LENGTH-WEIGHT RELATIONSHIP AND RELATIVE CONDITION FACTOR OF A MIGRATORY AND A RESIDENT HILL STREAM FISH FROM HYDROPOWER-IMPACTED TRIBUTARIES OF PUNATSANGCHHU RIVER, BHUTAN

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ABSTRACT: This study aimed to determine the length-weight relationship (LWR) and access the relative condition factor (K_n) of migratory fish, *Schizothorax richardsonii* and resident fish, *Garra gotyla* from the tributaries of Punatsangchhu River within the vicinity of the hydropower development area. The sampled tributaries were categorized into three zones: upstream, fragmented or impacted zone and downstream zone. Both species indicated significant negative allometric growth. The allometric form of LWR for *S. richardsonii* and *G. gotyla* is established as $W = 0.0155L^{2.6806}$ and $W = 0.0161L^{2.7506}$, respectively. The K_n of both species were > 1 at the downstream zone only. One-way ANOVA and Tukey HSD test indicated significantly lower K_n in the fragmented zone and upstream zone as compared to the downstream zone for *S. richardsonii*. An unpaired two-sample t-test indicated insignificant differences in K_n of *G. gotyla* between the downstream and fragmented zone. This study reflects the greater impacts of hydropower development on fragmented zone and migratory fish as compared to resident fish. The present study will help in the improvement of long-term monitoring of the impacts of hydropower development on fisheries and standardization of fisheries monitoring programs in Bhutan.

Keywords: allometric growth; *Garra gotyla*; hydropower; migratory; *Schizothorax richardsonii*

1. INTRODUCTION

The Bhutan Himalaya with a rich network of perennial water resources coupled with steep topography offers enormous techno-economically feasible hydropower potential (26,760 MW) attracting the attention of hydropower developers (NORAD 2017). As a result, six hydropower plants (HPPs) with a total installed capacity of 2,326 MW are currently under operation. However, > 91.31% of hydropower potential (i.e., 24,437 MW) remains untapped. To capitalize on the resources, HPPs of various capacities are planned and prioritized across different hydrological basins (HB). The Punatsangchhu River and its tributaries constitute the Punatsangchhu Hydrological Basin (PHB) with an estimated hydropower potential of 7250 MW (NORAD 2017). Two HPPs with an overall installed capacity of 2,220 MW i.e., the 1200 MW Punatsangchhu – I Hydropower Plant (PHPP – I) and 1020 MW Punatsangchhu – II Hydropower Plant (PHPP – II) are currently under advanced stage of construction on the Punatsangchhu River, Wangdue Phodrang. Moreover, PHPP – I and PHPP II are among the first major

HPPs to commence simultaneously on the same river. Among others, the 64 MW Basochhu Hydropower Plant consisting of 24 MW Upper Stage and 40 MW Lower Stage HPs are operational (since 2001 and 2004 respectively) on Basochhu and Rurichhu, tributaries of Punatsangchhu between the two dams of Punatsangchhu HPPs.

Although HPPs bring socio-economic benefits, hydropower dam fragments habitats, restrict the bidirectional movement of fishes, alter the natural hydrological regime of rivers, and in long-run, hydropower operation can change the structural and functional composition of fisheries resulting in the species expatriation due to invasions and changes in habitat conditions (Li et al. 2013; Santos et al. 2017; Santos et al. 2022). The operation of HPPs is known to negatively influence the growth and subsequently reproductive process of both migratory and non-migratory fishes (Arantes et al. 2011; Gomes et al. 2020). However, in Bhutan, studies related to the assessment of the impacts of hydropower development on fisheries are scanty and need attention.

The length-weight relations (LWRs) are a valuable tool in fisheries assessment and management, however, their importance is often undermined and is lacking for most species and regions (Froese 2006; Froese et al. 2011). This is significant for a region like Bhutan where established LWR is available only for the Golden mahseer, that too under the aquaculture conditions (Dorji et al. 2018). The importance of LWR depends on the determination of the relationship between the weight and length of a fish. The established LWR is used to determine the weight from length data and compare if the growth is isometric or allometric (Froese et al. 2011; Tyagi et al. 2014; Sharma et al. 2021). The condition factor also gives an idea of the well-being of the fishes under different conditions, based on assumption that heavier fishes at particular lengths are under good growing conditions (Froese 2006). This allows the comparison of the growth of a particular species among seasons (Çetinkaya et al. 2020), months (Sharma et al. 2021), different areas and ecological conditions (Sharma et al. 2021). Based on the above-mentioned applications, these parameters can be used for the assessment of the growing condition of migratory and non-migratory fish in hydropower-impacted areas.

Schizothorax richardsonii (Gray 1832) is the most abundant migratory species in the Punatsangchhu HB and is vulnerable in accordance with IUCN Red List Status. They have high commercial importance in the capture fisheries program in Bhutan and are the main species used to prepare the "*Nya-Dosem*", a traditional smoked fish from fisher communities of Rukha-Samthang and Lawa-Lamga of the Harachhu sub-basin of Punatsangchhu HB. *Garra gotyla* (Gray 1830) classified as least concern by IUCN Red List Status is the most abundant among the resident fish, but with limited commercial importance in inland fisheries. Therefore, this study is aimed to determine the LWR and relative condition factor (K_n) of migratory fish, *Schizothorax richardsonii* and resident fish, *Garra gotyla* from the tributaries of Punatsangchhu River adjoining the Punatsangchhu Hydropower Project (PHPP) area (PHPP – I and PHPP – II), and answer the following research questions: (i) Does the species-specific parameter "b" of LWR relationship differ with the isometric value (b = 3)? (ii) Does species-specific K_n vary among tributaries found in different zones during the hydropower construction stage.

2. MATERIALS AND METHOD

2.1 Study area and sampling design

The study covers eight tributaries of Punatsangchhu River (within ~ 40 km river length) between Hesothangkha and Taksha at Wangdue Phodrang, Bhutan within the vicinity of construction sites of two major

HPs: i.e., 1200 MW PHPP – I and 1020 MW PHPP – II (Figure 1). The tributaries joining the mainstem Punatsangchhu considered for the study include (from upstream): Nahichhu (Hesothangkhachhu), Basochhu, Baychhu, Phenrechhu, Kamerongchhu, Dikchhu, Harachhu and Nyerachhu (Figure 1, Table 1). One site was sampled from each tributary, giving a total of eight sampling sites. The tributaries of Punatsangchhu were categorized into three zones: upstream zone (i.e., tributary above PHPP – I Dam, namely Nahichhu), fragmented zone or impacted zone (i.e., tributary between PHPP – I and PHPP – II Dam, namely Basochhu) and downstream zone (i.e., tributaries below PHPP – II Dam, namely Nyerachhu, Harachhu, Dikchhu, Kamerongchhu, Phenrechhu, Baychhu. Please refer "limitations paragraph" in the results and discussion section which resulted in this sampling design). The Punatsangchhu River (discharge upstream of PHPP – I Dam: 58 – 2195 m^3/s; NCHM 2018) was diverted through a diversion tunnel in November 2011 for the construction of PHPP – I Dam and in May 2013 for the construction of PHPP – II Dam. As both the HPPs are currently under the advanced stage of construction, the stretch of Punatsangchhu River between PHPP – I and PHPP – II Dams, i.e., the impacted zone (Figure 1) remained fragmented for more than a decade. In addition, Basochhu is diverted for hydropower generation to the 24 MW Basochhu Upper Stage Project and then to the 40 MW Basochhu Lower Stage Project before draining into Punatsangchhu River at Rurichhu. The Basochhu sampling site thus represents dewatered stretch (Figure 1).

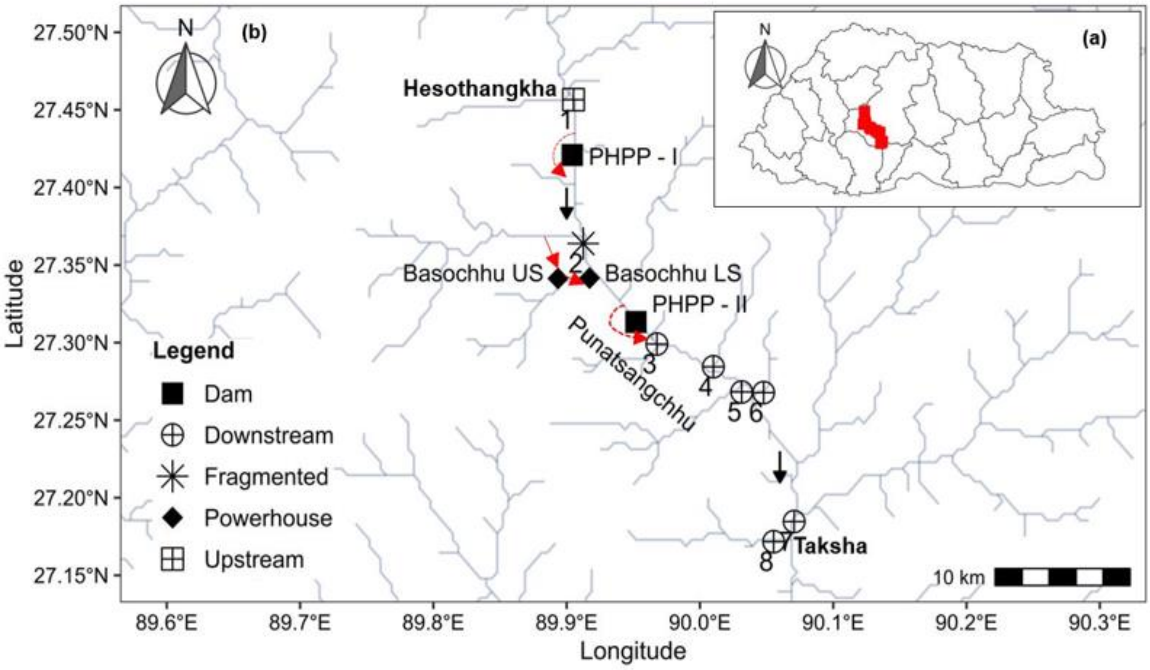

Figure 1. (a) Location of studied portion of Punatsangchhu River, Bhutan (red), and (b) location of sampled tributaries of Punatsangchhu River at Wangdue Phodrang District (data: HydroSHEDS). **Note:** Red arrow indicates water conveyance system (approximated) of Basochhu Hydropower Plants [i.e., diversion of Basochhu for Basochhu Upper Stage (Basochhu US) and Basochhu Lower Stage (Basochhu LS) Hydropower Plants], dashed arrow along the Punatsangchhu Dam sites indicate the water diversion tunnels (approximated) and black arrow indicates direction of flow of Punatsangchhu River. Refer site id from Table 1.

2.2 Specimen collection

The fishes were sampled from 25th to 26th November 2021 with 20 minutes of electrofishing (Model: ELT62-2D;

Table 1. Tributaries sampled along the mainstem Punatsangchhu River and its location.

Site ID	River	Zone	Latitude (DD)	Longitude (DD)	Altitude (m)
1	Nahichhu	Upstream	27.45660	89.90512	1186
2	Basochhu	Fragmented	27.36389	89.91241	1004
3	Baychhu	Downstream	27.29909	89.96768	755
4	Phrenchu	Downstream	27.28446	90.01007	716
5	Kamerongchhu	Downstream	27.26820	90.03131	668
6	Dikchhu	Downstream	27.26779	90.04778	661
7	Harachhu	Downstream	27.18469	90.07068	480
8	Nyerachhu	Downstream	27.17171	90.05533	581

GRASSL, Germany; DC 3KV) at each site as part of the post-monsoon assessment. The total length (TL) and weight (W) of each specimen were measured to the nearest 1 mm (measuring board) and nearest 1 g (Model: I-2000, Newpro Electronics, China).

2.3 Data analysis

The pooled samples (male and female, and from all sites/zones combined) of *S. richardsonii* and *G. gotyla* were considered for the determination of LWR to maintain recommended sample size under the current sampling design ($n > 30$; Froese 2006; Froese et al. 2011). The LWR was determined by applying the log transformation of the allometric growth model: $\mathbf{W = aL^b}$ (Le Cren 1951) as **Log W = Log a + b Log L** where, 'W' is the weight of the fish (g), 'L' is the total length of fish (cm), and 'a' and 'b' are intercept and slope of the regression line, respectively. To confirm the type of growth, the parameter 'b' obtained in the linear regression was compared with the isometric value (b = 3) using Student's t-test. The relative condition factor (K_n) was calculated as $\mathbf{K_n = W/W'}$, where "W'" is the expected weight determined from the established length-weight relationship (Le Cren 1951). To compare the well-being of fishes, the K_n of *S richardsonii* was compared with one-way ANOVA followed by Tukey's multiple comparison test. As only two specimens of *G. gotyla* were obtained from upstream, the upstream zone was eliminated from statistical analysis and thus K_n of *G. gotyla* was evaluated with an unpaired two-sample t-test (i.e., between downstream vs fragmented zone). Before the statistical analysis of K_n, all the outliers were removed from the datasets and were assessed for the requirements of statistical analysis (i.e., normality test, and homogeneity of variance test). All statistical analyses were performed using R Statistical Software (Version 4.1.2) at a significance of $p < 0.05$.

Table 2: Total length and weight data, and regression parameters of fishes from tributaries of Punatsangchhu River

Species	*n*	Total length (cm)		Weight (g)		r^2	Regression Parameters			
		Min-max	Mean ± SD	Min-max	Mean ± SD		a	b	95% CI of a	95% CI of b
SR	178	5.60 – 29.60	11.84 ± 4.00	1.28 – 136.20	15.75 ± 19.39	0.89	-1.8096	2.6806	-1.9556 to -1.6637	2.542 to 2.818
GG	74	5.20 – 19.00	11.06 ± 2.93	1.30 – 62.10	14.71 ± 12.97	0.90	-1.7942	2.7506	-2.019 to -1.569	2.533 to 2.968

SR = Schizothorax richardsonii, *GG = Garra gotyla*

3. RESULTS AND DISCUSSION

The LWR, its parameters and K_n are useful tools in assessing the growth and physiological condition of fishes, which are usually affected by overlying ecological and environmental factors (Froese 2006; Yadav and Dhanze 2018; Sharma et al. 2021; Arafat and Bakhtiyar 2022). During this study, a total of 178 individuals of *S. richardsonii* and 74 individuals of *G. gotyla* were collected from the assessed tributaries of the Punatsangchhu River. The details on regression coefficient (b), intercept (a), 95% confidence limits of a and b, and coefficient of determination (r^2) of the length-weight relationships of two species as required by Frose (2006) and Frose et al. (2011) are represented in Table 2. The LWR results showed a high and significant coefficient of determination (Table 2, Figure 2) for both species, indicating it is a good model. The allometric form of the LWR equation determined from Logarithmic LWR is given in Table 3 for future applications.

The values of “b” obtained for *S. richardsonii* (b = 2.681, Table 2) and *G. gotyla* (b = 2.751, Table 2) from the tributaries of Punatsangchhu River along the Punatsangchhu Hydropower Project area were within the expected range of 2.5 to 3.5 as proposed by Froese (2006). However, the observed parameter “b” differed significantly from the isometric value (b = 3) and showed negative allometric growth for *S. richardsonii* and *G. gotyla* (Table 4). This indicates less increment in weight than as predicted by an increase in length i.e., in the present case fish elongates and becomes less roundish as it grows (Froese et al. 2011). The value of “b” observed in this study for *G. gotyla* differed greatly from Jammu and Kashmir, India (b = 2.171; Awas et al. 2020) and Khyber Pakhtunkhwa, Pakistan (b = 3.435; Khan et al. 2021). In contrast, the value of “b” recorded in this study for *S. richardsonii* falls within the observed range

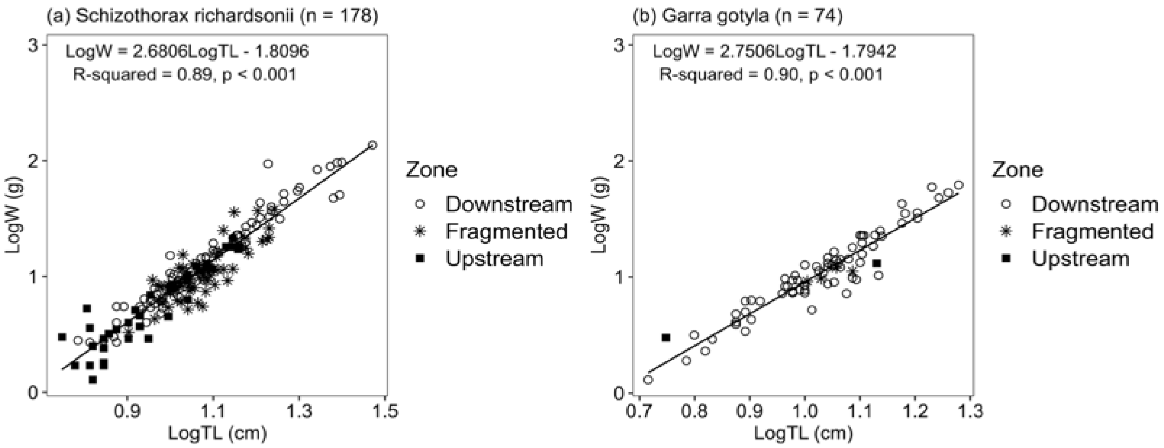

Figure 2. Scatter plot of natural log of total weight against the natural log of total length for (a) *Schizothorax richardsonii*, (b) *Garra gotyla* from tributaries of Punatsangchhu River

Table 3. Logarithmic and allometric LWR equation for fishes from tributaries of Punatsangchhu River

Species	Logarithmic LWR equation	Allometric LWR equation
Schizothorax richardsonii	LogW = 2.6806LogTL – 1.9634	$W = 0.0155L^{2.6806}$
Garra gotyla	LogW = 2.7506LogTL – 1.7942	$W = 0.0161L^{2.7506}$

Table 4. Results of student’s t-test ($p < .05$) comparing observed “b” from isometric value (b = 3)

Species	b	Isometric “b”	*t*	*df*	*p*	Remarks
Schizothorax richardsonii	2.681	3.00	-4.580	176	< 0.001	Negative allometric
Garra gotyla	2.751	3.00	-2.286	72	0.025	Negative allometric

at different spatial scales, i.e., at a smaller spatial scale from hill streams of Uttarakhand Himalaya (b = 2.4 to 3.08; Lohani and Ram 2018) and Indian Himalaya at a greater spatial scale (b = 2.44 to 3.11; Tyagi et al. 2014). Tyagi et al. (2014) attributed this deviation to differences in habitat conditions, life stage and type of length measured.

As highlighted by Froese (2011), $r^2 < 0.95$ and the lower size range of fishes in our case may reflect differences in the life stages of captured fishes [compare Table 2 of this study with Table 1 of Tyagi et al. (2014)]. However, our findings are comparable to those recorded for *S. richardsonii* from a tributary of Koshi River, Uttarakhand (b = 2.653; Goel et al. 2011) where the sample could have possibly been collected during a single fishing expedition as in our case. Considering this, the differences could result from the analysis of samples from a single sampling expedition which could have overlooked the seasonal variation (Froese 2006) as compared to multiple seasons considered by other authors (Tyagi et al. 2014; Lohani and Ram 2018; Khan et al. 2021).

The K_n derived from the LWR reflects the interaction of biotic and abiotic factors that determine the well-being of fish, indicating the quality of existing growing conditions (Froese 2006; Dorji et al. 2018). One-way ANOVA showed a significant difference in K_n among zones for *S. richardsonii* ($F_{2, 160}$ = 26.57, $p < 0.001$, Figure 3a, Table 5). The lowest K_n for *S. richardsonii* observed from the fragmented zone (K_n = 0.88 ± 0.20) followed by the upstream zone (K_n = 0.91 ± 0.20) and their significant variation with the downstream zone (K_n = 1.10 ± 0.16) as indicated by Tukey's HSD test for multiple comparisons (Figure 3a) shows "poor growing conditions" or "deteriorating conditions" upstream of PHPP – II Dam. Although the growth condition of *G. gotyla* was poor in the fragmented zone ($K_n < 1$, Table 5, Figure 3b) and good in the downstream zone ($K_n > 1$, Table 5, Figure 3b), the differences were not significant (t = 1.734, df = 66, p = 0.088, Figure 3b).

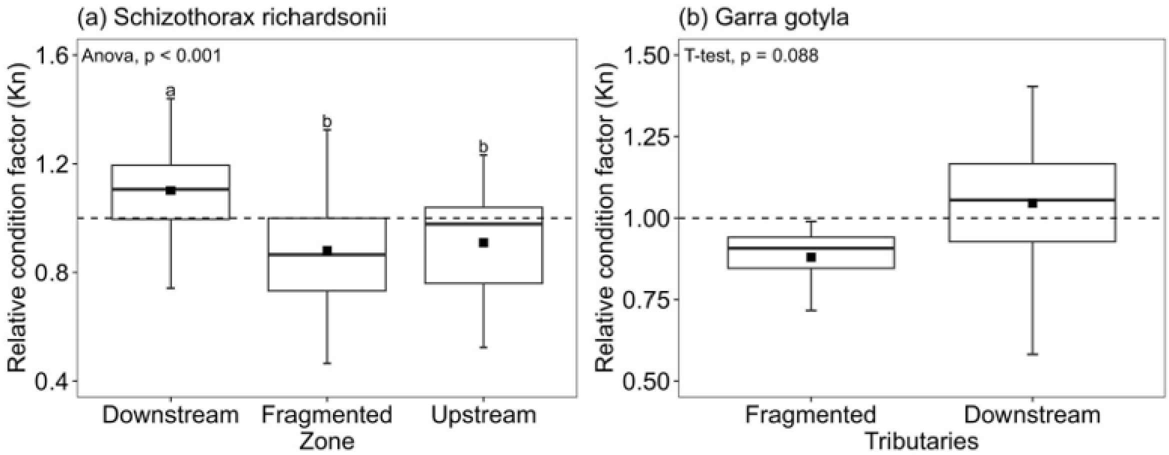

Figure 3. Boxplot showing the distribution and comparison of relative condition factors of fishes among different tributaries zone of Punatsangchhu River: (a) *Schizothorax richardsonii* [Different letters indicate statistically significant differences ($p \leq 0.05$)], and (b) *Garra gotyla*. The horizontal dashed line indicates $K_n = 1$ [$K_n < 1$ = poor growth condition, $K_n > 1$ = good growth condition].

The mainstream Punatsangchhu River between the PHPP – I and PHPP – II Dam is impacted and has been fragmented for more than a decade. Moreover, the tributary from the fragmented zone, i.e., Basochhu is an impacted area having existed as dewatered reach for more than two decades. Such modification of rivers alters flow regime, and habitat conditions and restricts migration of fish to life-stage specific habitats (Bunn and Arthington 2002; Benejam et al. 2016; Zhang et al. 2018; Moreno-Arias et al. 2021) resulting in the decrease of K_n over the years in hydropower impacted areas (Steffensen and Mestl 2016; Nyanti et al. 2021) due to the reduced food availability, increased temperature and increased energy demands of fish under reduced flow condition in dewatered reach (Steffensen and Mestl 2016; Poletto et al. 2018; Nyanti et al. 2021).

Table 3. The relative condition factor of fishes (K_n, mean ± SD**)** from tributaries of Punatsangchhu River.

Species	Upstream	Fragmented	Downstream
Schizothorax richardsonii	0.91 ± 0.20	0.88 ± 0.20	1.10 ± 0.16
Garra gotyla	-	0.88 ± 0.12	1.05 ± 0.20

The results thus, clearly show the impact of a series of hydropower development on fish, with pronounced impact visible along the tributaries in the fragmented or impacted zone and on migratory fish. The greater impacts on migratory species are mainly attributed to life-stage specific habitats distributed across greater spatial scales in contrast to the availability of life-stage specific habitat requirements of resident fishes within smaller spatial scales. Therefore, implementation of minimum environmental flow and subsequent assessment of fish growth under improved flow conditions (Weisberg and Burton 1993; Jacquemin et al. 2014) is recommended for adaptive mitigation of ecological impacts. Our findings contradict to catch per unit effort (CPUE) based assessment made by Khanal et al. (2022b) at the mainstem Punatsangchhu River. Apart from the assessment metrics (CPUE vs K_n in this study), another key difference between the two assessment approaches is the demarcation of the impacted zone. This analysis considered the impacted zone as the currently fragmented zones of Punatsangchhu River (i.e., between dams of PHPP – I and PHPP – II) whereas they considered the stretch between the PHPP – I Dam and the tailrace tunnel (TRT) of PHPP – II (i.e., future dewatered stretch) as the impacted zone. In consideration of this, the hydropower assessment approach in Bhutan should be holistic considering multiple scenarios and assessment metrics. Therefore, we recommend the establishment of LWR and condition factors of fishes before hydropower development and use them as one of the impact assessment metrics. Moreover, we recommend increased sampling efforts to capture adequate specimens.

However, the biggest limitation of this study is the study design where two zones, out of three, were each represented by a single site (i.e., upstream and fragmented zone). Although some tributaries along the upstream and fragmented zones were accessible, the sudden breakdown of the electrofisher during the sampling limited the design of zones with near-uniform sampling sites across all the zones. Therefore, the single-sampling sites considered for the upstream and fragmented zone, the proximity of the upstream river to the PHPP – I Dam, and limited samples of *Garra gotyla* (n = 4) from the fragmented zone could influence the results. Furthermore, this study considers data from a single sampling season and hence comparison of data from multiple sampling seasons is recommended for future assessments.

4. CONCLUSION & RECOMMENDATIONS

Studies on accessing the impact of hydropower development on fisheries and LWR of fishes are scanty in Bhutan. Thus, this study established the first-ever LWR for *S. richardsonii* and *G. gotyla* sampled from tributaries of the Punatsangchhu River and analyzed their well-being. The established LWRs can be used as a reference for two species in Bhutan until LWR for these species from minimally impacted areas at broader scales are defined. The obtained condition factor can also serve as a baseline for long-term monitoring of hydropower impacts. Furthermore, LWR and Kn can be considered as metrics for assessing the hydropower impacts in future hydropower establishments.

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