



## Biostatistical Analysis Of Morphological Traits Of *Schizothorax Richardsonii* From Snow-Fed Rivers Of Kumaun Himalaya, Uttarakhand, India

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### **Abstract:**

Morphometric characteristics of fishes were observable characteristics that are set of largely statistical methods used for examining variations in body size and shape. The objectives taken for the current study were to examine the complete morpho-biology of *Schizothorax richardsonii* from three major snow-fed rivers of the Kumaun Himalaya in Uttarakhand, India, and to establish the statistical relationship among various morphological characteristics related to body size and fin shape. In this study 120 fish specimen including 66 male and 54 female were studied for one year. Twelve distinct morphometric and five meristic counts had been examined in percentage of their body length and head length respectively. For biostatistics, the data was analysed by using various statistical methods including summary statistics, correlation, PCA analysis, Barlett's test of sphericity, and multiple linear regression. Fish samples studied were found within the range of 17.7 – 42.2 cm in TL and BW from 60 - 780 gm. Morphological characteristics had high coefficient of correlation ( $r \geq 0.90$ ), indicating the characters' strong relationship, with the highest correlation ( $r = 0.99$ ). Additionally, the coefficient of determination ( $R^2$ ) value was found within the range of 0.62 to 0.99, which shows a significant relationship between the morphometric characteristics. PCA analysis revealed the major significance found among parameters, together with regression analysis. This study will provide a baseline data and a valuable insight into the population dynamics, and may contribute to the development of more effective strategies for the management.

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**Keywords:** *Schizothorax richardsonii*, Kumaun Himalaya, Morphometric, Meristic, Uttarakhand

### 1. Introduction:

Morphometric study of fish is an essential part to examine the biology of fish species. Growth of the body parts has relation with the body length therefore analysis of morphometric characters and estimating the statistical relationship among them are very important. *Schizothorax richardsonii*, known as snow trout, are a

significant fish species found throughout the rivers and streams of Kumaun Himalayan region of India. This fish is an important food source for local communities, and it has its own economic and ecological significant role for the local communities of Uttarakhand [1]. Morphological variations within and between populations of *S. richardsonii* have been a subject of interest in several studies.

Morphological variations within and between populations can reflect genetic diversity and local adaptation, which may influence factors such as growth rates, survival, and reproductive success [2]. Morphometric and meristic analysis are also very powerful tools for assessing differentiation of the same species [3]. The study of morphometric characteristics of fishes is significant as they can be utilised to differentiate taxonomic units [4]. [5], claims that the study of morphology and the study of the modes of existence of living organisms are inseparable. A proper morphometric study of *S. richardsonii* from Kumaun Himalayan streams was therefore attempted while keeping in mind the morphological disparity and food value of *S. richardsonii*.

A review of the existing literature on the fish species *S. richardsonii* had also revealed that there is little information on the morphology of this fish species from the rivers of the Kumaun Himalaya. The first step in examining the general development structure of species under natural conditions is to understand morphobiology. Therefore, taking into account the need for research, this study was aimed to generate complete baseline information for the morphometrics and meristic counts of *S. richardsonii*, and to establish any potential statistical relationships among morphometric parameters, which will help in understanding the major influences on their growth in Himalayan rivers and surrounding areas.

## 2. Material and methods:

**2.1. Study site and sampling.** Three major snow-fed rivers, the Eastern Ramganga River (29°82'23" to 80°20'29"E), the Kali River (30°14'32" to 81°01'19"E), and the Saryu River (29°66'56" to 80°14'89"E), that originate from the Namik Glacier, the Kalapani Glacier, and the Sarmal Glacier of the Kumaun Himalayan region were chosen as study site. A total 120 fish species of *S. richardsonii* were collected from them during a period of one year on monthly basis from the commercially available sources. Fish sample were identified by [6].

**2.2. Morphometric and meristic.** Fresh specimens of *S. richardsonii* were analysed using a total of 12 morphometric measurements and 6 meristic counts in percentage of their total body length and head length, respectively, according to the standard methods [7]. The fish's total length (TL) was calculated using a divider and measuring board, and was measured to the closest millimetre from the tip of the snout to the end of the caudal fin. All other morphometric parameters were also measured, including, fork length (FL), standard length (SL), eye diameter (ED), pre-dorsal length (PDL), pre-orbital length (POL), head length (HL), pre-pectoral length (PPL), pre-pelvic length (PPvL), pre-anal length (PAL), body depth (BD), and caudal length (CL) (Figure 1). Dorsal fin rays (DFR), pelvic rays (PFER), pectoral fin rays (PFR), anal fin rays (AFR), caudal fin rays (CFR), and lateral line scales (LLS) were counted as meristic counts. All the data were recorded separately according to sexes. Additionally, variability in external appearance across size groups was also observed during morphometric analysis through macroscopic examination.

**2.3. Biostatistics.** Summary statistics of fresh fish specimens were examined separately for both sexes of fresh fish specimens. Following that, the information was used to calculate the regression equation ( $Y = a + bx$ ), using the TL and HL as independent factors and the other variables as dependent variables. The significance of the results was examined using the coefficients of correlation ( $r$ ), coefficients of determination ( $R^2$ ), and regression (slope ( $a$ ) and intercept ( $b$ )), respectively. In addition, principal component analysis (PCA) has been used to determine the intraclass relationship between the morphometric parameters and their multivariate analysis, which was further explained by the multiple linear regression of these parameters. The dataset was analyzed by using MS Excel data analysis software [8] and PAST software 4.08 version [9].

## 3. Results:

### Morpho-biology.

**3.1. In table 1, 2 and figure 1:** Fish species examined was found to be dominated by young individuals with female specimens ranging length from 17.3 to 43.6 cm and male specimens from 17.4 to 23.6 cm. Summary statistics showed the variation in all morphometric parameters and were used to assess the annual variation between male and female (Table 1). The study focused on size group distributions ranging from 15-20 cm, 20-25 cm, 25-30 cm, 30-35 cm, 35-40 cm, to 40-45 cm (Table 2). Notably, the observed variations in external

appearance were consistent annually. This suggests a dynamic pattern in the morphological characteristics of the fish population within each size group over the course of the study period. These findings suggested that the population structure was stable and healthy.

**3.2. In table 3 and figure 2:** A total of nine morphometric characters were studied in relation with their TL separately for male and female. For males, maximum correlation value ( $r = 0.99$ ) was found among all parameters, while minimum correlation with CL ( $r = 0.79$ ). For females, maximum value ( $r = 0.99$ ) was found among all parameters, while minimum correlation with PPvL ( $r = 0.62$ ). High correlation value signifies a close relationship in the observed parameters. Other three morphometric parameters were studied in relation of HL separately for male and female showing a significant correlation. For males, maximum correlation was found among BD ( $r = 0.98$ ), and minimum in POL ( $r = 0.83$ ), while for females' highest correlation was found among POL, while minimum correlation was found in ED ( $0.59$ ). Therefore, there was a significant positive correlation between growths of all morphometric parameters with respect to TL and HL, respectively (Table 3).

Dataset was further analysed by using multiple linear regression to confirm the correlation and significant relation between various morphometric parameters (Figure 2). This analysis showed that all the morphometric characters show linear relationship and are highly correlated, maximum ( $r = 0.95$ ) for BD, and minimum correlation ( $r = 0.88$ ) for POL in percentage with TL and HL, respectively, which indicates that all the morphometric parameters are highly correlated and they increase in the direct proportion with each other (Figure 2).

**3.3. In table 4, 5 and figure 3:** Multivariate PCA analysis of 10 morphometric measurements were done to examine any phenotypic differences between the populations, in relation with TL extracted two factors with eigenvalues  $> 1$ , explains total variance of 98.7%. PC1 accounted was found positively correlated to all the other dependent morphometric variables. Similarly, another 10 components (PC2 to PC11) showed different degrees of variation (Table 4 & Figure 3).

The validity of the factor analysis of the data set was supported by Bartlett's test of sphericity ( $\chi^2 = 680.77$ ;  $p \leq 0.001$ ) for total length relation (Table 5), showing highly significant relation among them. This test of sphericity examined the overall significance of the correlation matrix for the morphometric traits of fish species.

**3.4. In table 6, 7 and figure 4:** Multivariate PCA analysis of 3 morphometric measurements in relation with HL extracted one factors with eigenvalues  $> 1$ , explains total variance of 97.006%. PC1 accounted for this was also found positively correlated to all the other variables, showing high intra-relation among them due to body shape, respectively (Table 6 & Figure 4).

The validity of the factor analysis of the data set was supported by Bartlett's test of sphericity for head length ( $\chi^2 = 176.42$ ;  $p \leq 0.001$ ) (Table 7), showing highly significant relation among them. This test of sphericity examined the overall significance of the correlation matrix for the morphometric traits of fish species.

**3.5. In table 8:** For meristic, dorsal fin ranges (7 - 10), pelvic fin (8 - 10), pectoral fin (12 - 16), anal fin (5 - 7), and lateral line scale were in the range of (91 - 104), respectively (Table 8).

#### 4. Discussion:

In our study, we observed that the morphometric characters of male and female fish specimens were not statistically different, which was similar to the findings of [10] and [11], who had analyzed morphometric features and state that no significant differentiation observed in both sexes for *Schizothorax* species. This study also found a strong and significant correlation ( $r = 0.99$ ) between various morphometric parameters, which was comparable to the findings of [12], as well as [13] and [14], who had observed a positive and significant correlation among all morphometric characteristics. Body depth ( $r = 0.9504$ ) and Pre-orbital length ( $r = 0.8843$ ) were the body parameters with the highest and lowest correlations, respectively, to total length. [1] had also examined the morphometric characteristics of *S. richardsonii* and found that practically all of them exhibit highest levels of correlation ( $r \geq 0.90$ ).

Multivariate PCA analysis shows a significant differentiation between the morphological characteristics of *S. richardsonii* populations and their intra-relation (Tables 4 and 5). The PCA analysis's findings revealed a high significance rate ( $p \leq 0.001$ ). [15] and [1], had also found similar results and had reported that genetic variation

accounts for 90% of variance, whereas environmental variation accounts for 10%, even if current *S. richardsonii* populations have not yet shown evidence of genetic differentiation.

The findings of the present study for meristic counts showed slight variation in all the characters examined. According to [16], *S. richardsonii* fin ray variation falls within a certain range. Both [17] and [18] had described variations in silver carp meristic counts, which was similar with our finding. The slight variations in *S. richardsonii* morphometry may also be the consequence of geographic and topographic variances, since fishes are quickly adaptable to environmental changes by changing their physiology and behavior, which results in a change in morphometry.

This study indicates maximum correlation for all the morphometric characteristics and meristic counts of *S. richardsonii*, which revealed that they all showed linear relationship between dependent and independent characters. The results of statistical analysis of various morphometric characters showed highest significant relation among them, which provide important insights into the population dynamics of studied fish, and showed no significant differences among males and female fish specimens. Therefore, this study will help to understand the complete morpho-biology and their intra-relation among them, which is crucial for the development of sustainable management practices.

### Acknowledgements

The authors are highly thankful to Department of Zoology of SSJ University, LSM Campus Pithoragarh, Uttarakhand, India for providing laboratory facilities for carrying out this work. The support of people of local commercial market during the actual field sampling and data collection is duly acknowledged.

### Declaration

We affirm that this manuscript is entirely original, without any prior publication, and is not under consideration for publication elsewhere.

### Funding

This research received no specific grant from any funding agency.

### Compliance with Ethical standards

This article does not contain any studies with human participants performed by any of the authors.

### Conflict of Interest

There are no conflicts of interest related to this work. As the corresponding author, I attest that co-author have carefully reviewed and consented to the submission of this manuscript.

### Authorship contribution statement

The authors confirm contribution as follows: Corresponding author Rakesh Verma<sup>2\*</sup> designed and supervised the work, also performing the measurements, while Primary author Priyanka Sharma<sup>1</sup> processed the experimental data, performed analysis, and drafted the manuscript. Final draft of manuscript was designed and discussed by all authors.

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## TABLES

**Table 1. Summary statistics showing annual variation between morphometric parameters of male and female fish species of *S. richardsonii*.**

PARAMETERS Measurements cm	FEMALE			MALE		
	Min	Max	Mean± SD	Min	Max	Mean± SD
Total length TL	17.3	43.6	27.7±9.4	17.4	26.6	22.05±3.1
Standard length SL	14.2	37.6	22.8±7.8	14.1	21.9	18.2±2.7
Fork length FL	15.8	38.3	24.7±8.3	16.1	23.8	19.9±2.8
Head length HL	2.9	6.4	4.2±1.14	2.9	4.2	3.5±0.4
Pre-dorsal length PL	7.08	16.1	10.6±3.4	6.8	10.3	8.6±1.3
Pre-orbital length POL	1.2	3.2	1.9±0.6	1.2	2.2	1.5±0.3
Pre-pectoral length PPL	2.8	6.8	4.4±1.4	2.9	4.3	3.6±0.5
Pre-pelvic length PPvL	7.3	18.4	11.9±3.9	6.8	11.1	9.2±1.4
Body depth BD	3.1	8.8	5.5±1.8	3.2	5.2	4.3±0.7
Eye diameter ED	0.5	0.8	0.6±0.09	0.5	0.6	0.5±0.04
Anal length AL	10.8	27.2	17.14±5.6	11.2	16.5	13.6±1.7
Caudal length CL	3.8	13.8	7±3.5	3.4	7.2	5.4±1.2



**Table 2. Variability in External Appearance Across Size Groups observed during morphometric analysis**

Size group (cm)						
Parameters	15-20	20-25	25-30	30-35	35-40	40-45
Body colour	Dark golden	Light grey	Dark golden	Light grey	Light grey	Light grey
Head colour	Light grey	Dark grey	Dark grey	Dark grey	Grey	Dark grey
Lateral line Clarity	Clear	Very clear	Very clear	Very clear	Clear	Very clear
Lateral line colour	Light golden	Dark brown	Light brown	Light brown	Dark silver	Dark Brown
Dorsal	Dark golden	Dark grey	Golden	Light grey	Light grey	Light grey
Ventral	Silver grey	Silver	Silver	Silver white	Silver white	Silver white
Body spot	Present	Present	Present	Present	Present	Present
Fin colour	Light pink	Orange brown	Orange brown	Brown	Light brown	Orange brown
Fin texture	Smooth	Rough	Rigid	Rigid	Smooth	Rough
Abdomen	Normal	Normal	Bulged	Very bulgy	Bulged	Bulged
Tubicles	Present	Absent	Absent	Absent	Absent	Present

**Table 3. Regression analysis of Total length and Head length on different morphometric parameters of *S. richardsonii*.**

S.No	Parameters	Sex	Slope (a)	Intercept (b)	r	R <sup>2</sup>	Regression equation
In the %age of Total length							
1.	Fork length	M	0.89	0.09	0.99	0.99	FL = 0.8991TL + 0.0912
		F	0.87	0.64	0.99	0.99	FL = 0.8701TL + 0.6442
2.	Standard length	M	0.83	-0.11	0.90	0.99	SL = 0.8303TL - 0.1178
		F	0.82	0.03	0.99	0.98	SL = 0.8226TL + 0.0397
3.	Head length	M	0.15	0.15	0.97	0.95	HL = 0.1526TL + 0.1567
		F	0.11	1.10	0.95	0.90	HL = 0.1148TL + 1.1077
4.	Body depth	M	0.19	0.008	0.97	0.94	BD = 0.1947TL + 0.009
		F	0.19	0.03	0.98	0.96	BD = 0.1959TL + 0.0371
5.	Pre-dorsal length	M	0.39	-0.02	0.99	0.98	PL = 0.3923TL - 0.0286
		F	0.35	0.68	0.99	0.98	PL = 0.3573TL + 0.6791
6.	Pre-pectoral length	M	0.16	0.04	0.98	0.97	PPL = 0.1615TL + 0.0425
		F	0.13	0.56	0.97	0.94	PPL = 0.1381TL + 0.5653
7.	Pre-pelvic length	M	0.42	-0.14	0.99	0.99	PPvL = 0.4232TL - 0.1407
		F	0.39	1.19	0.93	0.86	PPvL = 0.3905TL + 1.1912
8.	Pre-anal length	M	0.60	0.33	0.99	0.98	PAL = 0.6008TL + 0.3354
		F	0.59	0.55	0.99	0.99	PAL = 0.5995TL + 0.5562
9.	Caudal length	M	0.22	0.55	0.79	0.62	CL = 0.219TL + 0.5514
		F	0.36	-3.03	0.97	0.94	CL = 0.3625TL - 3.0308
In the %age of Head length							
10.	Body depth	M	1.25	-0.13	0.98	0.96	BD = 1.2594HL - 0.136
		F	1.55	-1.22	0.94	0.89	BD = 1.5598HL - 1.224
11.	Eye diameter	M	0.14	0.04	0.96	0.92	ED = 0.1486HL + 0.0399
		F	0.05	0.44	0.59	0.84	ED = 0.0509HL + 0.4485
12.	Pre-orbital length	M	0.39	0.13	0.83	0.69	POL = 0.3952HL + 0.1324
		F	0.53	-0.38	0.96	0.93	POL = 0.5353HL - 0.3823

Note: All length were measured in centimetre, r is correlation coefficient, ( $Y = a + bx$ ) is regression equation, where a = slope, and b = Intercept, and R<sup>2</sup> is coefficient of determination, respectively.

**Table 4. Result of Multivariate PCA analysis of morphometric data in relation with Total length (TL) of fish.**

	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6	PC 7	PC 8	PC 9	PC 10	PC 11
TL	0.553	0.272	-0.266	0.020	0.198	0.545	-0.155	-0.340	-0.076	0.025	-0.253
FL	0.479	0.070	0.023	0.343	-0.004	0.335	-0.588	0.269	0.129	-0.081	0.303
HL	0.070	0.147	-0.101	0.317	0.135	0.328	0.396	0.750	0.098	0.008	-0.073
CL	0.203	-0.737	0.388	0.223	0.105	0.371	0.009	-0.094	0.227	0.058	0.001
SL	0.437	-0.321	0.004	0.547	0.266	0.305	0.132	0.280	-0.232	0.251	-0.163
BD	0.111	-0.032	0.091	0.213	0.576	0.099	0.396	-0.274	-0.204	-0.273	0.491
AL	0.333	-0.107	-0.413	0.133	0.507	0.131	0.380	-0.075	0.238	-0.436	0.128
PDL	0.196	0.063	0.136	0.508	0.328	-0.285	0.248	-0.144	-0.504	0.182	-0.346
POL	0.043	0.044	-0.159	0.069	0.298	0.221	0.065	0.0006	-0.213	0.603	0.643
PPL	0.083	0.153	-0.035	0.136	-0.174	0.292	0.249	-0.239	0.673	0.500	-0.100
Eigen value	165.592	1.376	0.371	0.141	0.078	0.073	0.053	0.017	0.010	0.008	0.0002
% variance	98.728	0.820	0.221	0.084	0.047	0.043	0.032	0.010	0.006	0.005	0.0001

**Table 5. Barlett's test of sphericity showing significant relation between Total length (TL) with other morphometric parameters.**

Bartlett's sphericity test	
Determinant:	4.60E-37
chi2:	680.77
df:	65
p (spherical):	2.02E-103

**Table 6. Result of Multivariate PCA analysis of morphometric data in relation with Head length (HL) of fish.**

	PC 1	PC 2	PC 3	PC 4
HL	0.517	0.645	-0.562	0.020
BD	0.799	-0.596	0.049	-0.047
ED	0.026	-0.042	0.012	0.998
Eigenvalue	3.245	0.072	0.026	0.0008
% variance	97.006	2.176	0.791	0.026

**Table 7. Barlett's test of sphericity showing significant relation between Head length (HL) with other morphometric parameters.**

Bartlett's sphericity test	
Determinant:	4.41E-08
chi2:	176.42
df:	9
p (spherical):	2.83E-33

**Table 8. Meristic counts of fish *S. richardsonii* specimens (n = 120) captured from rivers of Kumaun Himalayan region of Uttarakhand.**

S.No.	Parameters	Abbreviations	Range	Tilak (1987)	Yadav <i>et al.</i> (2014)
1.	Dorsal fin rays	DFR	7-10	II-III/8	III/8
2.	Pelvic rays	PFER	8-10	I/9-10	I/9
3.	Pectoral fin rays	PFR	12-16	I/15-16	I/15-16
4.	Anal fin rays	AFR	5-7	II-III/5	III/5
5.	Lateral line scales	LLS	91-104	94-110	85-110

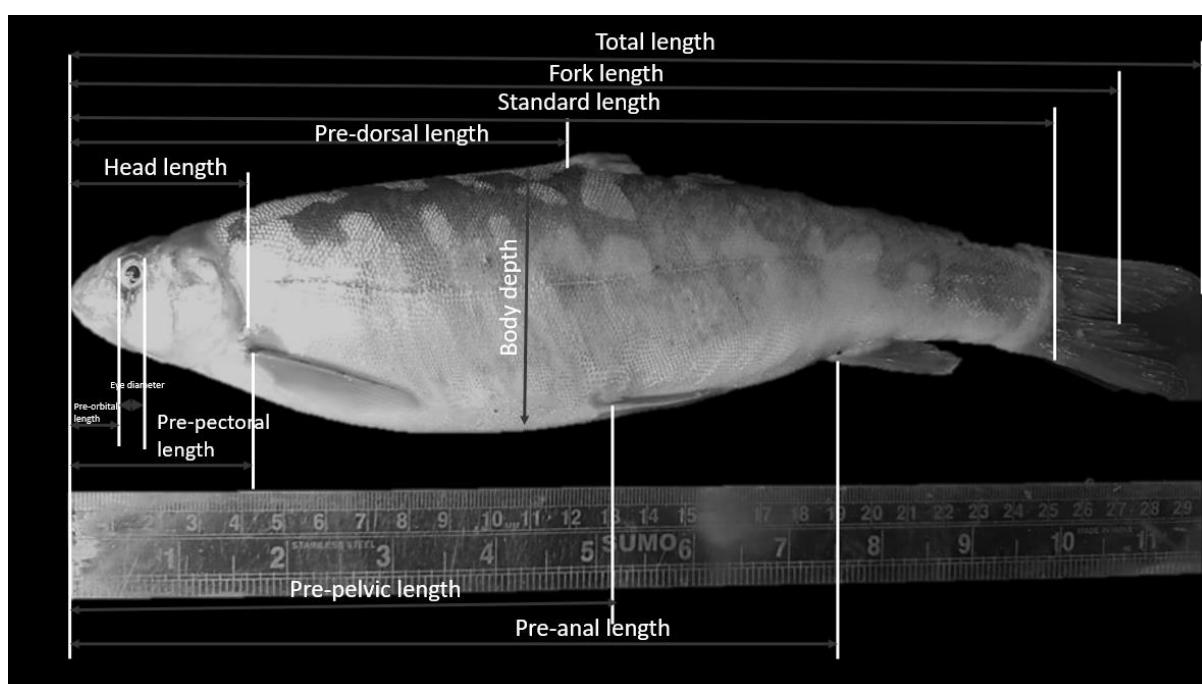
**FIGURE CAPTIONS**

**Figure 1.** Measurements of the different morphometric parameters of fish *S. richardsonii*.

**Figure 2.** Graph showing Multiple linear regression between various parameter (a) In % with TL, and (b) In % with HL.

**Figure 3.** Multivariate PCA relation between Total length (TL) of fish with other morphometric parameters.

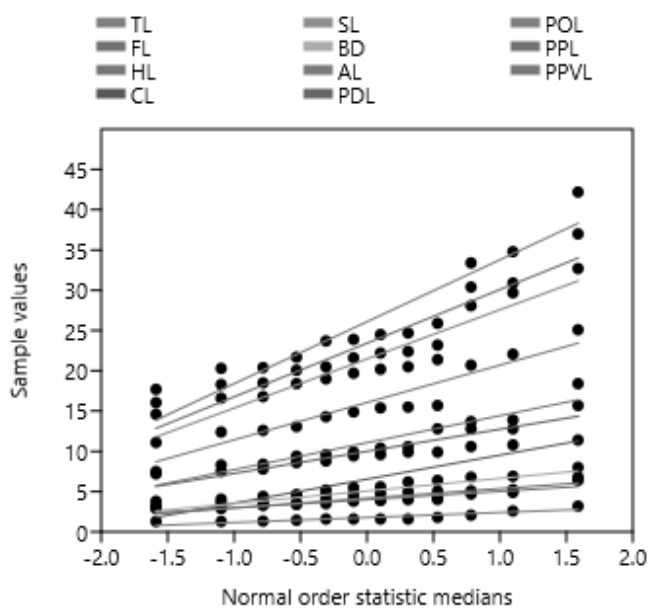
**Figure 4.** Multivariate PCA relation between Head length (HL) of fish with other morphometric parameters.

**Figure 1. Measurements of the different morphometric parameters of fish *S. richardsonii*.**

(a)

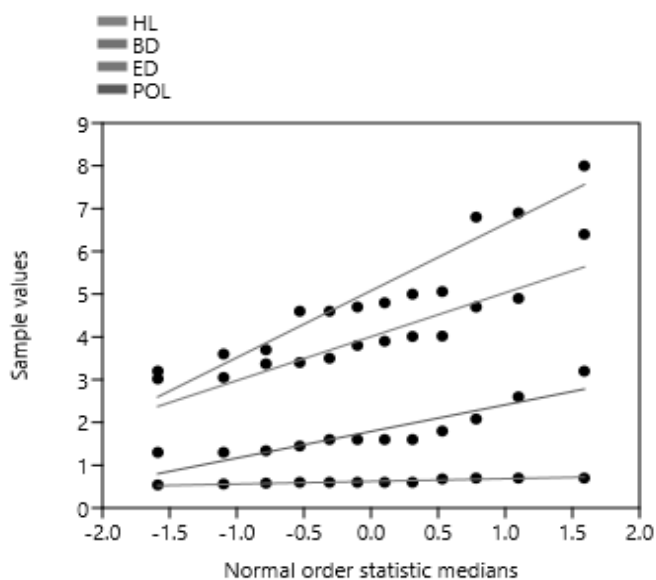
CORRELATION COEFFICIENT	
TL	0.9295
FL	0.9324
HL	0.9200
CL	0.9073
SL	0.9378
BD	0.9504
AL	0.9347
PDL	0.9371
POL	0.8843
PPL	0.9414
PPVL	0.9396



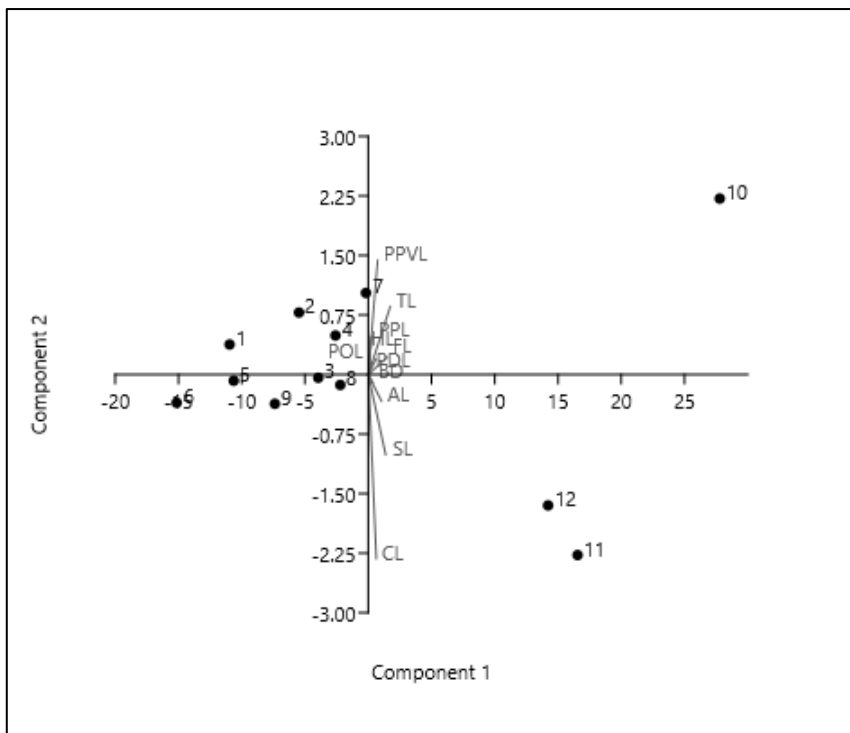


(b)

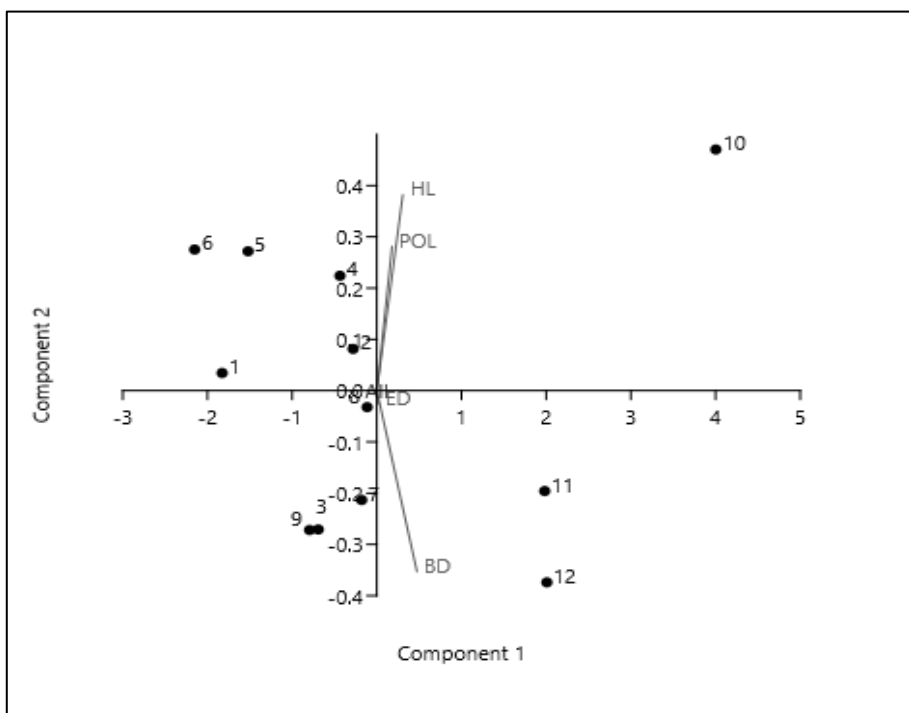
CORRELATION COEFFICIENT	
HL	0.92
BD	0.9504
ED	0.928
POL	0.8843



**Figure 2. Graph showing Multiple linear regression between various parameter (a) In % with TL, and (b) In % with HL.**



**Figure 3. Multivariate PCA relation between Total length (TL) of fish with other morphometric parameters.**



**Figure 4. Multivariate PCA relation between Head length (HL) of fish with other morphometric parameters.**