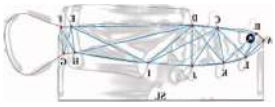
CHAPTER 8

Truss Networking: A Tool for Stock Structure Analysis



In fisheries management, the term 'stock' refers to a sub-set of a particular fish or shellfish species inhabiting a particular geographical area with the same growth and mortality parameters (Gulland, 1983). Stock structure means the contribution of stock units that represent the entire population. Fish stocks may be considered as subpopulations of a particular species of fish, for which intrinsic variables (growth, recruitment, mortality and fishing mortality) are the only significant factors in determining stock dynamics, while other factors, particularly immigration and emigration, are considered to have limited effect. Each population stocks usually characterized by the specific biological attributes (Secor, 2014). The differences can be seen through phenotype, genetic (Aini et al., 2020), or both simultaneously (Hollander and Butlin,2010). Stock identification is a field of fisheries science which aims to identify these sub-populations, based on a number of techniques involving an interdisciplinary approach (Cadrin et al., 2005).

Information on stock identity and spatial structure provide the basis for understanding fish population dynamics and enable reliable resource assessment for fisheries management (Reiss et al., 2009). Attempt to manage fisheries resources cannot be generalized in each region. Each stock may have unique demographic properties and responses or rebuilding capabilities when faced with exploitation. The biological attributes and productivity of the species may be affected if the stock structure considered by fisheries managers is erroneous (Smith et al*.,* 1991).

The major objective of stock assessment programs is to manage fishery resources by providing advice on the optimum exploitation (Sparre and Venema, 1998). Thorough knowledge of the stock structure of the target species in commercial fisheries forms the basis to formulate resource management strategies (Shaklee and Bentzen, 1998). If the stock structure is not considered while formulating plans for fisheries management, it can lead to the collapse of the population due to the changes in biological attributes and loss in productivity rates (Begg et al., 1999; Cadrin, 2005). Stock structure analysis is, therefore, a pre-requisite for developing fishery management plans to understand the existing levels of recruitment that may replenish the population (Cardrin et al., 2005).

A variety of body shapes developed because of variability in growth, development and maturation in individuals belonging to one or different populations of a species of fish (Cadrin, 2000). There are reports of multiple stock compositions in fish populations (Pepin and Carr,

1993; Serajuddin et al., 1998). Environmentally induced phenotypic variation provides rapid information on stock or subpopulation identity (Clayton, 1981). The study of morphometrics using truss network is a quantitative method to represent the complete shape of the fish (Strauss and Bookkstein 1982).

This representation is formed by interlinking the measurements between morphometric landmarks that give rise to a systematic pattern of connected cells covering the entire body structure (Turan 1999) which has been successfully used for population and taxonomic studies (Lin et al. 2005; Mevlut et al. 2006). Stock identification by truss network analysis is practically useful and an effective strategy for the description of the body shape in comparison to the traditional morphometric method (Cadrin 2005). It is effectively used to discriminate the stocks and differentiate between the population's shapes (Stratuss and Bookstein 1982).

A large number of studies using the box-truss network method gave better results in categorizing individuals accurately and classifying them to their intraspecific groups (Turan, 1999). In particular, the truss is a landmark-based technique that poses no restriction on the direction and localization of change in shape and is highly effective in capturing data on the shape of the organism (Cavalcanti et al., 1999). Phenotypic characters have been successfully used for stock differentiation in many shrimps, *Macrobrachium vollenhovenii* (Konan et al., 2010), *Macrobrachium nipponense* (P-C Chen et al., 2015) and fish species *viz*., *Decapterus russelli* (Sen et al., 2011), *Harpadon nehereus* (Pazhayamadom et al., 2015), *Sardinella longiceps* (Remya et al., 2015), and *Nemipterus japonicus* (Sreekanth et al., 2015) while homogeneity was reported in the population of *Farfantenaeus notialis* at Caribbean sea (Paramo and Saint-paul, 2010). Homogenous fish populations are often composed of discrete stocks which may have unique demographic properties and responses to exploitation, which should be managed separately to ensure sustainable fishery benefits and efficient conservation (Kinsey et al., 1994; Begg and Brown, 2000; Stransky et al., 2008; Neves et al., 2011).

A Case Study: Deep-sea Shrimp: *Aristeus alcocki*- Penaeid shrimp

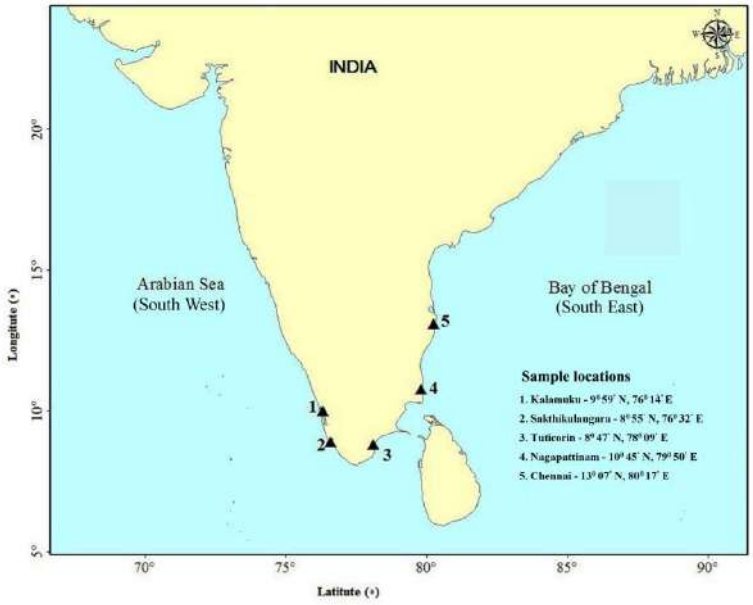
A. alcocki Ramadan, 1938 (Decapoda, Aristeidae), commonly known as Red Ring or Arabian red shrimp is distributed along the southern Indian coast at a depth range of 200-1000 m (Silas 1969; Suseelan 1989; Madhusoodana 2008; CMFRI 2015). It forms a commercial fishery confining only along the southeast and southwest coast, and it's not recorded along the northern coast of India (Mohamed and Suseelan, 1973). The catch landed between 2008 and 2015 indicate that the *A. alcocki* is the prime species in order of biomass among the deep sea penaeid catch accounting to about 36% from the whole Indian coast and the trend in catch rates indicates a decline of these deep-sea shrimps (CMFRI 2008-2015). In this study we aim to investigate the effectiveness of the truss variables in differentiating the populations of *A.alcocki* along the Indian coast using truss morphometry, to provide management advisory for fisheries sustainability.

Sampling

Samples of *A. alcocki* were collected from five different fishing harbors i.e., Tuticorin (SEN), Chennai (SEC), Nagapattianam (SEN) on the southeast, and Sakthikulangara (SWS), Kalamuku (SWK) on the southwest Indian coast (shown in figure below). The sampling sites were chosen such that they are distantly apart in latitudinal aspect to reduce the chances of mixing specimens from the same population. In total, 1842 specimens were collected from the selected sampling sites i.e., from commercial fishing harbors where the catch is landed by

multiday trawlers along the southern coast during December 2014 and January 2015. The samples were collected during peak breeding season (November to January) to ensure that they represent to their parent population.

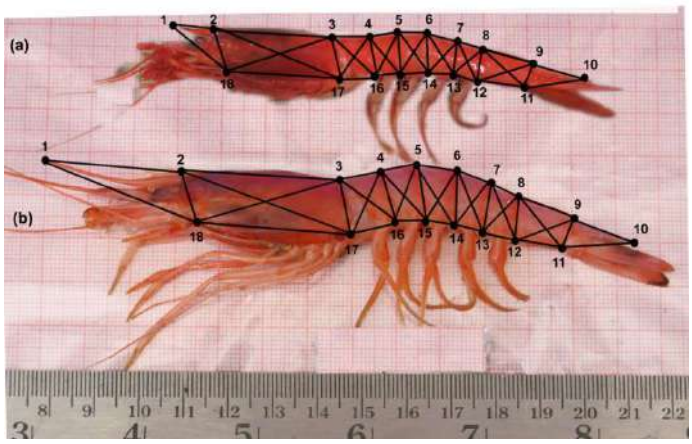
The matured specimens (carapace length: female>3.5 cm; male: >2.0 cm) were sorted from the samples collected from each fishing location and used for truss morphometric analysis. The species exhibit a high degree of sexual dimorphism where males were identified by the presence of petasma and females were sorted based on the presence of thelycum. Specimens showing physical damage *viz*., broken rostrum or any other body parts may distort the shape characteristics and hence they were not included in the samples for the study.



Digitization of specimens and fixing anatomical landmarks

Shrimp samples were first cleaned with running water, allowed the water to drain, wiped with tissue paper and finally placed on a graph paper (shown in figure below). Each specimen was placed on a flat platform with a graph paper over a thermofoam, appendages (pereiopods and pleopods) and telson were erected by positioning the rostrum portion towards the left side, telson on the right by assuming symmetry between left and right side of the shrimps and was labeled with a specific ID code. This helps us in identifying specimens if more landmarks are required to be fixed or if the morphometric measurements are to be repeated. Digital images of the specimens were captured using a camera (Canon G-15) which was fixed on a tripod stand directly above the specimen and the lens was adjusted so the margins of viewfinder align with margins of the graph paper in X-Y directions and each image included a scale to standardize the individual sizes and further scaling was applied in tpsdig utilizing the millimeter grid in graph paper. These images were used further in fixing the anatomical landmarks and measuring linear distances between them i.e., truss variables. In many previous studies, it has been found that differences in sex are likely to contribute to shape differences affecting total variance in morphometric distances (Reiss and Grothues, 2015; Sajina et al., 2011; Pazhayamadom et al., 2015). In the present analysis, both males and females were included to accommodate the effect

of sex on their morphometry. The extraction of numeric truss distances from the digital images of specimens were carried out by using two software platforms, 1) tpsDig2 V2.1 for marking the landmark coordinates on the digital images (Rohlf, 2006 and 2) paleontological statistics (PAST) for extracting the values pertaining to the marked distances (Hammer et al., 2001). The data extracted by this method ensures stability, accuracy, and repeatability.



Analysis of truss morphometric data

MANCOVA was carried out in order to study the statistically significant differences among sex, location using log-transformed data and carapace length (CL) was incorporated into the models as a covariate. Data sets were standardized by log transformation and tested for normality by SAS PROC UNIVARIATE procedure for removing outliers. An allometric method was adopted to remove size-dependent variation in morphometric characters.

The normality and homogeneity variance assumptions were verified with the log-transformed data, using the SAS PROC UNIVARIATE procedure (SAS 2014), and the data rows with outliers (7-10%) were removed from each location, before proceeding further for analysis. MANCOVA was used to establish significant differences among sex, location using log-transformed data and carapace length (CL) was incorporated into the models as a covariate. Therefore, the whole truss measurements were transformed to size-independent shape variables using an allometric method as suggested by Reist (1985) in Equation 1.

$M_{trans} = \log M - \beta (\log CL - \log CL\ mean)$ Equation 1,

Where M_{trans} is the truss measurement after transformation, M is the original truss measurement, CL is the carapace length of the shrimp which is reported to be more reliable than using total length (TL) in the case of crustaceans (FAO 1974), CL mean is the overall mean carapace length, and β is the slope regressions of the log M against log CL.

Correlation coefficients were checked between each pair of variables before and after the size effect removal. In such analysis, the absolute values of correlation coefficients were expected to decrease after size effect removal (Murta, 2000). Mean (X), standard error (SE), standard deviation (SD), maximum and minimum of all measurements were recorded for each population. The percentage of coefficient of variation (CV%) was computed as CV% =

100×SD/X of morphometric variables in each population. Multivariate analysis used in this study consisted of principal component analysis (PCA), discriminant functions (DF) and hierarchical cluster analyses.

PCA was used to evaluate morphometric variation among specimens and identify variables contributing substantially to that variation. DF was run to test the effectiveness of variables in predicting different group locations (Tomovi´c and D˘zuki´c 2003; Loy et al., 2008). The stepwise inclusion procedure was carried out to reduce the number of variables and identify the combination of variables that best separates the groups (Jain et al., 2000; Poulet et al., 2005, Hair et al. 1996). Hierarchical cluster analysis (HCA) based on Mahalanobis distances matrices determined with DF, was used to evaluate population relationships, as implemented by Slabova and Frynta (2007) and Ferrito et al. (2007). All the analysis in the present study was done by using Statistical Analysis System software (SAS 2014).

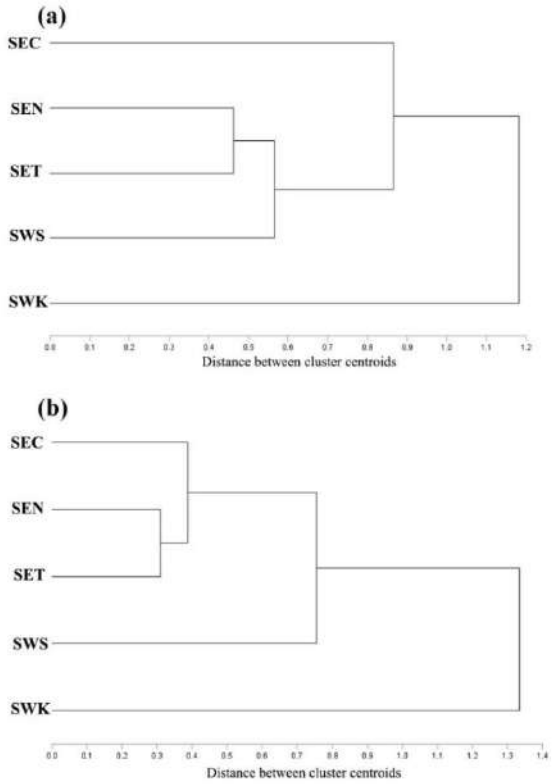
Results

Descriptive statistical results showed less coefficients of variation (CV) (<25%) in all the truss variables for both the sex at five different locations (Table 2). The range of CV for female varied from 7.6 to 20% and for male was 4.9 to 21.6%. The morphometric variability within populations was low for all the locations.

Correlation coefficients between the morphometric variables were estimated before and after the size effect removal. Before the size effect removal coefficient values were highly significant while it was reduced after the correction which suggested that the effects of size had been effectively removed from the morphometric data. The mean carapace length specifies that the males are much smaller than females, a significant difference on sex and location was observed.

The results of PCA analysis indicate that the first two components cumulatively explained >70% (female: 72.1%; male: 71.5%) of the total morphometric variation. A few truss distances loaded heavily on PC1 (1-2, 1-18, 2-18, 3-17, and 5-15) which alone explained >63% of the entire variance. The loadings of two variables i.e., the 1-2 distances that correspond to the rostral length and the 1-18 distances that connects the rostrum tip to the pterygostomian spine contributed a substantial proportion of the total variance. PC2 explained 8.21% of the total variation, and 3 distance variables (3-4, 15-16, and 4-17) corresponding to the abdominal region of the shrimp loaded heavily on this component. The distances with high loadings on both PC1 and PC2 characterize the rostrum and 2nd to 3rd abdominal segment portion of the shrimp and they all were found to be positive, signifying the positive correlation between the variables within a component i.e., these attributes grow in proportion with one another. A scatter plot between PC1 and PC2 resulted in the separation of SWK from other populations.

The results of hierarchical cluster analysis showed two distinct groups from five populations of both sexes. The group-I included SWK population and SWS, SET, SEN, SEC populations clustered in group-II. This analysis showed that SWK samples constituted phenotypically a separate population, while the morphometric resemblance between SWS, SET, SEN and SEC stocks were found to be high. The analysis of the present study revealed that the variables used in this study were capable to clearly differentiate SWK population from the other group.



Genetic Characterisation of the species

Genetic variation is considered to be an important feature of the population to reveal not only the short term fitness of individuals but also the long term survival of the population, through allowing adaptation to the changing environmental conditions. Information deduced from molecular markers can provide insight into genetic structure and geographical boundaries (*i.e.* breeding stock) and vulnerability (*i.e.* genetic diversity) of the species (Buchholz-Sørensen & Vella, 2016).

Molecular markers have been proved to be an effective indicator of genetic variation within and between fishery populations of shrimp species; *Aristeus antennatus* (Maggio et al., 2009; Cannas et al., 2012; Fernández et al., 2011b; Brutto et al., 2012), *Aristaeomorpha foliacea* (Fernández et al., 2011a), *Penaeus monodon* (Mandal et al., 2012; Sekar et al., 2014) and *Fenneropenaeus indicus* (Sajeela et al., 2015). Microsatellite markers are characterized as co-dominant and highly polymorphic in nature and addition to their abundance, even genomic distribution, small locus size, have quickly become useful molecular markers with great discriminatory power for the evaluation of genetic diversity in various species (Powell et al., 1996). Analyses of microsatellite nuclear markers were used to describe the differences and distribution patterns of natural populations of this species.

DNA extraction, amplification and genotyping of microsatellite loci

The total genomic DNA was extracted from the pleopod of the each individuals using DNeasy® Blood & Tissue Kit (Qiagen Inc.) according to the manufacturer's protocol. The cells were

lysed by incubating at 56^0C for 2 hrs and all other steps were followed as per the protocol. The primers for nine nuclear microsatellite loci were taken from Cannas et al. (2008), were originally designed for the *Aristeus antennatus.* The microsatellite loci were optimised for genotyping by following the general protocols of Palumbi (1996), and Cannas et al. (2008). The amplification of microsatellite markers were performed in 25 µl reaction cocktails containing genomic DNA (0.5 µg μl^{-1}), *Taq* DNA polymerase (0.05 U μl^{-1}), 1X buffer, MgCl2 (1.5 mM), 10 pM μl^{-1} of each primer and dNTPs (200 µM). The PCR thermal profile used was 94^0C for 5 min for initial denaturation, followed by 35 cycles of 94^0C for 1 min, annealing at 52–54^0C for 1 min, extension at 72^0C for 1.5 min, and a final extension at 72^0C for 5 min (Table 1). Amplification of PCR products were confirmed by electrophoresis on a 1.5% agarose gel containing ethidium bromide and visualized under UV transilluminator (Lark, India). Analysis of fragment size was carried out by ABI prism genetic analyser (Applied Biosystems, USA) at AgriGenome Labs, Scigenom, Cochin, India.

Data analyses

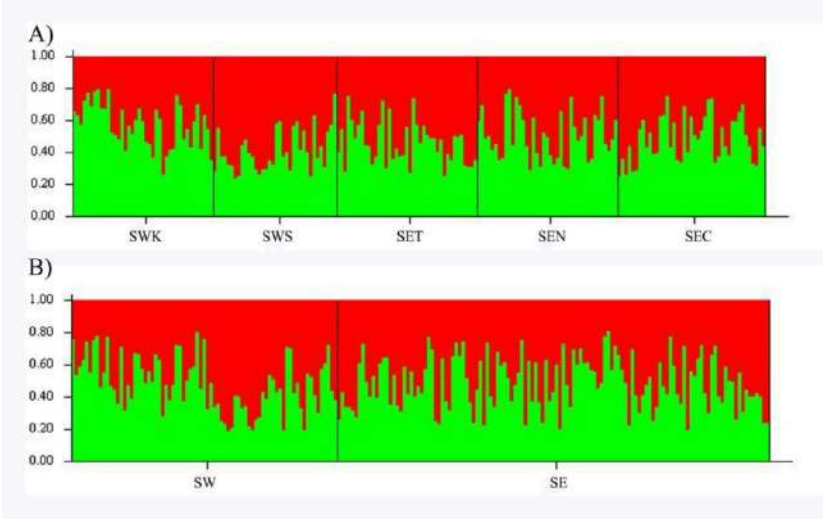
Allele frequency, the number of alleles (Na), observed (Ho), expected (He) heterozygosity and unbiased expected heterozygisity (UHe) per locus and locations were calculated with the computer program GenAlEx v. 6.41 (Peakall and Smouse, 2006). GENEPOP 4.0 package (Raymond and Rousset, 1995) was used to calculate deviations from Hardy-Weinberg equilibrium (HWE) for each locus and linkage disequilibrium between pairs of loci by using Fisher's exact test, under Markov Chain Monte Carlo (MCMC) algorithms (Guo and Thompson, 1992), with 1000 dememorizations, 100 batches (treatments per location) and 10000 iterations per batch. Significance levels for both determinations were adjusted with the Bonferroni test for multiple comparisons with a significance level of $p < 0.05$ (Rice, 1989). F_{IS} (Weir and Cockerham (1984) was calculated in GENEPOP 4.0 (Raymond and Rousset, 1995) with significance values for each locations. The presence of null alleles was tested with MICROCHECKER v 2.2.3 (Van Oosterhout et al., 2004). The F_{ST} values, relative to the null alleles and confidence intervals with and without correction were estimated with FREENA program (Chapuis and Estoup, 2007), if comparison of estimated F_{ST} values denoted significant difference, then any locus shows presence of null alleles in the sample should be

discarded. Polymorphism information content (PIC) for each locus and locations were calculated using PIC –Calc 0.6 software (Nagy et al., 2012). ANOVA F statistic was used to detect the differences among the locations with the means values of Ho and UHe.

To assess the genetic variation on among the populations and between the locations, pairwise Fst values were calculated and followed by statistical assessment of significance with 10,100 permutation steps for every comparison. Hierarchical analysis of molecular variance (AMOVA) was carried out using the program ARLEQUIN 3.5 (Excoffier & Lischer, 2010) to assess the presence of differential genetic structure. We performed a Bayesian cluster analysis to infer population structure and estimate the number of genetically distinct populations, using STRUCTURE v.2.2.3 (Pritchard, Stephens, & Donnelly, 2000) to determine the probabilistic assignments of samples based on genotypes to K sub populations. K estimation was completed using 20 independent simulations for K=1 to 5 with 100,000 MCMC iterations and 10,000 batches. The most probable estimation of groups in the current dataset was done by using the ad hoc statistic DK method proposed by Evanno et al. (2005) and the value of K best fitting the data was selected using the log posterior probability of the data for a given K, Ln Pr (XjK) (Pritchard & Wen, 2004).

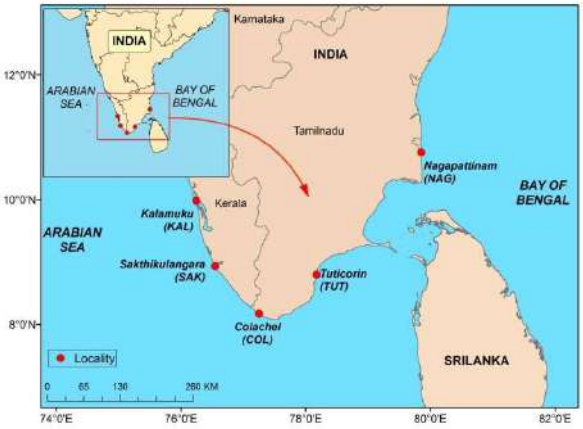
Molecular Results

The pairwise F_{ST}, Nei, and AMOVA values calculated from microsatellites indicated the absence of significant variation among the samples of *A. Alcocki* collected from the South west (Arabian Sea) and South east (Bay of Bengal) coast of India. Moreover the results of AMOVA also indicated the proportion of genetic variation was mainly associated to differences among the individuals (99.2%) with F_{st}=0.0078 which is further confirmed by the cluster analysis performed using STRUCTURE (shown in figure below) directed towards the presence of homogeneous groups due to the absence of specific allelic variation in the sampled localities. The present study was in agreement with the results reported in *A. Antennatus* (among individual difference 99.3%; Fst=0.0067) using same markers in the Mediterranean Sea (Cannas et al., 2012) where no genetic differentiation was noticed between the localities.

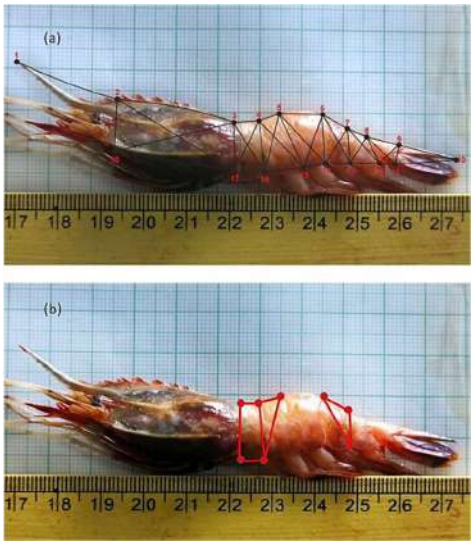


Case Study –II: *Heterocarpus chani*: A caridean deepsea shrimp

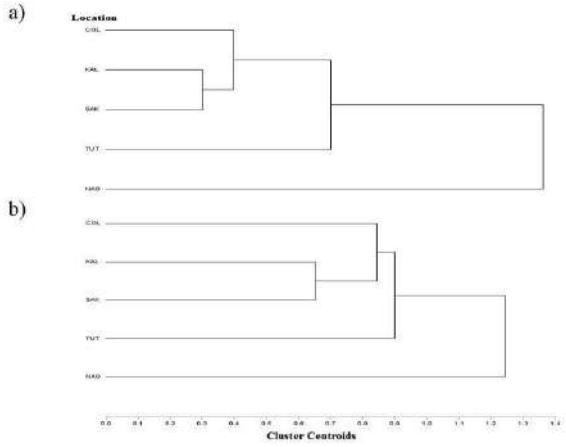
The samples of *H. chani* were collected from deepsea trawl shrimp catches obtained from five major fishing harbours along the southern coast of India. The sampling sites are Kalamuku (KAL), Sakthikulangara (SAK), Colachel (COL) on the southwest coast and Tuticorin (TUT), and Nagapattinam (NAG) on the southeast coast. Information on study sites, geographical coordinates, shrimp sex and the sample size from each location. A total of 1879 specimens of *H. chani* including 984 males and 895 female individuals were used in this study.



Digitisation of samples



The results of HCA showed three clear clusters from five populations of both sexes as shown below figure. The group-I included populations from NAG, group-II consisted of the TUT and group-III with SAK, KAL, and COL populations. Theinterpretation of resultsindicated that the samples obtained from the locations NAG and TUT represented a phenotypically distinct population while the morphometric resemblance between SAK, KAL, and COL stocks were observed to be high.



Conclusion

The truss morphometric characters in *A. alcocki* and *H.chani* can be efficiently used in the discrimination of populations as studied in other species of freshwater and marine environments. The major discriminating variable to differentiate the populations into two groups was attributed to the abdominal measurements, suggesting a need to adopt separate management strategies for the resource sustainability and policy regulations. Further, studies

based on the genetic markers in *A. alcocki* indicated the presence of single population. However, in *H.chani* molecular studies can be used to validate the findings of this study.

References

- A.O.Anyanwu and O.A. Ugwumba, *The Zoologist* 2: 70-77 (2003). 1997).
- Aini NK, Mashar A, Madduppa HH, Wardiatno Y. (2020). Genetic diversity of horseshoe crabs (*Carcinoscorpiusrotundicauda*and *Tachypleus gigas*) in Demak, Madura and Balikpapan waters based on Random Amplified Polymorphic DNA marker. *Journal of Natural Resources and Environmental Management.* 10(1): 124-137. doi: 10.29244/jpsl.10.1.124-137
- Bhosale MM, Pawar RA, Bhendarkar MP, Sawant MS, Pawase AS. (2018). Truss based morphometric approach for the analysis of body shape in portunid crabs (*Charybdis feriatus, P. pelagicus*and *P. sanguinolentus*) along Ratnagiri coast, India. *Journal Entomology Zoology Studies*. 6(2): 2641-2648.
- Booke, H.E., (1981). The conundrum of the stock concept—are nature and nurture definable in fishery science? Can. J. Fish. Aquat. Sci. 38, 1479–1480.
- Bookstein, F.L., Chernoff, B., Elder, R.L., Humpheries, J.M., Smith, G.R., Strauss, S.E., 1985. Morphometrics in Evolutionary Biology, vol. 15. Acad. Natl. Sci. Philadelphia Spec. Pub, p. 277.
- C. Turan, S. Yalcin, F. Turan, F. Okur and I. Akyurt, *Folia.Zool.* 54: 165-172 (2005).
- Cadrin, S. X., Friedland, K. D. & Waldman, J. R. (2005). *Stock Identification Methods: Applications in Fishery Science*. Amsterdam: Elsevier Academic Press.
- Cavalcanti, M.J., Monteiro, L.R., Lopez, P.R.D., (1999). Landmark based morphometric analysis in selected species of Serranid fishes (Perciformes: Teleostei). Zool. Stud. 38 (3), 287–294.
- Clayton, J. W. (1981). The stock concept and the uncoupling of organismal and molecular evolution. *Canadian Journal of Fisheries and Aquatic Sciences* 38, 1515–1522. doi: 10.1139/f81-204
- FAO (Food and Agriculture Organization). (1995). *Code of Conduct for Responsible Fisheries*. Rome (IT): FAO Technical Guidelines for Responsible Fisheries.
- H.J. Meng and M. Stocker, *Can.J.Fish. Aquat.Sci.* 41: 414-422 (1984).
- Hart PJB, Reynolds JD. (2002). *Handbook of Fish Biology and Fisheries: Fish Biology.* Oxford (UK): Blackwell Publishing.
- Hollander J, Butlin RK. (2010). The adaptive value of phenotypic plasticity in two ecotypes of a marine gastropod. *BMC Evol Biol.* 10(1): 333-340. doi: 10.1186/1471-2148-10-333.
- J. E. Eyo, *The Zoologist* 2: 1-17 (2003).
- J.A. Fowler, *Quart. Res. Bio.* 45, 148-167 (1970).
- L.A. Krumhoitz and S.H. Cavanah, *Trans.Am.Fish.Soc.* 97: 429-441 (1968).
- Ganesan, Kuberan & Chakraborty, Rekha & Paramasivam, Purushothaman. (2020). Phenotypic variation using truss network system in the deep-sea shrimp Heterocarpuschani Li, 2006 (Caridea: Pandalidae) off Arabian Sea and Bay of Bengal. 1839-1847.
- M. J. Cavalcanti, L.R. Monteiro and P.R.D. Lopez, *Zool Stud* 38(3):287–294(1999)
- M.C. Fabrizio, *Trans. Am. Fish. Soc.* 116: 728-736 (1987).

- Marini M, Suman A, Farajallah A, Wardiatno Y. (2017). Identifying *Penaeus merguiensis*de Man, 1888 stocks in Indonesian Fisheries Management Area 573: a truss network analysis approach. *AACL Bioflux*. 10(4): 922-935.
- Mojekwu TO, Anumudu CI. 2015. Advanced techniques for morphometric analysis in fish. *J Aquac Res Develop*. 6(8): 1-6. doi: 10.4172/2155-9546.1000354.
- of marine fish: mismatch between biological and fisheries management units. *Fish and Fisheries* 10, 361–395. doi: 10.1111/j.1467-2979.2008.00324.x
- P. Pepin and S.M. Carr, *Can.j.Fish.Aquatic Sci.* 50: 1924-1933 (1993).
- Pawson, M. G. & Jennings, S. (1996). A critique of methods for stock dentification in marine capture fisheries. *Fisheries Research* 25, 3–4.
- Pazhayamadom DG, Chakraborty SK, Jaiswar AK, Sudheesan D, Sajina AM, Jahageerdar S. (2015). Stock structure analysis of "Bombay duck" (*Harpadon nehereus*Hamilton, 1822) along the Indian coast using truss network morphometrics. *J ApplIchthyolo.* 31: 37-44. doi: 10.1111/jai.12629.
- Rawat S, Benakappa S, Kumar J, Naik K, Pandey G, Pema CW. (2017). Identification of fish stock based on truss morphometric: a review. *J Fish Life Sci.* 2(1): 9-14.
- Rebello VT, George MK, Paulton MP, Sathianandan TV. (2013). Morphometric structure of the jumbo tiger prawn, *Penaeus monodon* Fabricius, 1798 from southeast and southwest coasts of India. *J Mar Biol Assoc India*. 55(2): 11-15. doi: 10.6024/jmbai.2013.55.2.01784-02.
- Reiss, H., Hoarau, G., Dickey-Collas, M. & Wolff, W. J. (2009). Genetic population structure
- Sajina AM, Chakraborty SK, Jaiswar AK, Pazhayamadam DG, Sudheesan D. 2011. Stock structure analysis of *Megalaspiscordyla*(Linnaeus, 1758) along the Indian coast based on truss network analysis. *Fish Res*. 108: 100-105. doi: 10.1016/j.fishres.2010.12.006.
- Secor DH. (2014). *The Unit Stock Concept: Bounded fish and fisheries. In: Stock Identification Methods.* San Diego (US): Academic Press.
- Sen S, Jahageerdar S, Jaiswar AK, Chakraborty SK, Sajina AM, Dash GR. (2011). Stock structure analysis of *Decapterusrusselli* (Ruppell, 1830) from east and west coast of India using truss network analysis. *Fish Res*. 112: 38-43. doi: 10.1016/j.fishres.2011.08.008.
- Smith, P. J., Francis, R. I. C. C. &McVeagh, M. (1991). Loss of genetic diversity due to fishing pressure. *Fisheries Research* 10, 309–316. doi: 10.1016/0165-7836(91)90082-Q
- Strauss, R.E., Bookstein, F.L., (1982). The truss: body from reconstructions in morphometrics. *Syst. Zool*. 31, 113–135.
- Turan, C., (1999). A note on the examination of morphometric differentiation among fish populations: the Truss System. *Turk. J. Zool*. 23, 259–264.