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Intraspecific phenotype variations in olive barb *Systomus sarana* (Hamilton, 1822) population from different rivers is possibly linked to locomotive adaptations in caudal fin

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ABSTRACT

Systomus sarana (Hamilton, 1822) is an economically important food fish species occurring throughout Indian rivers, which also has ornamental value. This study focused on morphological variations in *S. sarana* from five river basins across India, *viz.,* Godavari, Mahanadi, Krishna, Middle Ganga and Lower Ganga. A truss network was constructed by interconnecting 12 landmarks to generate 65 morphometric variables extracted from digital images of specimens sampled from the study locations. Transformed truss measurements were subjected to Principal component analysis (PCA), Canonical discriminant function analysis (CDFA), Box plot and Thin plate spline (TPS) analyses. PCA identified eight truss variables with significant loadings, while CDFA designated two truss variables with potential for explaining discrimination between populations. Anterior attachment of dorsal membrane from caudal fin was identified to be the most important variable that presented variations across the river basins studied. Discriminant analysis correctly classified 70.5% of the specimens into their original populations. Thin plate spline for morphometric shape variation analysis indicated highest specimen-shape variations (warping) in Mahanadi basin. TPS-principal strain ratio on principal components (PC-1, PC-2) further revealed significant divergence among the populations in five river basins studied. Results of the study revealed variation in stocks of the species, on the basis of shape morphometry. The four significant parameters differentiating specimens from different basins, were linked to caudal fin origin at dorsal side and the centre and possibly indicate plasticity in response to locomotive adaptations.

Keywords: Plasticity, Populations, *Systomus sarana,* Truss morphometry

Introduction

Stock being the "subset of species having the same growth and mortality parameters and inhabiting a particular geographical area sharing a common gene pool" (FAO, 1998), is considered fundamental for species conservation. Stocks are evolutionarily significant units adapting to the local environmental conditions during the course of evolution, after separation from the common ancestor. The unique spatial, temporal, serological, biological and genetic characteristics of fish stocks necessitates cataloguing, for strategic conservation programs as well as to make use of variations in such parameters for aquaculture programs (Rawat *et al.,* 2017). Morphological variations between stocks could also reflect variation in growth and mortality patterns (Cadrin, 2000). The phenotypic evolution, in response to local adaptation can be attributed to divergent selection along the different environmental pressures (Kawecki and Ebert, 2004) and these phenotypic diversity or plasticity can be attributed to organism's functions (Camarillo *et al.,* 2020). Effective fishery management programmes need stock variation as critical input (Smith *et al.,* 1991). Several fishery biologists have utilised the potential of shape morphometry for discriminating groups or populations (Park *et al.,* 2004; Siddik *et al.,* 2016). Truss network profile generated through the use of landmarks extending across the entire fish to capture shape information and transforming into geometric morphometrics, provides a quantitative method to assess morphometric differences between the specimens from different geographical locations (Strauss and Bookstein, 1982; Turan, 1999; Bhosale *et al.,* 2018; Kaka *et al.* 2019; Ethin *et al.,* 2019; Mahfuj *et. al.,* 2019).

Systomus sarana (Hamilton, 1822), commonly known as "olive barb" is one of the commonly available barbs in the Indian subcontinent (Nahiduzzaman *et al.*, 2011). It is a tropical freshwater fish belonging to the family

Cyprinidae under the order Cypriniformes and has a good market demand, due to its high nutritional value (Akter *et al.,* 2010). In India, it is prevalent in all river basins except in peninsular India (south of Krishna River) (Talwar and Jhingran, 1991). The taxonomic ambiguity of this species with the peninsular *S. sarana subnasutus* (Valenciennes, 1842) was resolved recently, through the use of integrated taxonomic approaches (Biswal *et al.,* 2018). The whole mitogenome of *S. sarana* has been mapped, annotated and its phylogenetic status was addressed on the basis of concatenated mitochondrial genes (Biswal *et al.,* 2017). Studies have indicated serious decline in the population of *S. sarana* due to environmental degradation, aquatic pollution, destruction of breeding grounds, introduction of exotic fishes as well as in response to changes in the ecological habitat (Hossain *et al.,* 2009) and the species has been categorised under vulnerable group by Mijkherjee *et al.* (2002) while studying the local fishes of West Bengal.

The present study examined the body shape differences to identify phenotypic variations and divergence using truss network system based on morphometric characteristics, in the population of *S. sarana* in five river basins of India. The study also attempted to define significant morphometric characteristics to differentiate stocks of the species.

Materials and methods

Sample collection

A total of 207 intact specimens of *S. sarana* were collected from commercial catches in gillnet, cast net and traps across 14 sampling locations belonging to five different river basins namely, Godavari, Mahanadi, Krishna, Middle Ganga and Lower Ganga of India (Fig. 1). Samples of Godavari basin (n=53) comprised those from Adilabad (n=25), Nirmal (n=23) and Rajahmundry (n=5). Mahanadi basin sample (n=67) was inclusive of specimens collected from Jobra Barrage (n=17), Banki (n=15), Naraj Barrage (n=10) and its distributaries *viz.,*; Daya (n=7), Luna (n=4) and Birupa (n=14). Specimens collected from Ibrahimpatnam (n=30) constituted the Krishna basin sample (n=30). Middle Ganga basin collection (n=26) included samples from tributaries Betwa (n=11) and Bansagar (n=15), while Lower Ganga (n=31) constituted samples from Farakka (n=19) and Hooghly River, a major distributary of river Ganga sampled from Nababdwip (n=12) (Table 1).

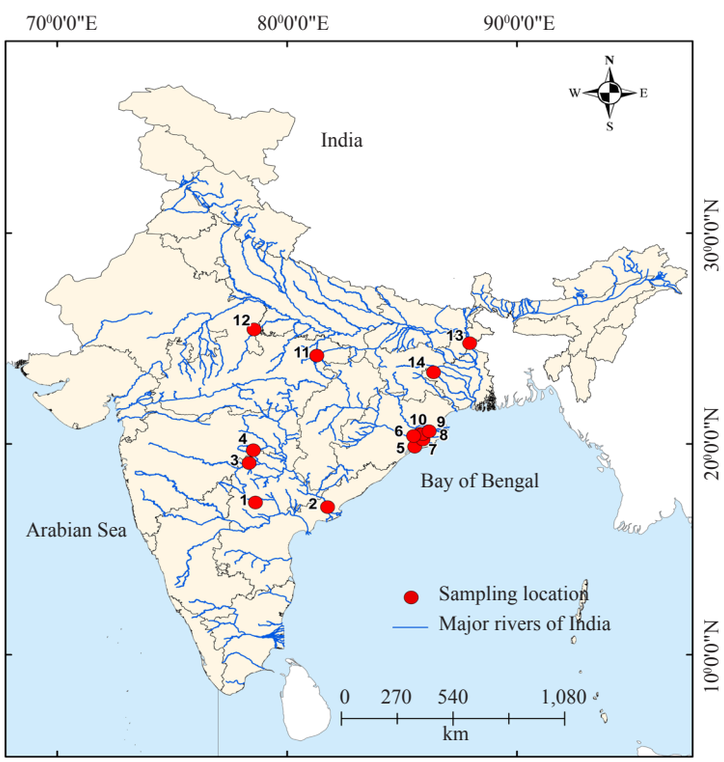

Fig. 1. Sampling locations of *S. sarana.* Krishna Basin: 1. Ibrahimpatnam; Godavari Basin: 2. Rajahmundry, 3. Nirmal, 4. Adilabad; Mahanadi Basin: 5. Daya River, 6. Luna River, 7. Jobra Barrage, 8. Naraj Barrage, 9. Birupa, 10. Banki; Middle Ganga Basin: 11. Bansagar, 12. Betwa River, Lower Ganga Basin: 13. Farakka, 14. Nababdwip

Table 1. Description of sampling localities and number of samples (N) of *S. sarana*

River basin	Sampling location	Month and year of collection	Latitude (Decimal Degree)	Longitude (Decimal Degree)	No. of specimens
Godavari	Rajahmundry, East Godavari, Andhra Pradesh.	Jan. 2015	17.00297	81.758517	05
	Nirmal, Telangana	Apr. 2019	19.09474	78.342624	23
	Adilabad,Telangana	Jan. 2011	19.70093	78.529206	25
Krishna	Ibrahimpatnam, Ranga Reddy, Telangana	Jan. 2015	17.22095	78.620756	30
Mahanadi	Daya River, Odisha	Jan. 2015	19.87126	85.550265	07
	Naraj Barrage, Cuttack, Odisha	Jan. 2015	20.47693	85.781462	10
	Luna River, Kendrapara, Odisha	Jan. 2015	20.21243	85.90705	04
	Jobra Barrage, Cuttack, Odisha	Apr. 2015	20.470000	85.900000	17
	Birupa River, Cuttack, Odisha	Apr. 2015	20.58935,	85.95711	14
	Banki, Cuttack, Odisha	Apr. 2015	20.37782	85.24837	15
Middle Ganga	Betwa, Jhansi, Uttar Pradesh	Nov. 2016	25.43	78.56	11
	Bansagar, Madhya Pradesh	May 2015	24.19166	81.287509	15
Lower Ganga	Farakka, West Bengal	Nov. 2016	24.78073	87.938392	19
	Nababdwip, West Bengal	Apr. 2016	23.4	86.37	12
Total					207

Digitisation of sample images

Fish specimens were cleaned, wiped dry and photographed at the collection site itself. Specimens were placed with mouth facing left, on a level platform with water resistant graph paper having vertical and horizontal grids, such that, an area of 1 cm^2 covered one square unit (Fig. 2). A specific code was used to label each specimen for easy identification. The fins of the specimens were stretched out, so that the origin and insertion points were perceptible. For digitising images of the samples, a Sony Cyber-shot DSC-W300 digital camera was used (Cadrin and Friedland, 1999). Calibration of reference scale for each individual specimen was possible with tpsUtil software (Rohlf, 2008a), as the graph paper was digitally imaged from the same height, with same resolution and focus as that for the specimen. Considering the size range in the samples, the photographs were not captured at same height for different specimens; which was compensated by calibration of the reference scale.

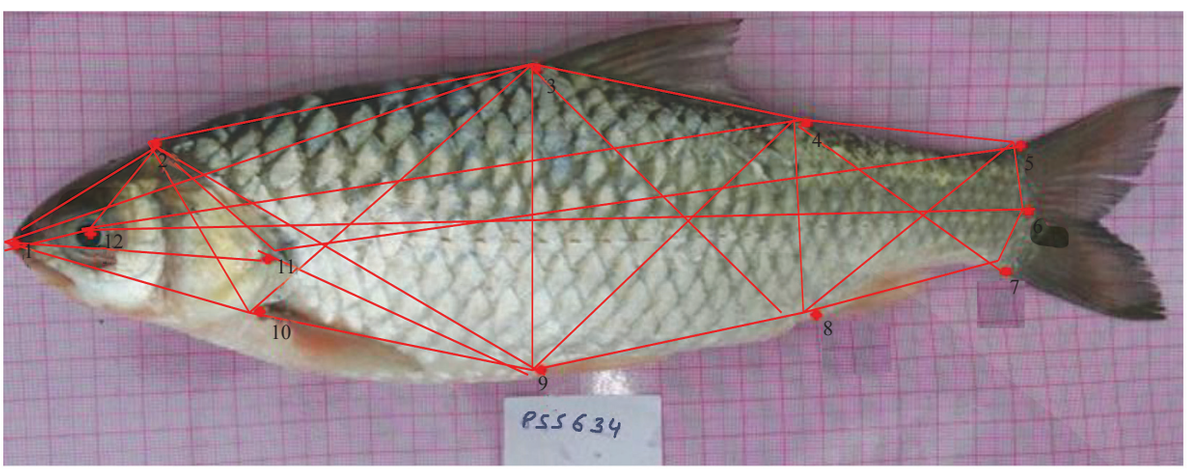

Fig. 2. Truss network of *S. sarana* showing the truss variables extracted from 12 landmarks (Landmarks: 1. Snout tip at upper jaw; 2. Posterior aspect of neurocranium (beginning of scaled nape); 3. Dorsal fin origin; 4. Line perpendicular to anal fin origin; 5. Anterior attachment of dorsal membrane from caudal fin; 6. Posterior end of vertebral column; 7. Anterior attachment of ventral membrane from caudal fin; 8. Anal fin origin; 9. Pelvic fin insertion; 10. Pectoral fin insertion; 11. Operculum end; 12. Eye centre)

Morphometric data generation

In the present study, 12 landmarks representing the developmental and anatomical features among specimens were selected. Landmarks were digitised using tpsDig2 ver. 2.31 and data was encrypted to tps files in X-Y coordinate form (Rohlf, 2008b). Truss network was generated by interconnecting these landmarks to form a total of 66 truss distances. Entire extent of the morphology of species was represented by the network extending across the fish (Fig. 2). Paleontological Statistics software (PAST) (Hammer *et al.,* 2001) was used to measure truss distances between the landmarks for each specimen (Sreekanth *et al.,* 2015).

Morphometric analysis

All the truss measurements from PAST were log transformed (Strauss, 1985) and size effect was eliminated as described by Elliott *et al*. (1995):

$$M_{adj}=M*(L_S/L_0)^b,$$

where; M is the original measurement, M_{adj} the size adjusted measurement, L_0 standard length of fish and L_S the overall mean standard length.

Standard length (character code 1_6) was not included in analysis, as it was used as the basis for transformation (Mamuris *et al.*, 1998). Univariate analysis of variance (ANOVA) was carried out for the other 65 morphometric characters retained; to evaluate whether any significant difference existed among the five river basins studied. The morphometric characters that exhibited significant variations ($p<0.01$), were further used to attain the ratio of number of samples (N) to the parameters included (P), for multivariate analysis (Johnson, 1981; Kocovsky *et al*., 2009) employing principal component analysis (PCA) and canonical discriminant function analysis (CDFA). PCA was carried out in 207 x 65 truss data matrix for identification of significant principal components (PCs) and contribution of components. In PCA, number of components was determined by applying Kaiser's (1960) criterion of retaining eigen values greater than one (Jolliffe, 2002). Thin plate spline (TPS) image analysis on mean score linked PCA (relative warps) was performed to analyse geometric shape variations (Rohlf, 2008c). CDFA was also carried out for identifying specimen distribution pattern and important discriminant functions. A cross validation step was done to estimate the probable error rates of the classification functions. Box plot was used for displaying the role of truss variables in discriminating distribution pattern of fish specimens across different locations. All truss morphometric data were analysed using SPSS version 16 (SPSS inc., Chicago, USA), SAS version 9.3 (SAS Inc., North Carolina, USA), TPS software package and Excel (Microsoft Office 2007).

Results and discussion

Multivariate analysis

The PCA provided 12 significant principal components as per Kaiser's (1960) criterion and contributed significantly up to 94.65% of total variation of data matrix (Table 2). The loading matrix of truss variables on PC-1 and PC-2 identified eight truss variables with significant and highest loading, namely 1_4 (snout tip to line perpendicular from origin anal fin); 1_8 (snout tip to anal fin origin); 1_10 (snout tip to pectoral fin origin); 1_11 (snout tip to operculum end); 1_12 (snout tip to eye centre); 4_12 (line perpendicular from anal fin origin to eye centre); 5_11 (anterior attachment of dorsal membrane from caudal fin to operculum end) and 6_12 (posterior end of vertebral column to eye centre) (Table 3). The PCs (PC-1 and PC-2) accounted for 48.84% of total variation with PC-1 accounting for 28.66% variation leading to the identification of two most important truss variables 5_11 (anterior attachment of dorsal membrane from caudal fin

Table 2. Principal component analysis (PCA) in five natural populations of *S. sarana* for truss analysis

Principal components	Eigen value	Percentage of variance	Cumulative variance
1	18.63	28.66	28.66
2	13.12	20.19	48.84
3	6.10	9.38	58.22
4	5.68	8.74	66.96
5	4.08	6.28	73.23
6	3.49	5.37	78.60
7	2.58	3.96	82.57
8	2.44	3.75	86.32
9	1.94	2.98	89.31
10	1.23	1.89	91.19
11	1.17	1.80	92.99
12	1.08	1.65	94.65
Total		94.65	

to operculum end) and 6_12 (posterior end of vertebral column to eye centre) with significant loadings (Table 3).

In CDFA, out of the four functions, two functions (Functions 1 and 2) explaining 73.68% of total variation of data, were found to be significant as per Wilks' lambda; for discriminating specimens between basins. The highest contribution in total variation was by Function-1 (44.82%) followed by Function-2 (29.86%) (Fig. 3, Table 4). CDFA identified two most important truss variables; 1_5 (snout tip to anterior attachment of dorsal membrane of caudal fin) and 2_5 (posterior tip of neurocranium to anterior attachment of dorsal membrane of caudal fin) with discrimination coefficient ranging from 761.83, (-) 653.22, for Function-1 and 710.74, (-) 562.12 for Function-2.

Classification results from predicted group membership showed that correct classification of individuals into their original population varied between 40 and 82.09% and cross validated classification between 30 and 77.61% by CDFA. Overall classification rate was estimated as 70.5% (Table 5). Krishna basin samples exhibited highest misclassification rates, *i.e.,* 70%. Godavari and Mahanadi basin samples clearly separated from rest of the populations, with 62.71 and 77.61% group membership, respectively, while overlapping was noticed with Krishna and Mahanadi samples, having small discriminant scores (0.65 and 0.83 respectively) on Function 1 and 2 (Fig. 3, Table 4 and 5). It may also be noted that Function-1 has similar score for Krishna and Mahanadi basins. Middle and Lower Ganga basin samples showed overlapping with each other as well as significant overlapping with Mahanadi population. In the present investigation, correctly classified cross validation (58.9%) indicated medium level differentiation and morphological homogeneity in samples of *S. sarana* from five river basins. The discriminant function analysis and the DF1 and DF2 scores demonstrated higher distinction for Godavari, Middle Ganga and Lower Ganga populations.

Box plot on truss variables distribution

Eight truss variables were identified from PCA which when arranged in ascending order*, i.e.,* from lower to higher loading, came out as 1_4 (snout tip to line perpendicular from origin anal fin), 1_8 (snout

Table 3. Truss morphometric relationship through mean, standard deviation (SD), minimum, maximum and range values of eight identified truss variables in *S. sarana*

Truss variable	Truss variable loading on two principal components		Truss variables, M-trans data matrix (after eliminating the effect of standard length)				Truss variables Log-morphometric data matrix (without eliminating the effect of standard length)			
	PC1*	PC2*	Mean	SD	Min, Max	Range	Mean	SD	Min, Max	Range
1_4	0.03	0.87	1.03	0.01	1.00, 1.08	0.07	1.03	0.19	0.29, 0.54	0.25
1_8	-0.05	0.87	1.04	0.01	1.00, 1.07	0.06	1.04	0.19	0.29, 0.54	0.25
1_10	-0.86	0.24	0.51	0.04	0.38,0.62	0.25	0.51	0.19	0.29,0.54	0.25
1_11	-0.89	0.16	0.46	0.05	0.34, 0.58	0.23	0.47	0.19	0.29,0.54	0.25
1_12	-0.85	-0.02	-0.07	0.09	-0.50, 0.11	0.61	-0.06	0.21	0.29,0.54	0.25
4_12	0.48	0.78	1.00	0.01	0.97, 1.04	0.08	1.00	0.20	0.29,0.54	0.25
5_11	0.86	0.02	1.06	0.01	1.03, 1.10	0.07	1.06	0.2	0.29,0.54	0.25
6_12	0.88	-0.03	1.14	0.01	1.12, 1.16	0.03	1.14	0.19	0.29,0.54	0.25

*Significant (≥0.35)

Table 4. Canonical discriminant function analysis (CDFA) in *S. sarana*

Centroid for Location/River	Function-1	Function-2
Godavari	-1.57	0.10
Mahanadi	0.68	0.36
Krishna	0.65	0.83
Middle Ganga	0.36	-2.23
Lower Ganga	0.65	-0.34
(%) Variance	44.82	28.86
Cummulative variance	44.82	73.68
Canonical correlation	0.71	0.63
Wilks Lamda	0.18	0.36
Significant ($p<0.05$)	0.00	0.00
Truss variable and Higher coefficient in function	1_5: 761.83 2_5:(-)653.22	1_5:710.74 2_5:(-)562.12

Table 5. Predicted group memberships in *S. sarana* from five river basins

	River basin	Predicted group membership					Total
		Godavari	Mahanadi	Krishna	Middle Ganga	Lower Ganga	
Original (Count)	Godavari	47	9	1	0	2	59
	Mahanadi	4	55	3	2	3	67
	Krishna	4	13	12	0	1	30
	Middle Ganga	1	2	0	13	4	20
	Lower Ganga	2	8	0	2	19	31
Original (%)	Godavari	79.66	15.25	1.69	0.00	3.39	100
	Mahanadi	5.97	82.09	4.48	2.99	4.48	100
	Krishna	13.33	43.33	40.00	0.00	3.33	100
	Middle Ganga	5	10	0	65	20	100
	Lower Ganga	6.45	25.81	0.00	6.45	61.29	100
Cross validated (Count)	Godavari	37	13	2	2	5	59
	Mahanadi	5	52	4	3	3	67
	Krishna	5	14	9	1	1	30
	Middle Ganga	2	3	0	9	6	20
	Lower Ganga	3	11	0	2	15	31
Cross validated (%)	Godavari	62.71	22.03	3.39	3.39	8.47	100
	Mahanadi	7.46	77.61	5.97	4.48	4.48	100
	Krishna	16.67	46.67	30.00	3.33	3.33	100
	Middle Ganga	10	15	0	45	30	100
	Lower Ganga	9.68	35.48	0.00	6.45	48.39	100

a: Cross validation is done for 5 cases in the analysis. In cross validation, each case is classified by the functions derived from all cases other than that particular case

b: 70.50% of original grouped cases correctly classified

c: 58.9% of cross validated grouped cases correctly classified

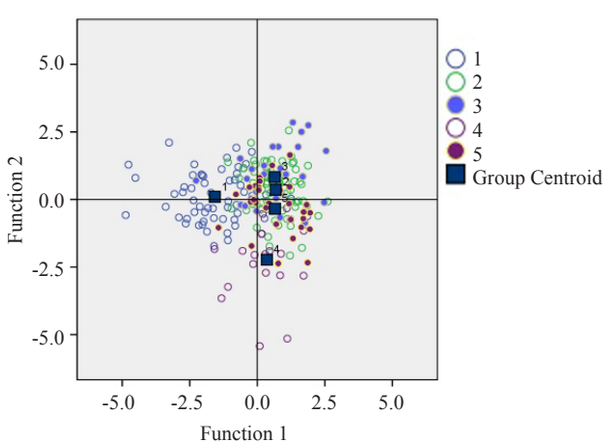

Fig. 3. *S. sarana* distribution in five river basins as per Canonical discriminant function: Function - 1 and 2 (1. Godavari; 2. Mahanadi; 3. Krishna; 4. Middle Ganga; 5. Lower Ganga)

tip to anal fin origin), 1_10 (snout tip to pectoral fin origin), 1_11 (snout tip to operculum end), 1_12 (snout tip to eye centre), 4_12 (line perpendicular from anal fin origin to eye centre), 5_11 (anterior attachment of dorsal membrane from caudal fin to operculum end) and 6_12 (posterior end of vertebral column to eye centre) (Table 3). CDFA identified two truss variables namely, 1_5 (snout tip to anterior attachment of dorsal membrane of caudal fin) and 2_5 (posterior aspect of neurocranium to anterior attachment of dorsal membrane from caudal fin) with higher coefficient in function, indicating capacity to differentiate populations (Table 4). Thus, among these ten truss variables listed, four variables were found to be most important, *viz.,* 5_11; 6_12 from PCA and 1_5; 2_5, from CDFA analysis. The box-plot graph on median truss value was used for all these four variables (Fig. 4.). Variable 5_11 over five locations indicated that Ganga, Godavari, Mahanadi, Krishna, Middle and Lower Ganga basins had differences over the median value indicating its significance in differentiating stocks. This analysis displayed the role of the four important truss variables identified in determining the distribution of specimens within and between locations with reference to median value.

Relationship between truss and morphometric variables

The distribution of truss variables on M-trans data and in Log-morphometric data displays mean, standard deviation and range values. The mean for Truss variables in M-trans data varied from -0.07 to 1.14 while in Log-morphometric data it varied from -0.06 to 1.14 (Table 3).

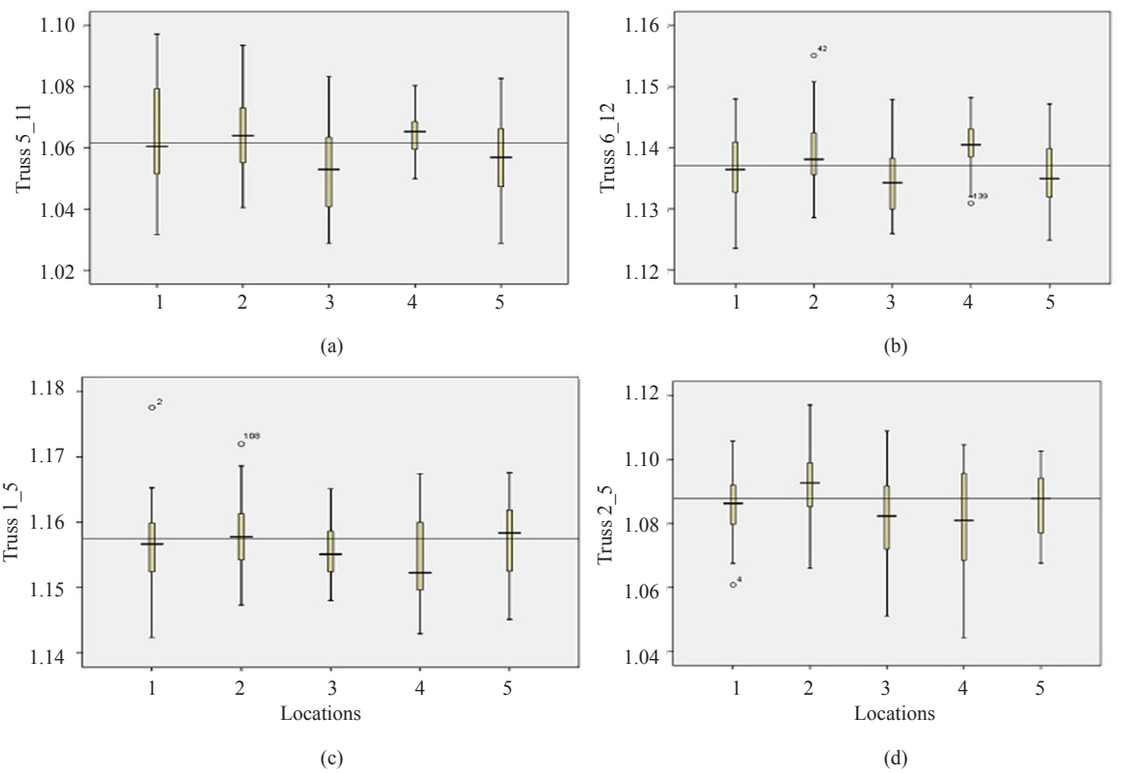

Fig. 4. Box plot display on role of four important truss variables in deciding the distribution pattern of *S. sarana* in the river basins (1. Godavari; 2. Mahanadi; 3. Krishna 4;. Middle Ganga; 5. Lower Ganga). Box plot for truss variable, (a) Dorsal origin of caudal fin to operculum end (5_11); (b) Posterior end of vertebral column to eye centre (6_12); (c) Snout tip to dorsal origin of caudal fin (1_5) and (d) Posterior aspect of neurocranium to dorsal origin of caudal fin 2_5

If we consider the truss variables, 5_11 (dorsal origin of caudal fin to operculum end) and 6_12 (posterior end of vertebral column to eye centre), which are variables having maximum loading on principal components, it can be seen that there is variation in the standard deviation and range value interpreted for M-trans data and Log-morphometric data. For instance, in variable 6_12, a mean of 1.14, standard deviation of 0.01 and range value of 0.03 in M-trans data was observed (Table 3), while Log-morphometric data, though had same mean of 1.14, exhibited higher standard deviation (0.19) and higher range value (0.25). Similar pattern can also be observed in truss variable 5_12. Thus, it can be inferred that improved morphometry based identification using the truss variables is possible in Log-morphometric data rather than M-trans data due to higher standard deviation and higher range value.

The truss linked PCA loading values observed on the river basins under study, indicated that the identified truss variables played a key role in identification and discrimination of specimens over different locations/ rivers/basins.

Thin plate spline analysis for variations in geometrical shape of fish specimens

The geometric shape variation through mean linked PCA (relative warps) revealed that the specimens from Mahanadi and Middle Ganga basins had higher relative warps (deformation) as 1.58 and 1.42 respectively, whereas other river basins had deformation (relative warps) in the range of 1.01-1.10. Further, with respect to the four important truss variables identified, Godavari and Mahanadi indicated different relative warps (1.01 and 1.58 respectively) along with differences in TPS image linked mean score on PC (PC-1). The identification of truss-specimens linked rivers with maximum and minimum score on principal component (PC-1), which was highest for Mahanadi (4.98; 1.22) followed by Godavari (3.21; 2.22), Middle Ganga (3.07; 1.15), Lower Ganga (3.03; 2.12) and Krishna (2.57; 2.06). Similarly, the maximum and minimum score on PC-2 was highest for Godavari (3.01; 2.09) followed by Mahanadi (2.15; 1.99), Lower Ganga (2.12; 1.38), Krishna (2.11; 1.90) and Middle Ganga (2.10; 1.49) (Table 6, Fig. 6).

The thin plate spline - PCA (relative warps) analysis for principal strain ratio values over maximum and minimum scores on principal components (PC 1 and 2) indicated distinctness in populations of Mahanadi (4.08; 1.08), Middle Ganga (2.67; 1.41), Lower Ganga (1.55; 1.54); while limited resolution between Godavari (1.45; 1.44) and Krishna (1.25; 1.11) (Table 6; Fig. 5).

Previous studies from several researchers have highlighted the role of genetic and environmental factors in shaping fish populations (Poulet *et al.,* 2004; Hossain *et al.* 2010). Franssen *et al.* (2013) suggested that the selective pressure of the environmental conditions leading to genetic-environmental interactions influence the pattern of phenotypic variations at intraspecific level. The results revealed 4 most significant morphological parameters

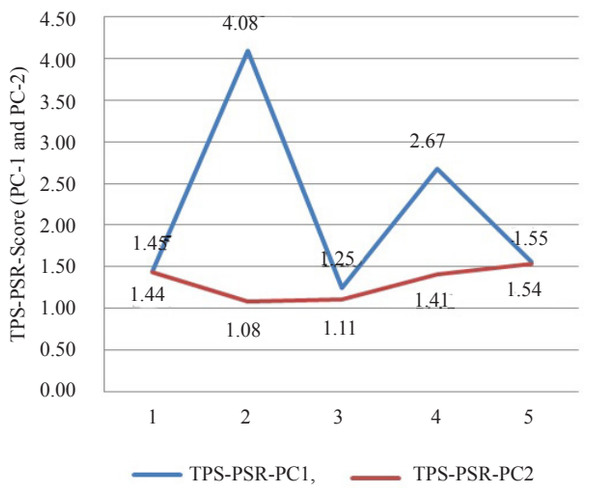

Fig. 5. Thin plate spline (TPS) in geometric shape variation analysis, on principal strain ratio (PSR) over maximum and minimum score on principal components (PC-1, PC-2) for discrimination of specimens from five locations (1. Godavari; 2. Mahanadi; 3. Krishna; 4. Middle Ganga; 5. Lower Ganga)

(5_11; 6_12; 1_5 and 2_5) which differentiate samples from different river basins. It is very interesting to note that, these are linked to caudal fin attachment to the body and are involved in the movement of fish. The landmark 5 to which three parameters are associated is the origin of dorsal lobe of the caudal fin. The landmark 6 which is linked to another significant parameter is the posterior end of the vertebral column and is the centre point of attachment of caudal fin. Lauder (2000) suggested that, though the homocercal tail exhibits symmetrical dorsal and ventral lobes, their movement is significantly more complex. He also suggested the existence of varied functional patterns in homocercal tail that bear significant consequences on the force balance of the fish body. As expected, the dorsal and ventral lobes of caudal fin do not function symmetrically. Rather, the dorsal lobe moves more swiftly and undergoes greater lateral excursions compared to the ventral lobe as observed in bluegill tuna (Lauder, 2000). Franssen *et al.* (2013) indicated more likelihood of plasticity of the caudal areas than anterior regions of the body, due to strong flow induced changes. He also suggested that variations in body shape is strongly influenced by habitat related variations, rather than genetic variation among basins. Lauder (2000) also stated that the assumption of horizontal reaction forces solely in the caudal region seems to be incorrect, as during swimming homocercal tail also generates lift forces perpendicular to body even during horizontal movements. The significant acute angle to the horizontal plane observed at dorsal lobe enables swift swimming aided by the hypochordal longitudinalis muscle present within the tail. Caudal region also happens to be the posterior region of the vertebrate axis where water, accelerated by anterior movement of the body, is discarded into the surrounding medium. Lauder (2000) was of the opinion that homocercal tail, postero-ventrally generates tilted and linked vortex rings. The changes in water currents, depths and habitats affect the musculature of this region leading to adaptational changes and thus,

Table 6. Distribution of mean, standard deviation (SD), minimum, maximum and range on scores over principal component PC1 (PC2) along with Thin plate spline (TPS) geometric shape variation through PCA (relative warps) analysis of *S. sarana*

River basins	Mean, SD, minimum, maximum and mean score linked PCA (relative warps) for specimens of locations on Principal component PC1 (PC2)					Thin plate spline (TPS) - PCA (relative wraps)			
						Principal strain of TPS image of specimens over PC1 & 2		Principal Strain Ratio over PC-1, PC-2	
	Mean	SD	Min.	Max.	Mean score linked PCA (relative warps)	PC1 Max (Min)	PC2 Max (Min)	PC1	PC2
Godavari	-0.15 (0.22)	1.01 (1.10)	-2.53 (-2.52)	1.72 (3.98)	1.01 (1.11)	3.21 (2.22)	3.01 (2.09)	1.45	1.44
Mahanadi	0.45 (-0.21)	0.88 (0.99)	-1.25 (-2.39)	3.08 (2.30)	1.58 (1.07)	4.98 (1.22)	2.15 (1.99)	4.08	1.08
Krishna	-0.57 (-0.41)	1.03 (0.93)	-2.40 (-2.19)	2.00 (2.22)	1.10 (1.15)	2.57 (2.06)	2.11 (1.90)	1.25	1.11
Middle Ganga	0.33 (0.39)	0.75 (0.86)	-0.89 (-1.28)	1.61 (2.21)	1.42 (1.19)	3.07 (1.15)	2.10 (1.49)	2.67	1.41
Lower Ganga	-0.35 0.18)	0.88 (0.75)	-2.32 (-1.00)	1.58 (2.25)	1.06 (1.09)	3.03 (1.95	2.12 (1.38)	1.55	1.54

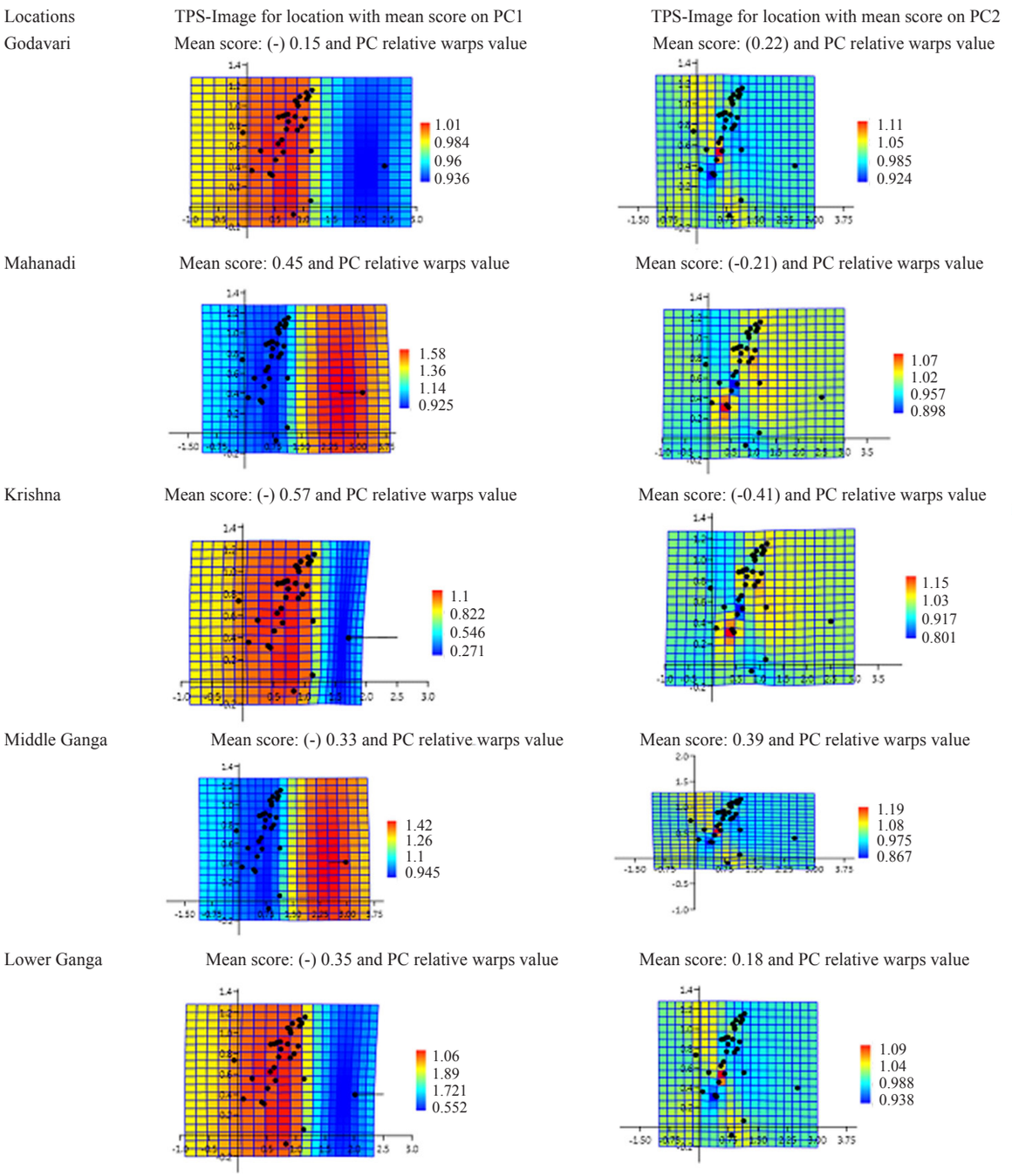

Fig. 6. Thin plate spline geometric shape variations (deformations) observed through principal component analysis - PCA (relative warps) on mean score linked principal components (PC-1, PC-2) for *S. sarana* from five river basins of India

plasticity. All these observations from the present study indicate wide functionality and variation in the caudal fin region between different stocks.

The findings of the present investigation indicated two phenotypically distinct populations of *S. sarana*, from five Indian river basins employing truss variables. However, further molecular studies are needed to correlate the phenotypic variations observed, with genetic variations in *S. sarana* populations.

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