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Research Article

MORPHOLOGICAL DIVERGENCE OF SNOW TROUT (*SCHIZOTHORAX RICHARDSONII*, GRAY 1932) FROM RIVERS OF NEPAL WITH INSIGHTS FROM A MORPHOMETRIC ANALYSIS

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Abstract

Asala or snow trout (*Schizothorax richardsonii*, Cyprinidae), one of highly valued freshwater fish of Transhimalayan regions, is distributed in upper reaches of all major river systems of Nepal. Morphometric diversification between six river populations of *S. richardsonii* was examined to identify intraspecific unit for enabling better management of the resources. Significant differences were observed in 17 measured morphometric characters of 207 specimens among the six river populations. Multivariate analysis of variance (Wilks' test) indicated a significant difference for mean vectors of mophometric measurements (λ =0.012, F85, 731 = 19.999, P<0.0001) among populations. Principal component and discriminant functions (DFs) analysis of morphometric measurements revealed high seperation of the stocks. The analysis showed that most of the shape and size variation among these populations occurs in the head region, body depth and fin length. Apparent morphometric divergence among *S. richardsonii* samples showed the existence of three differentiated groups viz., the Indrawati and Khudi populations, the Melamchi and Phalaku Rivers, and the Sabha and Tadi River populations of Nepal. The results of this study may be useful in fisheries management and potential exploitation of this species in coldwater aquaculture.

Keywords: Coldwater; fisheries management; morphometric; river population; *S. richardsonii*

Introduction

The capacity of fish populations or stocks to adapt and evolve as independent biological entities is limited by the exchange of genes among populations. Geographical isolation may result in prominent morphometric and genetic differences among stocks within a species (Carvalho and Hauser, 1994), and the morphometric differences among stocks of a species are recognized as an important tool for evaluating the population structure and as a basis for identifying stocks (Turan, 1999). In general, a 'fish stock' is a local population adapted to a particular environment, having genetic differences from other stocks (MacLean and Evans, 1981). Although morphometric characters may be influenced by environmental conditions, they can be valuable in indicating stock discreteness associated with more genetically related features (Ihssen *et al.*, 1981; Szlachciak, 2005).

Morphological study is one of the frequently employed and cost-effective methods of phenotypic characterization for fish stock identification. Traditional multivariate morphometrics, accounting for variation in size and shape, have also successfully been discriminated between many fish stocks (Turan, 1999).Multivariate morphometrics have

successfully been employed in aquaculture studies, in assessing fish health (Loy *et al.*, 2000), estimation of biomass (Hockaday *et al.*, 2000) and population discrimination (Pakkasmaa *et al.*, 1998). Such studies have significance on the relative importance of stock origin and rearing habitat in the determination of gross body morphology. Studies of morphological character variation are, therefore, vital in order to elucidate patterns observed in phenotypic and genetic character variation among fish populations (Beheregaray and Levy 2000).

Schizothorax richardsonii (Gray, 1832) is a coldwater fish, commonly known as Asala or Snowtrout, formed a substantial natural fishery in the major riverine ecosystem of Nepal. The distribution of this cyprinid species is confined to the rivers and streams of Himalayan foot hills across the country. Besides Nepal, this species is distributed in India, Bhutan, Pakistan and Afghanistan (Talwar and Jhingran, 1991). Although *S. richardsonii* is widely distributed along the Himalayan foothills, its populations have been declined from many areas due to introduction of exotic species, damming and overfishing (Negi and Negi, 2010). In view of the conservational value and the aquaculture potential of *S. richardsonii*, there has been a concerted effort to artificially propagate this species (FRD,

2014). Of the aquaculture interest, their inherent biological features such as slow growth, maturity at small size are the main constraints hindering their growth and population increase (Mir *et al.*, 2012). Establishment of founding stock with wider genetic variation and better shape size followed by selective breeding program would help to improve the performance of these traits.

Before going in for a breeding program, it is important to know the genetic make-up of the stocks as it would help in identifying the traits for which the stocks may be superior or inferior. Establishment of founding stock and its improvement through selective breeding requires superior character in shape and size and high level of genetic variation for traits of interest. Species widely distributed in a heterogeneous environment may be expected to exhibit differentiation in genetic or phenotypic characters or both. Among fishes, the likelihood of such character variation increases if the species has limited powers of dispersal (Planes 1998). Morphometric differences among stocks of a species are recognized as important for evaluating the population structure and as a basis for identifying stocks (Turan, 2004). In some cases pattern of morphometric

variation is consistent with differences in the genetic constitution of the stocks (Corti *et al.*, 1988). Therefore, presently reported investigation on stock identification of *S. richarsonii* from different rivers of Nepal through morphometric measurements using the concept of size and shape was undertaken as a step towards the successful development and management of this species.

Materials and Methods

Specimens of *S. richardsonii* were collected using gill net, cast net and local traps from Sabha River, Indrawati River and Melamchi River of Koshi River Systems and Khudi River, Phalaku River and Tadi river of Narayani Rivers Systems of Nepal (Fig. 1). Minimum of thirty specimens from each habitat used for body measurement were collected during September 2013 to February 2015. Samples were bagged individually and placed on dry ice for transport to the laboratory where they were stored in a 4 ºC refrigerator until thawed for measurements and counts. The size of specimans was in between 8.8-31.5 cm in total length and 4.9-443 g in weight. Geographic coordinates of sampling sites, the sample size, mean total length, and weight are given in Table 1.

Fig. 1: Map of Nepal, with sites where populations of *Schizothorax richardsonii* were sampled: (1) Sabha Khola, (2) Indrawati River, (3) Melamchi River, (4) Tadi River, (5) Phalaku River and (6) Khudi River

Table 1: GPS coordinates, altitude (masl; meters above sea level), number of samples, min-max. length and weight of *Schizothorax richardsonii* across the hill stream of Koshi and Narayani River Systems, Nepal

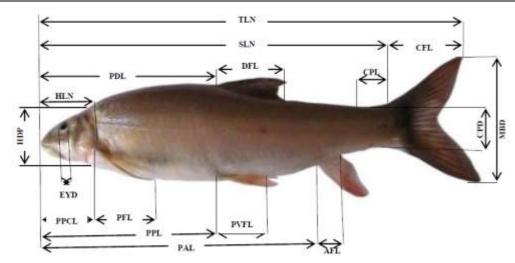| Parameters	Rivers (sites)					
	(1) Sabha, Sakhuwasabha	(2) Indrawati, Kavre	(3) Melamchi, Sindhupalchowk	(4) Tadi, Nuwakot	(5) Phalaku, Nuwakot	(6) Khudi, Lamjung
Latitude N	27º21'49"	27º38'27"	27º52'56"	27º55'29"	27º58'35"	28º11'07"
Longitude N	87º10'47"	85º42'23"	85º32'29"	85º23'06"	85º11'24"	84º27'30"
Altitude (masl)	335	630	985	921	625	638
Number of samples	30	30	30	30	47	40
Min-max TL, cm	11.1-21.0	16.1-31.5	14.3-25.2	8.8-13.0	10.7-24.3	15.0-27.5
Min-max BW, g	9.6-61.6	53.4-443.0	25.0-135.0	4.9-16.4	10.0-125.0	-

Fig. 2: External morphology of *Schizothorax richardsonii* showing the morphometric measurements, taken for each specimen. Abbreviations are given in the texts.

On each specimen 18 point to point measurements were taken using dial and vernier calipers. Definitions of most measurements were obtained from Zafar *et al.* (2002) and Teugels *et al.* (1998). They are (Fig. 2): (1) Total length (TLN), (2) Standard length (SLN), (3) Fork length (FLN), (4) Maximum body depth (MBD), (5) Caudal peduncle depth (CPD), (6) Head length (HLN), (7) Head width (HDP), (8) Eye diameter (EYD), (9) Predorsal length (PDL), (10) Prepelvic length (PPL), (11) Prepectoral length (PPCL), (12) Preanal length (PAL), (13) Dorsal fin length (DFL), (14) Pectoral fin length (PFL), (15) Pelvic fin length (PVFL), (16) Anal fin length (AFL), (17) Caudal fin length (CFL), and (18) Caudal peduncle length (CPL).

Univariate analyses (ANOVA) was conducted to examine body size differences between habitats. Since size distributions were highly overlapping between habitats, the data obtained were entered in a database for subsequent factor analysis. Significant correlations were observed between size and morphometric characters of the samples. An allometric method was used to remove size-dependent variations from all of the characters. The allometric methods are a significant help in achieving the size and shape separation and reasonably meet the statistical assumption (Swain and Foote 1999). All measurements were standardized following Elliott *et al.* (1995), to eliminate any variation resulting from allometric growth.

$$M_{adj} = M(\text{Ls}/\text{L0})^b$$

Where, M is the original measurement, M_{adj} is the size-adjusted measurement, L_o is the TL of the fish, and L_s is the overall mean of the TL for all fish from all samples. Parameter b was estimated for each character from the observed data as the slope of the regression of log M on log L_o, using all fish in all groups. This method effectively removes allomectric variation due to differences in fish size (Pakkasmaa *et al.*, 1998). The transformed data were used for multivariate analysis and the total length was excluded from the final analysis.

Seventeen morphometric characters subject to univariate analysis of for the evaluation of significant difference among the six locations. The transformed data were subjected to principal component analysis (PCA) and discriminant analysis to examine any phenotypic differences between the populations. Principal component analysis (PCA) based on the correlation matrices was done to create uncorrelated principal factors from the original variables. The data were further analyzed with discriminant function analysis (DF) exploring the variables most useful for discriminating *S. richardsonii* populations' habitats. This procedure predicts the habitat of origin for each individual by chance. PCA and DF were computed using STATISTICA (StatSoft Inc. ver 5.0) and SPSS (ver 20), respectively.

Results

There was no significant correlation between any of the transformed measured morphometric variables and standard length (P > 0.001), demonstrating that the size effect was successfully removed.

Principal components (PC) with eighenvalues higher that 1.00 of importance were considered (e.g. Chatfield and Collins, 1983). Based on this criterion, two components were extracted which explained about 89.8% of the variation of the original size-adjusted body morphology variables. PC1 and PC2 accounted for 80.24% and 9.54% of total variance, respectively, and was positively correlated to some variables and negatively correlated with others, showing that there is variation due to body shape (Table 2). The first component (PC 1) was composed mainly of the body depth, head region and fins length. The high component loadings in PC 2 were from the characters which mostly contributed to caudal peduncle region and eye size. Thus, the PC 1 characters are best associated with the swimming capacity of the fish while the second component (PC 2) characters associated with feeding and foraging.

Table 2: Principal component loadings for morphometric characters in *Schizothorax richardsonii* collected from rivers of Nepal. The PCA loadings are listed together with the variables correlations (r) with the component scores. The highest component loadings are indicated in boldface

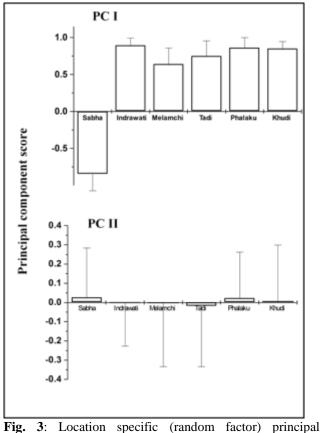| Component	PC 1	r	PC 2	r
Standard length (SLN)	**0.930**	0.063	0.269	0.447
Fork length (FLN)	**0.912**	0.062	0.300	0.498
Maximum body depth (MBD)	**0.955**	0.065	0.000	0.000
Caudal peduncle depth (CPD)	-0.003	-0.005	**0.895**	0.061
Head length (HLN)	**0.947**	0.065	-0.108	-0.179
Head width (HDP)	**0.939**	0.064	-0.085	-0.142
Eye diameter (EYD)	-0.568	-0.943	**0.726**	0.050
Predorsal length (PDL)	**0.980**	0.067	0.096	0.160
Prepelvic length (PPL)	**0.962**	0.066	0.120	0.199
Prepectoral length (PPCL)	**0.975**	0.066	-0.025	-0.041
Preanal length (PAL)	**0.951**	0.065	0.221	0.367
Dorsal fin length (DFL)	**0.945**	0.064	-0.069	-0.114
Pectoral fin length (PFL)	**0.973**	0.066	-0.051	-0.085
Pelvic fin length (PVFL)	**0.958**	0.065	-0.081	-0.134
Anal fin length (AFL)	**0.952**	0.065	-0.035	-0.059
Caudal fin length (CFL)	**0.941**	0.064	-0.103	-0.172
Caudal peduncle length (CPL)	0.202	0.003	**0.614**	0.055
Eigen values	14.06		1.20	
% of variance	80.24		9.54	
Cumulative % of variance	80.24		89.78	

Fig. 3: Location specific (random factor) principal component scores (mean with 95% confidence limit) for the five population of *S. richardsonii* studied.

Location specific (random factor) principal component scores clearly separate the population of *S. richardsonii* of Sabha River from the population of other rivers (Fig. 3). Average of component loadings was higher for Indrawati River population at PC1. Average of component loadings for location specific populations separates *S. richardsonii* populations of Indrawati, Melamchi and Tadi Rivers from the populations of Sabha, Phalaku and Khudi Rivers (Fig. 3). PCA graphs of PC1 and PC2 scores for each sample revealed clear separation among six populations of S. richardsonii.

Discriminant function analysis (DF) was used to look for, in more detail, the body shape variables which are most explicitly differentiating among the six populations of *S. richardsonii* originating from rivers. Wilks λ tests of discriminant analysis indicated significant differences in morphometric characters of all populations ($P < 0.001$). The DF was analysed based on the correlation matrix of the size-adjusted variables, thus provides equal weight for variation in all variables. The two discriminating functions were produced from the 17 morphometric variables. The first canonical discriminant function of the discriminant analysis explained 63.2% of the total variance while the second one accounted for 20.9% of the total variance. However, the functions emphasize the body-shape variables more than the principal component does (Table 3). The morphometric truss measurements, SLN, FLN, MBD, HDP, PDL, PPL,

PPCL, PAL, DFL, PFL and AFL contributed to DF1 while CPD, EYD, CFL and CPL characters contributed to DF2 (Table 3), showing that these characters were the most important in differentiating of the populations.

The multiple scatter plots of discriminant function (DF) axes DF1 vs. DF2 explained 84% of total variance among the samples and demonstrated significant distinction among *S. richardsonii* groups from the Rivers of Nepal (Fig. 4). The DF1 comprised of 63.2% of total variance completely separated Indrawati and Khudi populations from Sabha and Tadi populations, whereas Phalaku and Melamchi populations showed slight intermingling with those two groups of population at this axes. DF2 accounted for 20.9% of the total variance discriminated Indrawati and Melamchi populations and other four populations intermingled between these two populations.

Because the PCA and DF showed that the populations of *S. richardsonii*, separated by habitats, differed with one another, a further analysis was performed. Multivariate (Wilks' λ) and univariate F-test run for each habitat as the independent variable and all morphological charac-ters revealed differences in several traits (Table 4). In Indrawati and Khudi Rivers, the population had wider eye (P<0.001), longer fins with farther apart (P<0.001) than other four populations. They also tended to have longer and wider head (P<0.001), large caudle peduncle area (P<0.001) amongst *S. richardsonii* populations. Tadi River population followed by Sabha populations had closer and short fins, head region and caudle area (P<0.001). The length and apartness of most of the morphometric characters in Melamchi and Phalaku populations was intermediate (Table 4).

Table 3. Canonical discriminant function (DF), standardized by within variances, and correlations (r) with the size adjusted morphometric variables. Largest coefficients (absolute values) for each variable are indicated by asterisk

Component	DF1	r	DF2	r
Standard length (SLN)	0.721*	0.420	0.037	0.243
Fork length (FLN)	0.692*	0.875	0.009	0.176
Maximum body depth (MBD)	0.670*	0.706	0.063	0.080
Caudal peduncle depth (CPD)	-0.077	-0.235	0.541*	0.137
Head length (HLN)	0.563*	-0.243	0.341	1.861
Head width (HDP)	0.502*	-0.454	0.007	-0.984
Eye diameter (EYD)	0.228	0.249	0.299*	-0.016
Predorsal length (PDL)	0.716*	0.717	0.023	-0.140
Prepelvic length (PPL)	0.662*	0.126	0.045	0.021
Prepectoral length (PPCL)	0.650*	0.304	0.099	-0.357
Preanal length (PAL)	0.617*	-1.335	-0.001	-0.965
Dorsal fin length (DFL)	0.535*	-0.108	0.130	-0.032
Pectoral fin length (PFL)	0.641*	0.256	0.067	-0.503
Pelvic fin length (PVFL)	0.589	0.119	0.065	-0.066
Anal fin length (AFL)	0.512*	-0.506	0.088	-0.360
Caudal fin length (CFL)	0.314	1.243	0.544*	0.042
Caudal peduncle length (CPL)	-0.028	-0.117	0.446*	0.008
Eigen values	6.364		2.107	
Canonical correlation	0.929		0.823	
Cumulative variance explained	63.15		84.06	

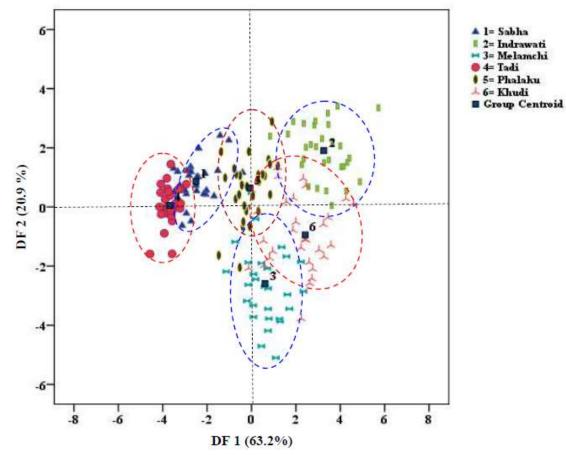Wilks' λ = 0.012, $F_{85, 731}$= 19.999, P<0.0001

Fig. 4. Discriminant function analysis scores (DF) of morphometric characters of *Schizothorax richardsonii*

Table 4. Mean size and variation of measured morphological characters of *S. richardsonii* among habitats. Statistical differences between habitats are based on multivariate (Wilks' λ) and univariate F-tests. Differences of morphological characters between habitats were determined by pairwise comparison (Tukey's test).

Morph character	*S. richardsonii* population						Tukey's test	P value
	Sabha	Indrawati	Melamchi	Tadi	Phalaku	Khudi		
SLN	11.7±2.1	20.1±0.8	15.8±0.9	8.5±0.2	14.7±0.6	18.7±4.8	ID=KD>PK=MH>SB>TD	0.0001
FLN	12.1±2.5	21.8±0.6	17.1±0.5	9.3±0.2	16.1±0.5	20.7±5.7	ID=KD>MH=PK>SB>TD	0.0001
MBD	2.4±0.4	4.6±0.9	3.5±0.5	1.7±0.2	3.2±0.6	4.2±0.6	ID>KD>MH=PK>SB>TD	0.0001
CPD	1.1±0.2	2.2±0.5	1.8±0.4	1.8±0.1	1.5±0.3	1.9±0.4	ID=KD≥MH>PK>SB>TD	0.0001
HLN	2.6±0.4	4.2±0.6	2.8±0.4	2.0±0.2	3.1±0.4	3.4±0.7	ID>KD=PK≥MH>SB>TD	0.0001
HDP	1.8±0.3	2.8±0.5	2.3±0.4	1.4±0.1	2.1±0.4	2.6±0.4	ID=KD≥MH≥PK>SB>TD	0.0001
EYD	0.6±0.1	0.8±0.1	0.6±0.1	0.5±0.1	0.7±0.1	0.7±0.1	ID=PK≥KD>MH>SB≥TD	0.0001
PDL	5.3±1.0	9.1±1.1	7.2±0.7	4.3±0.3	6.9±0.8	8.7±1.4	ID=KD>MH=PK>SB>TD	0.0001
PPL	6.1±1.3	10.1±1.2	7.9±1.3	4.5±0.3	7.6±0.9	9.6±1.6	ID=KD>MH=PK>SB>TD	0.0001
PPCL	2.6±0.4	4.3±0.6	3.4±0.4	2.0±0.2	3.2±0.5	3.8±0.5	ID>KD>MH=PK>SB>TD	0.0001
PAL	6.1±1.3	10.1±1.2	7.9±1.3	4.5±0.3	7.6±0.9	9.6±1.6	ID=KD>MH=PK>SB>TD	0.0001
DFL	2.8±0.6	4.3±0.8	3.4±0.5	1.8±0.3	3.4±0.5	3.8±0.7	ID>KD>PK=MH>SB>TD	0.0001
PFL	2.1±0.3	3.4±0.6	2.7±0.4	1.5±0.2	2.8±0.4	3.2±0.5	ID=KD>PK=MH>SB>TD	0.0001
PVFL	2.0±0.4	3.2±0.6	2.6±0.4	1.4±0.1	2.7±0.4	3.1±0.5	ID=KD>PK=MH>SB>TD	0.0001
AFL	2.2±0.5	3.9±0.8	2.8±0.4	1.5±0.2	2.9±0.7	3.6±0.8	ID=KD>PK=MH>SB>TD	0.0001
CFL	3.7±0.7	5.7±0.7	3.9±0.6	2.6±0.3	4.3±0.7	4.9±0.9	ID>KD>PK=MH≥SD>TD	0.0001
CPL	1.4±0.4	2.5±0.5	2.2±0.4	1.1±0.2	1.9±0.6	2.1±0.5	ID>MH=KD=PK>SB=TD	0.0001
Multivariate P value based on Wilks' Lamda								<0.0001

Discussion

Differences in river ecology and habitats like flow regime and foraging opportunities might create selective pressure in morphological divergence in the intraspecific popolations. Morphological characteristics of *S. richardsonii* populations in different rivers are best discriminaed by multivariate analysis (PCA and DF) (Table 2 and 3). The observations of morphological characters indicated that *S. richardsonii* of Indrawati and Khudi Rivers demonstrates differences in the complexes of morphological features from the Tadi and Sabha Rivers of Nepal. Divergence of stock structure appear from the reflection of the first and second principal component and discriminant factors. This study showed that the variation is evident in the head region (head length and width), body size and position of fins, and the caudal region (caudal fin length and caudal peduncle length), which were useful for the stock separation. Those characters reflect the swimming, feeding and foraging ability of the fish. The variation in the caudal region of specimens from the six water bodies could be a consequence of phenotypic plasticity of fish in response to uncommon hydrological conditions (Mir *et al.*, 2013).

The differences in the morphometry of *S. richardsonii* between these rivers could also be because of their geographic and topographic differences. The sampling sites on Indrawati and Khudi Rivers are more wider (100-200 m) and deeper (3-5m) with relatively low human influences towards upstream whereas, the site on Sabha and Tadi Rivers are narrow (<50m) and shallow (<2m) with extensive human interruptions towards upstream. The sites on Phalaku and Melamchi Rivers are characterized by narrow and high slope gradients producing turbulent water conditions. Within this hydrological set-up, these pressures can result in more resistance on fish body during the swimming. Most fish in rivers and streams are presumably habitat specialists that could evolve various morphological and behavioral adaptations to exploit specific habitat types (Wood and Bain 1995).

Body depth and fin size are the two important morphological characters of stream fish which affect static location and moving manipulation (Douglas and Matthews, 1992). On the basis of morphological data, Indrawati and Khudi River populations were the most divergent. Compared to the six river populations of *S. richardsonii*, the two populations from relatively deeper Indrawati and Khudi Rivers had the most large fins, head, wider eyes and deeper body (Table 4). This pattern of deeper body is consistent with the observation made on *Atherinops affinis*, in lakes of California (Reilly and Horn 2004) that the body depth of fishes increases in response to warmer water temperature. Fishes in low velocity and deep water are more often deeper bodied with larger caudal areas for improved burst-swimming performance and increased maneuverability

(Langerhans, 2008). Shorter pectoral fin length of *S. richardsonii* populations measured in Tadi, Sabha and Melamchi Rivers associated with colder water temperature and faster flowing did support the findings of Barlow (1961). Fishes that evolved in faster flowing water tend to be more streamlined to reduce drag (Langerhans, 2008). Bagherian and Rahmani (2009) also reported that high water velocity leads to slender body shape in a Caspian cyprinid (Alburnus chalcoides, Güldenstädt 1772). Thus, the different current pattern of these water bodies may have been playing an important role in modifying the morphology of *S. richardsonii* among these water bodies. A more cylindrical body shape paired with short pectoral fins length of *S. richardsonii* in Sabha and Tadi Rivers measured in this study might have been the fish plasticity to allow individuals to better confer swift flowing habitats with high substrate heterogeneity. Environmental parameters such as water temperature, conductivity and substrate heterogeneity influence morphological traits of *S. richardsonii* and other fish systems have been well documented (Chuang, *et al.*, 2006; Langerhans *et al.*, 2003; Rajput *et al.*, 2013).

The head morphology reflects a species' feeding habits (Skúlason *et al.*, 1989). *S. richardsonii* is known to be planktivore species, as adult it feeds upon aquatic plants, algal slime, and slimy deposits on rocks (Shrestha 1979). The first principal component consisted of head region, being a strong classificator, indicate the foraging habits of the studied populations. Relatively large heads of Indrawati and Khudi River populations found in the present study may enhance the capture of small prey (Baumgartner *et al.*, 1988).

The eye diameter can reflect the light conditions where the fish are living (Pakkasmaa *et al.*, 1998). Visual acuity of fish is limited by the amount of light available and the depth at which an individual lives. One of the most common visual adaptations of fish seen in the deeper zone of water, where the amount of light diminishes exponentially with depth, is the enlargement of the eye and pupil area (De-Busserolles, 2013). In this respect, the *S. richardsonii* populations in two large and relatively deeper rivers (Indrawati and Khudi) have large eyes. The fish live in glacial fed perennial and shallow Sabha and Tadi rivers, where water is quite clear, have small eyes. Baumgartner *et al.* (1988) suggested that the eye size may as well be related to feeding behavior.

The adaptation of *S. richardsonii* populations from the relatively large rivers Indrawati and Khudi reflects their body morphology: they are relatively strong with long and distant apartness of fins, which are related to slow and precise movement (Ehlinger, 1990); large fins are also of advantage in maintaining one□s position in the river (Riddell and Leggett, 1981). These river populations are more streamlined. That kind of body shape allows for efficient cruising, foraging for patchily distributed prey in

large volumes of torrential open water, and migration (Baumgartner *et al.*, 1988; Robinson and Witson, 1996).

Phenotypic variation among natural populations sometimes reflects genetic adaptation to local selective pressures (Schluter, 2000). At other times, it reflects plastic responses to local environmental conditions (James, 1983). Most of the time, populations could diverge via alternative, genetically based morphologies, or through environmentally induced phenotypes (Langerhans *et al.*, 2003). Morphological and genetic characters of fishes have been shown in some cases to co-vary (Dynes *et al.*, 1999; Houser *et al.*, 1995). Partitioning of the relative contributions of genetic and environmental effects is particularly important for understanding the factors that promote or constrain evolutionary diversification. Interactions between natural selection and gene flow are often invoked to explain patterns of phenotypic variation in the wild. Whether the observed morphological patterns were produced in this study through genetic differences or phenotypic plasticity is unknown. Crabtree (1986) found substantial morphological variation associated with genetic variation in *Atherinops affinis*. Similarly, greatest differences in genetic, morphometric and meristic data are known between wild and cultured tilapia (*Oreochromis* spp.) with high and low levels of genetic variation, respectively (Barriga-Sosa *et al.*, 2004). Indeed, low genetic diversity is commonly reported for natural population of fishes, perhaps largely because of high gene flow in the continuous water environment (Grant and Bowen, 1998). The analyses of the present study revealed variation among *S. richardsonii* populations in several morphological characters: body depth, head size, eye diameter, fins positioning and length. This apparent plasticity may be an adaptive response (Scheiner and Callahan, 1999) to a lesser extent and more genetically controlled. In species which have a wide range of zoogeographical distribution, most of the characters are strongly influenced by the environment and for the species showing restricted distribution the morphometric characters are genetically controlled (Vladykov, 1934). In a study, Negi and Negi (2010) reported that 90% variation in morphometric characters of *S. richardsonii* populations from Uttar Kashi, India are genetically controlled and environmentally controlled characters are a few (10%). Even though genetic differentiation so far has not been demonstrated in present *S. richardsonii* populations, molecular characters may yet be discovered that would explain more of the observed morphological variation.

Conclusion

In conclusion, the analyses of the present study revealed variation among *S. richardsonii* populations in Nepal in several morphological characters. These morphological variations observed in *S. richardsonii* should be considered in its biodiversity conservation and should also be used as a preliminary step towards exploitation of this species in aquaculture and any stock enhancement program. Nevertheless, further studies on genetic differentiation among the populations of *S. richardsonii* using molecular markers would help to elucidate the observed morphological discrepancy.

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