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Biochemical diversity and genetic distance within and between Tiv and Fulani Local chickens of Nigeria

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Abstract: *The study was undertaken to investigate biochemical diversity and genetic distance within and between Tiv and Fulani local chicken ecotypes of Nigeria. The experimental birds were randomly sourced from ten locations. The locations (1-5) for the Tiv ecotype were Uikpan, Daudu, Kadarko, Yelwata and Cohor (in Benue and Nasarawa States) while that of the Fulani ecotype were Lafia, Akurba, Adogi, Asakio and Namu (in Nasarawa and Plateau States). At maturity, four (4) male and four (4) female birds were randomly selected from each location per ecotype to give a total of eighty (80) adult birds (40 birds each for Tiv and Fulani ecotype) and used for blood protein characterization study. Blood protein loci, namely haemoglobin, albumen, transferrin and carbonic anhydrase were analyzed using electrophoresis. Data collected from the biochemical analysis were analyzed using popgene version 1.31. The results obtained indicate that, the genetic distance based on identity and distance were significant and generally positive in both the Tiv and the Fulani ecotypes. The F-statistics (F_{is}) values were positive in the two ecotypes except at alleles B and C in the Albumen locus of the Tiv ecotype and allele B in the Carbonic Anhydrase locus of the Fulani ecotype. The dendogram showed that the two chicken ecotypes originated from a common ancestry but diverge from one locality to another in each ecotype. The genotypic frequency varied across locations in both ecotypes. Haemoglobin genotypes AA and AB were prevalence in the Tiv (0.875) and the Fulani (0.538) ecotypes, respectively. Genotype AC for albumen was most prevalent in both ecotypes. However, albumin genotype AA and AB was not found in the Tiv and Fulani ecotypes, respectively. In transferrin, genotype BD (0.378) and AA (0.568) was most prevalent in the Tiv and the Fulani ecotypes, respectively. Genotype AC in carbonic anhydrase was prevalenth in both ecotypes while genotype BB only found in the Tiv ecotype. From the findings of this study it was concluded that, the genetic diversity within and between the Tiv and the Fulani chicken ecotypes as observed in this study should be exploited through selection within each ecotype and subsequent crossing between birds from different locations/ecotype to take advantage of heterosis.*

Keyword: *Allele, blood protein, dendogram, electrophoresis, genotypic frequency, locations*

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I. Introduction

Quantifying the structure of genetic diversity in different African chicken populations is important in optimizing genetic improvement, conservation and utilization strategies. Many technologies including DNA based technology are employed in diversity studies. DNA-based technologies are now the methods of choice for genetic characterization of livestock (Arora et al., 2011); but its applications in developing countries are limited due to its complex nature, facilities and high cost. Nevertheless, other biotechnological techniques has opened up molecular techniques, such as electrophoresis being employed for the detection of polymorphism at protein and enzyme loci as well as other serological and immuno-genetic procedures for measurement of genetic variations (Salako et al., 2007). Proteins or allozyme polymorphs remain tremendously useful in developing countries because of their lower complexities, cost, simplicity of data interpretation and amount of genetic information accessed (Rege and Okeyo, 2006). A major advantage of biochemical characterization is that they do not depend on environmental factors, stable throughout ontogenesis and have a simple type of inheritance (Lee et al., 1995). The experiment was aimed at determining the genetic distance within and between the two ecotype populations based on biochemical characteristics.

II. Materials and Methods

Experimental Procedure

Two local chicken ecotypes comprising of the Tiv (long legged) and the Fulani birds from five locations each were used for this experiment. The locations (1-5) for the Tiv ecotype were Uikpan, Daudu, Kadarko, Yelwata and Cohor (in Benue and Nasarawa States) while that of the Fulani ecotype were Lafia, Akurba, Adogi, Asakio and Namu (in Nasarawa and Plateau States). At maturity, four (4) males and four (4) females were randomly selected from each location per ecotype to give a total of fourty (40) adult birds (20 males and 20 femals) per ecotype and used for the blood protein characterization study.

Whole blood (3-5 ml) was collected from the wing vein of each of the selected healthy birds into correspondingly labelled heparinized tube. Heparin acted as anti-coagulant and blood contamination was prevented by using separate syringes and needles for individual birds. These samples were kept refrigerated in ice packs and transported to Animal Science Laboratory of the University of Ibadan, Ibadan, Oyo State for electrophoresis analysis.

The blood samples were centrifuged at 4 °C for 20 mins at 3 000 rpm in order to separate plasma and erythrocyte. Erythrocyte was washed with 9 percent NaCl to free them from plasma proteins and was then lysed with four fold of cold distilled water in order to release the haemoglobin. Then plasma and haemolysates aliquots were stored at 4^{o}c prior to electrophoresis analysis. The lysed red blood cells were used to determine Haemoglobin (Hb). However, the blood plasma was used to determine albumin (Alb), carbonic anhydrase (Ca) and transferrin (Tf) genotypes. Electrophoresis was performed using cellulose acetate strips as described by Akinyemi and Salako (2012). After electrophoresis, strips of bands were stained for 25 – 30 minutes. The stained bands were thereafter washed with 1 percent HCl for Ca and 5 percent HCl for Hb, Alb and Tf (Akinyemi and Salako, 2012). After washing, the bands were covered by distaining solution (1% and 5% HCl) until the electrophoresis bands became visible, then air dried and scored.

Parameters measured

The blood protein parameters such as haemoglobin, albumin, carbonic anhydrase and transferrin were determined using electrophoresis.

Experimental Design and Data Analysis

The design of the experiment was Completely Randomized Design (CRD). Stratified random sampling technique was employed in assembling the experimental bird's population. Four female and male birds of each of the Tiv and the Fulani ecotypes were randomly sampled from each of the five localities for blood protein analysis. Biochemical variability for blood protein (haemoglobin, transferrin, carbonic anhydrase and plasma albumin) within and between the ecotype and location was estimated using popgene statistical software version 1.31.

III. Result

Genetic Distance

Table 1 shows the pair-wise population matrix of Nei genetic distance based on identity and distance between the locations of the Tiv and the Fulani ecotypes. Generally, the values of genetic distance based on identity were generally higher than values of genetic variability base on distance. Genetic distance based on identity and distance were all positive in both ecotypes except between location 1 and 2 which was -0.044 in the Tiv ecotype and locations 1 and 2 (-0.013) as well as locations 3 and 4 (-0.011) in the Fulani ecotype. Distance ranges from 0.670 (between locations 1 and 5) to 1.045 (between locations 1 and 2) in the Tiv ecotype. In the Fulani ecotype, genetic distance ranged from 0.657 (between locations 3 and 5) to 1.013 (between locations 1 and 2). The shortest distance was -0.013 between locations 1 and 4 ecotype while the farthest distance was 0.420 between locations 3 and 5 in the Fulani ecotype.

Genotypic frequency

The genotypic frequencies at the haemoglobin, albumin, transferrin and carbonic anhydrase loci in the two Nigerian local chicken ecotypes are presented in Table 2. Two haemoglobin genotypes (AA and AB) were expressed in both the Tiv and the Fulani ecotypes. Haemoglobin genotype AA was prevalence in the Tiv (0.875) ecotype while haemoglobin AB was prevalence in the Fulani (0.538) ecotype. Genotype AC for albumen was most prevalent in both ecotypes. However, albumin genotype AA and AB was not found in the Tiv and Fulani ecotypes, respectively. In transferrin, genotype BD (0.378) and genotype AA (0.568) was most prevalent in the Tiv and the Fulani ecotypes, respectively. Genotype AB, BB and BD were only found in the Tiv ecotype. Genotype AC in carbonic anhydrase was prevalenth in both ecotypes with frequencies of 0.525 and 0.711 for the Tiv and the Fulani ecotypes, respectively while genotype BB only found in the Tiv ecotype.

Table 3 presents the effect of location on genotypic frequencies at blood protein loci of the two ecotypes of Nigerian local chicken. The genotypic frequencies vary from location to location in both the Tiv and the Fulani ecotypes. For haemoglobin locus in the Tiv ecotype, genotype AB was only found (1.000) in birds from Uikpan, Daudu, Kadarko and Yelwata (locations 1, 2, 3 and 4) while genotype AA overwhelmingly dominated genotype AB (0.625 vs 0.375) in birds from Cohor (location 5). However, in the Fulani ecotype, genotype AA was prevalent in birds from Lafia, Akurba and Namu (locations 1, 2 and 5). Genotype AB was however the most prevalent in birds from Adogi and Asakio (locations 3 and 4). Genotype AA was not found at in location 4 while genotype AB was not noted in location 2. In the albumin locus, genotype AC was only found (1.000) in birds from Daudu, Kadarko, Yelwata and Cohor (locations 2, 3, 4 and 5) for the Tiv ecotype while in the Fulani ecotype, genotype AC was only observed with frequency of 1.000 in birds from Lafia, Asakio and Namu (location 1, 4 and 5). Genotype AB was only found with a very negligible frequency (0.125) in the Tiv birds from Uikpan (location 1) and Fulani birds from Akurba and Asakio (locations 2 and 3).

Transferrin locus indicated six types of genotypes (AA, AB, AD, BB, BD and DD) in the Tiv ecotype and three genotypes (AA, AD and DD) in the Fulani ecotype. In the Tiv ecotype, genotype BD dominated in birds from Uikpan, Daudu and Kadarko (locations 1, 2 and 3) while genotype AD was prevalent in birds from Yelwata and Cohor (locations 4 and 5). In the Fulani ecotype, genotype AA dominated in all the locations except in birds from Namu (location 5) were allele DD was prevalent. Carbonic anhydrase showed 4 genotypes (AA, AB ACand BB) in the Tiv ecotype and 3 genotypes (AA, AB and AC) in the Fulani ecotype. In the Tiv ecotype, allele BB dominated in birds from Uikpan (location 1) while allele AB was prevalent in birds from Daudu (location 2). Similarly, genotype AC dominated in birds from Kadarko and Yelwata and Cohor (locations 3 and 4). Genotype AA and AC had equal frequency (0.500 each) in birds Cohor (location 5). In the Fulani ecotype, apart from location 2 (Akurba) were genotype AA dominated with frequency of 0.625, genotype AC dominated all other locations. Genotype AB was negligibly (0.125) found in location 2 (Akurba) but not found at all in birds from Lafia, Adogi, Asakio and Namu (locations 1, 3, 4 and 5).

F- Statistics (F_{is}) of alleles at four loci of the Tiv and the Fulani local chicken ecotypes

The F-statistics of alleles at haemoglobin, albumin, transferrin and carbonic anhydrase loci in two ecotypes of Nigerian local chicken population are shown in Table 4. The F_{is} values were positive in the two ecotypes except at alleles B and C in the Albumen locus of the Tiv ecotype and allele B in the Carbonic Anhydrase locus of the Fulani ecotype where the values were negative. The F_{is} values for the entire population range from -0.0046 to 0.4459 and -0.0046 to 0.4694 for the Tiv and the Fulani ecotype, respectively.

Dendogram of the relationships between the two ecotypes

The dendogram or un-weighted pair group mean analysis (UPGMA) trees of the Tiv (population 1) and the Fulani (population 2) local chicken ecotypes based on blood protein analysis are shown in Figure 1, 2 and 3. The trees showed that the two chicken ecotypes originated from a common ancestry (figure 1). The tree for the Tiv ecotype based on locations (figure 2) showed that birds from Kadarko (location 3) are more diverge and genetically distinct. Those birds in Uikpan and Daudu (locations 1 and 2) are more closely related genetically to each other while birds in Yelwata and Cohor (locations 4 and 5) are closely related to one another but not as close as that of Uikpan and Daudu. Figure 3 showed the dendogram tree for the Fulani chicken ecotype. From the tree, it was evident that though the birds were all from the same ecotype, birds from Namu (location 5) are most genetically diverge. The birds from Lafia and Akurba (locations 1 and 2) as well as those from Adogi and Asakio (locations 3 and 4) were close to one another genetically while those of Lafia and Akurba were more closely related.

Table 1: Genetic Distance base on Identity (lower diagonal) and Distance (upper diagonal) between the five Locations of the Tiv and the Fulani Local Chicken Ecotypes using Biochemical Parameters: Haemoglobin, Albumin, Transferin and Carbonic Anhydrase

Location	1	2	3	4	5
Tiv ecotype					
1	*****	-0.044	0.107	0.311	0.401
2	1.045	*****	0.048	0.188	0.255
3	0.898	0.953	*****	0.010	0.093
4	0.733	0.828	0.990	*****	0.008
5	0.670	0.775	0.912	0.992	*****
Fulani ecotype					
1	*****	-0.013	0.041	0.018	0.104
2	1.013	*****	0.063	0.107	0.214
3	0.96	0.939	*****	-0.011	0.420

4	0.982	0.899	1.011	*****	0.267
5	0.902	0.808	0.657	0.766	*****

Table 2 : Genotypic Frequencies at Haemoglobin, Albumin, Transferrin and Carbonic Anhydrase Loci in two Ecotypes of Nigerian Local Chickens

		Tiv ecotype		**Fulani ecotype**	
Locus	**Genotype**	**N**	**Genotypic frequency**	**N**	**Genotypic frequency**
Haemoglobin	AA	05	0.125	21	0.538
	AB	35	0.875	18	0.462
Albumin	AA	-	-	03	0.077
	AB	01	0.026	-	-
	AC	37	0.974	36	0.923
Transferrin	AA	02	0.054	21	0.568
	AB	01	0.027	-	-
	AD	11	0.297	09	0.243
	BB	06	0.162	-	-
	BD	14	0.378	-	-
	DD	03	0.082	07	0.189
Carbonic anhydrase	AA	08	0.20	10	0.263
	AB	07	0.175	01	0.026
	AC	21	0.525	27	0.711
	BB	04	0.100	-	-

N = Sample size, L = location, A, B, C, D = haemoglobin, albumin, transferrin and carbonic anhydrase allele respectively

Table 4: Effect of Location on Genotypic Frequencies at some blood protein Loci in two Ecotypes of Nigerian Local Chickens

Locus	Genotype	Tiv ecotype L1 (N = 8)	L2 (N = 8)	L3 (N = 8)	L4 (N = 8)	L5 (N = 8)	Fulani ecotype L1 (N = 8)	L2 (N = 8)	L3 (N = 8)	L4 (N = 8)	L5 (N = 8)
Haemoglobin	AA	0.000	0.000	0.000	0.000	0.625	0.750	1.000	0.125	0.000	0.875
	AB	1.000	1.000	1.000	1.000	0.375	0.250	0.000	0.875	1.000	0.125
Albumin	AA	-	-	-	-	-	0.000	0.125	0.250	0.000	0.000
	AB	0.125	0.000	0.000	0.000	0.000	-	-	-	-	-
	AC	0.875	1.000	1.000	1.000	1.000	1.000	0.875	0.750	1.000	1.000
Transferrin	AA	0.000	0.000	0.000	0.000	0.333	0.500	0.625	1.000	0.625	0.000
	AB	0.000	0.125	0.000	0.000	0.000	-	-	-	-	-
	AD	0.000	0.000	0.286	0.750	0.500	0.5000	0.375	0.000	0.375	0.000
	BB	0.375	0.375	0.000	0.000	0.000	-	-	-	-	-
	BD	0.625	0.500	0.714	0.000	0.000	-	-	-	-	-
	DD	0.000	0.000	0.000	0.250	0.167	0.000	0.000	0.000	0.000	1.000
Carbonic anhydrase	AA	0.000	0.000	0.000	0.500	0.500	0.250	0.625	0.429	0.000	0.000
	AB	0.125	0.750	0.000	0.000	0.000	0.000	0.125	0.000	0.000	0.000
	AC	0.375	0.250	1.000	0.500	0.500	0.750	0.250	0.571	1.000	1.000
	BB	0.5000	0.000	0.000	0.000	0.000	-	-	-	-	-

N = Sample size, L = location, A, B, C, D = haemoglobin, albumin, transferrin and carbonic anhydrase allele respectively

Table 4: F – Statistics of the Alleles at at some blood protein Loci in two Ecotypes of Nigerian Local Chicken

Loci	Allele	Tiv ecotype (N = 40)	Fulani ecotype (N = 40)
Haemoglobin	A	0.074	0.2586
	B	0.0714	0.2586
Albumin	A	0.000	0.0037
	B	-0.0046	-
	C	-0.0002	0.0037
Transferrin	A	0.2635	0.4694
	B	0.4459	-
	C	-	-
	D	0.0612	0.4694
Carbonic anhydrase	A	0.1498	0.0579
	B	0.3776	-0.0046
	C	0.0695	0.0889
Over all loci		0.1412	0.1949

N = Sample size, A, B, C, D = haemoglobin, albumin, transferrin and carbonic anhydrase allels respectively.

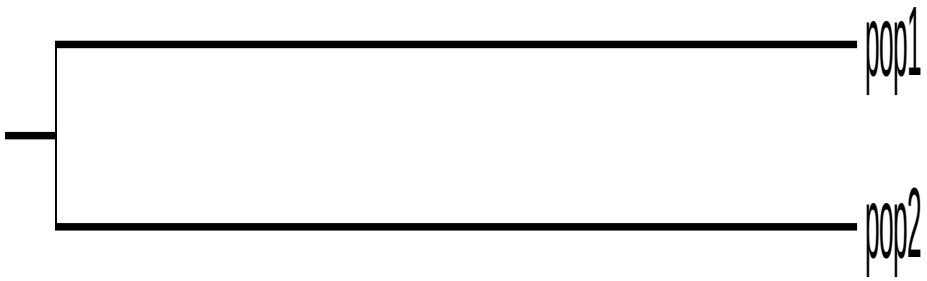

Figure 1: Dendogram representing the genetic relationship between the Tiv (population 1) and the Fulani (population 2) chicken ecotypes

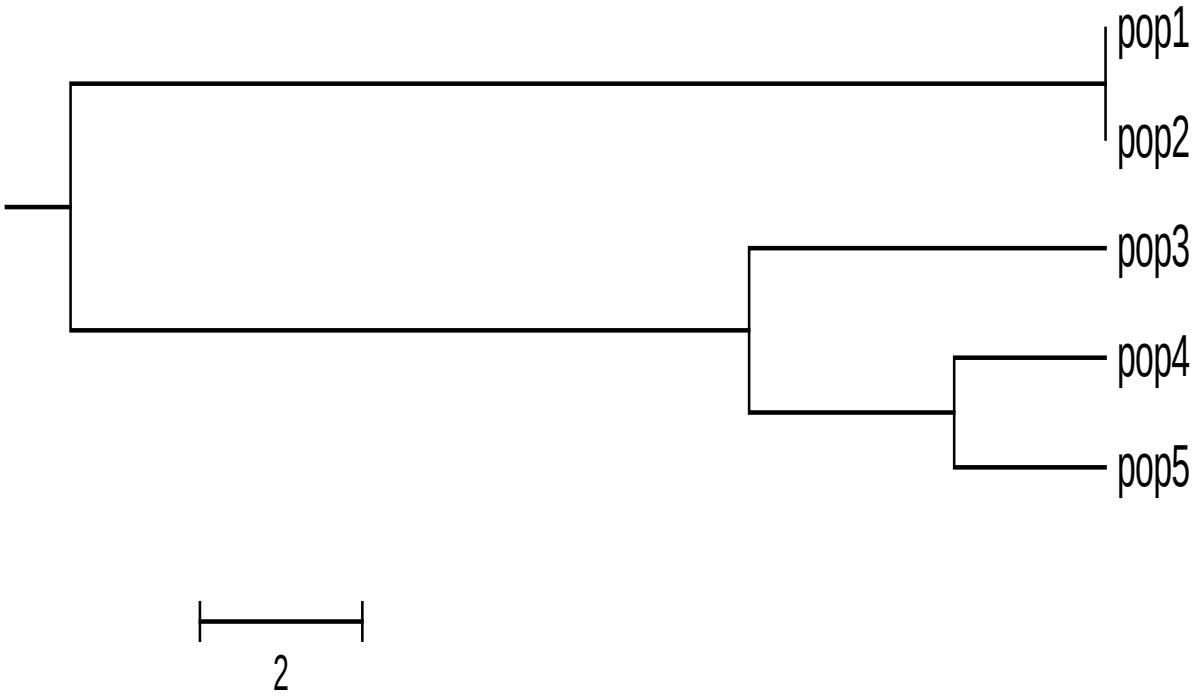

Figure 2: Dendogram representing the genetic relationship between the five locations of the Tiv local chicken ecotype

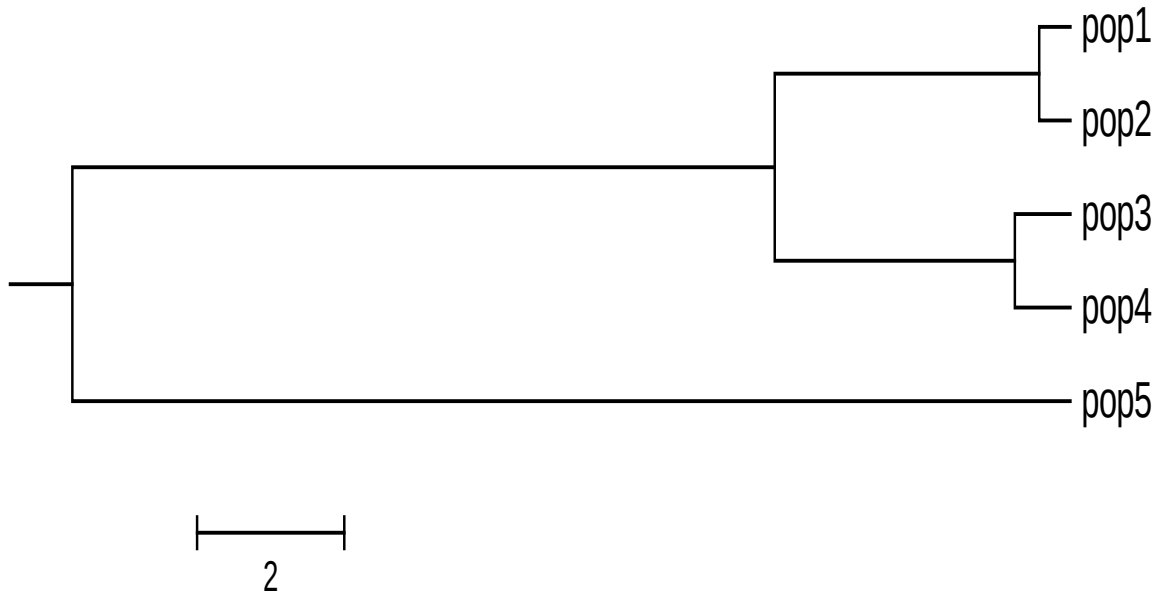

Figure 3: Dendogram representing the genetic relationship between the five locations of the Fulani local chicken ecotype

IV. Discusion

Genetic Distance

Though there are scarce literatures to compare the result on genetic distance due to biochemical parameters, however the all positive genetic distance based on identity and on distance in both the Tiv and the Fulani ecotypes is similar to the report of Adebambo (2003), Okpeku et al. (2011) and Ojo (2014) for Nigerian goats from different geographical regions. The value obtained for genetic distance based on distance is very similar to 0.39 and 0.27 reported by Okpeku et al. (2011) but higher than 0.09 reported by Adebambo (2003) as the distance between Red Sokoto and West African Dwarf goat. Also the distance between the Tiv and the Fulani chicken ecotypes (9.13) is higher than value report by aforementioned authors. The values for genetic distance based on identity in both ecotypes were far higher than value reported by above authors. The observed differences may be due to specie differences.

Genotypic frequency

The two haemoglobin allele A and B obtained in this study is consistent with those reported for chuckars and pheasants by Ugur et al. (2006). The genotype frequency observed in this study is in agreement with the earlier report of Yakubu and Aya (2012). Higher frequency of homozygous genotype Hb AA obtained in this study for the Fulani ecotype strongly agreed with homozygouse Hb AA reported for Muscovy ducks (Oguntunji and Ayorinde, 2015) and three varieties of Nigerian indigenous chicken (Yakubu and Aya, 2012). However, higher genotypic frequencies Hb AA and AB obtained in this study respectively for the Tiv and the Fulani ecotypes is in contrast to higher genotype frequency of Hb BB been reported for two ecotypes of Nigerian local chickens (Ige et al., 2013) and Nigerian domestic guinea fowl (Abel et al., 2016). The agreement in gene and genotype frequencies of Hb in the population under study with some previous reports on indigenous poultry could probably be attributed to adaptability to natural environments since most local birds are highly adaptable. However non-agreement with other authors for local chicken and guinea fowl could be due to species differences. This could possibly be an indicator of adaptive physiological modification in bird to survive in area where they are mostly found in Nigeria. The higher genotype frequency obtained in this study for Alb AC fairly agree with the findings of Oguntunji and Ayorinde (2015) who reported higher frequency of Alb CC in Muscovy ducks.

F- Statistics (F_{is}) of Alleles at four Loci of Tiv and Fulani Local Chicken Ecotype

The mostly positive F_{is} values in both ecotype is similar to the report of Barker et al. (2001), Li et al. (2002) and Okpeku et al., (2011) for Asian goats, Chinese goat and Nigerian goat. F_{is} values for the entire population at the four loci which ranged from low to moderate (-0.0046 to 0.4459) and (-0.0046 to 0.4694) in the Tiv and the Fulani ecotypes, respectivelyare in consonance with 0.14, 0.105, 0.11 and 0.58 reported by Barker et al. (2001), Li et al. (2002), Mujibi (2005) and Okpeku et al. (2011) for Asian goats, Chinese goat,

Kenya goats and Nigerian goats, respectively. Low F_{is} value implies that genotypic frequencies among location of each ecotype were not randomly distributed, meaning that the chicken population in both the Tiv and the Fulani ecotypes have some genetic similarity due to gene flow between sampled populations. According to Laval et al. (2000), migration, inbreeding and genetic drift may exert a greater effect on the reduction in genetic differentiation between populations.

Dendogram of Relationships between the two local chicken populations

Generally, the UPGMA trees have shown that the Tiv and the Fulani local chicken ecotypes genetically vary from location to location. From the dendogram, birds from adjacent locations were observed to be more genetically related. These trends are similar to the report of Ige et al. (2013) for Fulani and Yoruba Chicken ecotypes.

Genetic similarity as measured by dendogram supported high genetic flow between locations of the two ecotypes. The wide distance between the birds in location 5 of the Fulani ecotype may be due to genetic admixture. This could occur due to the crossbreeding with non Fulani chicken ecotypes that had stabilized to create and diverge the population genetic structure. This also indicated that, where the Fulani cattle rearers has stoped for a long time, the genetic quality of their birds could be altered by genetic mixture through crossbreeding.

V. Conclusion

The value of the estimated genetic diversity for the Tiv and the Fulani chicken ecotypes indicated that the two populations are variable in their genome and that there are chances of genetic improvement when crossed between themselves across location or with exotic breeds. The genetic diversity within and between the Tiv and the Fulani chicken ecotypes observed in this study should be exploited through selection within each ecotype and subsequent crossing between birds from different locations/ecotype to take advantage of heterosis. It was recommended that the observed uniqueness/distinctness of the Tiv and the Fulani ecotypes should be preserved through such conservative measures as in-situ and ex-situ conservation techniques.

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