

Research Article

## A Case Study of the Phenotypic Variations in *Barilius bendelisis* (Hamilton) from a Perennial Stream and a Fish Pond of Garhwal Himalayan Region of Uttarakhand, India

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*Article history:*

Submission May 2020

Revised June 2020

Accepted August 2020

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**ABSTRACT**

A comparative study on the morphometric and meristic variables of *Barilius bendelisis* (Ham.) from two different water bodies, i.e., Khanda Gad, a perennial spring fed stream and fish pond in the Garhwal Himalayan region of Uttarakhand was made during the present study. A total of 100 specimens were analysed for 26 different morphometric variables and 14 meristic counts. All morphometric variables showed linear relationship when expressed in relation to total length and head length (except for caudal length in Khanda Gad's fishes). As observed by meristic counts for different length of fishes, it remained constant with increasing body length. Standard length was found to be the highly correlated character in samples from both sites. Principal Component Analysis of 10 significant morphometric variables yielded three components accounting for 73.38% of the total variation. Principal Component Analysis of 3 meristic variables yielded single component accounting for 62.3% of total variation. Principal Component Analysis plots show that characters like, base of caudal fin, least height of caudal peduncle, pre-orbital distance, post-orbital distance, eye diameter, inter orbital distance, numbers of dorsal fin ray, scales from lateral line to pelvic fin and scales from lateral line to anal fin distinguishes the fish stock from fish pond and Kanda Gad. Discriminant Function Analysis for morphometric and meristic variables showed that 98% and 83% of individuals were allocated into their original populations respectively. The cluster analysis for morphometric characters showed of fish populations from both sites formed two major clades, thus significantly differentiating the two stocks of fish population. These morphometric variations could be attributed to the difference in the ecological and the rearing condition of fish stock from the stream and pond. The information on phenotypic variation between the natural stream and hatchery reared fish would be important for development of aquaculture.

*Keywords:* Discriminant function analysis, Morphometric measurement, Phenotypic characters, Principal component analysis

### Introduction

Morphometry plays an important role in identification and establishing the systematic position of fishes. Such taxonomic tools are also useful in detecting variations in the fish population. The morphometric features of a fish are subjected to adaptations under various habitat conditions.

Change in the hydrological conditions leads to the morphometric variations in the fish stocks [1]. Instead of direct genetic control, the morphometric variation has been suggested to influence by the environmental factors [2, 3]. Handful knowledge of about environmentally influenced variation in

*How to cite:*

Singh P, Nautiyal S, Swami N et al. (2020) A Case Study of the Phenotypic Variations in *Barilius bendelisis* (Hamilton) from a Perennial Stream and a Fish Pond of Garhwal Himalayan Region of Uttarakhand, India. Journal of Tropical Life Science 10 (3): 185 – 196. doi: 10.11594/jtls.10.03.02.

fish population can be gathered through the study of morphometric variation between fish stocks, which can further help in management of fisheries practices [4, 5]. Therefore, morpho-meristic measurements and counts were considerably used to recognize the variations among fish populations [6, 7]. The morphological plasticity because of the environmental inconsistency is commonly found among many fish species and the phenotypic variation due to the environmental variation has been extensively used to study the different population stocks [8, 3]. The morphological variability is significant adaptive strategy for populations experiencing environmental variation and the development of response towards the environment and learning leads to the phenotypic variations between wild and cultured fishes [8, 9].

*Barilius bendelisis* (Ham.) belongs to the order cypriniformes and commonly known as Indian Hill Trout [3]. It dwells in shallow clear water in streams of the Himalayan region including Uttarakhand (India). The species distribution has also been reported from Pakistan, Nepal, Bangladesh, Sri Lanka, Bhutan and Myanmar [10, 11, 12]. The species is characterised by 12 black bands running from dorsal to ventral on the body surface, and a single black spot on the base of each body scale [13]. *B. bendelisis* (Ham.) is in the least concerned category of IUCN, however, it is a popular ornamental species and a food source for the local population which leads to its reduction [14], although it has no commercial value because of its small size, it has an important place in the food chain of hill streams. The objective of the present study was to analyse the variability in the morpho-meristic characteristics of freshwater fish *B. bendelisis* (Ham.) from two different aquatic systems i.e., a perennial stream and a fish pond in the Garhwal region of Uttarakhand. This study should provide information that relates habitat and ecological conditions with the phenotypic variation.

## Material and Methods

### Sample collection

A total of 50 specimens of *B. bendelisis* (Ham.) were collected from Khanda Gad (30°6'42"–30°13'23" N; 78°41'48"–79°5'4" E), originating from Mandakhal ridge (2143m above sea level), a left side spring fed tributary of River Alaknanda, 21 Km from the Srinagar township in the Pauri district of Uttarakhand, India, during

November 2015 to April 2016. The fishes were collected randomly using 'Thali' from upstream and downstream at Khanda Township. 'Thali', a local method employs a meshed towel cloth wrapped around a rim of a stainless-steel plate (thali). An opening on the cloth surface is made for the fishes to enter to eat the bait of roasted flour. Also, 50 specimens of *B. bendelisis* (Ham.) were collected by the dip net from a pond in Fish Hatchery at the Chauras campus of HNB Garhwal University, Tehri district of Uttarakhand, India (latitude 30°13'36.43"N and longitude 78°48'08.74"E) during the same time period. These fishes were collected from Khanda Gad during November 2014 and stocked in hatchery. The species identification has been done by following the different keys [10, 13, 15]. The morphometric measurements and meristic counts of all the 100 specimens were recorded as outlined in [15]. The morphometric measurements were taken manually with the help of ruler and divider, whereas the counts for meristic characters were taken with the help of lens.

### Data analysis

The measurements were subjected to different statistical analyses using Microsoft Excel, SPSS 20 and PAST. The morphometric variables were analysed for correlation and regression and coefficient of correlation ( $r$ ) and regression ( $b$ ) were tested for significance. The regression equation was computed for each dependent variable to fit the straight-line equation:

$$Y = a + bx$$

Where, Y = dependent variable; a = intercept; b = slope of regression line; X = independent variable.

The values of mean, standard deviation, range of percentage and range difference were also calculated for various morphometric characters.

To avoid size variation errors in the specimens the morphometric variables were transformed and normalized according to equation [16]:

$$M_s = M_o \left( \frac{L_s}{L_o} \right)^b$$

Where,  $M_s$  = standardized measurement,  $M_o$  = measured character length (mm),  $L_s$  = overall (arithmetic) mean standard length (mm),  $L_o$  = standard length (mm)

of specimen,  $b$  = estimated for each character from the observed data using the non-linear equation

The univariate analysis of variance (ANOVA) was performed on normalized morphometric measurements to find the significant characters that vary between the two habitat sites. The significant variables were then subjected for Principal Component Analysis (PCA), Discriminant Function Analysis (DFA) and the cluster analysis (Jaccard's similarity index).

## Results and Discussions

Correlation coefficient ( $r$ ) and Regression coefficient ( $b$ ) of different morphometric characters of *B. bendelisis* (Ham.) was computed in relation to TL and HL at significance level of  $P < 0.01$  and

$P < 0.05$ . At Khanda Gad, SL and PrDL were the highly and CL was the least correlated character, while at fish pond the characters such as SL, PrDL, HL and PPD have higher degree of correlation in relation to TL. The HD shows high correlation in relation to HL at both the sites. The test of significance showed that all values were significant at  $P < 0.01$ . All the characters have been observed to follow linear relationship (Table 1).

The use of ' $r$ ' static indicated SL is most highly correlated morphometric parameter in both samples. Similarly, it has been also found that SL is the most highly correlated body part in *B. bendelisis* (Ham.) from hill streams of Himachal Pradesh [17]. The use of static ' $b$ ' seemed to support these findings. The highest value for regression coefficient ( $b$ ) was shown by SL in relation to TL

Table 1. Mean, Standard deviation, Correlation coefficient ( $r$ ) and Regression coefficient ( $b$ ) of different morphometric characters of *B. bendelisis* (Ham.) in relation to Total length and Head length in Khanda Gad and Fish Pond

S. No. Parameters	Khanda Gad			Fish Pond			Khanda Gad	Fish Pond
	Mean	S.D.	$r$	Mean	S.D.	$r$	b value	b value
<i>In relation to TL</i>								
1 Total length (TL)	9.70	1.91	–	12.88	1.79	–		
2 Standard length (SL)	7.77	1.65	0.993**	10.54	1.51	0.974**	0.85	0.81
3 Caudal Length (CL)	2.28	2.15	0.032	02.37	0.43	0.723**	0.21	0.17
4 Head length (HL)	2.12	0.43	0.934**	02.48	0.32	0.958**	0.21	0.17
5 Body depth (BD)	2.01	2.91	0.349*	02.37	0.10	0.920**	0.53	0.19
6 Pre-dorsal distance (PrDL)	4.49	0.96	0.987**	06.10	0.91	0.965**	0.49	0.49
7 Post-dorsal distance (PoDL)	2.44	0.48	0.970**	03.37	0.78	0.604**	0.24	0.26
8 Pre-pelvic distance (PPD)	4.02	0.88	0.969**	05.55	0.88	0.947**	0.44	0.46
9 Length of dorsal fin (LDF)	1.66	0.44	0.954**	02.31	0.52	0.916**	0.22	0.27
10 Width of dorsal fin (WDF)	1.00	0.29	0.876**	01.32	0.31	0.860**	0.13	0.14
11 Length of anal fin (LAF)	1.29	0.21	0.930**	01.51	0.26	0.849**	0.10	0.12
12 Width of anal fin (WAF)	0.94	0.34	0.820**	01.3	0.32	0.904**	0.14	0.16
13 Length of pelvic fin (LPF)	1.15	0.24	0.878**	01.43	0.29	0.784**	0.11	0.12
14 Width of pelvic fin (WPF)	0.39	1.22	0.785**	00.51	0.25	0.505**	0.05	0.12
15 Length of pectoral fin (Lpec.F)	1.62	0.36	0.958**	02.05	0.32	0.806**	0.18	0.14
16 Width of pectoral fin (Wpec.F)	0.50	0.15	0.868**	00.63	0.13	0.727**	0.06	0.05
17 Least height of caudal peduncle (LCPD)	0.85	0.22	0.924**	01.02	0.21	0.848**	0.10	0.10
18 Highest height of caudal peduncle (HCPD)	0.95	0.23	0.922**	01.22	0.21	0.849**	0.11	0.10
19 Base of Caudal fin (BCF)	0.85	0.22	0.924**	01.06	0.20	0.916**	0.10	0.10
<i>In relation to HL</i>								
20 Head depth (HD)	1.51	0.37	0.966**	01.90	0.37	0.88**	0.82	1.02
21 Pre-orbital distance (PrOD)	0.68	0.19	0.904**	01.33	0.18	0.849**	0.41	0.48
22 Post-orbital distance (PoOD)	1.14	0.35	0.886**	01.13	0.19	0.746**	0.71	0.44
23 Eye diameter (ED)	0.56	0.07	0.808**	00.55	0.09	0.521**	0.14	0.15
24 Inter orbital distance (IOD)	0.46	0.07	0.807**	00.46	0.06	0.845**	0.14	0.16
25 Snout length (SnL)	0.69	0.20	0.872**	00.77	0.13	0.821**	0.42	0.33
26 Inter nostril distance (IND)	0.41	0.06	0.710**	00.51	0.10	0.628**	0.09	0.20

\*\* Correlation is significant at the 0.01 level, \* Correlation is significant at the 0.05 level

Table 2. Mean and Range values of meristic counts of *B. bendelisis* (Ham.) collected from Khanda Gad and Fish Pond

S. No.	Meristic Character	Khanda Gad				Fish Pond			
		Mean	Min.	Max.	Range	Mean	Min.	Max.	Range
1	Lateral line scales count (Ltr.Ln.)	41.98	40	45	5	42.18	39	45	6
2	Pre-dorsal scales (Pr.Ds.)	22.14	21	24	3	21.98	20	25	5
3	Lateral transverse scales (Ltr.tr.s.)	07.68	07	09	2	07.52	06	09	3
4	Circumpenduncular scales (Circum.)	06.96	06	08	2	06.72	06	07	1
5	Dorsal fin ray (D.Fr.)	09.00	09	09	0	08.80	08	09	1
6	Pectoral fin ray (Pec.Fr.)	15.00	15	15	0	15.00	15	15	0
7	Ventral fin ray (V.Fr.)	09.00	09	09	0	09.00	09	09	0
8	Anal fin ray (A.Fr.)	09.88	09	10	1	09.74	08	10	2
9	Caudal fin ray (C.Fr.)	18.00	18	18	0	18.00	18	18	0
10	Scales from lateral line to pelvic fin (Lat.Pel.)	04.32	04	05	1	04.84	04	07	3
11	Scales from lateral line to dorsal fin (Lat.D.)	08.10	08	09	1	08.22	08	09	1
12	Scales from lateral line to anal fin (Lat.A.)	04.30	04	05	1	04.74	04	06	2
13	Pre-Anal scales (Pre.As.)	21.78	21	23	2	22.02	19	24	5
14	Number of Colour bands	11.48	10	12	2	12.70	08	15	7

at both Khanda Gad and the fish pond.

Meristic counts of fishes from Khanda Gad and Fish pond were recorded. At Khanda Gad, the range of lateral line scales, pre-dorsal scales, lateral transverse scales, circumpenduncular scales and pre-anal scales were 40–45, 21–24, 7–9, 6–8, and 21–23 respectively, while at fish pond it was 39–45, 20–25, 6–9, 6–7 and 19–24 respectively (Table 2). Percentage of various body parameters were computed in relation to TL and HL. At Khanda Gad, the CL and LPF shows the high and least degree of range difference respectively in relation to the total length TL. The maximum and least range of differences in relation to HL was observed for PoOD and IOD respectively. However, at the fish pond, PoDL has a higher range of difference in relation to the TL. The ED showed the higher range of difference followed by PoOD while IOD has the least range difference in relation to HL (Table 3).

According to the classification of [18] based on range differences, in the present study 16 characters were found to be genetic, 5 characters were found to be intermediate and 4 characters were

found to be environmental at Khanda Gad. Whereas, 15 characters were found to be genetic, 4 characters were found to be intermediate and 6 characters were found to be environmental in the fish pond (Table 3). Thus, the number of genetically controlled characters is large as compared to intermediate or environmental characters. Therefore, it can be presumed that *B. bendelisis* (Ham.) have restricted zoo geographical distribution because the most of their morphometric variables showed the narrow range difference and are controlled genetically. Evidently, these small fish in the Garhwal region occur mostly in small spring-fed streams and avoid larger snow-fed rivers.

#### Multivariate Analysis

The normalized morphometric data subjected to ANOVA showed that fish specimens from the Khanda Gad and fish pond differed significantly ( $P < 0.001$ ) in 10 standardized morphometric and 4 meristic variables (Table 4). Only the significant variables were further analysed and subjected to PCA. PCA of 10 morphometric variables yielded three components (Eigen value  $> 1$ ) accounting for

Table 3. Percentage of body parts of *B. bendelisis* (Ham.) in relation to total length and head length in Khanda Gad and Fish Pond

S. No.	Body parts	Khanda Gad		Fish Pond	
		Range of %	Range difference	Range of %	Range difference
<i>% age in relation to total length (TL)</i>					
1	SL	71.42–85.91	14.48	76.66–89.34	12.67
2	CL	14.08–20.48	06.40	12.62–23.33	10.71
3	HL	19.51–25.71	06.20	18.05–23.63	05.58
4	BD	11.68–17.46	05.78	16.05–20.68	04.63
5	PrDL	42.64–50.64	08.00	42.01–51.40	09.39
6	PoDL	21.10–28.16	07.06	11.42–38.88	27.46
7	PPD	33.70–46.82	13.11	36.36–48.59	12.22
8	LDF	14.45–22.76	08.30	13.08–22.72	09.64
9	WDF	07.50–17.02	09.52	08.18–13.63	05.45
10	LAF	11.47–15.95	04.48	08.40–13.84	05.44
11	WAF	03.48–12.06	08.58	07.27–13.07	05.79
12	LPF	10.00–15.00	05.00	08.40–17.29	08.88
13	WPF	02.59–08.82	06.22	02.63–10.88	08.25
14	Lpec.F	14.08–19.14	05.06	11.80–18.80	06.99
15	Wpec.F	03.89–07.93	04.04	03.73–09.09	05.35
16	LCPD	06.49–11.11	04.61	06.03–11.48	05.45
17	HCPD	06.32–11.90	05.57	07.27–12.16	04.88
18	BCF	06.49–11.11	04.61	06.03–10.13	04.10
<i>% age in relation to head length (HL)</i>					
19	HD	60.00–81.81	21.81	45.45–96.29	50.84
20	PrOD	23.52–41.37	17.84	42.30–62.96	20.65
21	PoOD	41.17–72.41	31.23	25.92–58.33	32.40
22	ED	20.68–33.33	12.64	02.30–29.16	26.85
23	IOD	16.66–26.66	10.00	16.66–25.00	8.33
24	SnL	23.52–52.38	28.85	19.23–40.74	21.50
25	IND	13.79–26.66	12.87	16.66–31.03	14.36

Standard Length (SL), Caudal Length (CL), Head Length (HL), Body Depth (BD), Pre-dorsal distance (PrDL), Post-dorsal distance (PoDL), Pre-pelvic distance (PPD), Length of dorsal fin (LDF), Width of dorsal fin (WDF), Length of anal fin (LAF), Width of anal fin (WAF), Length of pelvic fin (LPF), Width of pelvic fin (WPF), Length of pectoral fin (Lpec.F), Width of pectoral fin (Wpec.F), Least height of caudal peduncle (LCPD), Highest height of caudal peduncle (HCPD), Base of Caudal fin (BCF), Head depth (HD), Pre-orbital distance (PrOD), Post-orbital distance (PoOD), Eye diameter (ED), Inter orbital distance (IOD), Snout length (SnL), Inter nostril distance (IND).

73.38% of total variation. The first principal component (Eigen value = 5.021) accounted for 50.21% of total variation with the highest loading on HL, LAF, LCPD, BCF, HD, PoOD, ED, IOD and SnL variables. The first principal component is mainly a composition of HL, LCPD and BCF variables with significant loadings. Similarly, the second principal component (Eigen value = 1.254) explained 12.54% of total variation was mainly composed of BCF, PrOD, ED and IOD variables with maximum loadings (Table 5).

In the scatter plot of the PCA, the majority of the fish population of stream present on the right axis and the majority of the fish population from fish pond present on the left axis of the plot. So, the very first component has been fairly effective in separating the fish population of the stream from the fish population in the pond and only a very few samples were misclassified. The stream population is distinguished from the pond population by BCF, LCPD, ED, PrOD, PoOD and IOD (Figure 1). PCA of 3 meristic variables yielded

single component (Eigen value = 1.869) accounting for 62.3% of total variation with the highest loading on Lat.Pel. and Lat. A. variables. However, the second component (Eigen value = 0.985) explained 32.84% of total variation having D.Fr. variables with maximum loadings (Table 5). In the scattered plot of PCA, for meristic characters, D.Fr., Lat.Pel. and Lat.A. distinguish the two different fish population (Figure 2).

In Box's M test the null hypothesis of equal population covariance matrices were rejected with  $df_1 = 300$ ,  $df_2 = 29186.4$  and  $F = 3.39$  ( $P < 0.00$

0.001). The test for the prediction model found statistically significant ( $P < 0.001$ ) with Wilk's Lambda = 0.278 and Chi Square = 110.01. Similarly, for meristic variables the test for prediction model found statistically significant ( $P < 0.001$ ) with Wilk's Lambda = 0.487 and Chi Square = 66.842.

The single canonical discriminant function explained 100% of the variation in morphometry of samples at Eigen value 2.594. The maximum variation on this factor was due to ED, PrOD, PoOD, and IOD. Similarly, for meristic variables,

Table 4. F value (derived from analysis of variance) for morphometric and meristic characters of *B. bendelisis* (Ham.) collected from Khanda Gad and Fish Pond

Morphometric characters	Wilk's Lambda	F	Sig.	Morphometric characters	Wilk's Lambda	F	Sig.
CL	0.992	0.749	0.389	PoOD*	0.770	29.342	0.000
HL*	0.854	16.690	0.000	ED*	0.619	60.415	0.000
BD	0.990	0.956	0.330	IOD*	0.789	26.262	0.000
PrDL	1.000	0.002	0.962	SnL*	0.847	17.738	0.000
PoDL	0.994	0.572	0.451	IND	0.994	0.574	0.451
PPD	0.994	0.612	0.436	<b>Meristic characters</b>			
LDF	0.960	4.045	0.047	Ltr.Ln.	0.993	0.74	0.392
WDF	0.944	5.826	0.018	Pr.Ds.	0.991	0.859	0.356
LAF*	0.900	10.914	0.001	Ltr.tr.s.	0.984	1.593	0.210
WAF	0.944	5.825	0.018	Circum.	0.907	10.08	0.002
LPF	0.958	4.250	0.042	D.Fr.*	0.889	12.25	0.001
WPF	0.995	0.538	0.465	Pec.Fr.	a		
LPecF	0.974	2.635	0.108	V.Fr.	a		
WPecF	0.946	5.545	0.021	A.Fr.	0.975	2.541	0.114
LCPD*	0.854	16.693	0.000	C.Fr.	a		
HCPD	0.957	4.377	0.039	Lat.Pel.*	0.803	24.003	0.000
BCF*	0.888	12.333	0.001	Lat.D.	0.973	2.697	0.104
HD*	0.861	15.768	0.000	Lat.A.*	0.820	21.443	0.000
PrOD*	0.686	44.849	0.000	Pre.As.	0.976	2.452	0.121

Caudal Length (CL), Head Length (HL), Body Depth (BD), Pre-dorsal distance (PrDL), Post-dorsal distance (PoDL), Pre-pelvic distance (PPD), Length of dorsal fin (LDF), Width of dorsal fin (WDF), Length of anal fin (LAF), Width of anal fin (WAF), Length of pelvic fin (LPF), Width of pelvic fin (WPF), Length of pectoral fin (LPec.F), Width of pectoral fin (WPec.F), Least height of caudal peduncle (LCPD), Highest height of caudal peduncle (HCPD), Base of Caudal fin (BCF), Head depth (HD), Pre-orbital distance (PrOD), Post-orbital distance (PoOD), Eye diameter (ED), Inter orbital distance (IOD), Snout length (SnL), Inter nostril distance (IND), Lateral line scales count (Ltr.Ln.), Pre-dorsal scales (Pr.Ds.), Lateral transverse scales (Ltr.tr.s.), Circumpenduncular scales (Circum.), Dorsal fin ray (D.Fr.), Pectoral fin ray (Pec.Fr.), Ventral fin ray (V.Fr.), Anal fin ray (A.Fr.), Caudal fin ray (C.Fr.), Scales from lateral line to pelvic fin (Lat.Pel.), Scales from lateral line to dorsal fin (Lat.D.), Scales from lateral line to anal fin (Lat.A.), Pre-Anal scales (Pre.As.).

a: can't be computed (variables are same)

\* characters of fish samples from the two populations which differed significantly ( $P < 0.001$ )

Table 5. Components Loadings of two principal components for 10 morphometric and 3 meristic measurements of *B. bendelisis* (Ham.) collected from Khanda Gad and Fish Pond

Morphometric characters	PC1	PC2
HL	0.841	0.033
LAF	0.612	0.239
LCPD	0.813	0.355
BCF	0.804	0.418
HD	0.760	0.195
PrOD	-0.225	0.569
PoOD	0.705	-0.341
ED	0.601	-0.470
IOD	0.751	-0.438
SnL	0.760	0.040
Meristic characters		
D.Fr.	0.150	0.985
Lat.Pel.	0.694	-0.163
Lat.A.	0.703	-0.049

Head Length (HL), Length of anal fin (LAF), Least height of caudal peduncle (LCPD), Base of Caudal fin (BCF), Head depth (HD), Pre-orbital distance (PrOD), Post-orbital distance (PoOD), Eye diameter (ED), Inter orbital distance (IOD), Snout length (SnL), Dorsal fin ray (D.Fr.), Scales from lateral line to pelvic fin (Lat.Pel.), Scales from lateral line to anal fin (Lat.A.).

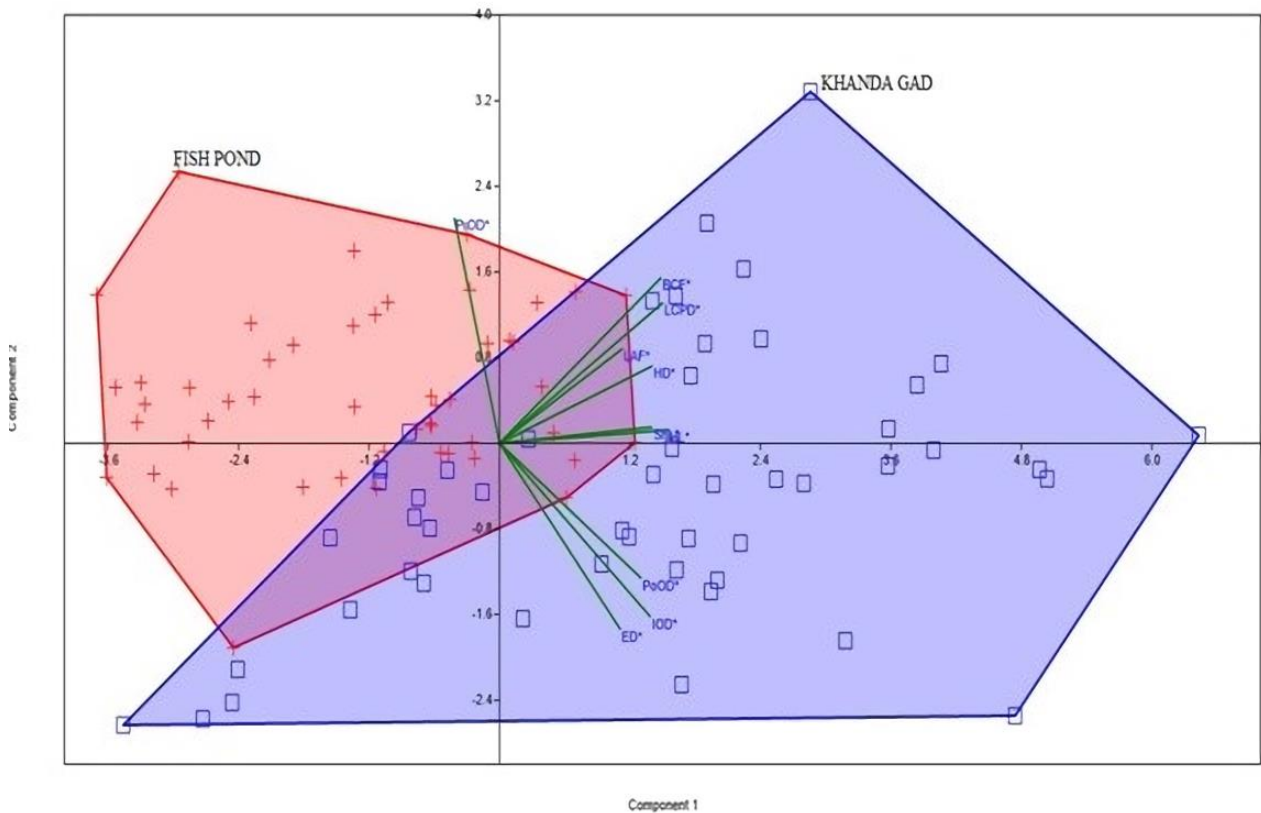


Figure 1. Principal component analysis of *Barilius bendelisis* based on 10 significant morphometric characters



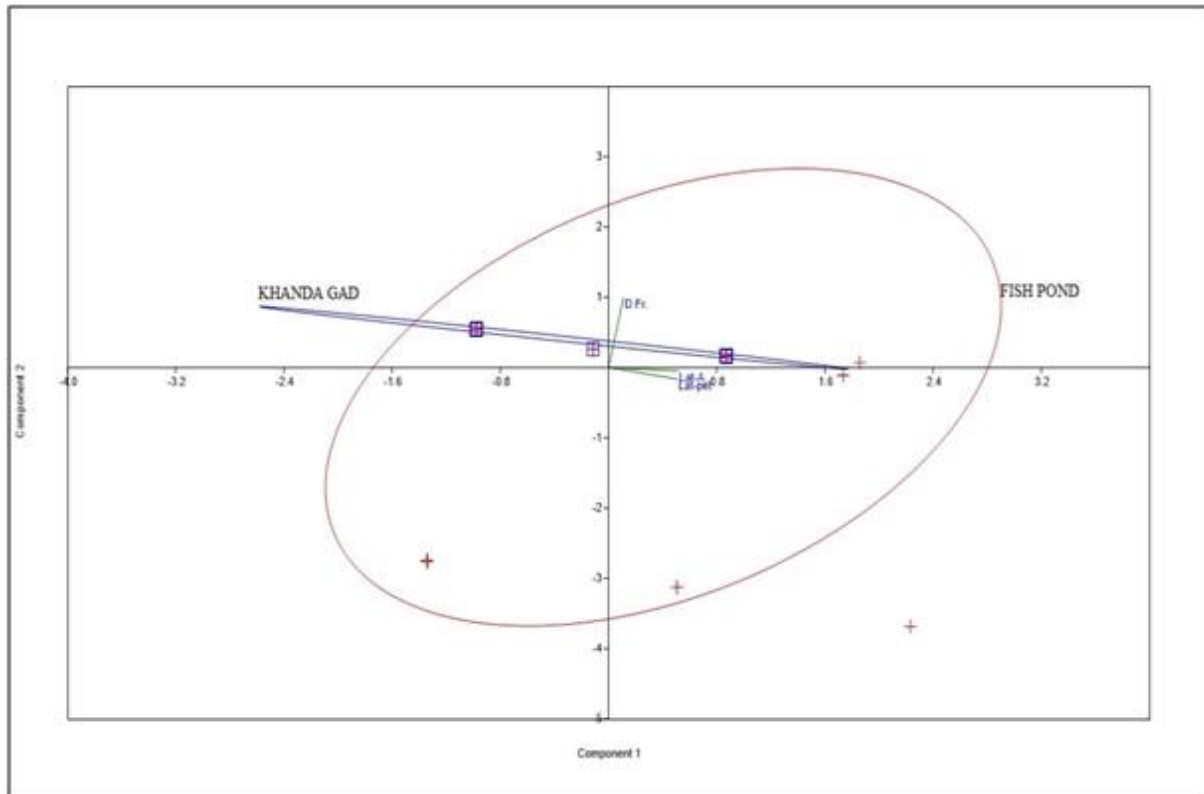


Figure 2. Principal component analysis (PCA) of *Barilius bendelisis* based on 3 significant meristic characters

the single function explained 100% variation at Eigen value 1.052. The Lat.Pel., Lat.A., D.Fr. Circum. are responsible for the maximum variation on this factor (Table 6). Principal component analysis (PCA) suggests the similar variables responsible for differentiating the fish population from the stream and fish pond.

Discriminant function analysis (DFA) for morphometric variables suggests that 98% of individuals were allocated into their original populations. In the cross-validation test 91% of samples were correctly classified. The percentage of the correctly classified fishes was similar in Khanda Gad and the fish pond with 98% correct classification and 2% individuals were misclassified. So, the sensitivity and specificity of the sample classification is 98% (Table 7). Similarly, for meristic variables, DFA showed 83% of individuals were allocated into their original population, while in the cross-validation test 81% of samples were correctly classified. The correctly classified fishes in the fish pond was 90% with 10% of misclassified individuals, and for Khanda Gad 76% of the samples were correctly classified and 24% of individuals were misclassified (Table 7). The results from PCA and DFA suggests that head and fins

dimensions, were the important characters in differentiating the two stocks from both wild and farmed individuals. Morphometric observations of fishes from two different sites showed that in comparison to the fishes of stream, cultured fish had slightly larger head depth, longer fins and wider abdomen. As in cultured condition, due to the decline in swimming performance and less chances of predation, the streamlining of fish body and head size reduces along with rise in depth of head and trunk [19, 20]. Furthermore, [21] observed the farmed fishes with wide abdomen and long head in comparison to wild stock, attributing these changes to the rearing condition, stock density, food availability, exposure of stress, fish mobility, swimming performance and local ecological condition [22, 23, 24, 25].

Phenotypic variations are not always suggestive of genetic segregation between populations, but might be affected by environmental conditions [26, 27]. The shape of the cultured fish, after released in wild habitat attains the wild like body shape after a considerable period of about half a year [24]. Therefore, the morphometric variations observed in this study between wild and cultured fishes could be partially explained by habitat and



Table 6. Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions (Variables ordered by absolute size of correlation within function)

Morphometric Characters	Function DF1 (100%)	Morphometric Characters	Function DF1 (100%)
ED	0.487	CL	0.054
PrOD	-0.420	PPD	-0.049
PoOD	0.340	IND	-0.048
IOD	0.321	PoDL	-0.047
SnL	0.264	WPF	0.046
LCPD	0.256	PrDL	-0.003
HL	0.256	<i>Eigen value</i>	2.594
HD	0.249	Meristic Characters	
BCF	0.220	Lat.Pel.	-0.483
LAF	0.207	Lat.A.	-0.456
WDF	0.151	D.Fr.	0.345
WAF	0.151	Circum.	0.313
WPecF	0.148	Lat.D.	-0.162
HCPD	0.131	A.Fr.	0.157
LPF	0.129	Pre.As.	-0.154
LDF	0.126	Ltr.tr.s.	0.124
LPecF	0.102	Pr.Ds.	0.091
BD	0.061	Ltr.Ln.	-0.085
		<i>Eigen value</i>	1.052

Eye diameter (ED), Pre-orbital distance (PrOD), Post-orbital distance (PoOD), Inter orbital distance (IOD), Snout length (SnL), Least height of caudal peduncle (LCPD), Head Length (HL), Head depth (HD), Base of Caudal fin (BCF), Length of anal fin (LAF), Width of dorsal fin (WDF), Width of anal fin (WAF), Width of pectoral fin (Wpec.F), Highest height of caudal peduncle (HCPD), Length of pelvic fin (LPF), Length of dorsal fin (LDF), Length of pectoral fin (Lpec.F), Body Depth (BD), Caudal Length (CL), Pre-pelvic distance (PPD), Inter nostril distance (IND), Post-dorsal distance (PoDL), Width of pelvic fin (WPF), Pre-dorsal distance (PrDL), Scales from lateral line to pelvic fin (Lat.Pel.), Scales from lateral line to anal fin (Lat.A.), Dorsal fin ray (D.Fr.), Circumpenduncular scales (Circum.), Scales from lateral line to dorsal fin (Lat.D.), Anal fin ray (A.Fr.), Pre-Anal scales (Pre.As.), Lateral transverse scales (Ltr.tr.s.), Pre-dorsal scales (Pr.Ds.), Lateral line scales count (Ltr.Ln.).

to predators which influences the changes in the shape of the body [28].

dietary shifts, swimming adaptation and exposure to predators which influences the changes in the shape of the body [28].

### Cluster Analysis

Significant morphometric characters of fish population from stream and fish pond were analyzed. Majorly two clades cluster were formed which significantly differentiate the two samples of fish population (Figure 3).

### Conclusion

The scatter plot of PCA for morphometric cha-

racters showed variation in stream population from the fish pond population by the characters of BCF, LCPD, ED, PrOD, PoOD and IOD; and the scatter plot of PCA for meristic character showed that D.Fr., Lat.Pel. and Lat.A. distinguish the two different fish population. Similarly, in DFA, single discriminant function explains 100% of variation in morpho-meristic characters of wild and cultured stock of *B. bendelisis* (Ham.). The cluster analysis for both the samples from stream and fish pond showed the significant difference among these two populations. Thus, it can be concluded that the fish samples of *B. bendelisis* (Ham.) from

Table 7. Counts and percentage of specimens classified in each group and after cross validation of morphometric and meristic measurements for *B. bendelisis* (Ham.) from Khanda Gad and Fish Pond

Morphometric measurements					
Classification		Site	Predicted Group Membership		Total
			Khanda Gad	Fish Pond	
Original	Count	Fish Pond	1	49	50
		Khanda Gad	49	1	50
	%	Fish Pond	2	98	100
		Khanda Gad	98	2	100
Cross-validated <sup>b</sup>	Count	Fish Pond	5	45	50
		Khanda Gad	46	4	50
	%	Fish Pond	10	90	100
		Khanda Gad	92	8	100

Meristic measurements					
Classification		Site	Predicted Group Membership		Total
			Khanda Gad	Fish Pond	
Original	Count	Fish Pond	5	45	50
		Khanda Gad	38	12	50
	%	Fish Pond	10	90	100
		Khanda Gad	76	24	100
Cross-validated <sup>b</sup>	Count	Fish Pond	7	43	50
		Khanda Gad	38	12	50
	%	Fish Pond	14	86	100
		Khanda Gad	76	24	100

b. Cross validation is done only for those cases in the analysis. In cross validation, each case is classified by the functions derived from all cases other than that case.

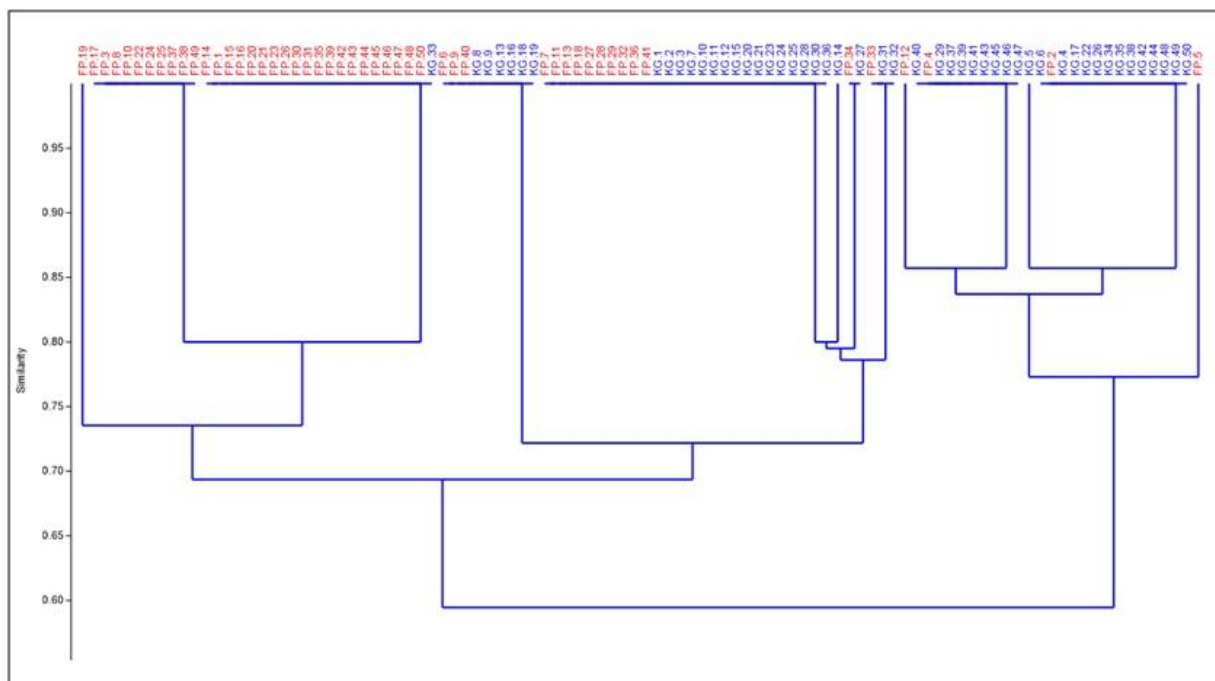


Figure 3. Cluster analysis of *B. bendelisis* from Khanda Gad and Fish Pond based on 10 significant morphometric characters. (KG: Khanda Gad; FP: Fish Pond)

the wild and cultured habitat differ significantly. These morphometric variations could be attributed majorly to the rearing condition in the pond, fish mobility and swimming performance of the fish.

### Acknowledgement

The authors are grateful to the Head, Department of Zoology and Biotechnology, Hemvati Nandan Bahuguna Garhwal University, Uttarakhand (India) for providing essential conveniences for the present study.

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