Chapter-1

INTRODUCTION

One of the six main groups of animals along with invertebrates, amphibians, reptiles, birds and mammals, fish are so plentiful in the world's oceans, lakes, rivers and many other waterbodies which is an inimitable source of animal protein. Fish now accounts for almost 17 percent of the global population's intake of protein and in some coastal and island countries it can top 70 percent (FAO, 2012). Fisheries and aquaculture support the livelihoods of 10–12 percent of the world's population (FAO, 2012). Bangladesh is gratified with rich extensive inland and marine fisheries potential resources with a wide variety of indigenous and exotic fish fauna. Fisheries sector represents one of the most productive and dynamic sectors in Bangladesh contributing 3.69% to the GDP of the country and 22.60% to the agricultural GDP (FRSS, 2016). There are 475 species of marine fishes and 260 species of freshwater fishes (DoF, 2013).

Morphological characters are most important in the identification and taxonomy of fishes, and the only known facts about many fishes. In addition understanding the function of a morphological structure is a stronghold for practical use in taxonomy and ecology (Bohlen, 2008). It is a widely used tool in the study of ichthyological systematics or taxonomy which looks at measurable components of fish anatomy such as body parts and fins and its ratio of body length. This technique is very useful for testing and graphically displays the differences in shape.

The measurement of morphometric and meristic characters are powerful tools which can be used for the stock identification, elucidating relationship among populations and to separate physically similar species. Information on the biology and population structure of any species is a prerequisite for developing management and conservation strategies (Turan et al., 2006) and may be applicable for studying short-term and environmentally induced variations, even for the genetic management of the population. Morphometric differences among stocks of a species are recognized as important for evaluating the population structure and as a basis for identifying stocks (Ihssen et al., 1981; Templeman, 1983; Smith and Jamieson, 1986; Turan, 2004; Turan et al., 2004b; Vishalakshi and Singh, 2008; Randall and Pyle, 2008). Intra and

interspecific patterns of variation of fishes can be obviously evaluated in terms of concept of size and shape (Mekkawy and Mahmoud, 1992b; Hajjej et al., 2011). Such concept is considered as the basic step in study of biometric variations in species especially in geometric terms (Jolicoeur and Mosimann, 1960; Bookstein, 1991; Akhter et al., 2003). The relative contribution of size and shape to the overall pattern of racial, geographic and inter-specific variations in species has long been investigated (Gunawickrama, 2007 and Hajjej et al., 2011). Such concept was found to be valid in identification of fish stock from a fisheries point of view (Cadrin, 2000; Monet et al., 2006). The traditional and geometric morphometric measurements are considered in univariate (allometric growth) and multivariate senses (Mekkawy, 1990) reflecting different patterns of size and shape variations. Analysis of these variations isolates specific morphometric indices and variants which have taxonomic potentials and discriminating powers away from the environmental and geographical influences. The meristic characters were also found to be valid in race and species identification and in turn in stock identification for fishery purposes (Mekkawy, 1991, 1997; Turan, 2004).

Morphometric and meristic characters of fish are the measurable or countable characters common to all fishes. Landmarks refer to some arbitrarily selected points on a fish's body, and with the help of these points, the individual fish shape can be analyzed. In other words, a landmark is a point of correspondence on an object that matches between and within populations (Barlow, 1961; Swain and Foote, 1999) and often subject to strong natural and sexual selection that may vary across a species range (Arnold, 1983; Bels et al., 2003). Landmark based measurements with the help of landmark points are powerful tools (Hossain et al., 2010) which can be used for the stock identification of fish species. Recently landmark based morphology data for gonia (Begum et al., 2013), kalibaus (Hossain et al., 2010), rohu (Hasan et al., 2007) and thai pangas (Khan et al., 2004) have been developed in home and abroad.

Hilsa fish, the national fish of Bangladesh is one of the most important tropical fish of the family clupeidae under the genus *Tenualosa* which is anadromous (available in the rivers, estuaries and the sea) in nature. Among the three kinds of *Tenualosa* sp. found in Bangladesh, the Padma ilish (*T. ilisha*) and the Chandana ilish (*T. toli*) are mostly well-known.

Hilsa is the largest single species fishery in Bangladesh. These species have a wide distribution in the Bay of Bengal and the rivers (Padma, Meghna) of Bangladesh. The production of Hilsa fish is 3.51 MT which contributes 11% to the total fish production and 1% to the national GDP of the country (DoF, 2014). Being an anadromous fish, hilsa populations from the Bay of Bengal mainly inhibits in marine water and migrates to the freshwater for spawning and returned to their original habitat (Hossain et al., 2016). Figure 01 shows the details of life history and migration pattern of hilsa shad (Hossain et al., 2016).

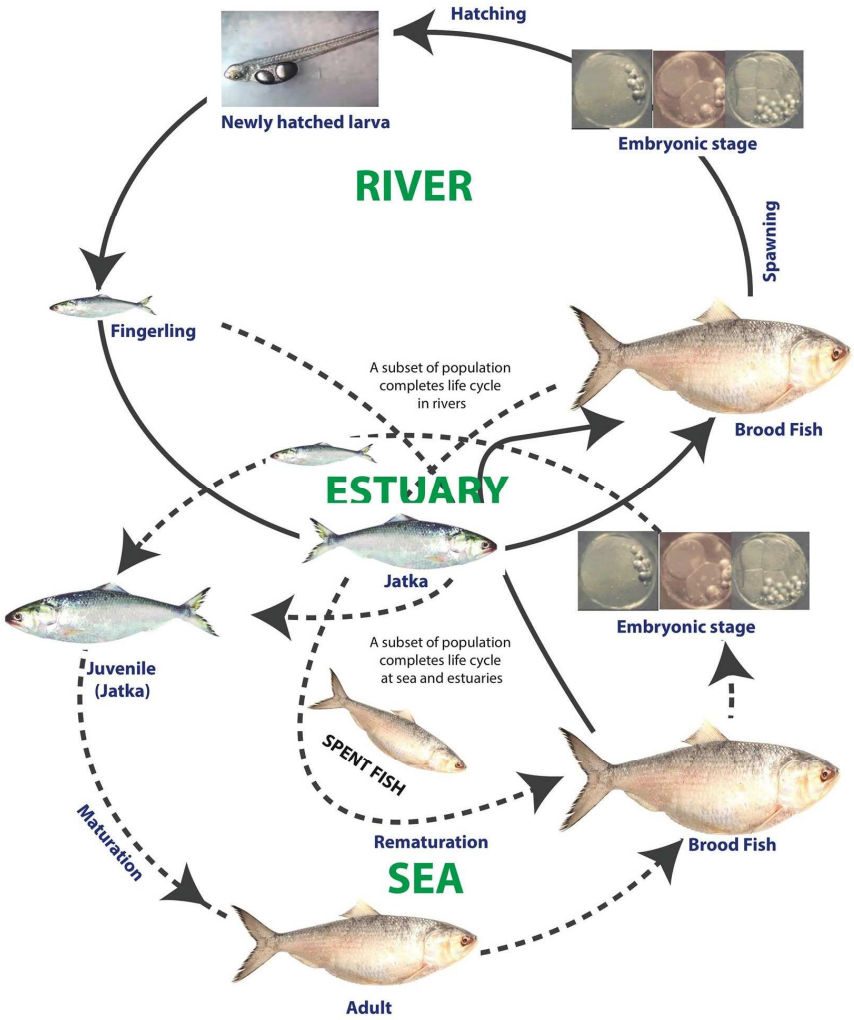

Figure 01: Schematic diagram of life cycle of hilsa shad in the Ganges-Brahmaputra-Meghna river system adjacent to the northern Bay of Bengal (Hossain et al., 2016).

Figure 01 shows that different subsets of hilsa populations from Bay of Bengal use coastal and estuarine habitats for spawning without entering freshwater. On the other hand, small subsets of populations complete their life cycle only within freshwater and does not migrate to sea at any stage of development. Therefore it can be hypothesized that there may be some morphological differences between freshwater or saltwater inhabitants of hilsha populations. For that, we collected fish from three different sources in consideration of spawning migration. The aim of this study was to determine the morphological differences between three different sources of *T. ilisha* populations and also in comparison with *T. toli* using land-mark based analysis.

1.1. Objectives of the research:

The objectives of the proposed research are as follows:

- To evaluate the morphometric and meristic variations between *T. ilisha* and *T. toli*
- To determine possible differences between separate unit stocks of the same species (variations among the stocks of *T. ilisha)*
- To differentiate closely related species of *Tenualosa* more precisely based on their morphology

Chapter-2

REVIEW OF LITERATURE

Different methods have been employed for studying genetic variability. Use of meristic, morphometric and landmark characteristics to study the variation among stocks of fish species is a common phenomenon in the world. Many studies have been carried out on biological aspects of different fish species in Bangladesh. This chapter is about a detailed review on the morphological studies carried in different fish species.

Hasan et al., (2007) studied the taxonomic variation of rohu (*Labeo rohita*) and mrigal (*Cirrhinus cirrhosus*) populations in Bangladesh based on the morphometric and meristic data of the populations and suggested that hatchery populations of rohu and mrigal might be deviated from its origin and morphological characters of these species could be used for the determination of purity of the species.

Hasan et al., (2005) made the taxonomic comparison within five populations of climbing perch (*Anabas testudineas*) collected from five regions of Bangladesh and found average total length, standard length, post orbital length, eye length, and length of base of the dorsal fin of the population of Khulna region higher than those of the other four populations.

Khan et al., (2004) studied morphological characters of four hatchery populations (Shambhuganj, Brahmaputra, Anudan and Bhai-bhai) of thai pangas (*Pangasius hypopthalmus*) from Mymensingh region in Bangladesh and found that four morphometric characters (BDA, PEL, FL, HL, and HW) and two meristic characters (AFR and CFR) in Anudan population were significantly ($p<0.0001$) higher than other three populations.

Ferrite et al., (2003) investigated morphological characters of four Italian populations of *Lebias fasciata* in order to assess the level of differentiation among populations. Fourteen meristic and twenty three morphometric characters, relative to the skull, vertebral column, and the rays of the dorsal and anal fins were examined. The morphological results showed a note worthy differentiation among the four populations which reflected their high degree of isolation but their morphological differentiation cannot be interpreted biogeographically.

Hossain et al., (2010) examined landmark-based morphometric characters along with truss network measurements and meristic counts to evaluate the population status of the endangered carp, kalibaus (*Labeo calbasu*) from two isolated rivers (the Jamuna and the Halda) and a hatchery and observed significant differences in four (maximum body height, pre-orbital length, peduncle length, and maxillary barbell length) of twelve morphometric measurements, two (pectoral fin rays and scales above the lateral line) of nine meristic counts, and four (8 to 9, 3 to 10, 2 to 10, and 1 to11) of twenty two truss network measurements among the stocks.

Turan et al., (2004a) worked with *Liza abu* stocks from the Orontes, Euphrates and Tigris rivers to know the genetic and morphometric structure. Simultaneously, allozyme electrophoresis for genetic comparison and the truss network system for morphometric comparison were applied to the same sample set and found highly significant morphological differences between the three *Liza abu* stocks and hence isolated Tigris stock from the other two stocks.

Swain et al., (1991) used the truss system for the identification of hatchery and wild populations of coho salmon (*Oncorhynchus kisutch*) and found significant morphometric variation.

Turan et al., (2004b) employed morphometric characters with the truss network system to know the population status of anchovy (*Engraulis encrasicolus* L.) in Turkish terrestrial waters and observed high degree of dissimilarity among the anchovy samples and thus identified as separated stocks.

Prakash and Verma (1982) studied morphometric characters and their relationship in *Notopterus notopterus* and found that the standard length, pectoral fin length, body height and head length (dependent variables) were highly correlated with the total length (independent variables), while the eye diameter and inter orbital width (dependent variables) were highly correlated with the head length (independent variables).

Islam et al., (1983) described morphological characters of maturing and non-maturing *Labeo rohita*. They studied nine morphometric and eight meristic characters of 44 maturing and 72 non-maturing fishes. Body depth, pre-dorsal, pre-pectoral, pre-ventral, pre-anal and head showed linear relationships with total length whereas eye

diameter and snout length showed linear relationship with head length. Slight variation was recorded in the meristic characters.

Hoque and Rahman (1985) reported morphometric characters and their relationship in *Gudusia chapra* and found that the fork length, dorsal fin length, pectoral fin length, pelvic fin length, body depth and head length of the fish were highly correlated with its total length. While the eyed diameter, snout length and post-orbital head length were highly correlated with the head length of the fish.

Devi et al., (1991) studied the morphometric characters of the catfish, *Rita rita* (Hamilton) from the river Yamuna in North India. Observations were made on the basis of total length, fork length, standard length, head length, depth of body at pectoral fin-base and at caudal peduncle. They found that males and females showed heterogeneity in characters. Standard length and depth of body at pectoral fin-base were different at 1% while forked length and head length were different at 5% level of significance. A linear relationship was obtained between body characters and total length.

Kohinoor et al., (1995) compared morphometric characters of red tilapia (mutant *O. mossambicus* and *O. niloticus*) and found that the standard length, pelvic fin length, pectoral fin length, dorsal fin length, anal fin length and head length of both of fishes were highly correlated with the total length of the fish.

Azadi and Naser (1996) reported morphometry of *Labeo bata* from Kaptai reservoir and commented that the relationship between the dependent variables (standard length, fork length, head length, pre dorsal distance, length of dorsal fin, depth of dorsal fin, pre anal distance, length of pectoral fin, length of pelvic fin, minimum body width, maximum body width, distance between pectoral and pelvic fin, distance between pelvic and anal fin, length of caudal peduncle, length of caudal fin) were found to be correlated with the independent variable (the total length of fish) significantly at 0.1 %.

Grobler et al., (1997) reported a significant positive correlation between heterozygosity and variation within the morphological parameters in case of *Clarias gariepinus*.

Rognon et al., (1998) distinguished two groups amongst the *Clarias gariepinus* populations, one containing Nilo-Sudanian populations and the other including Lake Victoria and Southern African populations on the basis of morphological and allozyme variations.

Narejo et al., (2000) studied morphometric and meristic characters of *Gudusia chapra,* collected from Keenjhar Lake (Pakistan) and found no significant morphological differences between the sexes. Regressions of length-weight did not deviate significantly from cube law indicating isometric growth.

Hoese and Allen (2009) described two new species of genus *Glossogobius* from southern New Guinea and a third related species from northeastern Australia. *G. bellendenesis* distinctive in having reduced pre-dorsal scale and fin ray count where *G. robertsis* distinctive in fin ray and scale count that species were confused with *G. giuris,* which generally occurs in lower reaches of the river.

The meristic and morphological characteristic data were used in the identification of fish stock (Murta, 2000; Saboridorey and Nedreaas, 2000), determining taxonomic groups (Marcua et al., 1996) and even to distinguish cohort of a single species (Austin et al., 1999).

High degree of variation was observed in morphological characteristics among three different stocks (the Meghna, Padma and Ichamoti) of *Rhinomugil corsula* due to their environmental variation and separate geographical location (Hossain et al., 2015).

Morphometric and meristic characters were used to differentiate two congeneric archer fish species *Toxotes chatareus* and *Toxotes jaculatrix* inhabiting Malaysian coastal waters (Simon et al., 2010).

The comparative study of two types of palla, *Tenualosa ilisha* from River Indus, Pakistan revealed significant intertype differences in six morphometric measurements (total length, standard length, fork length, head length, eye diameter and girth) and seven meristic characters (total number of scutes, pre pelvic scutes, post pelvic scutes, dorsal fin rays, pectoral fin rays, pelvic fin rays and anal fin rays) (Narejo et al., 2008).

Chapter-3

MATERIALS AND METHODS

Methodology is an indispensable and integral part of any research. In a scientific research the acceptability of the results depends to a great extent on the appropriate methodology. This chapter deals with the methods that are followed and materials that are used to achieve the objectives of the study. In this study a scientific and logical methodology has been followed by the researcher. This study is based on sample collections from different habitats and data are collected and analyzed for the interpretation of results.

3.1. Collection of samples

The comparative study of *T. ilisha* and *T. toli* was based on the morphological examination and analysis using 64 specimens collected from local fisherman from different habitats and immediately preserved in ice box. Samples were collected from all locations by considering the catching date and migration time of fish. Sixteen fresh and healthy fish samples from each group were chosen for further analysis. The descriptions of sampling area, sample size, total length are presented in table 01. Samples were then brought to the laboratory of Molecular Biology and Biotechnology, Faculty of Fisheries, Chittagong Veterinary and Animal Sciences University, Chittagong for morphometric, meristic and landmark studies.

Table 01: Summary of sampling area, sample size, total length and habitats of collected samples of *T. ilisha* and *T. toli.*

Species	Habitats	Source/location	Total length (cm)	Sample size
T. ilisha	Marine	Cox's Bazar 21°19N, 91°35E	25.68 ± 1.17	16
T. ilisha	River	Chandpur 23°12N, 90°37'E	31.65 ± 1.15	16
T. ilisha	Coastal	Chittagong 22°11N, 91°37E	26.43 ± 0.77	16
T. toli	Coastal	Chittagong 22°11N, 91°37E	28.57 ± 1.62	16

3.2. Measurement of morphometric characteristics

Sixteen general morphometric characters were measured (Figure 02) from each sampled fish following the conventional method described by Hubbs and Lagler (1958).

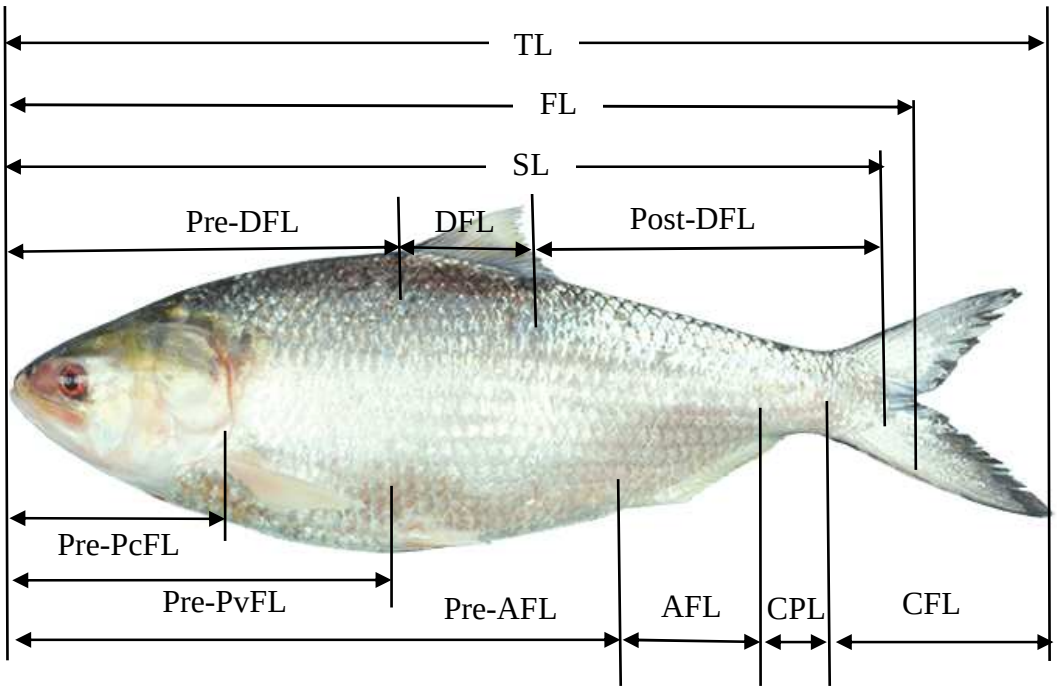

Figure 02: Overview of different morphometric indices of *Tenualosa* sp.

The morphometric characters were measured with an accuracy of 0.05 mm with the help of vernier calipers and metric scale. Table 02 shows the measured morphometric characters used in this experiment for morphological analysis with their descriptions.

Table 02: General morphometric characters and their descriptions used for the analysis.

SL. No	Characters	Description
01	Standard length (SL)	From the tip of the snout to the end of the vertebral column
02	Total length (TL)	From the tip of the snout to the longest caudal fin ray
03	Fork length (FL)	From the tip of the snout to the middle part of the fork of the tail

04	Pre-dorsal fin length (Pre-DFL)	From the snout tip to the origin of the dorsal fin
05	Dorsal fin length (DFL)	From base of first dorsal spine to base of last dorsal ray
06	Post-dorsal fin length (Post-DFL)	From posterior base of dorsal fin to the longest caudal fin ray
07	Pre-pelvic fin length (Pre-PvFL)	Front of the upper lip to the origin of the pelvic fin
08	Pelvic fin length (PvFL)	From base to tip of the pelvic fin
09	Pre-pectoral fin length (Pre-PtFL)	Front of the upper lip to the origin of the pectoral fin
10	Pectoral fin length (PtFL)	From base to tip of the pectoral fin
11	Caudal fin length (CFL)	From tail base to tip of the caudal fin
12	Pre-anal length (PAL)	Front of the upper lip to the origin of the anal fin
13	Anal fin Length (AFL)	From base of first anal spine to base of last anal ray
14	Highest body depth (HBD)	Vertical distance from the anterior part of the first dorsal fin and ventral part of the body
15	Least body depth (LBD)	Vertical distance at the end of the Vertebrae
16	Caudal peduncle length (CPL)	From the base of the anal fin to the base of the caudal fin

3.3. Measurement of meristic characteristics

Meristic characters such as dorsal fin rays (DFR), anal fin rays (AFR), caudal fin rays (CFR), pectoral fin rays (PcFR), pelvic fin rays (PvFR) were counted from each fish by using magnifying glass and used for comparative analysis.

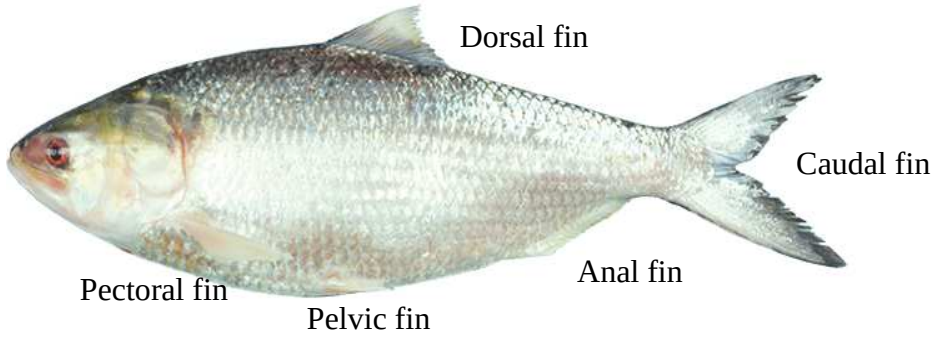

Figure 03: General indications of different meristic characters observed in *Tenualosa* sp.

3.4. Measurement of landmark distances

The truss network system was used to construct a network on fish body for measurement of landmark distances of the species. Eight landmarks outlining 14 distances were measured on the body. Landmark points were selected to make a homogeneous coverage of the total body plan in between two species based on the Strauss and Bookstein (1982). Each landmark was obtained by placing a fish on a graph paper and then the landmarks were detected with colored pointers for enabling accurate and consistent measurements. Finally the distances on the graph paper were measured using scale (Figure 04).

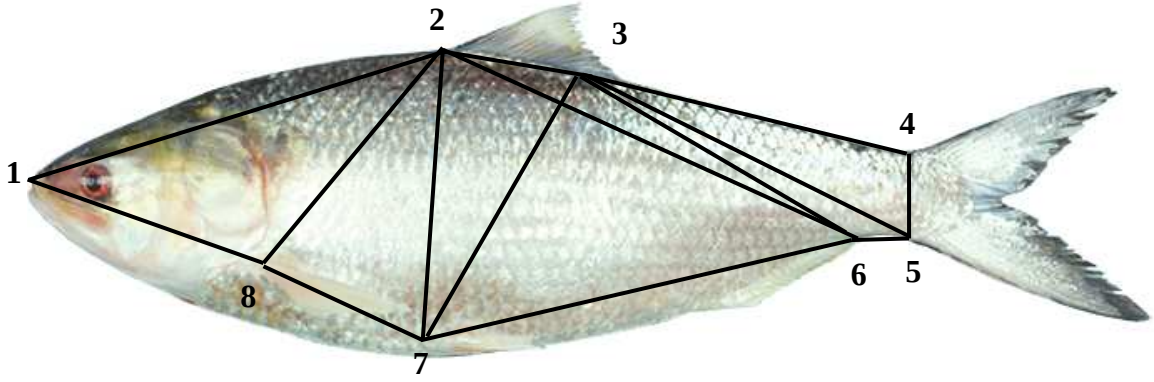

Figure 04: Randomly selected landmark points in fish body used in this study. The eight landmark points refers to (1) anterior tip snout of the upper jaw of mouth (2) base of origin of dorsal fin (3) end of dorsal fin (4) dorsal caudal fin base (5) ventral caudal fin base (6) ending of caudal fin base (7) base of pelvic fin (8) middle base of pectoral fin.

Table 03: Description of truss network characters used in the study.

Sl. No.	Character codes	Landmark points	Description of characters
01	A1	1-2	Anterior tip of snout to the origin of dorsal fin base
02	B1	2-3	Origin of dorsal fin to the end of dorsal fin base
03	C1	3-4	End of dorsal fin base to origin of caudal fin
04	D1	4-5	Upper to lower of caudal fin origin
05	E1	5-6	Origin of lower of caudal fin to end of the anal fin base
06	F1	6-7	Origin of anal fin to origin of pelvic fin
07	G1	7-8	Origin of pelvic fin to origin of pectoral fin
08	H1	8-1	Origin of pectoral fin to the end of snout tip
09	I2	2-7	Origin of dorsal fin to origin of pelvic fin
10	J2	2-6	Origin of dorsal fin to origin of anal fin
11	K2	3-7	End of dorsal fin base to origin of pelvic fin
12	L2	3-6	End of dorsal fin base to origin of anal fin
13	M2	3-5	End of dorsal fin base to lower caudal fin origin
14	N2	4-6	Origin of the upper caudal fin to end of the anal fin base

3.5. Statistical analysis

Prior to the analysis, size effects from the data set were eliminated. An allometric formula given by Elliott et al., (1995) with slight modification was used to remove the size effects from the data set.

$$M_{adj} = M\,(Ls/Lo)^{b}$$

Where,

M_{adj}: size adjusted measurement

M: original measurement

Ls: overall mean of standard length for all fish from all samples in each analysis

Lo: total length of fish

Parameter b was estimated for each character from the observed data as the slope of the regression of log M on log L_0, using all fish in all groups. The efficiency of the size adjusted values was then correlated with the TL and the transformed values.

In the first level of analysis, we compare among the collected samples of *T. ilisha* to show the morphological differences among habitats. In the second steps we compare between the *T. ilisha* and *T. toli* to observe the morphological distances in this two species. Univariate analysis of variance (ANOVA) was carried out to test the significance of morphological differences ($P<0.01$) on the basis of size adjusted morphological and landmark distance data. Meristic characters were compared using non-parametric Kruskal-Wallis test. In addition, all size adjusted morphological and landmark distance data were standardized and submitted to discriminant functional analysis (DFA) and principal component analysis (PCA). All statistical analysis was carried out using SPSS version 16.0 and MS excel 2010.

Chapter-4

RESULTS

This section is the simple descriptive part of the analysis of morphological data between two species of *Tenualosa* collected from Bangladeshi waterbodies. Result is an integral part of any research work. From our studies, here we present the details of systemic analytical observations of the morphology based on statistical approaches in two main parts.

4.1. Comparative studies on *T. ilisha* collected from three (03) different habitats

4.1.1. Analysis of meristic counts

Meristic counts of all samples of *T. ilisha* collected from three different habitats ranged from 17-21 for anal fin rays (M_e= 17), 18/19 for dorsal fin rays (M_e= 18), 7/8 for pelvic fin rays (M_e= 7), 14/15 for pectoral fin rays (M_e= 14), and 26-28 for caudal fin rays (M_e= 27). Number of branchiostegeal rays were fixed in all samples (B= VI). Though the number of scales on the lateral line (45-47) and lateral transverse (17-19) varied in between species, no significant differences were observed among three habitats. In the Kruskal Wallis (H) test the number of anal fin rays, dorsal fin rays, pelvic fin rays, pectoral fin rays and caudal fin rays were not statistically significant ($p>0.05$) among fish from three different habitats. Besides univariate statistics (ANOVA) also showed no significant differences ($P>0.05$) in meristic characters among fishes from three different habitats.

4.1.2. Analysis of morphometric and landmark distance measurements

There was no significant correlation ($p>0.05$) between the total length and adjusted morphological values which indicates that the size effects were successfully removed with the help of allometric transformations. Therefore, all the morphological and truss network measurements were considered for univariate analysis (ANOVA). Univariate analysis showed that eight [anal fin length (AFL), caudal peduncle length (CPL), highest body depth (HBD), least body depth (LBD), post-dorsal fin length (Post-DFL), pre-pectoral fin length (Pre-PcFL), pelvic fin length (PvFL), pre-pelvic fin length (Pre-PvFL)] of fifteen morphometric measurements were significantly different

in varying degrees ($p<0.05$ or $p<0.01$ or $p<0.001$) (Table 04) among three groups of populations of *T. ilisha*.

Table 04: Univariate statistics (ANOVA) of 15 of *T. ilisha* samples from three different habitats. *$p<0.05$, **$p<0.01$, and ***$p<0.001$ indicate degree of significance.

Morphometric characters	Wilks' Lambda	F value	Sig.
SL	0.973	0.632	0.536
FL	0.887	2.865	0.067
Pre-DFL	0.985	0.353	0.704
DFL	0.926	1.793	0.178
Post-DFL	0.865	3.526	0.038*
Pre-PvFL	0.868	3.425	0.041*
PvFL	0.528	20.112	0.000***
Pre-PcFL	0.835	4.450	0.017*
PcFL	0.906	2.324	0.110
Pre-AFL	0.927	1.768	0.182
AFL	0.669	11.123	0.000***
CFL	0.986	0.330	0.721
CPL	0.652	11.998	0.000***
HBD	0.660	11.612	0.000***
LBD	0.466	25.781	0.000***

In case of landmark distances, eight (1 to 2, 2 to 3, 3 to 4, 4 to 5, 6 to 7, 8 to 1, 2 to 6, 3 to 6) out of fourteen truss measurements were significantly different among samples in varying degrees ($p<0.05$ or $p<0.01$ or $p<0.001$) among three different groups of *T. ilisha* which was revealed through univariate statistics (Table 05).

Table 05: Univariate statistics (ANOVA) showing the differences among measurements of 14 truss networking (*p<0.05, **p<0.01, ***p<0.001) of *T. ilisha* from three (03) different habitats.

Landmark distance	Wilks' Lambda	F value	Sig.
1-2	0.646	12.333	0.000***
2-3	0.669	11.110	0.000***
3-4	0.822	4.887	0.012*
4-5	0.280	57.751	0.000***
5-6	0.905	2.354	0.107
6-7	0.823	4.833	0.013*
7-8	0.927	1.779	0.180
8-1	0.822	4.865	0.012*
2-7	0.996	0.089	0.915
2-6	0.724	8.585	0.001**
3-7	0.961	0.907	0.411
3-6	0.726	8.479	0.001**
3-5	0.923	1.865	0.167
4-6\	0.903	2.411	0.101

Discriminant function analysis (DFA) produced two sets of discriminant functions (DF1 and DF2) for both morphometric and landmark measurements. The first two DF analysis resolved 89.8% and 87.4% and the second DF accounted for 10.2% and 12.6% respectively of among group variability and together they explained 100% of the total variability for morphometric and landmark measurements.

Pooled within groups correlation between discriminating variables and discriminant functions revealed that among the 15 morphometric measurements, four measurements [least body depth (LBD), anal fin length (AFL), pre-pectoral fin length (Pre-PcFL), and pre-pelvic fin length (Pre-PvFL)] dominantly contributed to the first DF, while the remaining eleven [standard length (SL), fork length (FL), pre-dorsal fin length (Pre-DFL), dorsal fin length (DFL), post-dorsal fin length (Post-DFL), pelvic fin length (PvFL), caudal fin length (CFL), pre-anal length (PAL), highest body depth (HBD), caudal peduncle length (CPL)] contributed to the second DF (Table 06).

Table 06: Pooled within group correlation between discriminating variables and discriminant functions in case of general morphometric characteristics.

Morphometric characters	Discriminant function	
	DF1	DF2
LBD	-0.416*	0.169
AFL	-0.275*	0.044
Pre-PcFL	-0.174*	-0.010
Pre-PvFL	-0.152*	-0.054
PvFL	-0.284	0.706*
HBD	0.256	0.349*
Post-DFL	0.111	0.320*
CPL	0.269	0.291*
FL	-0.121	0.206*
Pre-AFL	0.085	0.206*
DFL	-0.095	-0.170*
PcFL	-0.117	0.137*
Pre-DFL	-0.022	-0.130*
CFL	0.030	-0.110*
SL	0.055	0.109*
Variables ordered by absolute size of correlation within function.		
*. Largest absolute correlation between each variable and any discriminant function		

In case of truss measurements, among the fourteen measurements two measurements (4 to 5 and 2 to 7) dominantly contributed to the first DF, while the remaining twelve measurements contributed to the second DF (Table 07).

Table 07: Pooled within group correlation between discriminating variables and discriminant functions in case of landmark distances among the samples of three different habitats.

Landmark distance	Discriminant function	
	DF1	**DF2**
D1	-0.513*	0.391
J2	-0.194*	0.179
A1	-0.014	0.649*
C1	-0.020	0.406*
H1	-0.050	0.387*
F1	0.078	0.351*
B1	-0.200	0.321*
L2	-0.175	0.281*
G1	-0.022	0.240*
M2	-0.046	0.221*
N2	-0.075	0.208*
E1	0.093	0.143*
K2	0.056	0.097*
I2	0.008	-0.051*

*. Largest absolute correlation between each variable and any discriminant function. Variables ordered by absolute size of correlation within function.

In discrimination space, morphometric measurements of fishes from the river sources were separated from other two populations (coastal and marine habitats). On the basis of morphometric measurement 81.3%, 75.0% and 100% of original group cases were correctly classified in case of coastal, marine and river habitats samples respectively and a total of 85.4% of original group cases were correctly classified for all three groups (Table 08). This suggested that *T. ilisha* of river populations were morphologically dissimilar to other groups. But the fish samples from the marine and coastal were not fully separated (Figure 05) which was revealed by principal component analysis (PCA).

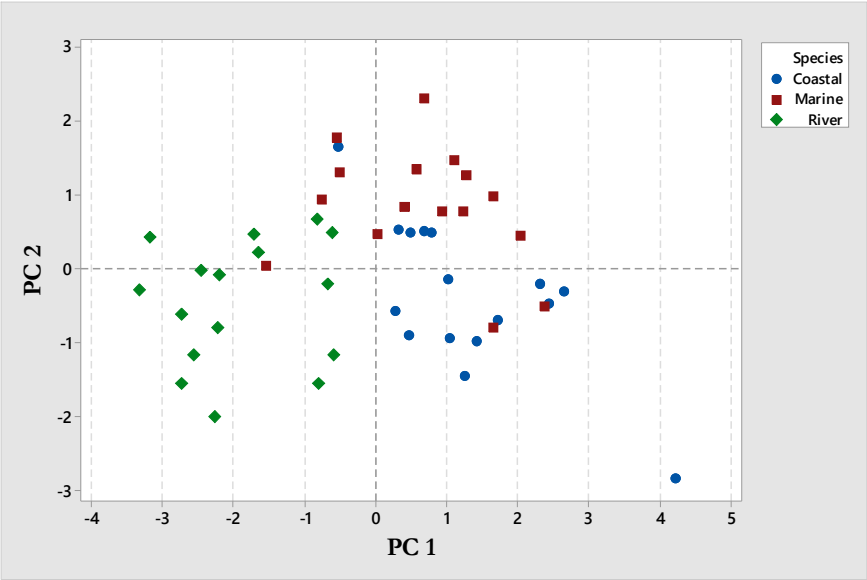

Figure 05: Scatterplot of the scores from PC1 and PC2 for morphometric characters of *T. ilisha* collected from three different habitats of Bangladesh.

Table 08: Classification results of canonical discriminant functions based on all morphometric measurements classification results.

		Species	Predicted Group Membership			
			Coastal	Marine	River	Total
Original	Count	Coastal	13	3	0	16
		Marine	3	12	1	16
		River	0	0	16	16
	%	Coastal	81.3	18.8	.0	100.0
		Marine	18.8	75.0	6.3	100.0
		River	0.0	0.0	100.0	100.0
a.85.4% of original grouped cases correctly classified.						

PCA based on the truss measurements data showed that the stocks were separated from each other specially fish stock of river originated was well separated from the fish stocks of other two sources. The truss measurement showed 100%, 93.8% and 100% of original group cases were correctly classified in case of coastal, marine and river populations respectively and a total of 97.7% original group cases were correctly classified for all three habitats (Table 09). This discriminant function scores based on

both morphometric and truss measurements suggested that fishes of river origin were isolated from the fish samples of coastal and marine inhabitants (Figure 06).

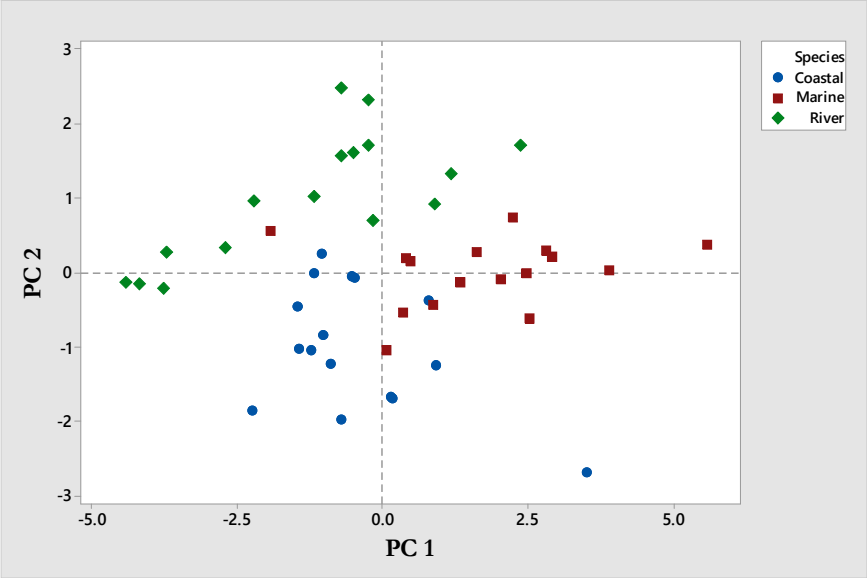

Figure 06: Scatterplot of the scores from PC1 and PC2 for truss measurements of *T. ilisha* collected from three different habitats of Bangladesh.

Table 09: Classification results of canonical discriminant functions based on all truss measurements classification results.

			Predicted Group Membership			
		Species	**Coastal**	**Marine**	**River**	**Total**
Original	Count	Coastal	16	0	0	16
		Marine	1	15	0	16
		River	0	0	16	16
	%	Coastal	100.0	.0	.0	100.0
		Marine	6.3	93.8	.0	100.0
		River	0.0	0.0	100.0	100.0
a.97.9% of original grouped cases correctly classified.						

4.1.3. Principal component analysis (PCA)

The significant traits (eight morphometric measurements and eight truss measurements) resulted from univariate analysis were further used for principal component analysis (PCA). To examine the suitability of the data for PCA, Bartlett's Test of Sphericity and the Kaiser–Meyer–Olkin (KMO) measurement were performed. The Bartlett's Sphericity test hypothesized that the values of the correlation matrix equal zero and the KMO measure of sampling adequacy tests, whether the partial correlation among variables is sufficiently high (Yakubu et al., 2011). The KMO statistics vary between 0 and 1 and the values greater than 0.5 are acceptable (Nimalathasan, 2009; Yakubu et al., 2011). The morphometric characters and landmark distances with an eigen value above 1 were included in this analysis. It is worth mentioning that a factor loading more than 0.30 is considered significant, 0.40 is considered more significant and factor loadings 0.50 or above is considered very significant (Lombarte et al., 2012).

PCA based on the morphometric measurements of *T. ilisha* from three different habitats showed the value of KMO for overall matrix was 0.685 and the Bartlett's Test of Sphericity was also significant ($P<0.01$). The results of KMO and Bartlett's Sphericity test suggested that the sampled data were appropriate to proceed with a factor analysis. The PCA based on eight morphometric measurements retained two components with eigen values>1, explaining 52.13% of the total variance. The first (PC1) and second (PC2) principal components accounted for 38.31% and 13.87% of the total variance respectively. All the eight morphometric measurements had significant loadings on PC1 and four [pelvic fin length (PvFL), highest body depth (HBD), least body depth (LBD) and post-dorsal fin length (Post-DFL)] on PC2 (Table 10).

Table 10: Component loadings of the first two principal components derived from the morphometric measurements of *T. ilisha*.

Morphometric characters	Component	
	PC1	**PC2**
Post dorsal fin length (Post-DFL)	-0.583	0.529
Pre-pelvic fin length (Pre-PvFL)	0.543	
Pelvic fin length (PvFL)	0.564	0.563
Pre pectoral fin length (Pre-PcFL)	0.577	
Anal fin length (AFL)	0.788	
Caudal fin length (CPL)	-0.715	
Highest body depth (HBD)	-0.545	0.347
Least body depth (LBD)	0.592	0.549
Eigen-values	3.065	1.106
% of variance	38.31	13.87
Cumulative variance %	38.31	52.13

The value of KMO for overall matrix was 0.72 and the Bartlett's Test of Sphericity was significant ($P<0.01$) based on the eight truss network data of *T. ilisha* from three different habitats. The results of KMO and Bartlett's test suggested that the sampled data was appropriate to proceed with a factor analysis procedure. The PCA based on eight truss measurements retained two components with eigen values>1, explaining 71.47% of the total variance. The first (PC1) and second (PC2) principal components accounted for 56.19% and 15.35% of the total variance respectively. All the eight truss measurement had significant loadings on PC1 and the most significant loadings on PC2 were 1-2, 2-3, 4-5, 6-7, 8-1 and 2-6 (Table 11).

Table 11: Component loadings of the first two principal components derived from the landmark distances of *T. ilisha.*

Landmark distance	Component	
	PC1	PC2
A1 (1-2)	0.612	0.502
B1 (2-3)	0.773	-0.317
C1 (3-4)	0.750	
D1 (4-5)	0.747	-0.458
F1 (6-7)	0.615	0.563
H1 (8-1)	0.740	0.393
J2 (2-6)	0.862	-0.377
L2 (3-6)	0.853	
Eigen-values	4.489	1.228
% of variance	56.118	15.352
Cumulative variance %	56.118	71.470

A dendrogram was drawn based on the landmark distances and morphological examinations among groups of centroids of *T. ilisha* populations collected from three different habitats. Two clusters were found based on the Squared Euclidean dissimilarity. The coastal and marine populations showed one cluster while the river group showed a distinct cluster (Figure 07).

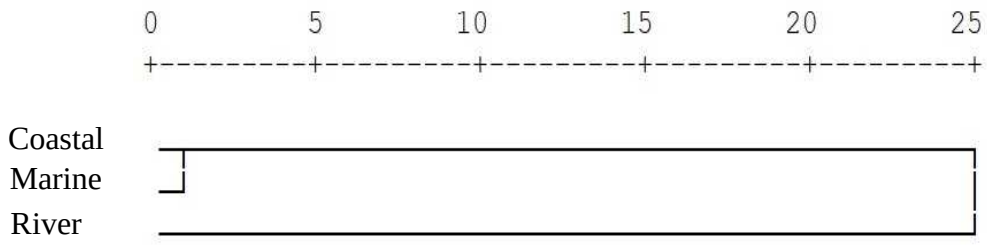

Figure 07: Dendrogram based on the morphometric and landmark distances of the coastal, marine and river samples of *T. ilisha.*

4.2. Comparison between *T. ilisha* and *T. toli*

4.2.1. Analysis of meristic counts

The number of dorsal fin rays (D 17/18; M_e =17), pelvic fin rays (V8; M_e=8), pectoral fin rays (P 14/15; M_e=15) and caudal fin rays (C 28/29; M_e=28), scales on lateral line (LL 40-41), scales on lateral transverse (LT 13-14) and the number of branchiostegeal rays (B= V) in *T. toli* were significantly (P<0.05) different from the *T. ilisha* in the Kruskal Wallis (H) test except anal fin rays (M_e=17). Besides univariate statistics (ANOVA) showed significant differences (P<0.01) between the two fish species (*T. ilisha* and *T. toli*) in case of caudal fin rays.

4.2.2. Analysis of morphometric and landmark distance measurements

Prior to analysis, correlation test between total length and adjusted morphometric characteristics were done for all data to confirm the removal of size effects. Out of fifteen morphometric characters, twelve characters [fork length (FL), pre-dorsal fin length (Pre-DFL), dorsal fin length (DFL), post-dorsal fin length (Post-DFL), pre-pelvic fin length (Pre-PvFL), pelvic fin length (PvFL), pre-pectoral fin length (PcFL), pectoral fin length (PcFL), anal fin length (AFL), caudal fin length (CFL), caudal peduncle length (CPL), least body depth (LBD)] showed significant differences in univariate analysis (ANOVA) between the populations of *T. ilisha* and *T. toli* in varying degrees (p<0.05 or p<0.01 or p<0.001) (Table 12).

Table 12: Univariate statistics (ANOVA) testing differences among samples from 15 morphometric measurements in *T. ilisha* and *T. toli*. Degree of significance were presented as *p<0.05, **p<0.01, ***p<0.001).

Morphometric characters	Wilks' Lambda	F value	Sig.
SL	0.998	0.046	0.831
FL	0.048	595.656	0.000***
Pre-DFL	0.041	696.546	0.000***
DFL	0.499	30.098	0.000***
Post-DFL	0.182	134.921	0.000***
Pre-PvFL	0.007	3970.950	0.000***
PvFL	0.030	953.827	0.000***
Pre-PcFL	0.036	795.143	0.000***

PcFL	0.285	75.159	0.000***
Pre-AFL	0.954	1.455	0.237
AFL	0.165	151.416	0.000***
CFL	0.871	4.440	0.044*
CPL	0.637	17.084	0.000***
HBD	0.989	0.348	0.559
LBD	0.056	506.605	0.000***

Univariate analysis (ANOVA) between populations of *T. ilisha* and *T. toli* using fourteen landmark distances showed significant differences in case of thirteen (1 to 2, 3 to 4, 4 to 5, 5 to 6, 6 to 7, 7 to 8, 8 to 1, 2 to 6, 2 to 7, 3 to 5, 3 to 6, 3 to 7, 4 to 6) truss measurements in varying degrees ($p<0.05$ or $p<0.01$ or $p<0.001$) (Table 13). Removal of size effects were also confirmed prior to analysis using correlation test between total length and the transformed truss-network characteristics.

Table 13: Univariate statistics (ANOVA) among samples from 14 truss measurements between *T. ilisha* and *T. toli*. Degree of varying effects were presented as *$p<0.05$, **$p<0.01$, ***$p<0.001$.

Landmark distance	Wilks' Lambda	F value	Sig.
A1	0.211	112.325	0.000***
B1	0.896	3.482	0.072
C1	0.247	91.484	0.000***
D1	0.026	1107.69	0.000***
E1	0.710	12.277	0.001**
F1	0.340	58.135	0.000***
G1	0.221	105.569	0.000***
H1	0.040	726.502	0.000***
I2	0.047	610.882	0.000***
J2	0.148	172.522	0.000***
K2	0.058	489.426	0.000***
L2	0.093	293.826	0.000***
M2	0.259	85.839	0.000***
N2	0.379	49.081	0.000***

4.2.3. Principal component analysis (PCA)

The significant traits resulted from univariate analysis (twelve morphometric measurements and thirteen truss measurements) were further used for principal component analysis (PCA). The morphometric characters with an eigen value above 1 were included and others were excluded in this analysis. In our present study, significant factors considered only those factors with loadings greater than 0.3. Principal Component Analysis (PCA) for the morphometric measurements of *T. ilisha* and *T. toli* showed that, the value of KMO for overall matrix was 0.906, and the Bartlett's Test of Sphericity was significant (P<0.01). The results of KMO and Bartlett's suggested that the sampled data was appropriate to proceed with a factor analysis procedure.

The PCA based on 12 morphometric measurements retained two components with eigen values>1, explaining 89.23% of the total variance. The first (PC1) and second (PC2) principal components accounted for 77.42% and 11.81% of the total variance respectively. All the twelve morphometric measurements had significant loadings on PC1 and two measurements [caudal fin length (CFL), caudal peduncle length (CPL)] were significant on PC2 (Table 14).

Table 14: Component loadings of the first two principal components derived from the morphometric measurements of *T. ilisha* and *T. toli.*

Morphometric Characters	Component	
	PC1	PC2
Fork Length (FL)	0.968	
Pre Dorsal fin length (Pre-DFL)	0.981	
Dorsal fin length (DFL)	-0.742	
Post Dorsal fin length (Post-DFL)	-0.901	
Pre Pelvic fin length (Pre-PvFL)	0.992	
Pelvic fin length (PvFL)	0.973	
Pre pectoral fin length (Pre-PcFL)	0.981	
Pectoral fin length (PcFL)	0.880	
Anal fin length (AFL)	-0.934	
Caudal fin Length (CFL)	-0.331	0.901

Caudal peduncle length (CPL)	-0.656	-0.650
Least body depth (LBD)	0.974	
Eigen-values	9.290	1.417
% of variance	77.421	11.811
Cumulative variance %	77.421	89.231

PCA for the landmark measurements of *T. ilisha* and *T. toli* revealed that, the value of KMO for overall matrix was 0.887 and the Bartlett's Test of sphericity was significant ($P<0.01$). The results of KMO and Bartlett's test suggested that the sampled data was appropriate to proceed with a factor analysis. The PCA based on 13 truss measurements retained two components with eigen values>1, explaining 88.29% of the total variance. The first (PC1) and second (PC2) principal components accounted for 78.603% and 9.691% of the total variance respectively. All the thirteen truss measurements had significant loadings on PC1 and the most significant loadings on PC2 were 2-6, 3-4, 3-5, 3-6, 4-6, 5-6 and 6-7 (Table 15).

Table 15: Component loadings of the first two principal components derived from the truss measurements of *T. ilisha* and *T. toli.*

Landmark distance	**Component**	
	PC1	**PC2**
A1 (1-2)	0.912	
C1 (3-4)	-0.866	0.444
D1 (4-5)	0.950	
E1 (5-6)	-0.660	0.332
F1 (6-7)	-0.692	0.429
G1 (7-8)	-0.936	
H1 (8-1)	0.983	
I2 (2-7)	0.983	
J2 (2-6)	0.907	0.337
K2 (3-7)	0.978	
L2 (3-6)	0.915	0.337
M2 (3-5)	-0.880	0.413
N2 (4-6)	0.788	0.516

Eigen-values	10.218	1.260
% of variance	78.603	9.691
Cumulative variance %	78.603	88.294

Based on PCA in both morphometric and landmark values it was clearly determined that scatter plots of specimens relating the first and second principal component (PC1 and PC2) revealed a visual differentiations between the two species. Dispersion in PCA plots showed a vast divergence in between *T. ilisha* and *T. toli* (Figure 08 and Figure 09).

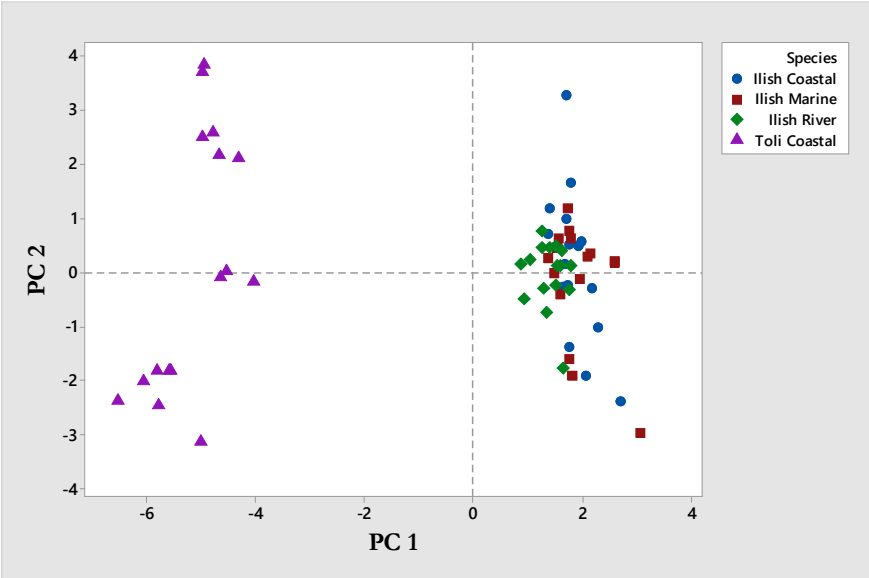

Figure 08: Scatterplot of the scores from PC1 and PC2 for morphometric characters of *T. ilisha* and *T. toli* collected from three different habitats of Bangladesh.

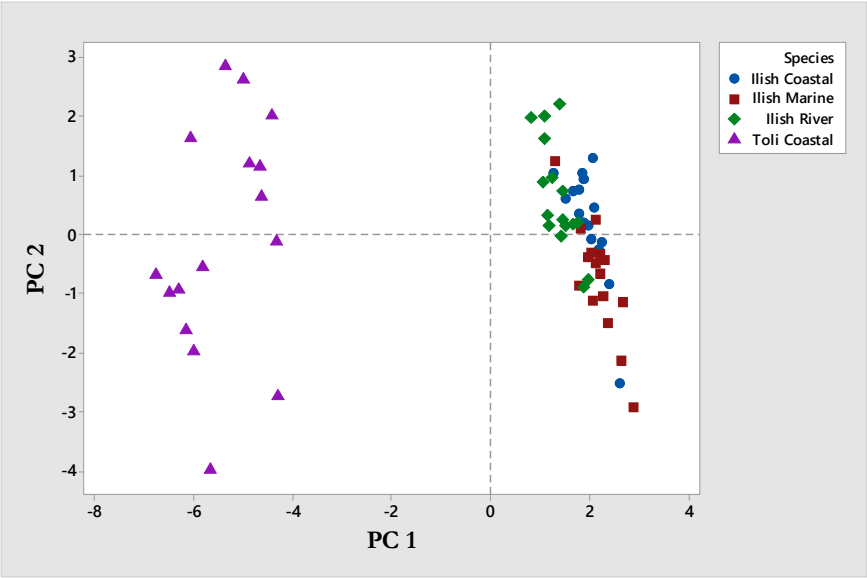

Figure 09: Scatterplot of the scores from PC1 and PC2 for truss measurement of *T. ilisha* and *T. toli* collected from three different habitats of Bangladesh.

Chapter-5

DISCUSSION

The phenotypic plasticity in fish is very high (Hossain et al., 2010) and the morphometric and meristic studies provide useful results for identifying fish stocks (Ihssen et al., 1981). In this study, morphometric and meristic characters with truss measurements have been used to analyze the potential differentiation of *Tenualosa sp.* populations collected from different habitats of Bangladesh. We used truss network system which is a powerful tool for identifying stocks of fish species (Turan, 2004). According to Dwivedi and Dubey (2012) the truss network is more useful and an effective strategies for describing the shapes, provides a better way of data collection, enables the data for the application in a diversified analytical tools in order to discriminate phenotypic stock in compared to that of traditional morphometric method because the configuration of the constructed landmarks covers the entire fish body with no loss of information and therefore it is more sensitive to change (Lim, 2008). This method has been also successfully utilized to differentiate and identify stock in many fish groups including the horse mackerel *Trachurus trachurus* (Murta et al., 2008); Japanese threadfin bream *Nemipterus japanicus* (Lim, 2008); Indian major carps (Hossain et al., 2010); mullet (Hossain et al., 2015); catfish (Parvej et al., 2014, Rahman et al., 2014); and gobies (Sabet and Anvarifer, 2013). To elucidate the differences, ANOVA (analysis of variance) and DFA (discriminant function analysis) with principal component analysis (PCA) were performed in this study.

Though no significant difference was observed among the populations of *T. ilisha* in case of meristic counts but highly significant morphometric differences were found among the coastal, marine and riverine populations of *T. ilisha*. Analysis of Variance (ANOVA) showed that out of 15 morphometric measurements, eight morphometric lengths [anal fin length (AFL), caudal peduncle length (CPL), highest body depth (HBD), least body depth (LBD), post-dorsal fin length (Post-DFL), pre-pectoral fin length (Pre-PcFL), pelvic fin length (PvFL), pre-pelvic fin length (Pre-PvFL)] were significantly different in varying degrees ($p<0.05$ or $p<0.01$ or $p<0.001$) among these three groups of populations of *T. ilisha*. Turan et al., (2004a); Hossain et al., (2015); Parvej et al., (2014); Hossain et al., (2010); Rahman et al., (2014) also found variations in morphological differences in diverse populations from different habitats

in *Liza abu*, *Rhinomugil corsula*, *Eutropiichthys vacha*, *Labeo calbasu* and *Heteropneustes fossilis* respectively.

The morphological variations among the populations of *T. ilisha* might be due to their separate geographical locations, high degree of existing environmental variation of their habitats or populations may be originated from different ancestors. Fish are very sensitive to environmental changes and quickly adapt themselves by changing their essential morphometrics with new environmental conditions (Allendorf et al., 1988). It is well known that, morphological characteristics can show high plasticity in response to differences in environmental conditions (Swain et al., 1991). Therefore, the distinctive environmental conditions of these habitats may underlie the morphological differentiation among the populations from different locations. Such kind of discrimination has been reported among six populations of *Capoeta capoeta gracilis* located in the Aras, Sefidrud, Shirud, Tonekabon, Haraz and Gorganrud river systems in Iran (Samaee et al., 2006). A similar study was conducted by Mir et al., (2013) and reported the variations among the *Labeo rohita* stocks of Ganga basin due to uncommon hydrological conditions such as differences in alkalinity, current pattern, temperatures, turbidity and the closeness among the stocks due to their similar habitat attributes and to environmental impacts. The environmental parameters, especially salinity in Tentulia and Meghna rivers were almost the same in comparison with Baleswar river. Dasgupta et al., (2014) reported that the salinity of Tentulia and Meghna river were 3.5 ppt and 6 ppt respectively while it was 0.6 ppt in Baleswar river which might be the possible cause for variation in *Labeo rohita*. Ferrito et al., (2007) stated that morphological discrimination in various populations was highly influenced by habitat differences.

Generally, fish show greater variances in morphological characters both within and between populations than any other vertebrates and are more vulnerable to environmentally induced morphological variations (Allendorf et al., 1987; Wimberger, 1992). As the phenotypic plasticity of fish is very high, they modify their physiology and behavior to adapt quickly to environmental changes which ultimately change their morphology (Stearns et al., 1983). Therefore, it might be impossible to detect small morphological differences in fish which are created due to small environmental differences by analyzing only gross morphometric and meristic

characters. For this constrains, truss network measurement method was implied in this research. In truss network, eight (1 to 2, 2 to 3, 3 to 4, 4 to 5, 6 to 7, 8 to 1, 2 to 6, 3 to 6) out of fourteen distances were significantly different ($p<0.05$ or <0.01 or <0.001) among the three populations of *T. ilisha*. Hossain et al., (2010) observed significant differences ($p<0.05$ or <0.001) in four of 22 truss network measurements in kalibaus (*Labeo calbasu*) populations collected from the Jamuna, the Halda and a hatchery in Bangladesh. The significant differences ($p<0.05$) were also found in 16 of 25 truss measurements in Anchovy (*Engraulisen crasicolus* L.) in Black, Aegean and Northeastern Mediterranean sea (Turan et al., 2004b). Parvej et al., (2014) found significant differences ($p<0.001$) in 4 of 17 morphometric traits and only 1 of 22 truss network measurements in *Eutropiichthys vacha* populations from Kaptai Lake, Meghna River and Tanguar Haor in Bangladesh.

Discriminant function analysis (DFA) could be a suitable method to differentiate different stocks of same species, which could be of concern to stock management programs (Karakousis et al., 1991). This discrimination was ensured by another multivariate analysis PCA (principal component analysis), where pictorial analysis of plotted PC1 and PC2 scores for every specimen was observed. Both discriminant function analysis (DFA) and principal component analysis (PCA) suggested that the river population of *T. ilisha* have high degree of phenotypic distinction from the coastal and marine habitat populations of *T. ilisha* in case of both morphometric characters and truss measurements. Scatter plotting from principal component analysis (PCA) based on both morphometric and truss measurements suggested that *T. ilisha* of river population was isolated from the coastal and marine habitats. This inter-population variation may be attributed due to separate geographical location as well as the environmental and physiological constrains like salinity, temperature, turbidity, water pressure, current flow and food availability experienced by each population (Allendorf, 1988; Swain et al., 1991; Wimberger, 2008). Konana et al., (2010) applied PCA on the populations of freshwater shrimp *Macrobrachium vollenhovenii* collecting from Côte d'Ivoire Rivers and reported notable morphometric variations due to distance and geographical location of rivers. Paugy and Lévêque (1999) also showed that populations of same species originating from different geographical areas were morphologically different.

In this study, DF analysis was conducted to determine the variations among the three stocks of *T. ilisha*. The canonical discriminant functions in DFA showed an overlapped in the coastal and marine stocks of *T. ilisha* whereas the river stock is totally isolated. In case of morphometric measurement, the first DF accounted for much more (89.8%) of the among group variability than did the second DF (10.2%) and in case of truss measurements the first DF accounted for much more (87.4%) of the among group variability than did the second DF (12.6%). From this both observations, it was obvious that the second DF explained much less of the variance than did the first DF. Therefore, the second DF was much less informative in explaining differences among the stocks.

The dendogram that was drawn based on the landmark distances and morphological examinations among groups of centroids of *T. ilisha* populations collected from three different habitats employed two main clusters: the coastal and marine samples in one and the river group in another. This demonstrated a high degree of separation of the river population from the marine and coastal habitats. These differences among the habitats might be happened due to environmental as well as genetic variations. A dendrogram based on data of the morphological characters shown in the population of Japanese charr, *Salvelinus leucomaenis* (Nakamura, 2003); Mullet, *Rhinomugil corsula* (Hossain et al., 2015); *Eutropiichthys vacha* (Parvej et al., 2014); *Labeo calbasu* (Hossain et al., 2010) from different habitats revealed separate stocks were possibly due to environmental condition, separate habitat as well as genetic variations.

Besides, this study also demonstrates comparative morphological differences between *T. ilisha* and *T. toli* collected from coastal water habitat. We found significant differences in case of morphometric, meristic characters and truss measurements between two species of *Tenualosa*. Meristic characteristics such as dorsal fin rays, pelvic fin rays, pectoral fin rays and caudal fin rays, scales on lateral line, scales on lateral transverse and the number of branchiostegeal rays in *T. toli* were significantly ($P<0.05$) different from the *T. ilisha*.

Twelve morphometric measurement [fork length (FL), pre-dorsal fin length (PreDFL), dorsal fin length (DFL), post-dorsal fin length (Post-DFL), pre-pelvic fin length (Pre-PvFL), pelvic fin length (PvFL), pre-pectoral fin length (PcFL), pectoral fin length (PcFL), anal fin length (AFL), caudal fin length (CFL), caudal peduncle

length (CPL), least body depth (LBD)] out of fifteen morphometric characters and thirteen truss measurements (1 to 2, 3 to 4, 4 to 5, 5 to 6, 6 to 7, 7 to 8, 8 to 1, 2 to 6, 2 to 7, 3 to 5, 3 to 6, 3 to 7, 4 to 6) out of 14 networking showed significant differences in univariate analysis (ANOVA) between the populations of *T. ilisha* and *T. toli* in varying degrees ($p<0.05$ or $p<0.01$ or $p<0.001$). The present study has uncovered some morphological (i.e., morphometric and meristic) variations between the *T. ilisha* and *T. toli* using multivariate techniques as reported for other marine vertebrates and invertebrates also (Fridriksson, 1958; Boetius, 1980; Pierce et al., 1994a; 1994b; Tudela, 1999; Bolles and Begg, 2000). The comparative study of two types of palla (*T. ilisha*) collected from river Indus revealed significant intertype differences in six morphometric measurements (total length, standard length, fork length, head length, eye diameter and girth) and seven meristic characters (total number of scutes, pre pelvic scutes, post pelvic scutes, dorsal fin rays, pectoral fin rays, pelvic fin rays and anal fin rays) reported by Narejo et al., (2008).

Wilk's Lambda values calculated by stepwise discriminant analysis showed greater values in both morphometric and truss- networking measurements. The Wilk's Lambda values were greater than 0.1 in eighteen cases of the total measurement in these two species which indicates that there was high degree of variations in between two species. Yakubu and Okunsebor (2011) showed significant morphological differences between *Oreochromis niloticus* and *Lates niloticus* where they found the values of Wilk's Lamba was greater than 0.1 in most measurement.

In both case of morphometric and landmark values *T. ilisha* and *T. toli* showed high degree of variations based on the PCA. The PCA with eigen values >1, shows 89.23% of the total variance. The PC1 and PC2 was 77.42% and 11.81% for the morphometric measurement and the truss measurements revealed 88.29% of the total variance and 78.603% and 9.691% for PC1 and PC2 respectively. This data clearly confirmed significant differences between these two species. Yakubu and Okunsebor (2011) found morphometric difference between two Nigerian fish species (*Oreochromis niloticus* and *Lates niloticus*) using principal components and discriminant analysis. Moreover scatter plotting from PCA revealed that, *T. toli* exhibited higher degree of variations from the marine and river habitat of *T. ilisha* in case of both morphometric characters and truss measurements. Pillay et al., (1962)

reported two separate populations of *T. ilisha* population from studying in rivers and coastal areas in India. Gosh et al., (1968) identified three varieties of *T. ilisha*, denoted as sub-populations (slender, broad and broader) from a part of the Gangetic system between Allahabad and Buxar. While Quddus et al., (1984a, b) reported meristic and morphometric difference and comparison of age and growth of two types of *T. ilisha* from Bangladesh waters.

From the above demonstration it is clearly revealed that, the river populations of *T. ilisha* is morphologically separated than the coastal or marine populations and the *T. toli* is also far more different from the *T. ilisha.*

Chapter-6

CONCLUSIONS

Fish and fisheries are the integral parts of Bangladesh. In Bangladesh, fish plays a central role in dietary patterns, livelihoods and culture. Fish is the most commonly consumed animal-source food across all population groups. But this sector is facing an increasing threat due to over fishing, habitat degradation, pollution in the rivers and the indiscriminate use of agrochemicals, introduction of exotic species, lack of suitable habitat, decreased fecundity and so on. To fulfill the demand of its increasing pressure, sustainable and efficient stock management is necessary.

For proper conservation and management of any population, it is needed to know about their biology and population structure. It is also essential to select genetically superior stocks along with better features for, the both successful aquaculture and open-water management. This study has provided important morphological information that can be used to differentiate this *Tenualosa* sp. more precisely among groups and species. This study was not designed to investigate the actual cause due to which morphological variation occurs in different stocks of same species and to determine whether the morphological variations are environmentally induced or due to genetic factors or both. Investigation to this regard may be initiated on the basis of the present findings. The findings of the study would serve as primary information of stock management and enable efficient management strategies for the distinct stocks of *Tenualosa* sp. populations in order to make its fishery sustainable and develop appropriate conservation plans in near future. The authors hope that the information obtained from the present study will be helpful for fisheries, biologists, and taxonomist concerned with these two fascinating fish species

Chapter-7

RECOMMENDATIONS AND FUTURE PERSPECTIVES

Since the identification of populations and their connectivity between each other is a major point for sustainable management and conservation of species, the use of morphological characters as baseline information appears promising in this region. The present study affords elementary information about the variation of *Tenualosa sp.* populations in different water habitats of Bangladesh and it recommends that use of morphometric characters and truss measurements generate reliable information for stock discrimination of *Tenualosa sp.*

However, present study had some limitations in terms of limited number of individuals and populations. The result of the present study might be used as a guideline for further study with more samples and for more clarification and conformation and finally following points might be considered for sustaining *Tenualosa* species in Bangladesh.

a) Systematic study might be conducted with more individuals from more different locations.

b) DNA level work (RAPD, RFLP, microsatellite etc.) might be conducted for more clarification and conformation of genetic variation.

c) Breeding ground of *Tenualosa* species should be protected.

d) Sperm cryopreservation of *Tenualosa* species should be approached for both conservation and aquaculture.

f) Finally proper conservation plans should be formulated.

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APPENDICES

Appendix I. Comparative studies on *T. ilisha* collected from three (03) different habitats for morphometric characters and truss measurements

Group Statistics for Morphometric Characters

Species	Characters	Mean	Std. Deviation	Valid N (listwise)	
				Unweighted	Weighted
Tenualosa ilisha (Coastal)	SL	20.3589	0.4723	16	16.000
	FL	26.1829	0.3407	16	16.000
	Pre-DFL	9.7434	0.2185	16	16.000
	DFL	2.5100	0.1456	16	16.000
	Post-DFL	10.7636	0.3609	16	16.000
	Pre-PvFL	12.1013	0.1442	16	16.000
	PvFL	0.6839	0.0316	16	16.000
	Pre-PcFL	5.2983	0.2153	16	16.000
	PcFL	0.9701	0.0592	16	16.000
	Pre-AFL	14.7756	0.2653	16	16.000
	AFL	2.0907	0.0978	16	16.000
	CFL	3.6797	0.1850	16	16.000
	CPL	1.5162	0.0528	16	16.000
	HBD	3.3259	0.1565	16	16.000
	LBD	2.4838	0.1259	16	16.000
Tenualosa ilisha (Marine)	SL	20.4420	0.2075	16	16.000
	FL	26.2807	0.3335	16	16.000
	Pre-DFL	9.6937	0.1713	16	16.000
	DFL	2.4599	0.1257	16	16.000
	Post-DFL	10.9484	0.1854	16	16.000
	Pre-PvFL	12.0504	0.1550	16	16.000
	PvFL	0.7157	0.0254	16	16.000
	Pre-PcFL	5.2662	0.1833	16	16.000
	PcFL	0.9789	0.0534	16	16.000
	Pre-AFL	14.9135	0.3620	16	16.000
	AFL	2.0782	0.0928	16	16.000
	CFL	3.6492	0.1720	16	16.000
	CPL	1.5555	0.0417	16	16.000
	HBD	3.4414	0.1189	16	16.000
	LBD	2.4840	0.1086	16	16.000
Tenualosa ilisha (River)	SL	20.4811	0.1729	16	16.000
	FL	26.0228	0.2380	16	16.000
	Pre-DFL	9.7039	0.1288	16	16.000
	DFL	2.4314	0.0736	16	16.000
	Post-DFL	10.9818	0.1526	16	16.000
	Pre-PvFL	11.9062	0.3139	16	16.000
	PvFL	0.6562	0.0216	16	16.000
	Pre-PcFL	5.1042	0.1920	16	16.000
	PcFL	0.9433	0.0272	16	16.000
	Pre-AFL	14.9561	0.2000	16	16.000

	AFL	1.9519	0.0851	16	16.000
	CFL	3.6921	0.0844	16	16.000
	CPL	1.6004	0.0506	16	16.000
	HBD	3.5408	0.0955	16	16.000
	LBD	2.2326	0.1073	16	16.000
Total	SL	20.4273	0.3116	48	48.000
	FL	26.1622	0.3196	48	48.000
	Pre-DFL	9.7137	0.1743	48	48.000
	DFL	2.4671	0.1209	48	48.000
	Post-DFL	10.8979	0.2634	48	48.000
	Pre-PvFL	12.0193	0.2296	48	48.000
	PvFL	0.6853	0.0357	48	48.000
	Pre-PcFL	5.2229	0.2113	48	48.000
	PcFL	0.9641	0.0500	48	48.000
	Pre-AFL	14.8817	0.2883	48	48.000
	AFL	2.0403	0.1101	48	48.000
	CFL	3.6732	0.1516	48	48.000
	CPL	1.5574	0.0589	48	48.000
	HBD	3.4361	0.1520	48	48.000
	LBD	2.4001	0.1638	48	48.000

Tests of Equality of Group Means

Characters	Wilks' Lambda	F	df1	df2	Sig.
SL	.973	.632	2	45	.536
FL	.887	2.865	2	45	.067
Pre-DFL	.985	.353	2	45	.704
DFL	.926	1.793	2	45	.178
Post-DFL	.865	3.526	2	45	.038
Pre-PvFL	.868	3.425	2	45	.041
PvFL	.528	20.112	2	45	.000
Pre-PcFL	.835	4.450	2	45	.017
PcFL	.906	2.324	2	45	.110
Pre-AFL	.927	1.768	2	45	.182
AFL	.669	11.123	2	45	.000
CFL	.986	.330	2	45	.721
CPL	.652	11.998	2	45	.000
HBD	.660	11.612	2	45	.000
LBD	.466	25.781	2	45	.000

Eigenvalues

Function	Eigenvalue	% of Variance	Cumulative %	Canonical Correlation
1	6.502[a]	89.8	89.8	.931
2	.740[a]	10.2	100.0	.652

a. First 2 canonical discriminant functions were used in the analysis.

Standardized Canonical Discriminant Function Coefficients

Characters	Function	
	1	2
SL	.575	.071
FL	-.761	.194
Pre-DFL	.365	-.029
DFL	.640	-.034
Post-DFL	.095	.380
Pre-PvFL	-.338	-.068
PvFL	-.219	.780
Pre-PcFL	.013	.194
PcFL	.392	.208
Pre-AFL	.296	.053
AFL	-.287	.036
CFL	.153	-.116
CPL	.225	.303
HBD	1.082	.431
LBD	-1.017	-.145

Unstandardized Canonical Discriminant Function Coefficients

Characters	Function	
	1	2
SL	1.829	.225
FL	-2.474	.631
Pre-DFL	2.063	-.166
DFL	5.384	-.285
Post-DFL	.380	1.517
Pre-PvFL	-1.545	-.309
PvFL	-8.244	29.369
Pre-PcFL	.064	.982
PcFL	8.059	4.282
Pre-AFL	1.043	.187
AFL	-3.112	.394
CFL	.992	-.756
CPL	4.614	6.234
HBD	8.575	3.412
LBD	-8.896	-1.268
(Constant)	-22.089	-80.197

Structure Matrix

Characters	Function	
	1	2
LBD	-.416*	.169
AFL	-.275*	.044
Pre-PcFL	-.174*	-.010
Pre-PvFL	-.152*	-.054
PvFL	-.284	.706*
HBD	.256	.349*
Post-DFL	.111	.320*
CPL	.269	.291*
FL	-.121	.206*

Pre-AFL	.085	.206*
DFL	-.095	-.170*
PcFL	-.117	.137*
Pre-DFL	-.022	-.130*
CFL	.030	-.110*
SL	.055	.109*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
Variables ordered by absolute size of correlation within function.
*. Largest absolute correlation between each variable and any discriminant function

Group Statistics Group Statistics for Truss Measurements

Species	Landmarrk distances	Mean	Std. Deviation	Valid N (listwise)	
				Unweighted	Weighted
Tenualosa ilisha (Coastal)	A1	8.3365	0.2346	16	16.000
	B1	2.4266	0.1350	16	16.000
	C1	6.6854	0.1917	16	16.000
	D1	3.0167	0.1336	16	16.000
	E1	1.3908	0.0757	16	16.000
	F1	5.6811	0.1781	16	16.000
	G1	3.0361	0.1633	16	16.000
	H1	6.2926	0.1874	16	16.000
	I2	5.3273	0.2176	16	16.000
	J2	11.2455	0.7099	16	16.000
	K2	4.8478	0.1749	16	16.000
	L2	8.0872	0.2618	16	16.000
	M2	7.5142	0.2623	16	16.000
	N2	2.6438	0.1060	16	16.000
Tenualosa ilisha (Marine)	A1	8.7290	0.1458	16	16.000
	B1	2.5200	0.1334	16	16.000
	C1	6.9448	0.2245	16	16.000
	D1	3.1340	0.1347	16	16.000
	E1	1.4190	0.0356	16	16.000
	F1	5.8921	0.1869	16	16.000
	G1	3.1316	0.1629	16	16.000
	H1	6.5253	0.2124	16	16.000
	I2	5.2985	0.1896	16	16.000
	J2	11.4706	0.4636	16	16.000
	K2	4.9041	0.1881	16	16.000
	L2	8.3286	0.3505	16	16.000
	M2	7.6721	0.2801	16	16.000
	N2	2.7040	0.1105	16	16.000
Tenualosa ilisha (River)	A1	8.4932	0.2748	16	16.000
	B1	2.3214	0.0811	16	16.000
	C1	6.7730	0.2893	16	16.000
	D1	2.6289	0.1485	16	16.000
	E1	1.4418	0.0792	16	16.000
	F1	5.8803	0.2701	16	16.000

	G1	3.0593	0.1172	16	16.000
	H1	6.3270	0.2744	16	16.000
	I2	5.3253	0.2375	16	16.000
	J2	10.6752	0.4689	16	16.000
	K2	4.9416	0.2277	16	16.000
	L2	7.8151	0.4268	16	16.000
	M2	7.5069	0.2772	16	16.000
	N2	2.6181	0.1233	16	16.000
Total	A1	8.5196	0.2739	48	48.000
	B1	2.4227	0.1425	48	48.000
	C1	6.8011	0.2576	48	48.000
	D1	2.9266	0.2571	48	48.000
	E1	1.4172	0.0684	48	48.000
	F1	5.8179	0.2324	48	48.000
	G1	3.0757	0.1518	48	48.000
	H1	6.3816	0.2457	48	48.000
	I2	5.3170	0.2116	48	48.000
	J2	11.1306	0.6434	48	48.000
	K2	4.8978	0.1978	48	48.000
	L2	8.0770	0.4051	48	48.000
	M2	7.5646	0.2783	48	48.000
	N2	2.6553	0.1169	48	48.000

Tests of Equality of Group Means

	Wilks' Lambda	F	df1	df2	Sig.
A1	.646	12.333	2	45	.000
B1	.669	11.110	2	45	.000
C1	.822	4.887	2	45	.012
D1	.280	57.751	2	45	.000
E1	.905	2.354	2	45	.107
F1	.823	4.833	2	45	.013
G1	.927	1.779	2	45	.180
H1	.822	4.865	2	45	.012
I2	.996	.089	2	45	.915
J2	.724	8.585	2	45	.001
K2	.961	.907	2	45	.411
L2	.726	8.479	2	45	.001
M2	.923	1.865	2	45	.167
N2	.903	2.411	2	45	.101

Eigenvalues

Function	Eigenvalue	% of Variance	Cumulative %	Canonical Correlation
1	8.992[a]	87.4	87.4	.949
2	1.296[a]	12.6	100.0	.751

a. First 2 canonical discriminant functions were used in the analysis.

Standardized Canonical Discriminant Function Coefficients

Landmark distances	Function	
	1	2
A1	.567	.524
B1	-.387	.647
C1	.701	1.209
D1	-1.400	.262
E1	.499	.635
F1	.583	.532
G1	.311	.349
H1	-.270	.055
I2	-.034	-.240
J2	.471	-.545
K2	1.050	-.150
L2	-.454	.033
M2	-.683	-1.236
N2	-.651	-.766

Unstandardized Canonical Discriminant Function Coefficients

Landmark distances	Function	
	1	2
A1	2.522	2.328
B1	-3.247	5.427
C1	2.936	5.066
D1	-10.062	1.885
E1	7.492	9.534
F1	2.702	2.463
G1	2.080	2.337
H1	-1.186	.241
I2	-.158	-1.113
J2	.843	-.974
K2	5.296	-.759
L2	-1.286	.094
M2	-2.499	-4.522
N2	-5.737	-6.743
(Constant)	-19.261	-37.694

Structure Matrix

Landmark distances	Function	
	1	2
D1	-.513*	.391
J2	-.194*	.179
A1	-.014	.649*
C1	-.020	.406*
H1	-.050	.387*
F1	.078	.351*
B1	-.200	.321*
L2	-.175	.281*
G1	-.022	.240*
M2	-.046	.221*
N2	-.075	.208*

E1	.093	.143*
K2	.056	.097*
I2	.008	-.051*

Pooled within-groups correlations between discriminating variables and standardized canonical discriminant functions
Variables ordered by absolute size of correlation within function.
*. Largest absolute correlation between each variable and any discriminant function

KMO and Bartlett's Test for Morphometric Characters

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		.685
Bartlett's Test of Sphericity	Approx. Chi-Square	82.964
	Df	28
	Sig.	.000

Total Variance Explained in case of Morphometric Characters

	Initial Eigenvalues			Extraction Sums of Squared Loadings		
Component	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	3.065	38.306	38.306	3.065	38.306	38.306
2	1.106	13.826	52.133	1.106	13.826	52.133
3	.888	11.100	63.232			
4	.813	10.160	73.392			
5	.752	9.394	82.786			
6	.631	7.884	90.670			
7	.479	5.989	96.659			
8	.267	3.341	100.000			

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component	
Morphometric characters	PC1	PC2
Post-DFL	-.583	.529
Pre-PvFL	.543	
PvFL	.564	.563
Pre-PcFL	.577	
AFL	.788	
CPL	-.715	
HBD	-.545	.347
LBD	.592	.549

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

KMO and Bartlett's Test for Truss Measurements

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		.716
Bartlett's Test of Sphericity	Approx. Chi-Square	275.770
	Df	28
	Sig.	.000

Total Variance Explained in case of Truss Measurements

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	4.489	56.118	56.118	4.489	56.118	56.118
2	1.228	15.352	71.470	1.228	15.352	71.470
3	.870	10.878	82.348			
4	.651	8.139	90.487			
5	.319	3.992	94.479			
6	.274	3.426	97.905			
7	.108	1.345	99.250			
8	.060	.750	100.000			

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component	
	PC1	PC2
A1	.612	.502
B1	.773	-.317
C1	.750	
D1	.747	-.458
F1	.615	.563
H1	.740	.393
J2	.862	-.377
L2	.853	

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

Appendix II. Comparison between *T. ilisha* and *T. toil* based on morphometric characters and truss measurements

Group Statistics for Morphometric Characters

Species	Morphometric characters	Mean	Std. Deviation	Valid N (listwise)	
				Unweighted	Weighted
Tenualosa ilisha	SL	20.3589	0.4723	16	16.000
	FL	26.1829	0.3407	16	16.000
	Pre-DFL	9.7434	0.2185	16	16.000
	DFL	2.5100	0.1456	16	16.000
	Post-DFL	10.7636	0.3609	16	16.000
	Pre-PvFL	12.1013	0.1442	16	16.000
	PvFL	0.6839	0.0316	16	16.000
	Pre-PcFL	5.2983	0.2153	16	16.000
	PcFL	0.9701	0.0592	16	16.000
	Pre-AFL	14.7756	0.2653	16	16.000
	AFL	2.0907	0.0978	16	16.000
	CFL	3.6797	0.1850	16	16.000
	CPL	1.5162	0.0528	16	16.000
	HBD	3.3259	0.1565	16	16.000
	LBD	2.4838	0.1259	16	16.000
Tenualosa toil	SL	20.3891	0.3022	16	16.000
	FL	22.6960	0.4587	16	16.000
	Pre-DFL	7.88856	0.1768	16	16.000
	DFL	2.8377	0.1890	16	16.000
	Post-DFL	11.9762	0.2099	16	16.000
	Pre-PvFL	8.5706	0.1715	16	16.000
	PvFL	0.4018	0.0183	16	16.000
	Pre-PcFL	3.2557	0.1938	16	16.000
	PcFL	0.7918	0.0571	16	16.000
	Pre-AFL	14.8961	0.2985	16	16.000
	AFL	2.6877	0.1675	16	16.000
	CFL	3.8363	0.2337	16	16.000
	CPL	1.8416	0.3104	16	16.000
	HBD	3.3789	0.3233	16	16.000
	LBD	1.6736	0.0696	16	16.000
Total	SL	20.3740	0.3903	32	32.000
	FL	24.4395	1.8154	32	32.000
	Pre-DFL	8.8161	0.9623	32	32.000
	DFL	2.6738	0.2352	32	32.000
	Post-DFL	11.3699	0.6810	32	32.000
	Pre-PvFL	10.3359	1.8003	32	32.000
	PvFL	0.5429	0.1455	32	32.000
	Pre-PcFL	4.2770	1.0578	32	32.000
	PcFL	.8810	0.1071	32	32.000
	Pre-AFL	14.83588	0.2844	32	32.000
	AFL	2.3892	0.3319	32	32.000
	CFL	3.7582	0.2223	32	32.000

	CPL	1.6789	0.2744	32	32.000
	HBD	3.35249	0.2513	32	32.000
	LBD	2.0787	0.4235	32	32.000

Tests of Equality of Group Means in case of Morphometric Characters

Characters	Wilks' Lambda	F	df1	df2	Sig.
SL	.998	.046	1	30	.831
FL	.048	595.656	1	30	.000
Pre-DFL	.041	696.546	1	30	.000
DFL	.499	30.098	1	30	.000
Post-DFL	.182	134.921	1	30	.000
Pre-PvFL	.007	3970.950	1	30	.000
PvFL	.030	953.827	1	30	.000
Pre-PcFL	.036	795.143	1	30	.000
PcFL	.285	75.159	1	30	.000
Pre-AFL	.954	1.455	1	30	.237
AFL	.165	151.416	1	30	.000
CFL	.871	4.440	1	30	.044
CPL	.637	17.084	1	30	.000
HBD	.989	.348	1	30	.559
LBD	.056	506.605	1	30	.000

Group Statistics for Truss Measurements

Species		Mean	Std. Deviation	Valid N (listwise)	
				Unweighted	Weighted
Tenualosa ilisha	A1	8.3365	0.2346	16	16.000
	B1	2.4266	0.1350	16	16.000
	C1	6.6854	0.1917	16	16.000
	D1	3.0167	0.1336	16	16.000
	E1	1.3908	0.0757	16	16.000
	F1	5.6811	0.1781	16	16.000
	G1	3.0361	0.1633	16	16.000
	H1	6.2926	0.1874	16	16.000
	I2	5.3273	0.2176	16	16.000
	J2	11.2456	0.7099	16	16.000
	K2	4.8478	0.1749	16	16.000
	L2	8.0872	0.2618	16	16.000
	M2	7.5142	0.2623	16	16.000
	N2	2.6438	0.1060	16	16.000
Tenualosa toli	A1	7.0395	0.4296	16	16.000
	B1	2.3105	0.2089	16	16.000
	C1	8.2331	0.6181	16	16.000
	D1	1.5948	0.1065	16	16.000
	E1	1.7993	0.4601	16	16.000
	F1	6.4564	0.3656	16	16.000
	G1	5.8673	1.0900	16	16.000
	H1	3.5748	0.3571	16	16.000
	I2	2.9289	0.3213	16	16.000

	J2	8.3945	0.4995	16	16.000
	K2	2.8935	0.3069	16	16.000
	L2	6.1730	0.3619	16	16.000
	M2	9.1865	0.6726	16	16.000
	N2	2.1897	0.2365	16	16.000
Total	A1	7.6880	0.7416	32	32.000
	B1	2.3685	0.1827	32	32.000
	C1	7.4593	0.9060	32	32.000
	D1	2.3058	0.7320	32	32.000
	E1	1.5951	0.3850	32	32.000
	F1	6.0688	0.4848	32	32.000
	G1	4.4517	1.6292	32	32.000
	H1	4.9337	1.4088	32	32.000
	I2	4.1281	1.2479	32	32.000
	J2	9.8202	1.5689	32	32.000
	K2	3.8707	1.0227	32	32.000
	L2	7.1301	1.0208	32	32.000
	M2	8.3504	0.9868	32	32.000
	N2	2.4168	0.2927	32	32.000

Tests of Equality of Group Means in case of Truss Measurements

	Wilks' Lambda	F	df1	df2	Sig.
A1	.211	112.325	1	30	.000
B1	.896	3.482	1	30	.072
C1	.247	91.484	1	30	.000
D1	.026	1107.697	1	30	.000
E1	.710	12.277	1	30	.001
F1	.340	58.135	1	30	.000
G1	.221	105.569	1	30	.000
H1	.040	726.502	1	30	.000
I2	.047	610.882	1	30	.000
J2	.148	172.522	1	30	.000
K2	.058	489.426	1	30	.000
L2	.093	293.826	1	30	.000
M2	.259	85.839	1	30	.000
N2	.379	49.081	1	30	.000

KMO and Bartlett's Test for Morphometric Characters

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		.906
Bartlett's Test of Sphericity	Approx. Chi-Square	682.411
	Df	66
	Sig.	.000

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	9.290	77.421	77.421	9.290	77.421	77.421
2	1.417	11.811	89.231	1.417	11.811	89.231
3	.545	4.542	93.773			
4	.250	2.087	95.860			
5	.202	1.682	97.543			
6	.129	1.079	98.621			
7	.064	.531	99.152			
8	.037	.308	99.460			
9	.025	.205	99.665			
10	.022	.180	99.845			
11	.012	.098	99.943			
12	.007	.057	100.000			

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component	
	PC1	PC2
FL	.968	
Pre-DFL	.981	
DFL	-.742	
Post-DFL	-.901	
Pre-PvFL	.992	
PvFL	.973	
Pre-PcFL	.981	
PcFL	.880	
AFL	-.934	
CFL	-.331	.901
CPL	-.656	-.650
LBD	.974	

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

KMO and Bartlett's Test for Truss Measurements

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		.887
Bartlett's Test of Sphericity	Approx. Chi-Square	1664.598
	Df	78
	Sig.	.000

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	10.218	78.603	78.603	10.218	78.603	78.603
2	1.260	9.691	88.294	1.260	9.691	88.294
3	.677	5.209	93.503			
4	.315	2.427	95.929			

5	.175	1.342	97.272			
6	.106	.819	98.091			
7	.077	.593	98.683			
8	.068	.525	99.208			
9	.042	.319	99.528			
10	.028	.217	99.744			
11	.016	.126	99.870			
12	.012	.089	99.959			
13	.005	.041	100.000			

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component	
	PC1	PC2
A1	.912	
C1	-.866	.444
D1	.950	
E1	-.660	.332
F1	-.692	.429
G1	-.936	
H1	.983	
I2	.983	
J2	.907	.337
K2	.978	
L2	.915	.337
M2	-.880	.413
N2	.788	.516

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

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