowest in the dam site (12) in Trisuli River. The same pattern was observed in the Marsyangdi River where the lowest mean value was found in the dam site (13), and the highest in the Reference site (129) (Fig.3).

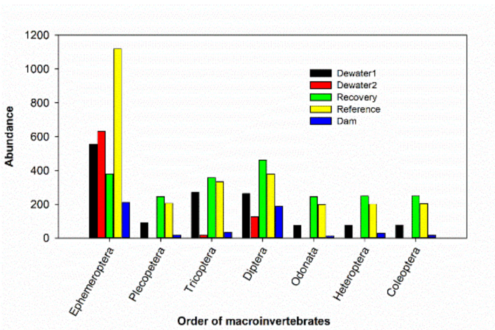

Figure 3. Abundance (number) of benthic macroinvertebrates by stations in two rivers (pooled data).

The one-way ANOVA *F* test revealed that there were significant differences in Ephemeroptera and sampling stations ($F_{(4,29)}$=5.711, p<0.002), Coleoptera and sampling stations ($F_{(4,29)}$=3.539, p<0.02), EPT and sampling stations ($F_{(4,29)}$=4.509, p<0.007) for both rivers.

In terms of abundance of Ephemeroptera, the pairwise post hoc LSD test revealed that there was a significant difference between the reference, and the other 4 sites: reference with damsite (p<0.000), reference and dewater1 (p<0.017), reference and dewater2 (p <0.011), reference and recovery (p<0.008). Similarly, the abundance of Plecoptera was found significantly different between dewater and other sampling stations (dewater1 and dam site (p<0.031), dewater1 and dewater2 (p <0.040), Dewater1 and recovery (p <0.032). Trichoptera was also found significantly different between dewater1 and damsite (p<0.047), and between dam site and recovery (p<0.031). Besides EPT, dipteral also found a significantly different between dewater1 and damsite (p<0.036) and dewater2 and damsite (p<0.025).

A total of 5 functional feeding groups, namely collector-gatherers, filters, scrapers, shredders, and predatorswere recorded in the study area (Fig. 4). Collector-gatherers were the most dominant taxonomic group comprised nearly 45% of overall macroinvertebrates assemblages followed by filters (30%). Scrapers accounted for less than 15% of overall benthic macroinvertebrates abundance across all three seasons (Fig. 4). Predators and collector-gatherer shared nearly 40% and 20% of taxonomic richness, respectively, for all seasons while scrapers and shredders shared less than 5% of the taxonomic richness of all seasons (Fig. 5).

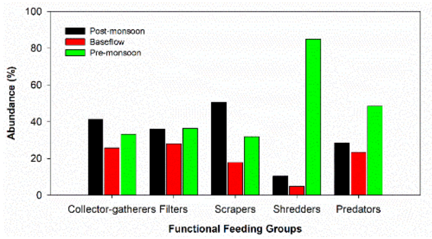

Figure 4. Relative abundance of Functional Feeding Groups across seasons.

The one-way ANOVA F-test revealed that there were significant differences in scrapers and season ($F_{(2,29)}$=13.37, p <0.001), collector-gatherers and sampling sites ($F_{(4,29)}$=4.317, p<0.009), filters and sample sites ($F_{(4,29)}$=4.471, p<0.003) and filters and discharge ($F_{(4,29)}$=4.460, p<0.020). The F test revealed that there was a significant difference between the composition of functional feeding groups (Fig. 6.) and sampling stations ($F_{(4,29)}$=4.410, p<0.008) whereas no significant differences were found between the functional feeding groups and seasons and both river's hydropower sites. In terms of abundance of collectors and gatherers, the pairwise post hoc LSD test revealed that there was a significant difference between damsite and reference (p<0.001), damsite and dewater (p<0.002), damsite and dewater2 (p <0.014), damsite and recovery (p<0.029). Likewise, an abundance of filters and gatherer feeding functional group (Fig.6) showed a significant difference between reference and dewater1 (p<0.042), reference and dewater2 (p<0.028), damsite and dewater1 (p<0.001), damsite and dewater1 (p<0.000), damsite and recovery (p<0.029).

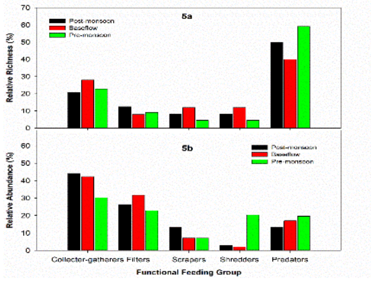

Figure 5. Relative richness (a) and relative abundance (b) of Functional Feeding Groups across seasons.

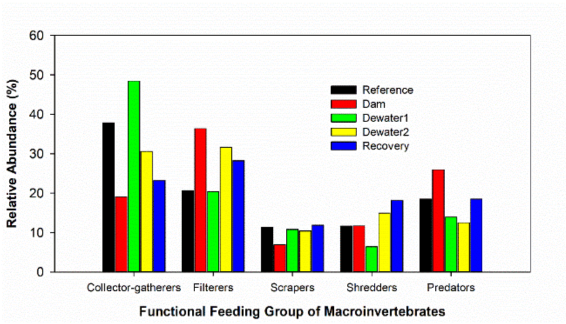

Figure 6. Relative abundance of FFGs across the sampling stations.

Variation in water discharge and macroinvertebrate composition

Water discharge for this study incorporated three categories: 1) up to 10% mean monthly flow (low flow), 2) 11-30% mean monthly flow (medium flow), and 3) above 30% mean monthly flow (high flow). The analysis between water discharge and macroinvertebrates abundance (Fig. 7) revealed that EPT was dominant in all three water discharge categories low flow, medium flow, and high flow followed by Diptera in all discharge categories. In the EPT index, Ephemeroptera comprised 90% in medium flow, 40% in low flow, and nearly 25% above high flow. The one-way ANOVA test revealed that there were significant differences between flow discharge and filter feeding functional group ($F_{(2,29)}$=4.888, p<0.015), and flow discharge and Diptera ($F_{(2,29)}$=3.956, p<0.031). The pairwise post hoc LSD test revealed that there were significant differences between low flow and medium flow (p<0.004) for Filter, between low flow and medium flow for Shredder (p<0.022),and between low flow and high flow (p<0.010) for Diptera. Flow discharge and sample sites also showed significant differences ($F_{(4,29)}$=17.218, p <0.000) primarily between dewaters (water abstraction sites) and all remaining sites: dewater1 and reference (p<0.000), Dewater1 and dam (p<0.000), dewater1 and dewater2 (p<0.001), dewater1 and recovery (p<0.000).

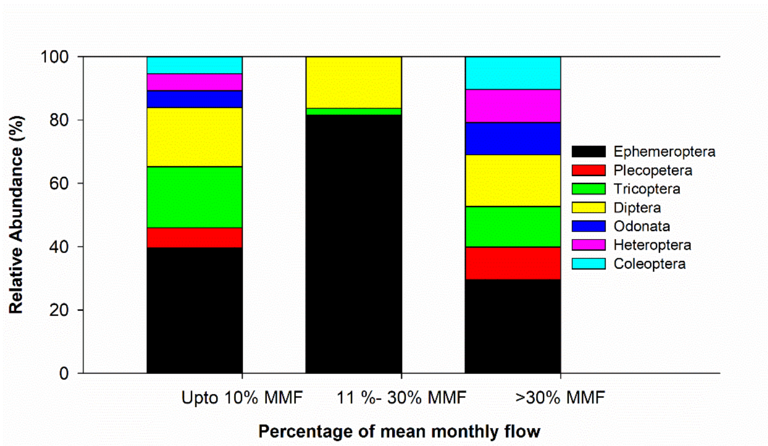

Figure 7. Percentage of mean monthly flow across macroinvertebrate order

Non-metric multidimensional Scaling (NMDS) ordination plot (stress=0.15, Figs. 8a & 8b) revealed a distinct clustering pattern representing macroinvertebrates according to rivers, and sampling stations. Performing adonis2 (PERMANOVA) showed differences inmacroinvertebrate compositions between rivers (F=2.3661, df=1p<0.025), seasons (F=2.971, df=2, p<0.004) and sampling stations (F=3.163, df=4, p<0.001).

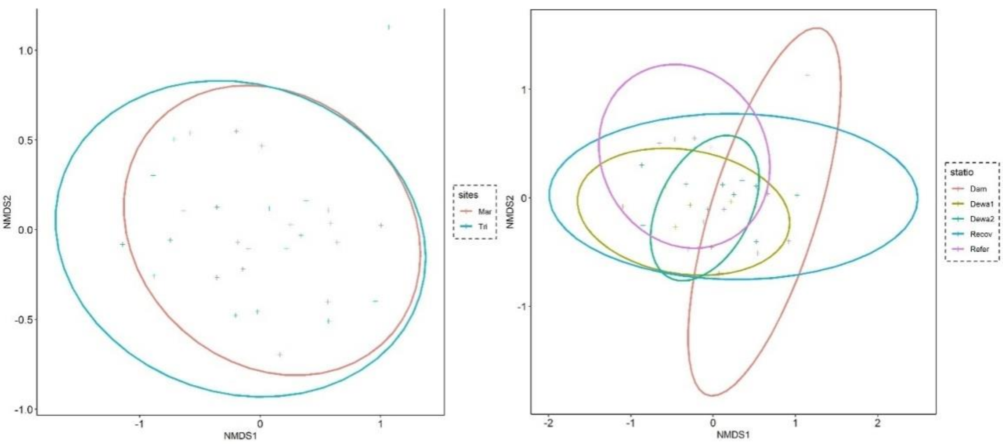

Figure 8. Non-metric multidimensional scaling (NMDS) ordination plot explains clustering of macroinvertebrates communities by river sites (Abbreviations are Mar = Marsyangdi, Tri = Trisuli), 8b. Clustering of macroinvertebrates communities by sampling stations in NMDS ordination plot.

The cluster analysis of macroinvertebrate taxa at the family level generated a dendrogram with 3 distinct clusters at a higher level and 5 distinct clusters at a lower level separated by rectangles with visible colors shown in Fig. 9. One of the main groups ordinated towards the left end of the plot was formed by Baetidae belonging to Ephemeroptera Order followed by Chironomidae and Blephariceridae belonging to Diptera Order. Many macroinvertebrate families formed a much wider third cluster ranging from Heptagenidae to the Gomphidae family. The last and fifth cluster on the extreme right comprised Rhyacophilidae belonging to Trichoptera Order, and Tipulidae belonging to Diptera Order (Fig. 9).

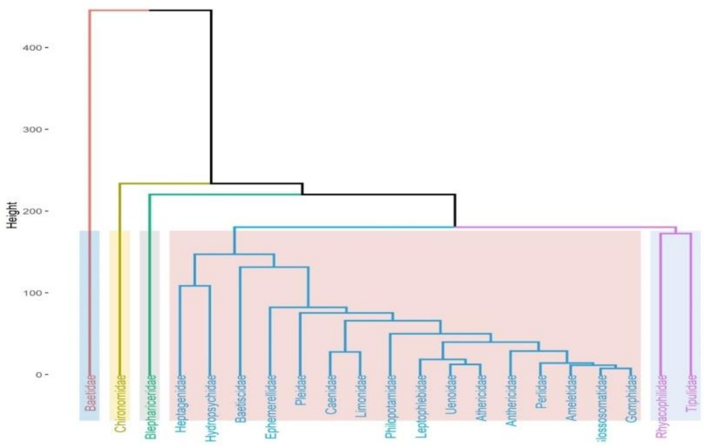

Figure 9. Hierarchical clustering plot of macroinvertebrates at a family level based on Bray-Curtis Similarity.

Environmental variables affecting benthic macroinvertebrates community composition in sampling sites in RDA

The RDA (Fig. 10) summarized the spatial trends in environmental gradients and macroinvertebrates' abundance. The RDA ordination plot showed a correlation between the environmental variables and macroinvertebrate assemblages. Macroinvertebrates at the family level were overlaid with environmental factors according to sampling stations in Fig. 10a, according to seasons in Fig. 10b, and according to rivers in Fig. 10c. In the plots, vector lines are radiated from the mean of all environmental variables. Similarly, the angle and length of the lineindicated the strength of that value.

RDA plots (Figs. 10a &10b) showed that the first axis's left side encompasses the interaction between environmental variables such as pH, temperature, discharge, and phosphate with Tupilidae and Tipulidae in reference, recovery, and dam site. The RDA second axis bottom shows strong interaction of environmental variables such as TDS, Chloride, conductivity, ammonia, total hardness, calcium hardness, nitrate, dissolved oxygen, and alkalinity with Blephariceridae andLimoniidae (belonging to Diptera Order);Ephemerellidae,Baetidae, andBaetiscidae (belonging to Ephemeroptera Order) in reference, dam, and dewater2 sampling stations. The second axis upper part shows an interaction of environmental factors with macroinvertebrate families such as Heptagenidae (belonging toEphemeroptera); Rhyacophillidae, Hydropsychidae, and Philopotamidae (belonging to Trichoptera); Perlidae (belonging to Plecoptera). RDA plot (Fig. 10c) axis 1 indicates that explanatory variables are more manifested in Marsyangdi River (r=0.23) than in Trisuli River (no r-value). This is also supported by no single explanatory variable is visible on axis2 where Trisuli River (legend green) is mostly placed.

RDA1 explained 28.52% of the taxonomic variance with eigenvalue (λ=0.12) whereas RDA2 explained 11.86% of the taxonomic variance with eigenvalue (λ=0.05). The RDA's 1st axis was found to be highly significant (RDA1, F = 29.881, df=1, p=0.029). The sum of the canonical eigenvalue was 0.41. Of the environmental variables, dissolved oxygen seemed highly significant with RDA1 (F=4.204, df=1, p=0.005). In terms of season, post-monsoon seemed significantly different with the RDA axis (F = 3.364, df=1, p=0.02) whereas dam (F = 8.911, df=1, p=0.001) and recovery site (F = 2.642, df=1, p=0.044) also found a significant difference with RDA axis.

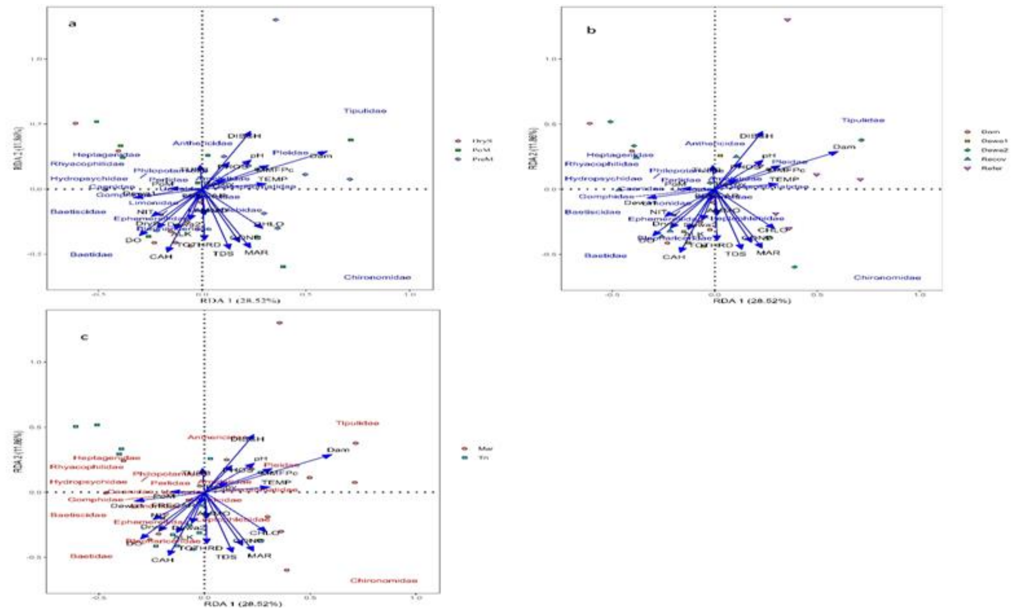

Figure 10. Redundancy Analysis (RDA) ordination plots (Axis1 explained 28.52% of the variation of taxa-environmental relations, axes1 and 2 together explained 40.38% of overall variation). The ordination of family level variance of benthic macroinvertebrates explained by environmental variables of 3 seasons (a). The biplot shows the variance in benthic macroinvertebrate communities at family level taxa explained by environmental variables in 5 sampling stations (b) of two rivers (c). Abbreviations are MAR = Marsyangdi, TDS = Total dissolved solids, COND = Conductivity, CHLO = Chloride, CAH = Calcium hardness, TOTHRD = Total hardness, DO = Dissolved oxygen, ALK = Alkanity, DryS = Dry season (baseflow), Dewa2 = Dewater zone2, NIT = Nitrate, Dewa1 = Dewater zone1, AMMO = Ammonia, PoM = Post-monsoon, TEMP = Temperature, DISCH = Discharge, PHOS = Phosphate, MMFPc = Mean monthly flow %, Recov = Recovery, Refer = Reference, TURB = Turbidity, PreM = Pre-monsoon.

A positive correlation existed between the first axis and pH (0.24), temperature (0.31), TDS (0.13), EC (0.18), chloride (0.30), total hardness (0.01), phosphate (0.13), ammonia (0.01), and discharge (0.23) whereas DO (-0.30), turbidity (-0.01), chloride (-0.26), alkalinity (-0.13), free CO_2 (-0.02), calcium hardness (-0.17), and nitrate (-0.24) were negatively correlated with axis1.

5. DISCUSSION

Macroinvertebrate community composition changes gradually from headwaters to downstream rivers as postulated by the theory of the River Continuum Concept (Vannote, 1980). Our results suggest that hydropower plants have a negative impact on water quality. This study showed that there is significant variation between most of the water quality variables between rivers, among sampling stations and seasons. There was a significant difference in pH, temperature, conductivity, Turbidity, Chloride, and Ammonia among sampling stations.Increasedconductivity and temperature range usually decreasesthe proportion of dissolved oxygen concentrationas water bodies dry out. The significant variation in water quality between the sampling stations of the two rivers could imply that the hydropower dam influences directly the water quality. As such the negative correlation of pH ($r=-0.26$, $p<0.000$) and positive correlation of temperature ($r=0.17$, $p<0.023$) with sampling stations were observed in the present study. Within the group, the study showed a significant difference between the upstream and downstream ($p<0.012$) of the two rivers.The water quality parameters alone explained over 40.38 % variations in macroinvertebrates composition of the two rivers (Fig. 5). These findings corroborate with the study of Vaikasas et al. (2013) that Hydropower power plan had a significant negative downstream impact on water quality.

Both seasons and sampling sites wise significant temperature differences, and between flow discharge and sample sites in the present study corroborate with the study of Munn and Brusven (1991) and MacNally and Wallis (2011) that the major factors contributing to the variation in macroinvertebrate community are the altered food supply, reduced habitat diversity, altered thermal regime, altered water levels among others. The significant difference between two rivers with different hydropower schemes ($F=2.366$, $df=1$, $p<0.025$), among seasons ($F=2.971$, $df=2$, $p<0.004$) and among sampling stations ($F=3.163$, $df=4$, $R^2=0.301$, $p<0.001$) for macroinvertebrate taxa in the present study corroborate with the finding of Greathouse et al. (2006) who showed significant difference for macroinvertebrate taxa between dammed and undammed sites. Furthermore, the seasonal variations in the macroinvertebrate community structure were also observed by Tachamo et al. (2020) in the Karnali River basin of the Nepal Himalayas.

Dam building has significant impacts on the composition of macroinvertebrates just above the dam site probably because of the deposition of inorganic materials within the reservoir and change in water velocity (Doyle et al., 2005; Sharma et al., 2005). In our study, we also found that there was a significant difference between the composition of functional feeding groups at different sampling stations, specifically between dams and below the dam which showed a low abundance of macroinvertebrates. In terms of the abundance of collectors and gatherers, a significant difference between the damsite and other stations was observed. Likewise, the lower abundance of filterers and gatherer functional groups in the damsite compared to dewater and recovery zones in the present study are also supported by the study of Jesus et al. (2004).

The dominance of Ephemeroptera was followed by Diptera and Trichoptera in all the stations (Fig. 2). Nevertheless, significant differences in Ephemeroptera, Coleoptera, and EPT were observed in the present study. Furthermore, the higher abundance of Ephemeroptera in the reference site compared to the other four sampling sites indicates the impacts of the dam on macroinvertebrate assemblages as was also observed by Vaikasas et al. (2013) that taxon number and total abundance of macroinvertebrates declined significantly both in HPP dam and below them in comparison to control sampling site in VirvyteRiver, Lithuania. A similar impact was seen in the dewater zone as indicated by a different abundance of Plecoptera compared to other sampling sites. Trichoptera was more affected in the damsite compared to the other locations. Besides, significant differences in the abundances of EPT and Diptera between dewater zone and dam sites might be the impact of dam construction.

In Canada, macroinvertebrates abundance was not different during high flow season (Patterson andSmokorowski, 2011). These findings showed that macroinvertebrate diversity varies temporally depending on the availability of food sources in different habitats. Collectors and gatherers were dominant in the pool substrate along withshredders, but collectors-filters were dominant in riffle in contrast to the scrapers in the hard substrate (Oliveira andNessimian, 2010). There is a direct relation between higher abundance and lower flow in the non-regulated river (Richards,1990). However, this correlation is less relevant in regulated rivers (Malmqvist andEnglund, 1996). In line with this, Fleituch (2003) found that the FFGresponds differently to variations in flow in regulated rivers. Collectors and gatherers were found abundant with high flow, and they decreased in the same way the predators did, other groups demonstrated different behaviors. The change from the shredder's dominance in headwaters to the collector's dominance in the downstream is evident large river is evident. This was observed in contrast to Vannote (1980). However, Heino et al. (2005) found that macroinvertebrate abundances were evenly distributed among FFGs in the main river. Flow reduction affects macroinvertebrates' abundance and composition owing to the alteration in nutrient flow, food availability, and dispersal (Dewson et al., 2007). The run of the river hydropower scheme in Trisuli, and the Peaking Run of the river scheme in Marsyangdi River diverted water via a pressure tunnel with the construction of the dam to obstruct water. In this study, the analysis between water discharge and macroinvertebrates abundance revealed that EPT was dominant in all three water discharge categories (low, medium, and high flow) followed by Diptera. In the EPT index, Ephemeroptera comprised 40% in low flow regime, 90% in medium flow, and ~25% in high flow implying low abundance in water abstracted site of hydropower. Fenoglio et al. (2007) showed that water abstraction (dewater area) creates an unfavorable condition for fast-moving macroinvertebrates (such as trichopterans). In dewater sites, more tolerant species replace sensitive macroinvertebrates (Death et al., 2009). Brittain and Salteveit (1989) found that scrapers of Ephemeroptera increased in regulated rivers whereas collectors/shredders decreased. Richness was less affected across seasons, between stations which might be because of the replacement of lotic taxa by lentic taxa between early seasons (Bogan and Lytle,2012). This situation was the same for Ephemeroptera gatherers in the present study area.

The significant variation of FFG and Diptera with flow discharge in this study is supported by the findings of Vallania and Corigliono (2007) that the collectors-filterers, scrapers, and predators increased whereas collectors-gatherers and shredders decreased. The significant difference in the abundance of Diptera during different flow regimes (low, medium, and high), particularly in water abstraction zones in the present study is in agreement with the finding of Vallania and Carigliano (2007) that the number of collector-gatherers and shredders decreased whereas the abundance of filters, predators and scrapers increased in the regulated site.

The study found that collector-gatherers were the most dominant taxonomic group comprised of ~45% of overall macroinvertebrates assemblages followed by filters (30%) for all seasons. Thomson et al. (2005) also found that downstream sites of the dam had poorer biotic index scores than upstream sites. Scrapers accounted for less than 15% of overall benthic macroinvertebrates abundance across all three seasons. The study found predators shared nearly 40% of taxonomic richness for all seasons while scrapers and shredders shared less than 5% of the taxonomic richness of all seasons. This is supported by the study of Cortes et al. (2002) that poor water quality and lack of litter inputs affected mainly the shredder group in the regulated portion of the river. This study also revealed significant differences in scrapers and season, collector-gatherers and sampling stations, filterers and sampling stations, and discharge. Our results support the finding that increased water diversion reduces river discharge in downstream river stretches affecting biological communities (Anderson et al., 2015). Kennedy and Turner (2011) found that 50% lower of benthic macroinvertebrates in channelized reaches attributed to water diversions.

The study showed the collectors were relatively higher in post-monsoon followed by filterers (Fig. 4). The relative abundance of collector-gatherers and filters in post-monsoon season implies that this season provides more food sources to feeding functional groups. Scrapers and shredders were found relatively higher in post-monsoon and pre-monsoon, respectively. The relative abundance of predators seems low in post-monsoon owing to the overwhelming increase in collectors in this season. All these temporal changes in the composition of FFGs were in corroboration with the finding of Vannote et al. (1980) and Cummins (2016). In both seasons, the relative richness and abundance of collectors were the highest (Fig. 5), but the codominance of collectors and shredders was not found as hypothesized by Vannote et al. (1980) and Cummins (2016). Bunn (1986) did not findthe codominance of shredders and collectors as well.

In summary, hydropower plants induce huge changes in the species composition of macroinvertebrates and the water quality of the rivers. Not only are the rheophilic species of macroinvertebrates extinct but also the distribution and abundance of the surviving species changes triggering the change in life quality of other vertebrates and invertebrate animals as well.

Way forward for Sustainable hydropower

Our results provide a basis for future studies on the benthic macroinvertebrates' composition and various environmental variables across the rivers already impacted and to be impacted by the hydropower. Of the aquatic species of rivers, macroinvertebrates are the neglected research topic in the hydropower developments of Nepal. In this regard, academicians, government agencies, and of course, hydropower developers will benefit from the findings and insights of this research to develop hydropower in a more sustainable way.

5. CONCLUSIONS

Dam building has significant impacts on macroinvertebrates in both Trisuli and Marsyangdi rivers as indicated by significant differences in their compositions amongdifferent seasons, and sampling stations. However, there was no difference in macroinvertebrates composition in two rivers. Thedifferences among sampling stations suggest a higher impact of hydropower on aquatic species, particularly just above and downstream of the dams. Furthermore, major factors for variability in benthic macroinvertebrates were dissolved oxygen (amongst the water quality variables), post-monsoon (among the seasons), and dam and recovery sites (among the sampling stations). The higher richness of predators implies that habitats are changing and feeding functional groups are declining owing to the unfavorable living conditions created by the disturbance of hydropower. Less abundance of macroinvertebrates in Marsyangdi indicates that the PRoR hydropower scheme has more impact than that of the RoR hydropower scheme as observed in Trisuli.

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Author Contribution Statement

Conceptualization: AB and CMS; Data collection from the field: AB, UTR, BR, JNR and CMS; Laboratory work: AB, UTR, BR and JNR; Data analysis: AB, CBB and CMS; Manuscript Draft: AB; Manuscript review and edit: AB, UTR, CBB and CMS.

Declaration of Interest Statement

The authors declare no conflict of interest.

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