STOCK IDENTIFICATION OF SELECTED FRESHWATER FISH SPECIES INHABITING THREE RIVERS OF THE GANGETIC RIVER SYSTEM

ABSTRACT

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ABSTRACT

The aquatic ecosystems are facing threat due to ever increasing demand for fish as source of white meat and the rapid overall development. Freshwater environment is degraded due to urbanization, construction of dams, abstraction of water for irrigation and power generation, pollution, siltation, and global warming which consequently put fishes in stressful condition and causing negative effects on its genetic biodiversity particularly in rivers. Catch statistics over many years show a declining trend both in quantitative and qualitative terms for riverine catches. The wild populations of the fish have shown declining trend over the years but there is paucity of published information on the population structure. Determination of the population structure of exploited fish species is an essential component in the successful management of fisheries. However, a comprehensive knowledge on the stock structure is necessary for proper management and conservation of the target fish species. Examination of phenotypic characters is very useful for exhibiting intraspecific variation within a population as the adaptation of a species to its environment is most easily comprehended by fish morphometry. Phenotypic characters are useful for studying short term environmentally induced variation in fisheries management and organism with the same morphometric characteristics are often assumed to constitute a stock and morphometric variations between stocks can provide a basis for stock structure. To overcome the inherent weaknesses of traditional morphometric methods, the truss network system has been increasingly employed for the purpose of stock identification. Otoliths, being metabolically inert and keeping permanent environmental record, have been increasingly used to identify the discrete stocks of the fish. The present study was undertaken to identify different stocks of the stinging catfish, Heteropneustes fossilis, snakehead fish, Channa punctata, walking catfish, Clarias batrachus, freshwater shark, Wallago attu and giant river catfish, Sperata seenghala inhabiting the Ganga, Yamuna and Gomti Rivers of India using truss network analysis of the entire body shape and the elemental profile of sagittal otoliths. In H. fossilis eleven morphometric landmarks were chosen which determined the 23 interlandmark distances while ten landmarks were chosen in C. punctata which determined the 21 interlandmark distances to construct truss network system on fish body. A total of 23 distance measurements
were taken in *C. batrachus* and *W. attu* using the 11 morphometric landmarks to construct the truss network on the fish body while a total of 28 distance measurements were taken using the 13 morphometric landmarks to construct the truss network in *Sperata seenghala*. Additional data such as total length (TL), standard length (SL), head length (HL), post orbital length, (POL), eye diameter (ED), body depth (BD), pectoral fin length (PFL) and ventral fin length (VFL) were also recorded. Inductively coupled plasma-atomic emission spectrometry (ICP–AES) was used to estimate concentrations of 12 trace elements (Ca, Na, Mg, Sr, Ba, Mn, Fe, Cu, Pb, Ni, Zn and K) in the otoliths of *H. fossilis, C. batrachus* and *S. seenghala*. However in *C. punctata*, the elements Mn, Fe, Ni and Zn were not analyzed. Inductively coupled plasma mass spectrometry was used to analyze twelve elements viz, Ca, Na, Mg, Sr, Ba, Mn, Fe, Cu, Pb, Ni, Zn and K in the otoliths of *W. attu*. In order to eliminate the size effect, all the truss morphometric measurements were standardized to eliminate any variation resulting from allometric growth. Further, the standardized data were subjected to appropriate statistical treatments (Univariate Analysis of Variance, Principal Component Analysis and Discriminant Function Analysis) in order to obtain clear inference. The otoliths chemistry data were standardized to remove the effect of otolith weight/fish length on the elemental concentrations of otolith. The transformed data were subjected to Univariate Analysis of Variance, Multivariate Analysis of Variance and Discriminant Function Analysis. In truss morphometry, the univariate ANOVA showed significant (P<0.001) differences between the means of four samples for 25 of 29 morphometric measurements in *H. fossilis*, 23 of 27 morphometric measurements in *C. punctata*, 24 of 30 morphometric measurements in *C. batrachus*, 27 of 31 morphometric measurements in *W. attu* and 30 of 36 morphometric measurements in *S. seenghala*.

In *H. fossilis*, seven principal components showed eigenvalues greater than 1 using standardized morphometric measurements and explained 82.57% of the total variance in the PCA. The first PC (PC-I) accounted for 39.1% of the total variance. The second PC (PC-II) accounted for 13.4% and the third PC (PC-III) for 10.1% of the total variation. The contribution to the PC-I was noted from the morphometric measurements ED, HL, 1-3, 1-2, 5-6, 4-5, BD, 3-6, and 5-8. The DFA revealed that the first DF (DF I) accounted for 59.7% of the total variation, while DF II and DF III, respectively accounted for 25.5% and 14.8% of the group variability among
populations. The morphometric measurements, 1-3, 5-8, 6-7, 5-6, 7-8, ED, BD, 1-2, HL and 4-6 contributed to DF I while 4-5, 8-9, 10-11, 7-9, 3-6, 8-10, 7-10, 5-7, 3-5 etc. contributed to DF II. The scatter plot of DF I vs. DF II explained 85.2% of the total variance among samples and formed three different groups. Samples from the Narora and Kanpur populations on the Ganga River formed one group, while the two other groups were separately comprised of fish from the Yamuna and Gomti Rivers. The overall allotment of individuals to their original population was high (72.3%). The percentage of correctly classified fish was found to be highest in the Yamuna River (83.3%) followed by those in the Gomti River (81.8%). Elemental concentrations in otoliths differed (MANOVA, P<0.01) among the fish samples collected from the four sites of the selected rivers. Eight of 12 elements analyzed exhibited significantly (ANOVA, P<0.001) different mean values. Otoliths from Kanpur fish population on the Ganga River showed significantly higher concentrations of Ca, Sr, Mn, Cu, and Zn compared to otoliths from the fish of other sites (ANOVA, P<0.001). A significantly higher (P<0.001) concentration of Ba was found in otoliths collected from Narora fish population on the Ganga River, while Pb was found to be significantly higher (P<0.001) in fish otoliths from the Yamuna River. Nickel was present in significantly higher (P<0.001) concentrations in otoliths of fish collected from the Gomti and Yamuna Rivers. The DF I accounted for 76.1% of the total variation. The DF II and DF III, respectively accounted for 17.9% and 6% of the group variability among populations. Allocation success was high for all populations with 98.7% of individuals being correctly classified into their original group. A graphical representation of the first two DFs revealed a clear separation of the Narora, Kanpur, Firozabad and Lucknow populations. Percentage classification accuracy showed significant variation (t = -3.92, P<0.05) between allocation success of H. fossilis population obtained by the two methods, truss morphometry and otolith chemistry.

In Channa punctata, six principal components showed eigenvalues greater than 1 using standardized morphometric measurements and explained 77.17% of the total variance in the PCA. The first principal component accounted for 47.75% of total variance. The second principal component accounted for 8.18%, and the third principal component accounted for 7.48% of the total variation. The high component loadings were from the characters POL, 1-2, 5-6, 3-5, HL, 2-4, 4-5, 3-4, 1-4, 5-7,
PFL and BD to the first principal component. The first discriminant function accounted for 64.8% of the total variation. The second discriminant function and third discriminant function accounted for 22 and 13.2%, respectively between group variability among the populations. The morphometric measurements, eye diameter, 3–5, 1–2, and PFL contributed to DF I while HL, 5–6, 3–4, 3–6, 5–8 contributed to DF II. The DF I vs. DF II plot explained 86.8% of total variance among the samples and formed three separate groups. The first group comprised of the samples from Narora and Kanpur sites of the Ganga River. The second and third groups were formed from the samples of the Yamuna and Gomti Rivers, respectively. DFA showed higher values (81.85%) for the overall allotment of individuals into their original populations. The percentage of correctly classified fishes was highest in the Yamuna River (87.8%), followed by the Gomti River (86.6%). Variations in elemental concentrations of otoliths were noted among C. punctata populations from different rivers with 7 out of 8 elements having significantly (P<0.001) different mean concentrations. Fish otoliths collected from Kanpur site of the river Ganga showed significantly (P<0.001) higher concentrations of Ca, Sr, Mn, Fe and K compared to those from other sites. Higher (P<0.001) concentration of Ba was obtained in the fish otoliths collected from Narora site of the river Ganga, while the concentration of Mg was found to be higher (P<0.001) in the otoliths of the fish collected from the river Yamuna. The DF I accounted for 92.81% of the total variation. The DF II and DF III accounted for 5.39% and 1.9%, respectively between group variability among the populations. The elements Ca, Mg, Sr, Ba and Mn contributed most to discriminating among the different populations. The overall random allotment of fishes was high for all populations with 96% of individuals being correctly classified into their original group. The data obtained from Lucknow site of the river Gomti and Firozabad site of the river Yamuna exhibited 100% classification success. The graphic representation of the first two discriminating functions revealed a clear separation of the Narora, Kanpur, Firozabad and Lucknow populations. Percentage classification accuracy showed significant variation (t = -10.82, P<0.001) between allocation success of C. punctata population by truss morphometry and otolith chemistry.

In Clarias batrachus, eight principal components showed eigenvalues greater than 1 using standardized morphometric measurements and explained 83.15% of the
total variance in the PCA. The PC I accounted for 40.32% of the total variance. The PC II accounted for 10.55% and PC III accounted for 8.09% of the total variation. The high component loadings were from the characters 5-8, 5-9, 7-8, 7-9, 5-7, 3-6, 9-11, BD, 1-3, ED, HL and 4-6 to the PC I. The characters 10-11, 8-11, 8-9 and 9-10 contributed to PC II. The first DF accounted for 49.5%, the second DF accounted for 30% and the third DF accounted for 20.5% of the between-group variability. Ganga River samples (Narora population and Kanpur population) together formed one group. Yamuna River samples (Firozabad population) and Gomti River samples (Lucknow population) were isolated forming different groups in discriminating space showing clear separation from other populations. DFA using cross-validation classification showed 68.38% correct classification of the individuals into their original populations. The proportion of correctly classified Gomti River sample to their original group was highest (80%), followed by Yamuna River sample (76.78%). Differences in mean elemental chemistry of otoliths among sites were significant (MANOVA, P<0.001). Nine out of 12 analyzed elements viz., Ba, Mn, Pb, Fe, Cu, Ni, Mg, K and Zn showed significantly (ANOVA, P<0.001) different mean values. No significant (ANOVA, P>0.05) differences were observed among locations for Na, Ca and Sr elements. Otoliths of fish collected from Kanpur site on the river Ganga exhibited significantly (ANOVA, P<0.001) higher values of Fe, Cu, Mn, K and Zn compared to those from Narora (Ganga River), Firozabad (Yamuna River) and Lucknow (Gomti River). Ba was significantly higher (ANOVA, P<0.001) in otoliths of fish from Narora site on Ganga River. A significantly higher (ANOVA, P<0.001) concentrations of Pb and Mg were found in otoliths of fish from Firozabad site on the Yamuna River. The first discriminant function accounted for 70.5% of the total variation. DF II and DF III accounted for 18.7% and 10.8% of the group variability among the populations. The elements, Cu and Pb contributed to DF I while Ba, Mn and Ni contributed to DF II in discriminating the populations. DF I and DF II accounted for 89.2% of the group variability among the populations. DF I vs. DF II graph depicted the presence of four distinct units and showed clear separated stocks. The fish populations from Lucknow, Firozabad, Kanpur and Narora showed classification success of 100%, 96%, 92% and 88%, respectively. Percentage classification accuracy showed significant variation (t = -7.213, P<0.005) between allocation success of C. batrachus population by truss morphometry and otolith chemistry.
discriminating the populations. DF I vs. DF II graph depicted the presence of four distinct units and showed clear separated stocks. The fish population at Lucknow site showed highest classification accuracy (88%), followed by Firozabad (84%), Kanpur (80%) and Narora (72%) populations. Percentage classification accuracy showed significant variation (t = -3.823, P<0.031) between allocation success of *W. attu* population by truss morphometry and otolith chemistry.

In *Sperata seenghala*, eight principal components showed eigenvalues greater than 1 using standardized morphometric measurements and explained 85.57% of the total variance in the PCA. PC I accounted for 48.75% of total variance. PC II and PC III accounted for 8.35% and 6.23%, respectively of the total variance. The high component loadings were from the characters 1-3, 2-5, 2-3, POD, 2-4, ED, 4-6, 7-9, 6-9, 4-5, 6-8, 5-6 and 8-11 to the first principal component and characters 10-12, 11-12, 11-13, and 10-13 to the second principal component. DF I accounted for 58.5% of the total variation. DF II and DF III accounted for 25 and 16.5%, respectively between group variability among the populations. The morphometric measurements, 2-4, 1-2, 5-6, ED, POD, 1-3 and 2-3 contributed to DF I while 4-5, 8-10, 5-7, 9-10, 3-4, 6-7, and 6-8 contributed to DF II. The DF I vs. DF II plot explained 83.5% of total variance among the samples and formed four separate groups. All the four populations (Narora, Kanpur, Firozabad and Lucknow) showed a clear separation from each other, and discerned the existence of different stocks in these rivers. Cross-validation classification showed higher values (79.165%) for the overall allotment of individuals into their original population. The percentage of correctly classified fishes was highest in the Gomti River (85%), followed by the Yamuna River (83.33%) and Ganga River (80%, Kanpur site). Differences in mean elemental chemistry of otoliths among sites were significant (MANOVA, P<0.001). Nine out of 12 analyzed elements Mg, Ba, Mn, Pb, Fe, Cu, Ni, K and Zn showed significantly (ANOVA, P<0.001) different mean values. No significant (ANOVA, P>0.01) differences were observed among locations for Ca, Na and Sr elements. Otoliths of fish collected from Kanpur site on the river Ganga exhibited significantly (ANOVA, P<0.001) higher values of Mg, Fe, Cu, K and Zn compared to those from Narora (Ganga River), Firozabad (Yamuna River) and Lucknow (Gomti River) while Ca, Sr and Na exhibited comparable (ANOVA, P>0.05) values. Ba was significantly higher (ANOVA, P<0.001) in otoliths of fish from Narora site on Ganga River. A significantly higher
(ANOVA, P<0.001) concentration of Pb was found in otoliths of fish from Firozabad site on the Yamuna River. Mean elemental concentration of Ni was significantly (ANOVA, P<0.001) higher in the otoliths of fish collected from Lucknow on the Gomti River. DF I accounted for 66.4% of the total variation. DF II and DF III accounted for 29.4% and 4.1%, respectively of the group variability among the populations. The elements Ba and Zn contributed most to discriminating among the populations in DF I while the elements Ni and Cu contributed to DF II in discriminating the populations. DF I and DF II accounted for 95.9% of the group variability among the populations. DF I vs. DF II graph depicted the presence of four distinct units and showed clear separated stocks. Highest classification success was observed for the fish from Lucknow site (88%) followed by Firozabad (88%), Kanpur (80%) and Narora (76%). A total of 83% fish were correctly classified to their site of capture. Percentage classification accuracy showed comparable variation (t = -2.393, P<0.09) between allocation success of S. seenghala population by truss morphometry and otolith chemistry.

Generated information on stock structure of the selected fish species may be useful in developing the strategies for sustainable utilization and conservation of fishery resources in the Ganges river system.