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A Morphometric Study on Mahseer (*Tor putitora*) From a Mid-Hill Lake and Rivers of Nepal

S.K. WAGLE[1*], N. PRADHAN[2], A.K. RAI[2]

[1]Fisheries Research Center
P.O. Box 274
Pokhara, Kaski, Nepal

[2]Fisheries Research Division
Godawari, Lalitpur, Nepal

Abstract

Mahseer or Sahar (*Tor putitora*, Cyprinidae), one of highly valued, mighty and giant freshwater fish of Transhimalayan regions, is distributed in all major river systems and mid hill lakes of Nepal. It has been hypothesized that differences between habitats (e.g. flow regime, foraging opportunities) might create selective pressures resulting in morphological divergence between intraspecific populations. Morphometric diversification between three river populations (Koshi, Trishuli, Kali Gandaki) and one lake population (Phewa lake) of mahseer was examined to identify intraspecific unit for enabling better management and perpetuation of the resources.

Morphometric analysis showed that most of the shape variation among these populations occurs in the head region, body depth and fin length. Lake population of mahseer was found to diverge most from river populations. The characters that best discriminated the river and lake population of mahseer were associated with locomotion patterns and foraging behavior of fish. The mahseer may be phenotypically plastic in response to the environmental conditions of the habitat of each population.

* Corresponding author: Tel.: +977-61-560089, 9846047454, Fax: +977-61-560825
E-mail address: waglesk@yahoo.com

Introduction

Among 184 indigenous fishes reported from Nepal (Shrestha 1995), the mahseer, *Tor putitora*, forms one of the most popular freshwater sport fish throughout the trans-himalayan mid hill waters. Mahseer is distributed in all major river systems and lakes of Nepal and serve as food fish supporting a substantial natural fishery (Swar 2002). Despite its significant contribution to the natural stock of fish in Southeast Asian region, the natural stocks of mahseer have declined to such an extent that they have been identified as a critically endangered species in the region (Chandra and Haque 1982; Islam 2002). The main causes were encroachment caused by industrialization, urbanization, agricultural development along water bodies, illegal methods of fishing, ecological alterations and physical changes in natural environment (Swar 2002). These have imposed deleterious long-term consequences on population dynamics of this species. Therefore, awareness has grown in recent years to conserve biological diversity of mahseer at all levels (from genes to ecosystems) in a sustainable manner.

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 notable morphological, meristic and genetic differences among stocks within a species, which may be recognizable as a basis for the conservation and management of distinct stocks. Meristic (Creech 1992) and morphometric (Shepherd 1991; Haddon and Willis 1995) characters have been commonly used as markers in fisheries biology for stock identification. Multivariate morphometrics have successfully been employed in aquaculture studies, in assessing fish health (Loy et al. 2000), estimation of biomass (Hockaday et al. 2000), conservation driven biogeographical studies (Haas and McPhail 2001) and population discrimination (Friedland and Reddin 1994; Pakkasmaa et al. 1998). Such studies raise questions on the relative importance of stock origin (genetics) and rearing habitat (environment) 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).

The water bodies of Nepal are characterized by complex climatic conditions and are known to support a rich flora and fauna. Species widely distributed in such 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). In a study of stock differences in the common carp *Cyprinus carpio* L., Corti et al. (1988) found the pattern of morphometric variation to be consistent with differences in the genetic constitution of the stocks, although the inheritance of shape-determining traits was not fully understood. Nevertheless, selection pressure on heritable traits governing shape would be expected to differ between fishes growing in different environments, leading to greater survival of some genotypes in some habitats (Cramon-Taubadel et al. 2005). Mahseer is an ideal species in which to study such variation in phenotypic characters because of its broad distribution in the lotic and lentic environment of Nepal. Body shape variation obtained from morphometric study can reflect ecological and behavioral differences and provide useful information on the ecology and evolution of fishes (Walker 1997; Douglas et al. 2001). Therefore, in the present study morphometric data were used to determine the morphological differences in mahseer populations originating from the geographically isolated rivers Kali Gandaki, Trishuli and Koshi and Phewa lake.

Materials and Methods

Specimens of mahseer were collected from three glacial fed rivers (Figure 1) (1) Kali Gandaki (27^o 58' N, 83^o 35' E), (2) Trishuli river (27^o 58' N, 84^o 52' E), (3) Koshi river (26^o 43' N, 87^o 20' E), and lake Phewa (28^0 13' N, 84^o 00' E). Thirty-five specimens from each habitat used for body measurement were collected during September 2004 to April 2005. After capture with gill net, umbrella net and local traps, specimens (Figure 2a) were bagged individually, and placed on dry ice for transport to the laboratory where they were stored in a -20^0 C freezer until thawed for measurements and counts.

On each specimen 20 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 (Figure 2b): (1) Total length (TL), (2) Fork length (FL), (3) Standard length (SL), (4) Predorsal length (PDL), (5) Head length (HL), (6) Head width (HW), (7) Dorsal fin length (DFL), (8) Caudal peduncle length (CPL), (9) Caudal fin length (CFL), (10) Body height (BH), (11) Eye diameter (ED), (12) Prepectoral length (PPEL), (13) Pectoral fin length (PEFL), (14) Prepelvic length (PPL), (15) Pelvic fin length (PFL), (16) Preanal length (PAL), (17) Anal fin length (AFL), (18) Caudal peduncle depth (CPD), (19) Maximum body depth (MBD) and (20) Inter orbital width (IOW) (not shown in figure).

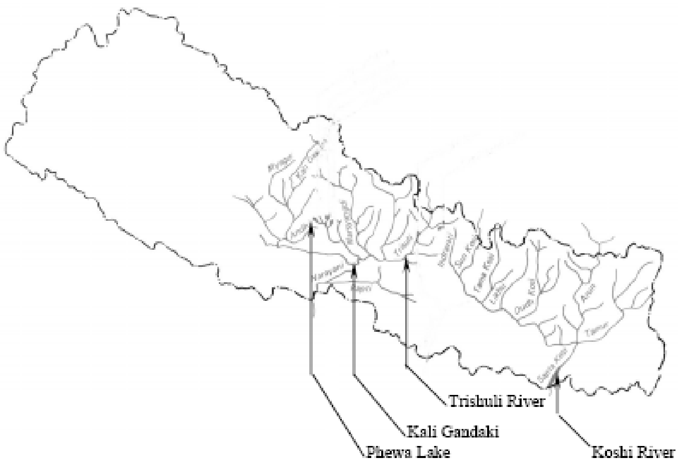
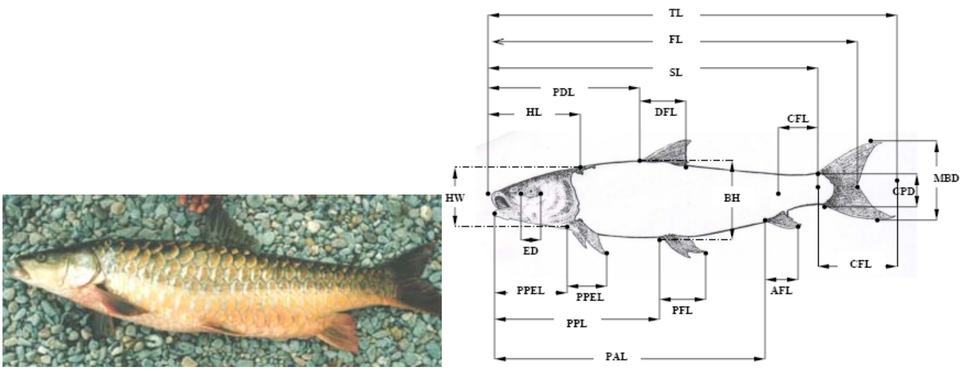


Figure 1. Map of Nepal, with sites where populations of mahseer (*T. putitora*) were sampled: (A) Phewa Lake, (B) Kali Gandaki River, (C) Trishuli River, and (D) Koshi River.



(a) (b)

Figure 2. (a) Photo of freshly caught mahaseer (*Tor putitora*), (b) external morphology of mahseer (*T. putitora*) showing the morphometric measurements, taken for each specimen. Abbreviations are given in the texts.

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. Because of differences in size (TL), size-adjusted values in data analyses were used. Thus, the first step in analyzing the data was to calculate linear regressions against TL of the fish for all the other measured characters. This method effectively removes allomectric variation due to differences in fish size (Pakkasmaa et al. 1998). The standardized regression residuals were then applied in statistical analysis.

Principal component analysis (PCA) based on the correlation matrices was done to create uncorrelated principal components from the original variables. The data were further analyzed with discriminant function analysis (DF) exploring the variables most useful for discriminating mahseer between habitats. This procedure predicts the habitat of origin for each individual by chance. Both PCA and DF were computed from regression residuals using STATISTICA (StatSoft Inc. ver 5.0).

Results

Principal components with eighenvalues higher that 1.00 of importance were considered (e.g. Chatfield and Collins 1983). According to this criterion, three components remained, explaining about 42% of the variation of the original size-adjusted body morphology variables (Table 1). The first component (PC 1) was composed mainly of the preanal fin length, predorsal fin length and fork length. The second component (PC 2) consisted of the dorsal, pectoral and anal fin length. Thus, the PC 1 and PC 2 pooled characters are associated with the swimming ability of the fish. The third component (PC 3) represents of the head width, head length and eye diameter, characters associated with feeding and foraging.

Table 1. Principal component analysis (PCA) form mahseer (*T. putitora*) from lake and rivers. PCA was computed from correlation matrix using regression residuals as the initial variables. 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	PC 3	r
Standard length	0.644	0.232	-0.057	0.005	0.148	0.004
Fork length	0.666	0.242	-0.088	-0.009	0.107	-0.016
Maximum body depth	-0.285	-0.122	0.162	0.076	0.244	0.127
Caudal peduncle depth	0.067	0.040	0.078	0.024	-0.162	-0.062
Head length	-0.004	-0.067	-0.058	0.030	0.828	0.315
Head width	0.125	-0.015	-0.009	0.053	0.808	0.301
Interorbital width	0.506	0.231	0.232	0.087	-0.402	-0.178
Eye diameter	0.122	-0.016	-0.273	-0.065	0.703	0.243
Predorsal length	0.396	0.105	0.073	0.084	0.631	0.217
Preanal length	0.755	0.277	0.061	0.059	0.160	0.008
Prepelvic length	0.682	0.251	-0.081	-0.008	0.075	-0.029
Prepectoral length	0.504	0.163	-0.190	-0.045	0.302	0.065
Dorsal fin length	-0.134	-0.021	0.702	0.284	-0.112	0.013
Pectoral fin length	-0.033	0.010	0.694	0.290	-0.018	0.041
Pelvic fin length	0.013	0.032	0.805	0.337	-0.029	0.041
Anal fin length	0.053	0.053	0.703	0.287	-0.151	-0.016
Caudal fin length	-0.253	-0.092	0.309	0.126	0.065	0.065
Caudal peduncle length	-0.057	-0.044	0.143	0.081	0.352	0.149
Body height	0.099	0.039	-0.125	-0.054	-0.064	-0.041
Eigenvalues	3.65		2.31		2.09	
% of variance	19.23		12.17		10.98	
Cumul. % of variance	19.23		31.40		42.38	

The PC 1 and PC 2 clearly separate the population of mahseer of Phewa lake from the river (Koshi, Kali and Trishuli) populations (Figure 3). Despite that the PC 3 characterizes weakly the four populations, the populations from Phewa lake and Trishuli river differ from the populations of Koshi and Kali rivers-in terms of body shape - longer head and smaller eye.

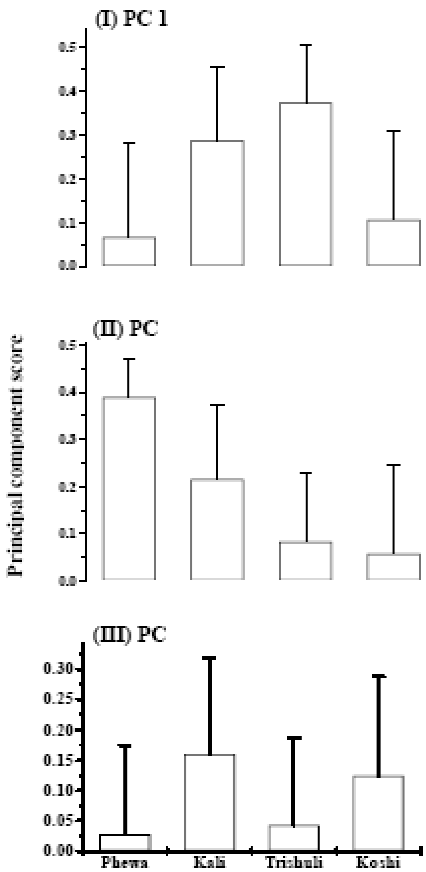


Figure 3. Location specific (random factor) principal component scores (mean with 95% confidence limit) for the four population of mahseer (*T. putitora*) studied.

Discriminant function analysis (DF) was used to look for, in more detail, the body shape variables which are most explicitly differentiating among the four populations of *T. putitora* originating from lake and rivers. The DF was based on the correlation matrix of the size-adjusted variables, thus giving equal weight for variation in all variables. However, the functions emphasize the body-shape variables more than the principal component does (Table 2).

The test of overall discrimination on morphometric data (Table 1) for the four populations of mahseer was highly significant ($P<0.001$). The multiple scatter plots of discriminant function (DF) axes 1 and 2 (Figure 4) showed nearly complete separation between lake and river populations on DF 1 and much overlap between the river populations on DF 2. These two axes accounted for 94% of the variation among the four populations.

Table 2. 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 in bold.

	DF1	r	DF2	r
Maximum body depth	0.418	0.129	0.019	0.050
Head length	-0.418	-0.387	0.900	0.449
Head width	-0.346	-0.401	0.022	-0.091
Eye diameter	-0.590	-0.630	-0.576	-0.346
Predorsal length	-0.154	-0.295	-0.164	-0.040
Prepectoral length	-0.088	-0.210	0.211	0.264
Dorsal fin length	0.350	0.268	-0.105	-0.014
Pectoral fin length	0.297	0.180	-0.071	-0.037
Pelvic fin length	0.360	0.216	0.150	0.175
Caudal peduncle length	0.196	0.041	-0.523	-0.237
Eigenvalue	3.34		1.26	
Canonical correlation	0.877		0.658	
Cumulative variance explained	76.5		94.0	

Wilks' λ =0.103 $F_{57,364}$ = 7.289, P<0.001

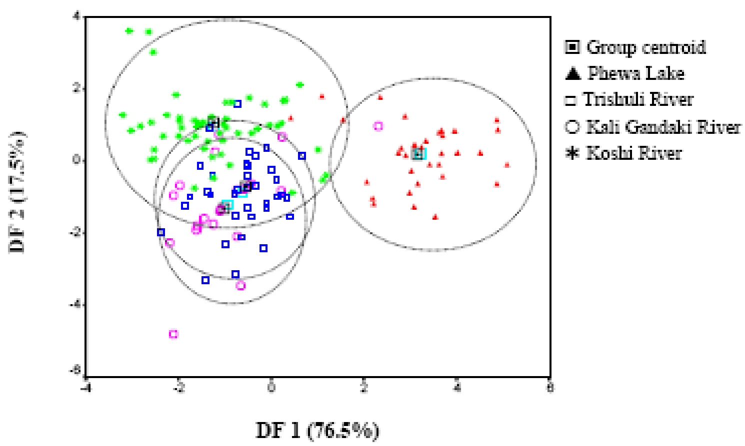


Figure 4. Discriminant function analysis scores (DF) of morphometric characters of mahseer (*T. putitora*).

Because the PCA and DF showed that the populations of mahseer, separated by lake and river habitat, 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 characters (regression residuals were used to equalize variances) revealed differences in several traits (Table 3).

Table 3. Mean size and variation of measured morphological characters of mahseer (*T. putitora*) among habitats (Pkewa lake, Koshi river, Kali river and Trishuli river). 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).

Traits	Mean ± SD				Differences among habitats (Tukey's test)	P-values
	Phewa (PL)	Kali (KA)	Trishuli (TR)	Koshi (KO)		
Standard length	18.16±9.01	17.63±5.57	17.90±2.94	21.17±6.31	KO>KA=TR=PL	0.046
Fork length	19.80±9.42	19.16±6.01	19.55±3.16	23.38±6.81	KO=KA=TR=PL	0.394
Maximum body depth	5.59±2.46	4.84±1.20	4.78±1.02	6.38±2.27	KO=PL>KA=TR	0.001
Caudal peduncle depth	2.01±0.89	1.81±0.55	2.03±1.69	2.26±0.67	KO=KA=TR=PL	0.779
Head length	3.54±1.85	4.09±1.10	4.44±0.77	5.33±1.10	KO=TR>KA=PL	0.000
Head width	2.71±1.20	3.03±0.85	2.91±0.53	3.64±0.97	KO=KA>TR=PL	0.000
Interorbital width	1.49±0.70	1.35±0.47	1.39±0.28	1.59±0.49	PL>KO=KA=TR	0.021
Eye diameter	0.54±0.18	0.96±0.20	0.79±0.11	0.95±0.21	KA>KO>TR>PL	0.000
Predorsal length	8.55±4.26	8.85±2.82	8.95±1.39	10.68±2.79	KO>KA=TR>PL	0.000
Preanal length	13.57±6.80	13.23±4.13	13.42±2.15	16.10±4.88	KO=KA=TR=PL	0.126
Prepelvic length	9.08±4.32	8.82±2.60	9.13±1.44	10.61±2.93	KO=KA=TR=PL	0.272
Prepectoral length	4.48±2.15	4.50±1.26	4.76±0.65	5.49±1.48	KO=KA=TR>PL	0.000
Dorsal fin length	4.70±1.49	4.12±1.06	4.19±0.51	4.92±1.27	KO=KA=TR>PL	0.000
Pectoral fin length	3.39±1.50	3.03±1.03	3.07±0.55	3.75±0.93	KO=KA=TR>PL	0.001
Pelvic fin length	3.01±1.22	2.63±0.84	2.75±0.45	3.38±0.85	KO=PL>KA=TR	0.000
Anal fin length	3.15±1.36	2.81±1.04	2.98±0.52	3.68±1.04	KO=PL=TR>KA	0.007
Caudal fin length	4.60±2.82	4.33±1.51	4.52±0.65	5.05±1.48	KO=KA=TR=PL	0.343
Caudal peduncle length	3.13±1.66	2.99±1.04	2.86±0.66	3.48±1.03	KO=KA=TR=PL	0.078
Body height	4.13±1.75	4.02±1.30	4.33±2.86	5.07±1.48	KO=KA=TR=PL	0.804
Multivariate P value based on Wilks' Lamda						<0.001

In lake Phewa, the population had closer eye (P<0.001) with farther apart (P<0.002) than the river populations. It also tended to have shorter and narrow head (P<0.001), shorter dorsal and pectoral fin lengths (P<0.001) than the river populations. However, mahseer in lake Phewa had large pelvic fin and highest body depth (P<0.001). Among river populations, the head region of mahseer was greatest (P<0.001) in Koshi river, while bigger eye size (P<0.001) was evident in population of Kali river.

Discussion

With multivariate statistics (PCA and DF) the morphological characters that best discriminated mahseer populations of lake and river origin were identified. Especially the head size and body depth as well as eye diameter, pectoral and dorsal fin size appeared to differentiate the populations. Those characters reflect the swimming, feeding and foraging ability of the fish. On the basis of morphological data, the Lake population was the most divergent. Compared to the three river populations of mahseer, the lake fish had the most shortest head, dorsal fin, pectoral fin and, smaller and narrow eyes. The lake fish also had large pelvic fin and deeper body. 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. However, shorter pectoral fin length associated with colder water temperature (Barlow 1961) did not support our present findings.

The head morphology reflects a species' feeding habits (Skúlason et al. 1989). Mahseer is known to be an omnivore species, as adult it feeds upon green filamentous algae, insect larvae, small mollusks and slimy deposits on rocks (Shrestha 1997; Dubey 1986). The third principal component consisted of head dimension, although being a weak classificator, indicate the foraging habits of the studied populations. Relatively large heads of 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). In this respect, the mahseer population in Phewa lake has close and small eye, because the lake is characterized as meso-eutrophic and light does not penetrate very deep (FRC 1999). Mahseer live in glacial fed perennial rivers, where water is quite clear, and they have large eyes. Baumgartner et al. (1988) suggested that the eye size may as well be related to feeding behavior.

The adaptation of the river populations of mahseer reflects their body morphology: they are relatively robust with long pectoral 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). The 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 migrating (Baumgartner et al. 1988; Robinson and Witson 1996).

Whether the observed morphological patterns were produced through genetic differences or phenotypic plasticity is unknown. Populations could diverge via alternative, genetically based morphologies, or through environmentally induced phenotypes (Langerhans et al. 2003). However, morphological and genetic characters of fishes have been shown in some cases to co-vary (Dynes et al. 1999; Houser et al. 1995). 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 mahseer populations in several morphological characters: body depth, head size, eye diameter, dorsal fin length, pectoral fin length. This apparent plasticity may be an adaptive response (Scheiner and Callahan 1999) to the complex and varied environmental conditions under which this widespread species exists. Nevertheless, even though genetic differentiation so far has not been demonstrated in mahseer (*T. putitora*), molecular characters may yet be discovered that would explain more of the observed morphological variation.

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