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Can the *Nemipterus japonicus* stocks along Indian coast be differentiated using morphometric analysis?

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This study has been carried out to identify the various stocks of the species, Japanese thread fin bream, *Nemipterus japonicus* (Bloch, 1791), based on their morphometric characters. Fish samples were collected from four locations, two each from west and east coast of India. Twenty one morphometric distances were measured from each individual and the canonical discriminant analysis showed that the body depth distances along with horizontal measurements associated with the head are useful for differentiating these fish stocks along the Indian coast. A discriminant function analysis classified more than 80% of the samples correctly to their respective locations. Results indicated that more than one stock is present in the west as well as east coasts of India. Hence the various spawning stock populations have to be identified and assessed separately in future while formulating management strategies for sustaining the catch from this resource.

[Keywords: Japanese threadfin bream, *Nemipterus japonicus*, morphometrics, stock structure**]**

Introduction

Stock is a subset of species which is defined as an intraspecific group of randomly mating individuals with temporal and spatial integrity[1]. For effective management of a stock and implementation of worthwhile stock rebuilding programmes, it is important to know the identity of the stock structure of that species, as each stock must be managed separately to optimize their yield[2]. Fisheries management can lead to changes in the biological attributes and productivity rates of a species when the stock structure is not taken into account while assessing the population abundances[3, 4, 5]. Consequently, this may lead to overexploitation of the species and could result in a stock collapse[6]. Hence identification of independent fish stocks is important prior to assessment on population abundances as most fisheries models assume that the group of individuals has homogenous vital rates and closed life cycle in which young fish in the group were produced by generations in the same group[7]. Morphometric analysis on fish samples can provide information on phenotypic stocks, groups of individuals with similar growth, mortality and reproductive parameters[8].

Nemipterus japonicus (Japanese threadfin bream) is the most dominant fish species among the threadfin bream resources (Family Nemipteridae) along the Indian coast[9]. This species contributes about 40% of the total production of threadfin breams in India[9]. The species has a continuous distribution along the Northwest, Southeast and Southwest regions of the Indian coast (Fig. 1.)[10]. Traditionally, the stock assessment of this species is carried out based on the spatial distribution of landed catch in different regions/zones[6]. However, no extensive study has been undertaken to identify the number of stocks of this species along the Indian coast and hence the stock assessments conducted along the Indian coast have not considered the stock structure of this species[6]. This may lead to erroneous results and distort the management plans adopted for fisheries through marine policies[11, 12]. Present study attempts to identify the stock structure of *N. japonicus* populations at four sites of the Indian coast using morphometric analysis so that the management strategies for fisheries in future can be designed based on assessments from independent fish stocks.

Material and Methods

Samples of *N. japonicus* from the Indian waters were chosen from four locations, one each from the maritime states of India based on their regional landings distribution. Two locations from the west Mumbai (State of Maharashtra), Cochin (State of Kerala), and two locations from the east coasts of India *viz*., Chennai (State of Tamil Nadu) and Kakinada (State of Andhra Pradesh) were used in the present study (Fig. 1)[13, 14]. Sampling sites were chosen from centres which are apart in latitudinal aspect to avoid mixing up of specimens from the same regions. A total of

389 fish samples were collected from commercial fish landing centres between October 2010 and December 2010 (Table 1).

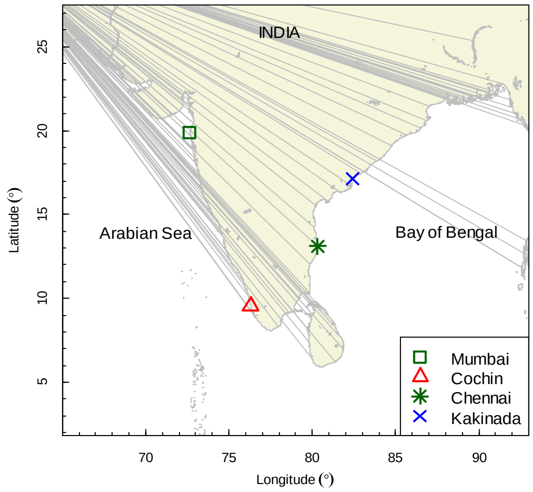


Fig. 1. The figure shows the four locations from which samples of Japanese thread fin bream, *N. Japonicus* were collected. *Source*: *GSHHS shoreline data plotted using PBS mapping R package*[13, 14].

An image of each fish specimen was captured using a cyber-shot DCS-S500 digital camera (Sony, Japan). A total of twenty one morphometric distances were measured along the entire body surface on the left side of the fish i.e., head, trunk and tail (Fig. 2)[15]. These distances were based on morphologically important anatomical locations or points called 'landmarks'. Landmarks were digitized on the image using the software "tpsdig2 V2.1" and the data were encrypted into tps files in the form of X-Y coordinates[16]. Further, the Paleontological Statistics software (PAST) was used to measure the morphometric distances between the pre-determined anatomical landmarks in each fish specimen[17, 18].

The distances measured were first tested for outliers and 20 observations were removed based on Cook's distance estimates using PROC ROBUSTREG procedure of SAS software as they may distort the general tendency in the size distribution[19]. Significant correlations were observed between the body size and the morphometric variables ($p<0.01$) and hence, the variation in the whole data may discriminate the populations based on size of the fish[20]. Therefore, the absolute morphometric variables were first transformed into shape variables that are size-independent. This was employed using an allometric approach using the following formula[1, 21].

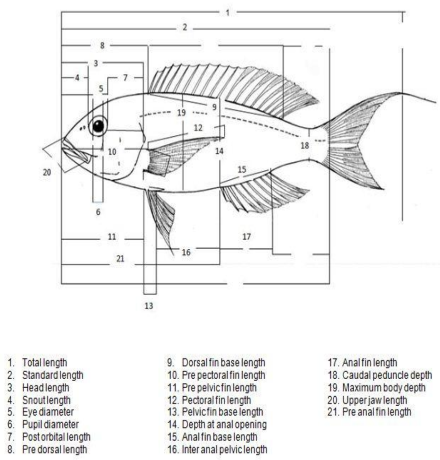


Fig. 2. Morphometric variables of *N. japonicus* used for the present study. *Source of the image: FAO*[15]

$$\mathbf{M_{adj} = M\ (SL_{mean} / SL)^{ß}}$$

Where,

M_{adj} : transformed morphometric measurement

M : original morphometric measurement

SL : standard length of fish

SL_{mean} : Overall mean standard length of the fish

ß : within group slope of the geometric mean regressions of the log M against log SL.

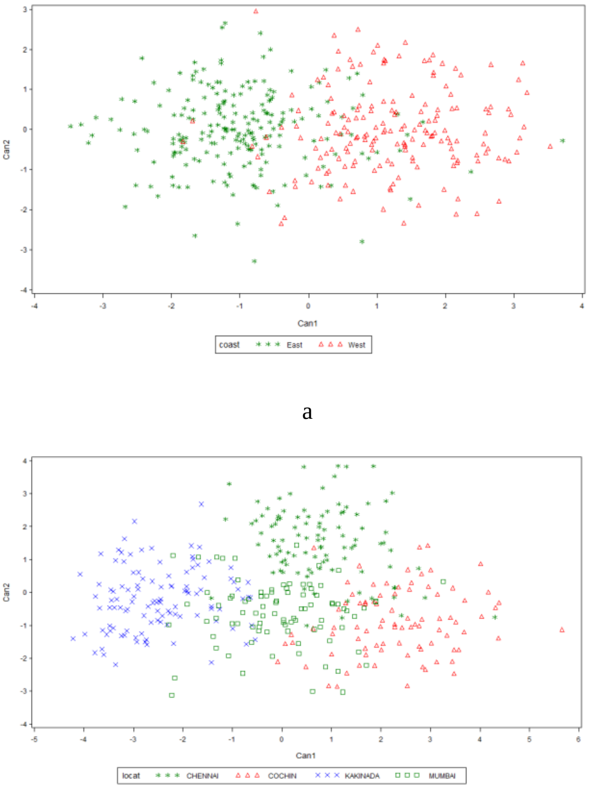
The multinomial Mardia's test was carried out to check whether the data follows a multivariate normal distribution[22]. This was done by subjecting the transformed morphometric measurements to PROC MODEL procedure in SAS[19]. Further, the Canonical discriminant analysis (CDA) was employed, which is a multivariate dimension-reduction technique that finds linear combinations of the quantitative variables and provides maximal separation between categories or groups[23]. Twenty one morphometric measurements were subjected to CDA using PROC CANDISC procedure of SAS, to test two aspects a) whether samples from east and west coast belong to two different stocks b) whether multiple stocks exist within each coast[19]. In the canonical discriminant analysis, the transformed morphometric distances loaded heavily on the first and second canonical axes and therefore they were used to carry out the discriminant function analysis. Variables were selected based on the Hatcher's scratching procedure and the (DFA) was employed using DISCRIM procedure in SAS[19, 24]. For a set of observations containing more than one quantitative variable and a classification variable defining groups of observations, the discriminant function analysis develops a discriminant criterion to classify each specimen into one of the groups which can be locations or coasts, based on the context of analysis[25].

Results

In the present study, the mean standard length of samples from all locations was significantly different from each other (Table 1). Canonical discriminant analysis revealed that there is a significant morphometric variation between individuals obtained from all the different sampling locations and the morphometric distances with high loadings on the first two canonical axes were found useful in distinguishing these samples.

The plot between first and second canonical scores indicates the morphometric discrimination of samples from west and east coast populations (Fig. 3a). Measurements loaded on the first canonical axis were associated to vertical body depth measurements. The distances; depth at anal opening, caudal peduncle depth and maximum body depth were found to have significant loadings on the first canonical axis which explained 65% of the total variation (Fig. 4a). Separation of clusters was on the first canonical axis and hence these distances are useful for differentiating individuals belonging to the west or east coast of India (Fig. 3a). Second canonical axis explained 19.1% of the total variation and represented the horizontal measurements associated with the head i.e., eye diameter, snout length, upper jaw length and head length (Fig. 4b).

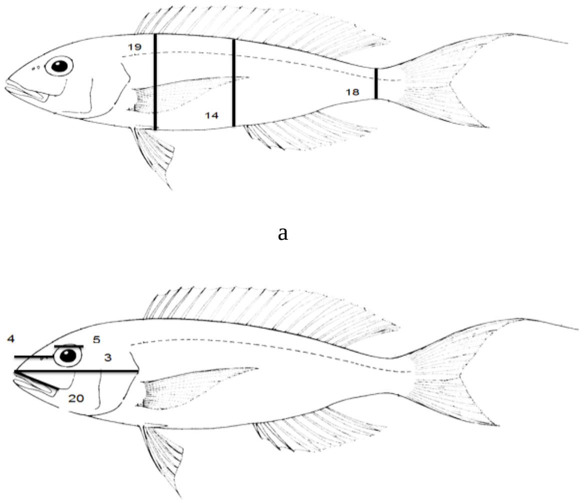
Populations within the west or east coast were found significantly different in CDA since the canonical scores of samples from each location show a clear separation (Fig. 3b). First and second canonical axes were explaining 79.8% (59.8% for first and 20% for second canonical axis) of the total morphometric variation. The variables with high loadings on first canonical axis explained 59.8% of the total morphometric variation and they represented the vertical body depth measurements i.e., depth at anal opening, caudal peduncle depth and maximum body depth (Figure 4a). The second canonical axis gave significant loadings on the horizontal measurements associated with the head i.e., eye diameter, snout length, upper jaw length and head length (Fig. 4b).



a

B

Fig. 3. Comparison of the first and second canonical scores obtained from morphometric measurements of *N. japonicus (*a) differentiation between east and west coast (b) differentiation across the sampling locations



a

b

Fig. 4. Morphometric variables with high loadings on the (a) first and (b) second canonical axes in CDA. *Source of the image: FAO*[15]

The variables loaded on the first and second canonical axes were taken for classifying individuals between the coasts and the four sampling locations. Coast wise discriminant function analysis has given 86.12% of accuracy in classifying the observations to various locations (87.85% for the east coast and 84% for the west coast samples) (Table 1). Location-wise analysis has given overall 80% of classification accuracy i.e., 93.75%, 82.56%, 82.35% and 70.79% for Kakinada, Cochin, Chennai and Mumbai samples respectively (Table 1).

Table 1

Cross-validation of individuals classified by discriminant analysis. Percentage of fish from each location or coast (in rows) classified by discriminant analysis to their respective groups (in columns), `n' indicates the total number of samples collected from the respective locations, SL indicate the mean standard length and the means bearing the same superscript within a subgroup are not significantly different from each other ($p \leq 0.01$).

Coast	n	SL	Coast				Total rate of classification (%)	Total rate of misclassification (%)
			East		West			
East	214	13.2^a	87.85		12.15		86.12	13.88
West	175	14.92^b	16		84			
Location			Location				83.03	16.97
			Chennai	Kakinada	Cochin	Mumbai		
Chennai	102	14.25^a	82.35	0.98	8.82	7.84		
Kakinada	112	12.15^b	0.89	93.75	0.89	4.46		
Cochin	86	16.03^c	10.47	0	82.56	6.98		
Mumbai	89	13.81^d	7.87	13.48	7.87	70.79		

Discussion

To understand the stock structure of *N. japonicus* along Indian coast, samples were collected from four locations along the coast of India i.e., Mumbai (west coast), Cochin (west), Chennai (east coast) and Kakinada (east coast). Morphometric analysis revealed significant phenotypic heterogeneity among the populations of *N. japonicus* and the variables related to the body depth measurements are useful for classifying populations between the different sampling locations. The results indicated that separate stocks of this species may exist along the east and west coasts. In the following sections, we discuss the reasons which could have led to the separation of these populations.

Different stocks may exist due to geographical separation of the fish populations[26]. Japanese threadfin bream has a non-migratory demersal life status and this might have resulted in limited mixing of their populations and thus the formation of geographically isolated fish stocks, particularly the populations from east and west coast of India. However, it is less likely to have several distinct stocks in the same ecological habitat as the probability of individuals sharing same spawning ground will be higher[27]. Previous studies have shown that the northern and southern parts of Arabian Sea as well as the Bay of Bengal hold significantly different ecological habitats[28, 29, 30, 31]. This is because marked variations have been observed in the composition of fish species in the trawl landings from these regions[32]. Thus, geographical isolation along with ecologically different environments can result in the development of significantly different morphological features between fish populations. The interactive effects of environment, selection, and genetics on individual ontogenies lead to morphometric differences within a species[33]. Moreover, previous studies have shown that *N. japonicus* populations along east and west coast differed in their feeding pattern[34]. Obviously, the isolation of the east and west coast stocks could be due to the geographical barrier that reserved the flow of genetic exchange[26]. However, more research is required to determine which ecological factors lead to the formation of distinct stock within the east and west coast of India.

The environmental conditions play a significant role in the development of unique morphometric characters or adaptations in fish populations[34, 35, 36]. Thus the organisms of same species may develop different morphological features in different environments. In the present study, the body depth measurements were found to be significant in discriminating the west coast and east coast individuals. Moreover, many fish samples from the west coast were found to be bigger in size when compared to the east coast[18, 37, 38] (Table 1). This difference may be attributed to the presence of the more extensive continental shelf area on the west coast where upwelling is a common phenomenon resulting in availability of sufficient food required for the growth of individuals[39]. The east coast of India has unique environmental characteristics such as lower salinity, high turbulence and comparatively strong water currents[40]. Previous studies have also confirmed that such environmental features could lead to the formation of phenotypically distinct fish stocks[18, 35, 37]. However, the meristic traits in *N. japonicus* populations didn't show significant difference between east and west coasts of India[41]. In the present study the morphometric characters belonging to the vertical body depth measurements were found to be significant in differentiating the samples from east and west coasts. Similar observations have been reported for other groups of species from the Indian coast. For example, the body depth measurements in *Megalaspis cordyla*[18] and *Decapterus russelli*[38] were significant in differentiating their populations between west and east coasts. Thus it is concluded that the environmental conditions prevailing in the west and east coast of India could have played an important role in the development of morphometric characters in this species.

An important limitation of the present study is the low spatial resolution in the number of sampling sites relative to the species distribution along the Indian coast. However, the chosen location represents most of the important fishing ports where majority of the *N. japonicus* landings occur throughout the year[9]. Hence the results are useful for providing guidelines for the assessment of populations exploited along the Indian coast. The present analysis identified distinct stocks of *N. Japonicus* along the east and west coasts of India, which indicates that these populations have to be managed separately and this will need collaborative efforts between state governments within India. Future studies may use other stock identification tools, such as life history traits, fatty acid signatures, otolith chemistry, osteology, molecular tools and tagging studies, to validate the results obtained in this study [42, 43, 44, 45, 46].

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