

Morphometric study of *Labeo nandina*, the fresh water endangered cyprinid from Brahmaputra River, Assam and a comparative analysis with four locally available *Labeo* species

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Abstract

Labeo nandina is an interesting component of the fish diversity of North-East India. It is an endangered cyprinid of the Brahmaputra river system. Due to its polyphyletic origin, this species has nomenclatural and taxonomic ambiguity. Individuals of the same genus living in variety of habitats may be subjected to different environmental conditions. As a result they may adapt to local conditions in those habitats which include changes in morphology from the common phenotype. 10 morphometric parameters like total length (TL), standard length (SL), head length (HL), eye diameter (ED) etc were taken for the morphometric study. Morphometric study was done on *Labeo nandina* and compared along with four locally available species of *Labeo*, viz *Labeo rohita*, *Labeo gonius*, *Labeo calbasu*, *Labeo bata*. The quantitative data were then analyzed by descriptive statistics obtaining dendrogram using SPSS version 16 software. The dendrogram thus obtained was used for a comparative analysis of *Labeo nandina* with the four other *Labeo* species mentioned above. The results showed specific morphological divergence among the species. From the study it was found that *Labeo nandina* has the highest eye diameter (1.64 cm) and *Labeo bata* has the lowest eye diameter (0.9cm). The pre orbital length of *Labeo nandina* is highest (2.7cm) than all the other *Labeo* species taken. The dendrogram obtained from the analysis of the average measurements of morphometric characters of *Labeo nandina* with the four locally available species taken, showed three major clusters. *Labeo rohita*, *Labeo calbasu*, *Labeo gonius* formed one cluster, *Labeo bata* formed another cluster with the above clustered species and finally, *Labeo nandina* formed the third cluster with the species of the above two clusters. *Labeo bata* was found to form the closest cluster with *Labeo nandina* than the species of other cluster formed. This result shows that *Labeo nandina*

has a greater metric similarity with *Labeo bata*. Next to it *L. rohita*, *L. calbasu* and *L. gonius* shows almost equal similarity with *Labeo nandina*, in terms of metric characters.

Keywords: Labeo, Taxonomy, Morphometric, Cluster

Running Title : Barman and Sharma

Introduction:

Morphometrics is the study of shape variation and its covariation with other variables (Bookstein 1991; Dryden and Mardia 1998). This approach, called traditional morphometrics (Marcus 1990; Reyment 1991) or multivariate morphometrics (Blackith and Reyment 1971) consisted of applying multivariate statistical analyses to sets of quantitative variables such as length, width, height etc.

Morphometrics refers to the quantitative analysis of form, a concept that encompasses size and shape. Morphometrics involves measuring the length of, or distance between physical features. Morphometric analyses are commonly performed on organisms, and are useful in analyzing their fossil record, the impact of mutation on shape, developmental changes in form, co-variances between ecological factors and shape, deduce something of their ontogeny, function or evolutionary relationships. A major objective of morphometrics is to statistically test hypothesis about the factors that effect shape. When combined with multivariate statistical procedures, they offer the most powerful tool for testing and graphically displaying differences in shape (Loy *et al.*, 1993; Rohlf and Marcus, 1993; Rohlf *et al.*, 1996).

The morphometric technique is an important technique that is frequently applied to detect species and population differences. It involves measuring distances between homologous landmarks over the entire length on the surface of the fish. It is able to detect differences in overall shape and size of individuals among species as well as populations. Both factors are known to be influenced genetically and environmentally. Morphometric study is usually based on the measurements of the fish taking into consideration different parameters like head length (HL), standard length (SL), total length (TL). Every species show different morphometric measurements and this in turn shows that each species is different from that of the other, morphologically.

Nomenclature ambiguity of *Labeo nandina*:

The various synonyms of *Labeo nandina* (Hamilton, 1822) given by the different authors are:

- (i) *Cyprinus nandina* Hamilton-Buchanan, 1822, *Fishes of Ganges*: 300, 388, pl.84 (type-locality: Mahananda river).
- (ii) *Labeo nandina*: Day, 1877, *Fishes of India*: 535, pl. 126, fig. 1; Day, 1889, *Fauna Br. India*, Fishes, 1: 258;

Labeo nandina var. *macronotus* (McClelland) Day, 1877, *Fishes of India*: 535, pl. 126, fig. 2.

Cirrhinus macronotus (McClelland,1839).

Rohita nandina (Hamilton,1822).

Common Names:

Nandi labeo ... English

Nandani ... Assam; Nandi ... West Bengal

Materials and Methods:

Materials:

Experimental animals:

In the present study on various aspects the animals used are *Labeo rohita*, *Labeo gonius*, *Labeo bata*, *Labeo calbasu* and *Labeo nandina*.

Site of collection and preservation:

Fish samples comprising 5 species viz *Labeo nandina*, *Labeo rohita*, *Labeo bata*, *Labeo gonius*, *Labeo calbasu* were collected from different localities. *Labeo nandina* was collected from some rivers Morigaon wetland. *Labeo bata* and *Labeo gonius* were collected from Barpeta

(market) and from Guwahati (market). *Labeo rohita* and *Labeo calbasu* were collected from Guwahati (market). Altogether 160-165 individuals were collected of each species in each season.. Digital photographs of all the samples were taken and stored at ice cold 80-90% ethanol (Merck) as soon as the samples were collected.

S1 = Morigaon

S2 = Barpeta

S3 = Guwahati

Table1: Sites with Geographical Coordinates:

Sites	Geographical coordinates
S1	Latitude = 26.15 degrees North and 26.5 degrees North and at the longitude of 92° East.
S2	Latitude = 26° 8' N to 26° 45' N and longitude of 90° 45' E to 91° E.
S3	Latitude = 26°10'45" N Latitude and 91° 45' 0" N longitude.

Field survey and collection procedure:

Field survey was done in the wetland along with fisherman by riding on a boat and fishing net was used to collect the fish samples.

Time of collection:

The fish species were collected from May 2012 to September 2015. Field survey was carried out throughout the year to investigate their refuge, and the accidental sightings if any.

For the smooth conduct of the survey a given year of the study period was divided into four seasons. These were, Pre-Monsoon (March to May), Monsoon (June to August), Post Monsoon (September to November) and Winter (December to February).

Table2: No. of Sightings of Labeo species recorded during the study period 2012-2015

Year	Seasons	Period of the year	No.of samples collected of each species
2011	Pre-Monsoon	March-May	10
	Monsoon	June-Aug	10
	Post-Monsoon	Sep-Nov	10
	Winter	Dec-Feb	10
	Total		40
2012	Pre-Monsoon	March-May	10
	Monsoon	June-Aug	10
	Post-Monsoon	Sep-Nov	10
	Winter	Dec-Feb	10
	Total		40
2013	Pre-Monsoon	March-May	10
	Monsoon	June-Aug	10
	Post-Monsoon	Sep-Nov	10
	Winter	Dec-Feb	10
	Total		40
2014	Pre-Monsoon	March-May	10
	Monsoon	June-Aug	10
	Post-Monsoon	Sep-Nov	10
	Winter	Dec-Feb	10
	Total		40
2015	Pre-Monsoon	March-May	10
	Monsoon	June-Aug	10
	Post-Monsoon	Sep-Nov	10
	Winter	Dec-Feb	10

	Total		40
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Identification of fish species:

All the fish species were identified morphologically according to the Fishes of North-East India published by Vishwanath et.al (2007), based on the authors like Talwar and Jhingran 2001, *Labeo rohita*, *Labeo bata*, *Labeo calbasu*, *Labeo gonius* were identified and *Labeo nandina* was identified based on author like Hamilton, 1822, Day, 1889.

Methods:

Limited information is available on *Labeo nandina* morphometry. There have been a few attempts to evaluate the population structure of *Labeo nandina*, using different methods based on phenotypical and genetic aspects. The data analysis was carried out without taking the sex of the sample into consideration. The fin formulae were studied from their physical count. A comparative study on the fin formula was made after comparing different reports (Talwar and Jhingran, 1991. Goswami *et al.*, 2012. Duttamunshi and Srivastava 1988; Nikolsky, 1963; Pillay, 1957).

A dial reading caliper is used for taking body measurements and also a ruler is used for accurate reading. Four local species of *Labeo* which are commonly available are taken for comparison with *Labeo nandina*. These species are *Labeo rohita*, *Labeo gonius*, *Labeo bata* and *Labeo calbasu*. From each species, including *Labeo nandina* 13 individuals were collected. Various body measurements were carried out according to Munshi and Shrivastava with certain modifications. 10 morphometric parameters like total length (TL), standard length (SL), head length (HL), eye diameter (ED) , pre dorsal length (PrDL), post dorsal length PDL), Peduncle length (PL), Body height(BH), Pre-orbital length (PrOL), Post-orbital length(POL) were taken for the morphometric study. The readings taken by the scale was expressed in centimeters. The average values of various external morphological characters studied in the four species of *Labeo* were taken out and their percentage values in relation to head length for the metric characters in head region and that of body region in relation to standard length were taken out . The quantitative data were then analyzed by hierarchial tree analysis, obtaining dendrogram using SPSS version 16 software. The dendrogram thus obtained was used for a comparative analysis of *Labeo nandina* with the four other *Labeo* species mentioned above.

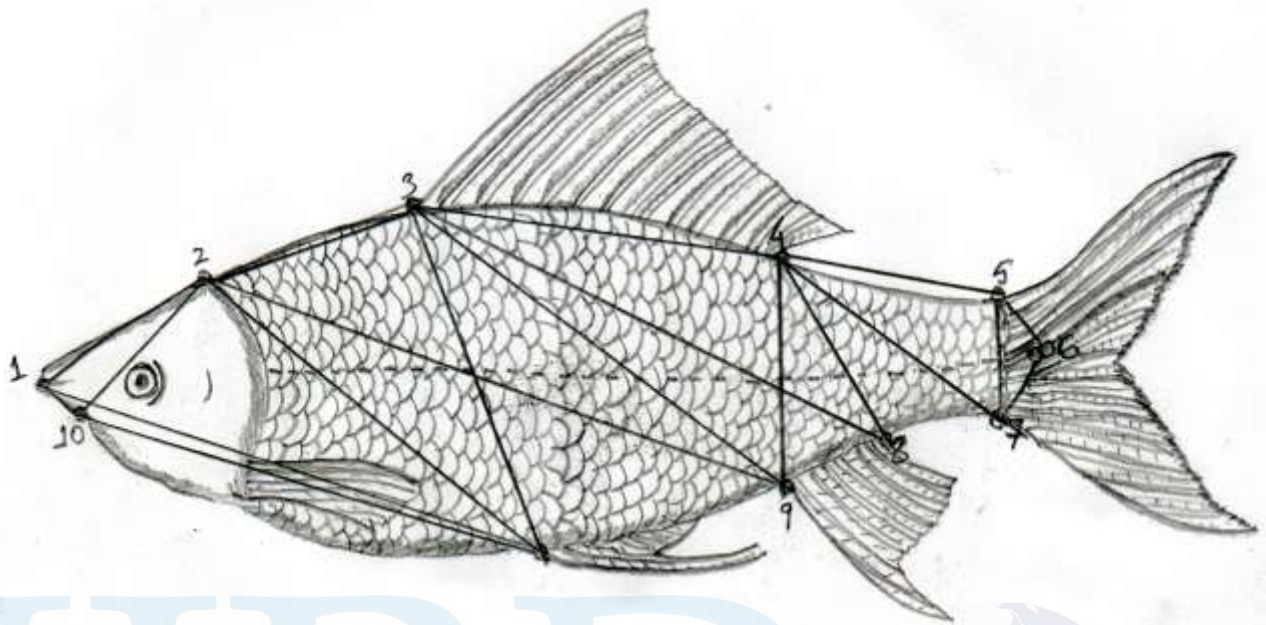


Fig 1: Locations of 10 landmarks used for the morphometric analysis of *Labeo* species

Table 3 : Morphometric characters used for analysis of *Labeo* species stock variations

Characters	Descriptions
Total length (TL)	Distance from the tip of the snout to the longest caudal fin ray
Standard length (SL)	Distance from the tip of the snout to the end of the vertebral column
Body height (BH)	Vertical distance from the anterior part of the 1 st dorsal fin and ventral part of the body.
Head length (HL)	Distance from the tip of the snout to the

	posterior margin of the opercula
Eye diameter (ED)	the maximum diameter cover by the eye.
Pre orbital length (PrOL)	Distance from the tip of the snout to the anterior margin of the eye
Post orbital length (POL)	Distance from the posterior margin of the eye to the end of the operculum.
Pre dorsal length (PrDL)	Distance from the snout tip to the anterior base of the dorsal fin.
Post dorsal length (PDL)	It was the distance between the structural base of the dorsal fin up to the base of the caudal fin.
Peduncle length (PL)	Length of the peduncle



Fig 2: *Labeo nandina* (Ham.)

Description of the Labeo species considered for the morphometric study:***Labeo nandina* (Hamilton-Buchanan, 1822)****Distinguishing Characters:**

D ii-iii 22-24; A ii 5; P i 15; V i 8. The same has been reported by Talwar and Jhingran 1991.

D.24-26 (2-3/22-24). P.15.V.9.A.7 (2/5). C.19.L.I.42-44.L.tr.6-7/8 (Day, 1875-78) Fishes-Vol.I.). Length of head $4\frac{1}{2}$ to 5 caudal fin $4\frac{1}{4}$ to $4\frac{1}{2}$, height of body 4 in the total length. Eye-diameter $4\frac{1}{2}$ to 5 in length of head, $\frac{1}{2}$ diameters from ends of snout, and $2\frac{1}{2}$ apart. Body elongate, its dorsal profile quite concave above eyes; interorbital region flat. Snout obtuse, slightly projecting beyond mouth, no lateral lobe, studded with a few pores. Eyes moderate, not visible from underside of head, the diameter 4.5 to 5 times in head. Mouth wide; lips thick and fringed, with a distinct inner fold above and below. Barbels two short pairs (rostral and maxillary). Dorsal fin inserted midway between snout-tip and caudal fin base. Caudal fin deeply forked. Scales small; lateral transverse scale-rows 5 between lateral line and pelvic fin base (Talwar and Jhingran 1991).

Colour: in life, dark greenish on back, becoming lighter on flanks and abdomen, with a few diffused blotches on flanks; centre of several scales reddish; iris red.

Geographical Distribution: India, West Bengal and Assam; Bangladesh; and Burma.

Fishery Information: This carp which attains a length of 26 cm, was fairly common in the catches in West Bengal and Assam. At present it is totally rare

***Labeo bata* (Hamilton-Buchanan,1822)**

Body is elongated, its dorsal profile more convex than the ventral. Snout slightly projecting beyond mouth, often studded with pores. Eyes large, not visible from underside of head. Mouth inferior; lips thin, lower, lower lip slightly fringed and folded back and joined to isthmus by a narrow bridge. Dorsal fin inserted nearer snout-tip than base of caudal fin. Black blotch on 5th and 6th scales on the shoulder. Its colour in life is golden-yellow above and on dorsal half of flanks, silvery on lower half of flanks and belly.

Geographical Distribution – India: Cauvery, Krishna and Godavari river systems in South India, Orissa, Madhya Pradesh, Uttar Pradesh, Bihar, West Bengal, Assam; Bangladesh; and Nepal (introduced).

IUCN- Red list status – Least Concern (LC)

***Labeo calbasu* (Hamilton-Buchanan, 1822)**

Its body is stout and rather deep. Head fairly large and conical, its length less than body depth. Snout depressed and fairly pointed, devoid of lateral lobe, studded with pores. Mouth inferior; lips thick and conspicuously fringed, both lips with a distinct inner fold. Eyes moderate. Barbels two pairs. Dorsal fin with a fairly long base, inserted midway between snout-tip and base of caudal fin. Colour in life is blackish-green, lighter below; flanks buff pink or with scarlet spots with dark edges which may form stripes. Fins black; upper lobe of caudal fin usually tipped with white.

Geographical Distribution – Pakistan, India, Bangladesh, Nepal, Burma, Thailand and Yunna (South China).

IUCN- Red list status – Least Concern (LC)

***Labeo gonius* (Hamilton-Buchanan,1822)**

Body is elongated and dorsal profile more convex than the ventral. Snout slightly projecting beyond mouth, devoid of lateral lobe, studded with numerous pores. Eyes moderate. Mouth narrow and subinferior; lips thick and fringed, with a distinct inner fold in their circumference. Barbels two very short pairs. Dorsal fin inserted nearer to snout-tip than to base of caudal fin. Scales small. Colour in life is greenish-black on back, becoming dull white on flanks and belly; scales darkest at their margins, and several scales with red lunules giving the impression of faint longitudinal lines.

Geographic Distribution – Pakistan, northern India, Bangladesh and Burma.

IUCN- Red list status – Least Concern (LC)

***Labeo rohita* (Hamilton-Buchanan, 1822)**

Body is moderately elongated and its dorsal profile more arched than the ventral profile. Snout fairly depressed, projects beyond mouth, devoid of lateral lobe. Eyes large, mouth small and inferior; lips thick and fringed, with a distinct inner fold to each lip. A pair of small maxillary barbels concealed in lateral groove. Dorsal fin inserted midway between snout-tip and base of caudal fin. Scales are moderate. Colour in life is bluish along back, becoming silvery on the flanks and beneath, with a reddish mark on each scale during breeding season; eyes reddish.

Fins grayish or dark; pectoral fins dusky. The body colour tends to vary in fishes living among weeds, exhibiting greenish black on back.

Geographical Distribution – Pakistan; north and central India; Bangladesh; Terai region of Nepal; and Burma.

IUCN- Red list status – Least Concern (LC)

Results:

Table 4: Comparative meristic characteristics of different *Labeo* species

Species	Distinguishing characters							
	D		A		P		V	
<i>Labeo nandina</i> (n=60)	ii-iv	9-10	ii-iii	5	i	15-27	i	8
<i>Labeo calbasu</i> (n=65)	iii-iv	13-16	ii-iii	5	i	16-18	i	8
<i>Labeo gonius</i> (n=65)	ii-iii	13-16	ii	5-6	i	16	i	8
<i>Labeo bata</i> (n=60)	iii-iv	22-26	ii	5	i	15	i	8
<i>Labeo rohita</i> (n=65)	iii-iv	12-14	ii-iii	5	i	16-18	i	8

In the table D = Dorsal fin; P = Pectoral fin; A = Anal fin; V = pelvic fin
ii-iii (indicate branching pattern of fin rays).

Collection of Male and Female samples: Male species = 30-35 samples of each species;
Female species = 30-35 samples of each species.

Table 5: Average measurements (cm) and standard deviation of different morphometric characters of *Labeo nandina* with other *Labeo* species.

Morphometric characters	<i>Labeo rohita</i>		<i>Labeo bata</i>		<i>Labeo gonius</i>		<i>Labeo calbasu</i>		<i>Labeo nandina</i>	
	Avg.	Std. dev	Avg.	Std. dev	Avg.	Std. dev	Avg.	Std. dev	Avg.	Std. dev
Total length	41.9	±3.2	27.9	±2.53	25.9	±2.83	33.5	± 3.1	20.4	±2.04
Standard length	35	±2.83	23.8	±2.32	20.93	±2.35	27.7	± 2.9	18.72	±2.09
Head length	8.3	±0.87	5.3	±0.67	5.23	±0.73	6.55	± 0.9	4.6	±0.5
Eye diameter	1.2	±0.15	0.9	±0.10	1.06	±0.13	1	± 0.14	1.64	±0.11
Pre dorsal length	15.4	±2.13	9.9	±1.38	9.53	±1.84	12.55	± 2.03	5.63	±1.23
Post dorsal length	10.6	±2.75	8.4	±1.32	6.8	±1.82	8.15	± 2.1	4.01	±1.34
Peduncle length	5	±0.84	3	±0.34	2.7	±0.45	3.9	± 0.5	4.1	±0.4
Body height	10.5	±0.83	6.7	±0.73	6.3	±0.85	8.7	± 0.93	7.29	±1.16
Pre orbital length	2.4	±0.18	1.7	±0.13	1.1	±0.14	2.3	± 0.20	2.7	±0.19
Post orbital length	4	±0.20	2.8	±0.12	2.3	±0.13	3.4	± 0.21	2.2	±0.19

Table 6: Showing ratio of average measurements of the morphometric characters of head region in relation to head length and that of body region in relation to standard length.

Morphometric characters	<i>Labeo rohita</i>	<i>Labeo bata</i>	<i>Labeo gonius</i>	<i>Labeo calbasu</i>	<i>Labeo nandina</i>
ED/HL	0.14	0.16	0.20	0.15	0.35
PrOL/HL	0.28	0.32	0.21	0.35	0.58
POL/HL	0.48	0.52	0.43	0.51	0.47
PrDL/SL	0.44	0.41	0.45	0.45	0.30
PDL/SL	0.30	0.35	0.32	0.29	0.21
PL/SL	0.14	0.12	0.12	0.14	0.21

Dendrogram using Average Linkage (Between Groups)

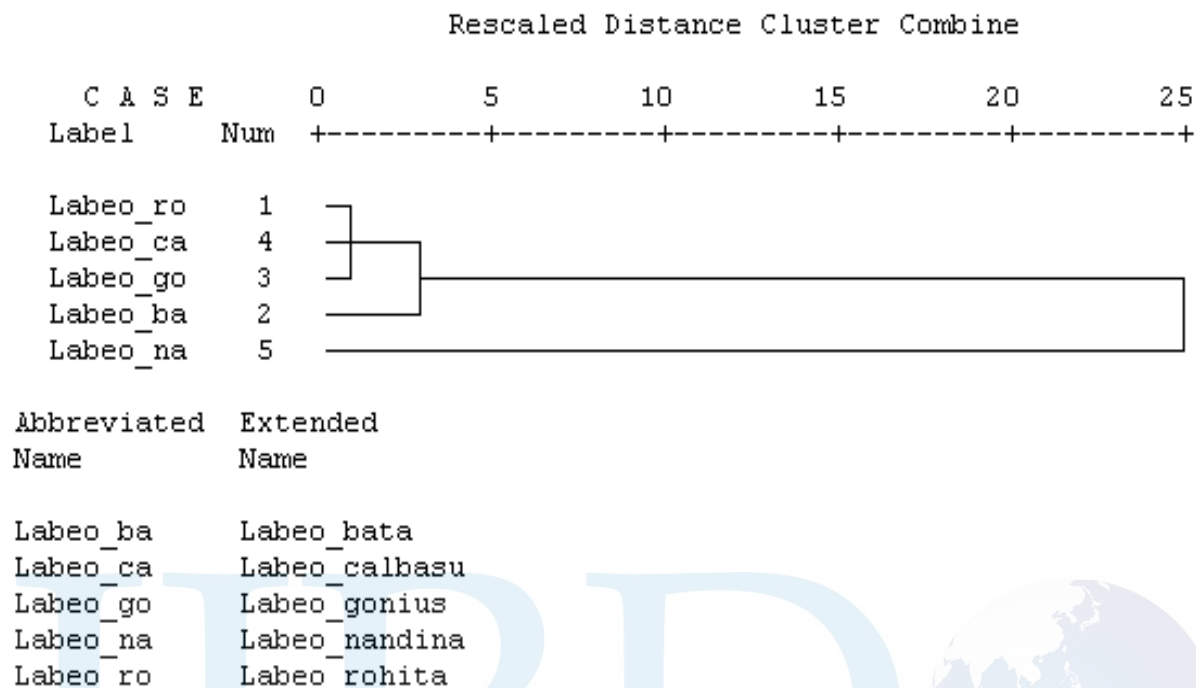


Fig3: Dendrogram of five *Labeo* species, *L. rohita*, *L. calbasu*, *L. gonius*, *L. bata* and *L. nandina* on the basis of the Morphometric parameters showing average linkage (between groups) of *Labeo nandina* with four other *Labeo* species.

Discussions

Different morphometric characters were measured for *Labeo nandina* along with the four other *Labeo* species taken for the comparative study. When the average measurements of *Labeo nandina* was compared with the other four species (Table 2, Fig1), it was found that *Labeo nandina* has the highest eye diameter (1.64cm) than the other four *Labeo* species while *Labeo bata* showing the lowest measurement (0.9cm). The peduncle length of *Labeo nandina* (4.1cm) is less than *Labeo rohita* (5cm) but more than *Labeo bata*, *Labeo gonius* and *Labeo calbasu*. On the other hand, the pre orbital length of *Labeo nandina* is highest (2.7cm) than all the other *Labeo* species taken. The lowest length was seen in *Labeo gonius* (1.1cm). The body height of *Labeo nandina* (7.29cm) was found to be more than *Labeo bata* and *Labeo gonius* but less than *Labeo rohita* and *Labeo calbasu*. The rest of the metric

characters of *Labeo nandina* was found to be less than the other four *Labeo* species taken. Even though of the same genus, the significant difference in their morphometry was evident.

The dendrogram obtained from the analysis of the average measurements of morphometric characters of *Labeo nandina* with the four locally available species taken, showed three major clusters (Fig 3). *Labeo rohita*, *Labeo calbasu*, *Labeo gonius* formed one cluster, *Labeo bata* formed another cluster with the above clustered species and finally, *Labeo nandina* formed the third cluster with the species of the above two clusters. *Labeo bata* was found to form the closest cluster with *Labeo nandina* than the species of other cluster formed. This result shows that *Labeo nandina* has a greater metric similarity with *Labeo bata*. Next to it *L. rohita*, *L. calbasu* and *L. gonius* shows almost equal similarity with *Labeo nandina*, in terms of metric characters.

Fish are very sensitive to environmental changes and quickly adapt themselves by changing necessary morphometrics. It is well known that morphological characters can show high plasticity in response to differences in environmental conditions such as food abundance and temperature (Allendorf and Phelps 1988, Swain *et al.* 1991, Wimberger 1992). In general, fish demonstrate greater variances in morphological traits both within and between populations than any other vertebrates and are more susceptible to environmentally induced morphological variations (Allendorf *et al.* 1987, Wimberger 1992). The morphometric differences observed between *Labeo nandina* and other *Labeo* species taken, may have been due to environmental as well as genetic variations.

As no study was done on *Labeo nandina*, particularly from this region i.e the Brahmaputra River System, the morphometric characters would be helpful in comparing the same species in different locations.

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