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Morphological Variation in *Oreochromis Niloticus* using Morphological Indices in Three Populations

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ABSTRACT: This study provides foundational information in terms of genetic variation and morphology of *O. niloticus* in three water bodies in the south-western region of Nigeria. A total of one hundred and nine individual samples of *Oreochromis niloticus* were obtained from the study locations for morphological studies. The study indicated that there was a high morphological variation in the fish from all locations and that, not all the morphological features of *O. niloticus* in the study areas contributed significantly to the variations observed. The PCA loadings for the entire region showed that the head height (HH) is the character that contributed most to the variance observed in the study. The scatter diagram, using 95% sensitivity, indicated that fish samples from Awara and Egbe reservoirs have morphometric features that are more related than those from Ogbese River as confirmed by Mann-Whitney U-test. The result from the PCA of meristic characters was different, the measured characters did not vary within the same population except scales count on two samples from Ogbese River. The observed variation in the meristic characters occurred mostly within the entire region. The results from the study indicated that the reservoirs have more diversified *O. niloticus.*

Keywords: Morphology; Morphometrics; Meristics; *Oreochromis niloticus;* Nile Tilapia

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INTRODUCTION

Southwest Nigeria is a well-watered region teemed with various Cichlids. These cichlids are important freshwater fishes of the tropics and are remarkably known for their high rates of speciation, which often leads to rapid radiation (Ndiwa *et al.,* 2016). They are phenotypically diverse in morphology, behaviour and colouration. They are characteristically marked by their high levels of genetic and morphological diversity, which in turn affect their morphology, ecology, behaviour and genomes (Nelson, 1994; Barlow, 2000; Chakrabarty, 2006).

The Nile tilapia, *Oreochromis niloticus*, is a widely known cichlid and one of the most cultured fish species in the world. It ranks second, after carp, of the most favoured farmed fish species in the world. The high choice of this fish is due to its low-fat content, good taste, high consumer demand and quick growth rate to a considerable size (Fitzsimmons, 2000; Abdelghany, 1993). As common among cichlids, variations have been reported to occur naturally among tilapia of the same species genetically and phenotypically. The cause has been attributed to various genetic modifications through the introduction of hybrid species, an unchecked mixture of such cultured species with the natural species (Ameen, 1999). Also, the environment had been noted to cause morphological changes in *O. niloticus.*

The variability study in the fish is important as it helps in the management of its stocks. This will also reveal how the fish is influenced morphologically by its environment. Population study in fish generally is important for effective fisheries management and conservation practices.

MATERIALS AND METHODS

Study Area

Egbe Reservoir in Ekiti State (7° 36'N and 7° 39' North and longitude 5° 32' E and 5°35), Awara Reservoir in Ikare Akoko (5° 30'and 6° 00' N 7° 30'and 8° 00'E) and River Ogbese, 6°43'N to 7°17'N and longitudes 5°26'E to 6°34'E both in Ondo State are important water bodies to the two States, they serve as sources of water and fishing sites for the people in the area.

Samples Collection and Preparation

A total of one hundred and nine individual samples of *Oreochromis niloticus* adult were obtained from three different water bodies. These were brought to the Department of Fisheries and Aquaculture Laboratory, Federal University of Technology, Akure where the morphometric and meristic characteristics of the fish were measured using calibrated measuring board according to Teugels (1982) aided with a ruler and a pair of divider/caliper. The characters measured in morphometrics are standard length (SL), total length (TL) caudal length (CL), head length (HL), body height (BH), caudal peduncle height (CPH), inter-orbital distance (IOD), ocular diameter (OD), pre-anal length (PL), predorsal length (PdL), and head height (HH). Other characters were the distances between dorsal and pectoral fins (DPD), pectoral and pelvic fins (PPD), pelvic and anal fins (PAD), and dorsal and anal fins (DAnD) (Figure 1).

Meristic characters consisting of the dorsal, anal, caudal, pectoral and pelvic fin rays were counted. Scales on the lateral lines were also considered. The fin rays fins were counted with the aid of a hand lens, as it magnifies the region to be counted.

Data Analysis

Data on morphometric and meristic characters were recorded on population basis. Morphometrics and meristic data were analysed separately using Paleontological Statistical Test (PAST) software (Hammer *et al*. 2001) because morphometric data changes continuously and is more susceptible to environmentally induced variability, unlike the meristic data which is discrete and fixed in early developmental stages (Simon *et al.,* 2010; Samaradivakara *et al.,* 2012). To remove any size effect and differences on the morphometric an allometric formula designed by Elliott *et al*., (1995) was used to remove the length effects in the samples thus:

$M_{adj} = M (L_s / L_o)^b$

Where M: original measurement, M_{adj}: size adjusted measurement, L_o: standard length of fish, L_s: overall mean of standard length for all fish from all samples in each analysis.

Parameter b was estimated for each character from the observed data as the slope of the regression of log M on log L_o, using all fish in each population. To clarify patterns of correlation among populations from various locations, Principal Component Analysis (PCA) was performed on the data. Cluster Analysis was performed on the morphometric and meristic data (Chiu, *et al.,* 2002; Madec *et al.,* 2003). Also, a canonical variate analysis was employed to observe if populations from various locations are significantly different and to identify morphometric and meristic characters by which this operational taxonomic unit could be diagnosed using PAST, a paleontological statistical software (Hammer *et al.,* 2006). Population centroids with 95% confidence ellipses derived from the PCA was used to visualize relationships among the populations. A comparative analysis of the variables between populations using the Mann-Whitney U-test was also performed. The meristic data were also analysed as with the morphometric data but were not transformed.

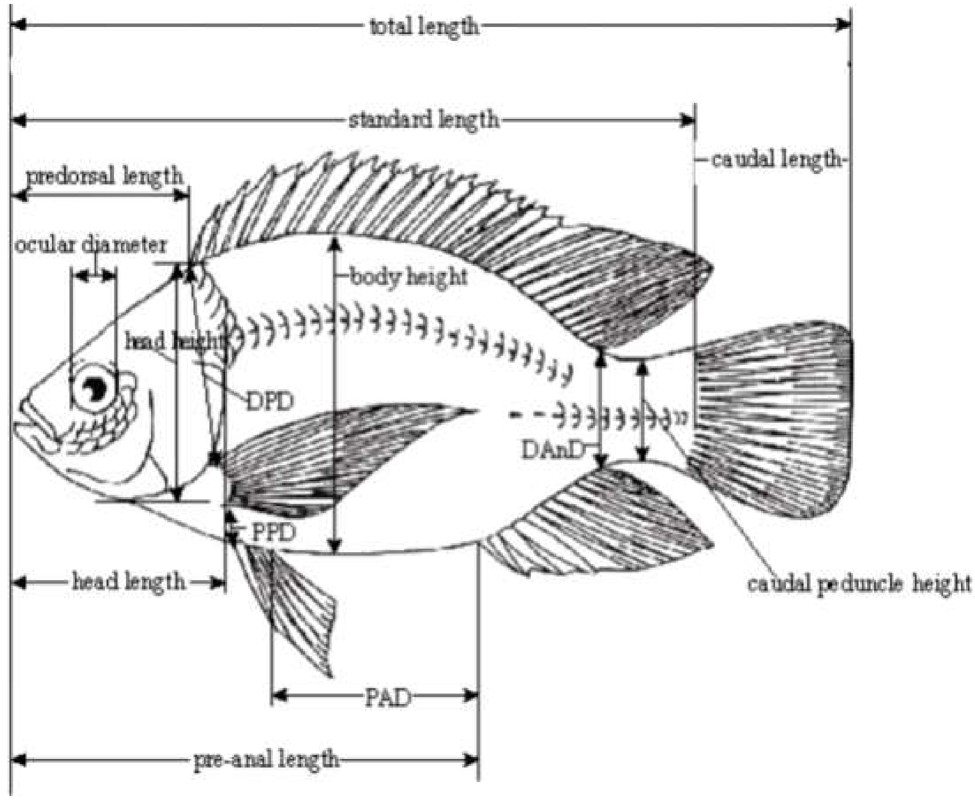

Figure 1: Measured morphometric characters (Adapted from Azua *et al*., 2017)

RESULTS

Morphological Identification of the Samples

All the fish samples were morphologically identified initially as *Oreochromis* sp. of the family Cichlidae according to Trewavas (1983) and Olaosebikan and Raji (2013). Some of the observable features include the possession of extended dorsal fin with long spines and soft rayed portions. The spiny part is anteriorly located while the soft ray portion is posteriorly located. The fish samples from Egbe and Awara reservoirs had a total of 28 dorsal fin rays of which 16 were spiny and 12 were soft; while those from Ogbese River had 29 of which 17 were spiny and 12 were soft rays. All had 12 soft rays in common on the dorsal fin. All the samples had 3 spiny anal fin rays and soft ray of either 9 or 10. A single nostril was present at each side of the head. Cycloid scales were present on the body. The scales on the ventral part of the samples, especially between the pectoral and pelvic fins were noticeably smaller than those on the flank. The caudal fin was truncated with vertical bar/stripes. The variations agree with the assertions of GISD (2018) on *O. niloticus*.

Morphometrics

The standard length (SL) of the sampled fish ranged from 80 mm - 119 mm (Egbe Reservoir), 88 mm - 141 mm (Awara Reservoir), 88 mm - 158 mm (Ogbese River). Thus, a range of 80 mm - 158 mm was the SL of the samples from all populations. Skewness and kurtosis measure the distribution pattern of variables in the data set.

The results of the Principal Component Analysis (PCA) conducted on 15 morphometric characters for 109 fish samples from the three water bodies in Southwest Nigeria, is shown in Table 1. The first five components accounted for the significant variance with a total of 86.060% variance, the first PC accounting for 57.998% of the total variance and the Jolliffe cut-off was 8.92.

Table 1: The Principal Component Analysis Summary Showing the Eigen Values and Percentage (%) Variance for *Oreochromis niloticus* sampled from the three Water Bodies

Principal Component (PC)	Eigenvalue	% variance
1*	103.492	57.998
2*	19.530	10.944
3*	11.371	6.373
4*	9.936	5.568
5*	9.236	5.176
6	6.062	3.397
7	5.574	3.124
8	4.835	2.710

Eigenvalue ≥ Jolliffe cut-off: 8.92 is significant.

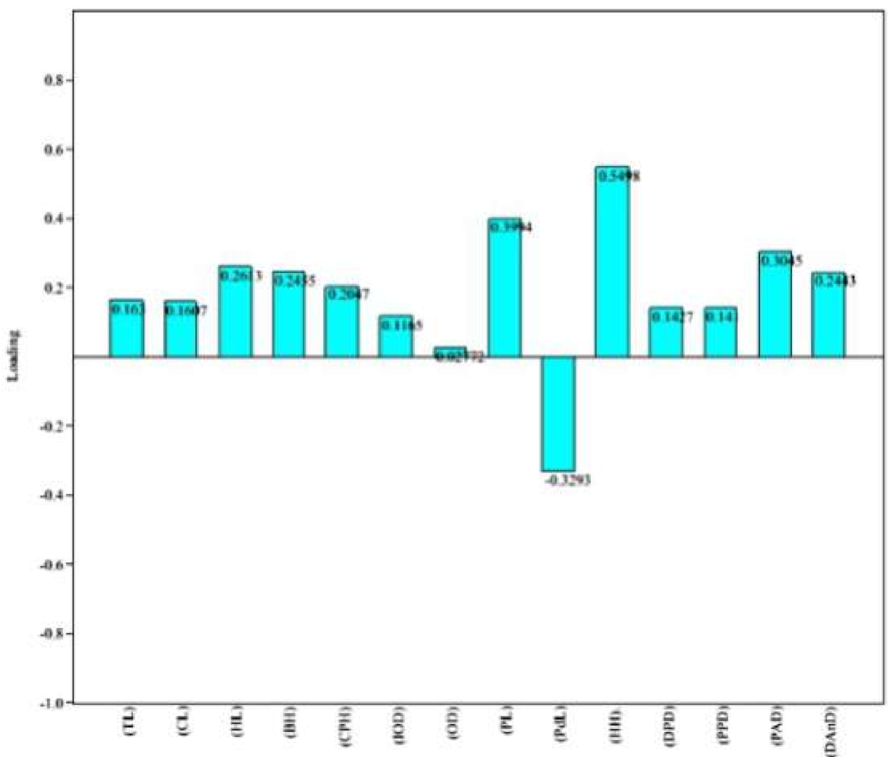

Figure 2: Loadings Plot for PC1 showing variability in different morphometric features for fish samples from all populations

Standard length (SL), total length (TL) caudal length (CL), head length (HL), body height (BH), caudal peduncle height (CPH), inter-orbital distance (IOD), ocular diameter (OD), pre-anal length (PL), predorsal length (PdL), and head height (HH), distances between dorsal and pectoral fins (DPD), pectoral and pelvic fins (PPD), pelvic and anal fins (PAD), and dorsal and anal fins (DAnD).

Variation in Morphometric Characters as Revealed by Loadings

Figure 2 contains the PCA loading of the three populations studied. From the Figures, each bar represents a morphometric character and its height gives a measure of its contribution to the variance in the component considered. The PCA loading (Figure 2) indicated that the head height (HH) is the character with the highest contribution to the variance followed by the pre-anal length (PL), then the predorsal length (PdL), while the ocular diameter (OD gave the least contribution to the variance.

Variation in Morphometric characters as shown by Scatter diagram

The Scatter diagram showing the variation and relationship among specimens is shown in Figure 3. Each population is marked out with an ellipse at 95% sensitivity. The three populations intersect, indicating the uniqueness of certain morphometric features, as such, shared common taxa. While some are closely placed, it can also be seen from the scatter that the samples differ significantly in morphometric features by the fair spread of some samples within the ellipses.

Hierarchical Clustering

Figure 4 shows the classical clustering of the samples across all populations using Euclidean Similarity Index (ESI). The result showed two taxa with a single main taxon consisting of virtually all the samples. The second taxon had just one sample from Awara as revealed by the morphometric data analysis. The main taxon was further branched into two with a branch containing some samples from Awara Reservoir, while the other contained the rest of the samples in the three water bodies of South-west Nigeria and they were all further branched into more clades. The clustering showed that they have a common root and that some are closely related in the upper region while the other few ones were not. It could be inferred from the diagram that the fish are closely related but differ from each other based on the differences in their morphometry. This could be seen from the third taxon.

The meristic characters of the sampled fish of each population vary slightly from the other. All measured meristic features were observed unvaried within the same population except scales count on two samples observed to be 21 and 22 from Ogbese River, while others had 23 each. Some of these characters were also observed unvaried across all populations, these include the soft dorsal fin ray which amounted to 12 each, pelvic fin ray totaling 6 on each fish; the caudal fin ray totaling 18 and the spiny anal fin ray observed to be 3 on all samples. All these are within the range previously recorded for *Oreochromis niloticus.*

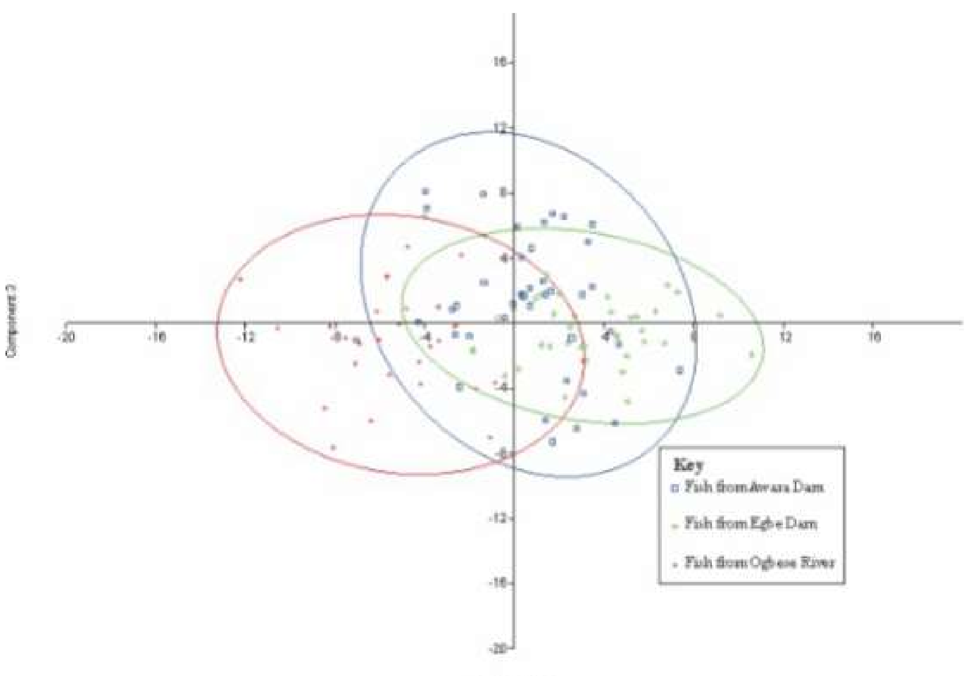

Figure 3. PCA Scatter Diagram (PC2 and PC3) showing the variability and linkage of *Oreochromis* sp. across all populations as revealed by their morphometric characters

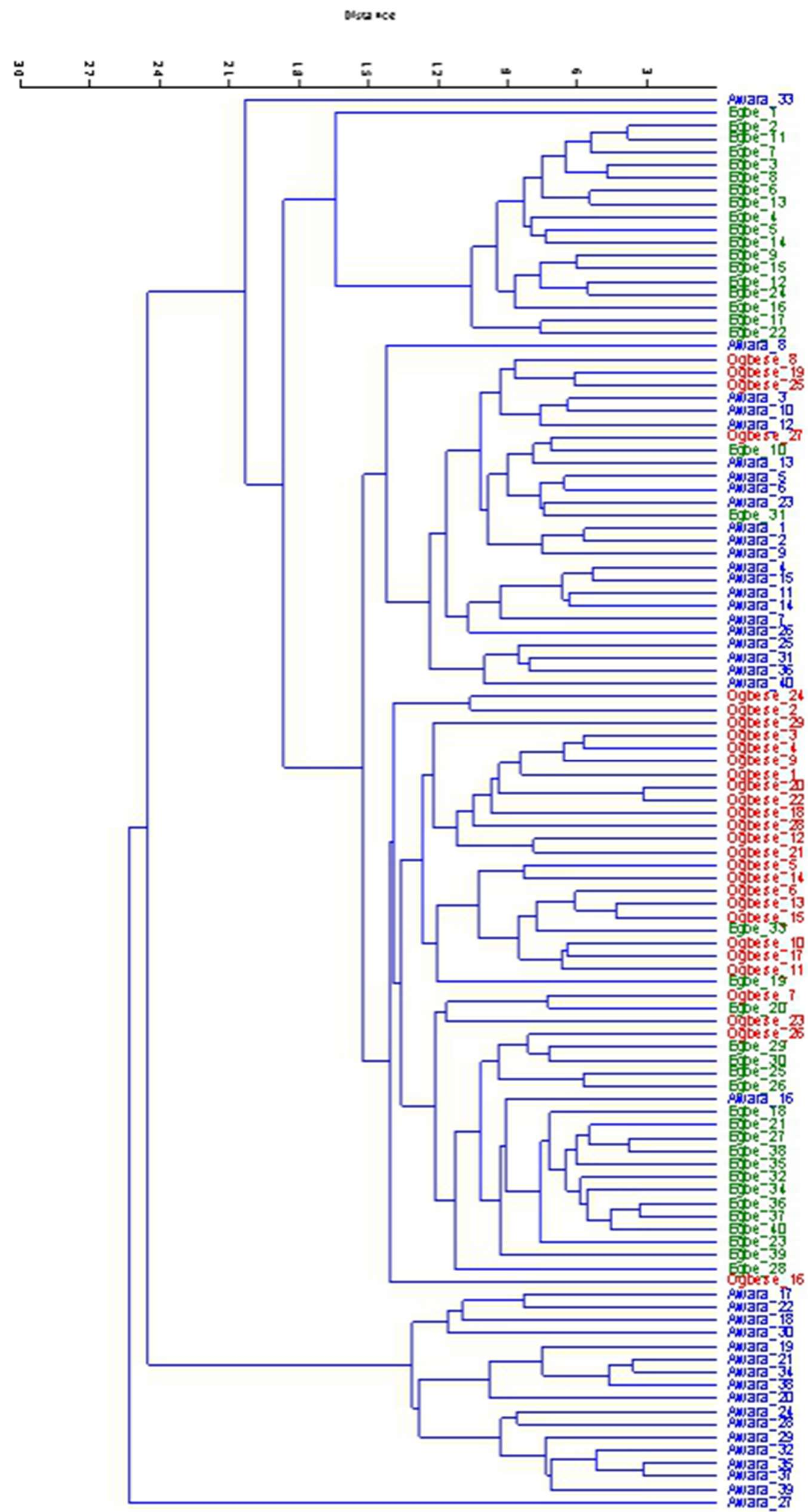

Figure 4: The Hierarchical Clustering of Samples based on their Morphometric Features Meristic Variability of Sampled fish.

Principal Component Analysis for Meristic Counts

There was observed variation in sampled fish across the water bodies. Table 2 contains the PCA summary of all the water bodies done while disregarding the group option. The Jolliffe cut-off was 0.053 accounting for the significance of only PC1 and PC2 with a total of 93.62% variance.

Variability according to PCA Loadings of Meristic features

Considering territorial partitioning between the water bodies, the Principal Component Analysis indicated that there was variation in some of the meristic characters in the region under study. The PC1 loading plot (Figure 5) indicated that the dorsal spiny fin ray and the anal soft fin ray contributed equally the same measure of variation to the observed percentage variation in the PC1, while the pectoral fin ray was next and the scale count indicated little contribution to the observed variation.

Table 2: The Principal Component Analysis summary showing the eigenvalues and Percentage (%) variance for values of meristic features of sampled tilapia in all water bodies.

PC	Eigenvalue	% variance
1*	0.469	69.382
2*	0.164	24.238
3	0.043	6.380

Jolliffe cut-off 0.053 * Significant

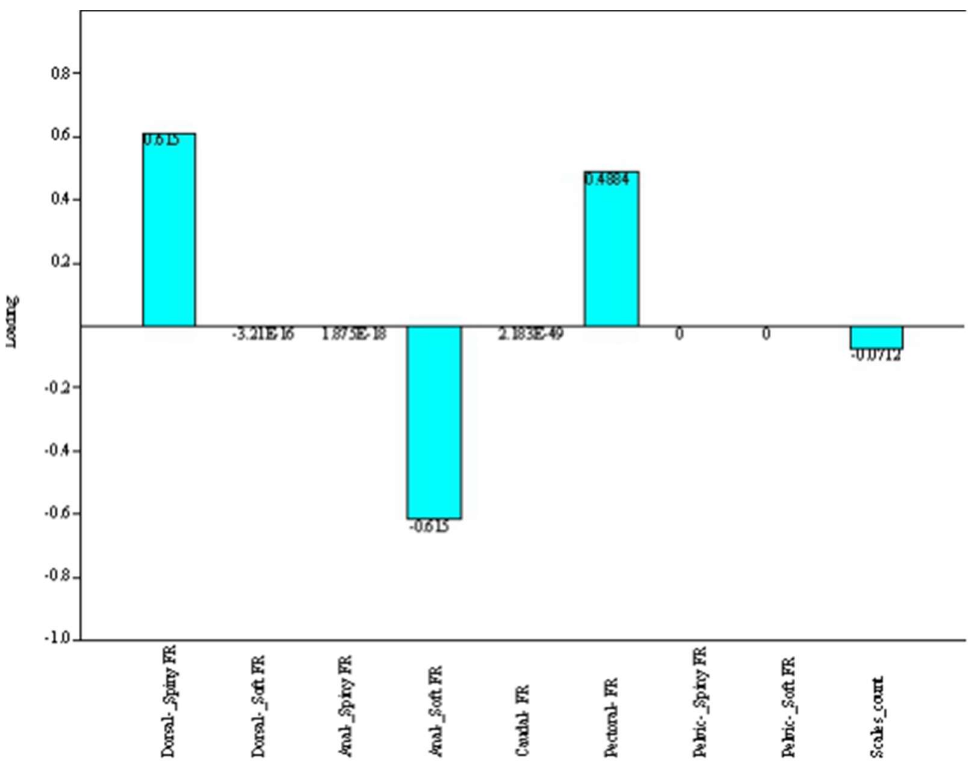

Figure 5. Loadings Plot for PC1 showing variability in different meristic features for fish samples from all populations (FR = fin ray)

Variation in Meristic characters

The scatter plot for the meristic count is shown in Figure 6. The plot indicated that each population is related. The biplot (shown as lines) revealed that the dorsal spiny, anal soft and pectoral fin rays had significant contributions to the observed variation. The biplot also revealed that scales count contributed to the variation only observed with the sample from Ogbese River. The scatter diagram also showed that there was no variation among samples within Egbe Reservoir and Awara Reservoir (each has single point on the plot while Ogbese has four) concerning their meristic features.

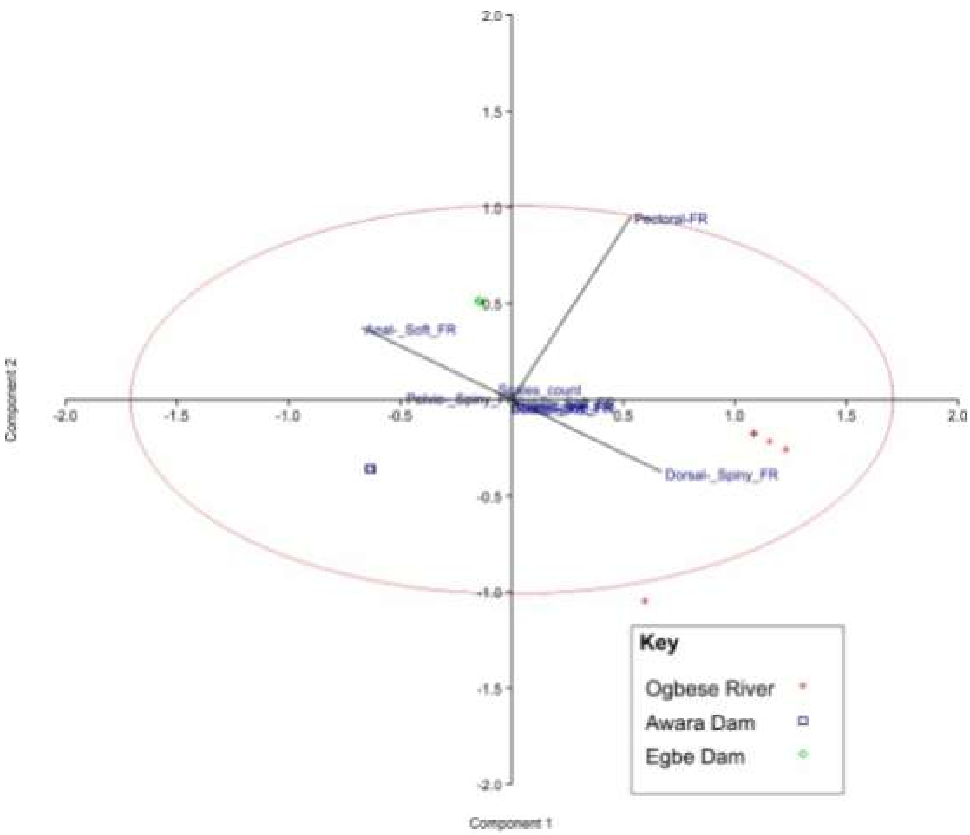

Figure 6. PCA Scatter Diagram (PC2 and PC3) showing the variability and linkage of *Oreochromis* sp. from all populations as revealed by their meristic characters

DISCUSSION

The contributions of morphometric and meristic features to observed variations

This study indicated that not all the morphological features of *O. niloticus* in the study areas contributed significantly to variations observed. There was a high morphological variation in the studied features of the fish which agrees with the assertions by many authors (Naeem and Salam, 2005; Ndiwa *et al*., 2016; Azua *et al*., 2017). Mwanja *et al*., (2016) noted that there was a high variation in the morphological features of *O. niloticus,* which was due more to size than shape. The result from this study indicated that the observed variations were both of size and shape. The head height (HH), the pre-anal length and pre-dorsal length were responsible for most of the observed morphological variations. The head height gave the highest contribution to the observed variations, which was true for all the water bodies. Agostinho *et al*. (2008) opined that reservoirs greatly affect the composition and structure of fish aggregation, hence present relevant spatiotemporal diversity. Discriminant analyses give the characters that contributed to the discrimination of the populations. In discriminant analyses (PCA), HH, PaL and PdL contributed majorly to principal component analysis 1. Morphometrics of the head and body depth have been observed to be the most important characters for fish population discrimination. Leslie & Grant (1990) observed this on angler fish (*Lophius vormernus*), and Haddon & Willis (1995) on Pacific herring (*Clupea pallasi*). However, fishes generally demonstrate greater variance in morphological characters both within and between populations compared with other vertebrates, and they are prone to environmentally induced morphological variation (Allendorf, 1988; Thompson, 1991; Wimberger, 1992), resulting in different feeding environment, prey types, food availability or other features.

The meristic features variations had a different trend. The meristic feature did not vary within all populations, the high variation recorded for within populations was only within Ogbese. The meristic feature within each of Awara and Egbe Reservoirs had zero variance which implies that both location are reservoirs and organisms found in the same environment exhibit similar morphological features. The result from the three water bodies indicated that tilapia tends to adapt to its environmental changes for survival leading to variations observed in this study, also, that the two reservoirs had more diversified fish. Popoola *et al.,* (2014) indicated that genetic diversity within a population is highly crucial for adaptation to changing environments, which will in turn enhance the survival of species.

CONCLUSION

The observed morphometric discreteness in relation to the sampling sites showed that environmental factors are affecting the phenotypic discreteness of *O.niloticus* populations, which revealed that the observed differences are caused by some intermingling between populations. The use of molecular genetic markers such as microsatellite (Turan *et al*., 1998) and mitochondrial DNA (mt-DNA) applications (Shaw *et al*., 1999) would be definitive methods of examining the genetic component of phenotypic discreteness between geographically isolated regions, which will help the proper management of this stock.

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