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Original Research Article

Testing of Hardy-Weinberg Equilibrium in Population of Two Nigerian Local Chicken Ecotypes Base on Location

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Abstract: Test of deviation from Hardy-Weinberg equilibrium expectation base on location of the Tiv and Fulani local chicken ecotypes of Nigeria was undertaken using eighty (80) matured birds (40 birds each for Tiv and Fulani ecotypes). Blood samples were collected from five locations each of the ecotype to determine their blood protein. Blood protein loci namely: haemoglobin, albumen, transferrin and carbonic anhydrase were determine using electrophoresis. Data collected were analyzed using popgene version 1.31. The result revealed significant deviations ($P<0.05$) from Hardy-Weinberg expectations in the two ecotypes except in the Fulani ecotype were haemoglobin did not deviate significantly. Based on location, there were no significant ($P>0.05$) deviation from Hardy-Weinberg expectations in the Fulani ecotype except in location 4 were Hb, Al and Ca as well as location 5 where Al and Ca were significant. Allelic frequencies of albumin showed significant ($P<0.05$) deviation from Hardy-Weinberg expectations at locations 1 and 5 while allelic frequencies of transferrin deviated significantly ($P<0.05$) from the Hardy-Weinberg frequency at location 5. In the Tiv ecotype, albumin showed the highest significant ($P<0.05$) deviation from the Hardy-Weinberg expectations in all locations. The allelic frequencies of transferrin did not differ significantly ($P>0.05$) from the Hardy-Weinberg expectations. From the finding of this research, it was concluded that the deviation from Hardy-Weinberg expectations at the four blood proteins loci of the two ecotypes indicated that the Tiv and the Fulani chicken populations are variable in their genome and that there are chances of genetic improvement when crossed between themselves across location or with exotic breeds. Generally, these findings provide the fundamental step in the direction of judicious decision-making before the development of genetic enhancement and preservation programmes without interfering with the uniqueness of the Tiv and Fulani chicken ecotypes in Nigeria.

Keywords: Albumin, carbonic anhydrase, haemoglobin, transferring, electrophoresis, polymorphism.

INTRODUCTION

Poultry keeping is an agricultural enterprise with a high potential in Nigeria. More than 40% of households keep poultry with indigenous chickens being the most preferred, accounting for approximately 80% of the reared chicken species (Bobbo *et al.*, 2016). Raising local chicken is preferred to exotic breeds because of their low cost of production, scavenging capacity and adaptability to harsh environmental conditions thus the need for their genetic enhancement and diversity. Genetic diversity in animals is the product of interaction between environment and genetic effects. This interaction leads to differentiation of morphological, physiological and productive traits vital to all production systems provide selection criterion for breed improvement needed for adaptation to ever changing environmental circumstances (Ceriotti *et al.*, 2003). Quantifying the structure of genetic diversity in different African chicken populations is of significance in optimizing genetic improvement, conservation and utilization strategies. Many technologies including

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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 have opened up molecular techniques, such as routine 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). Genetic enhancements need a resolute breeding objective, sustainable breeding plans, and an in-depth comprehension of the genetic diversity of prevailing genotypes and ecotypes (Ojo, 2014). Therefore, elucidating the genetic characteristics of the prevailing local stock will not only favour genetic enhancement but will also expedite their preservation. The objective of the study was to test for deviation from Hardy-Weinberg expectation in populations of the Tiv and Fulani chicken ecotypes base on location using biochemical analysis.

MATERIALS AND METHODS

Experimental Procedure

Two local chicken ecotypes comprising of the Tiv 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 females) 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.

RESULT

Table 1 reveals the observed and expected heterozygosity of the Tiv local chicken populations as well as their Hardy-Weinberg equilibrium (HWE) values. The observed and expected heterozygosity at locations in the Tiv local chicken populations as well their Hardy-Weinberg equilibrium (HWE) values revealed significant ($P<0.05$) deviations from the Hardy-Weinberg equilibrium. However, in the Fulani ecotype (Table 2) there were no significant ($P>0.05$) deviation from Hardy-Weinberg expectations except in locations 4 (Hb, Alb and Ca) and 5 (Alb and Ca) which were significant ($P<0.05$). Allelic frequencies of albumin showed significant ($P<0.05$) deviation from Hardy-Weinberg expectations at locations 1 and 5 while allelic frequencies of transferrin deviated significantly ($P<0.05$) from the Hardy-Weinberg frequency at location 5 in the Fulani ecotype. In the Tiv ecotype, albumin showed high significant ($P<0.05$) deviation from the Hardy-Weinberg expectations in all locations. The allelic frequencies of transferrin did not differ significantly ($P>0.05$) from the Hardy-Weinberg expectations.

Table 1: Hardy-Weinberg Equilibrium for the Tiv Local Chicken Ecotype base on Location

Location 1						Location 2						Location 3						Location 4						Location 5					
L	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**
Hb.	AA	00	2.00			Hb.	AA	00	2.00			Hb.	AA	00	2.00			Hb.	AA	00	2.00			Hb.	AA	0.5	5.28		
	AB	08	4.00	0.03	*		AB	08	4.00	0.03	*		AB	08	4.00	0.03	*		AB	08	4.00	0.03	*		AB	0.3	2.44	1.00	N
	BC	00	2.00				BB	00	2.00				BC	00	2.00				BC	00	2.00				BB	00	0.28		
Al.	AA	00	2.00			Al.	AA	00	1.75			Al.	AA	00	1.75			Al.	AA	00	2.00			Al.	AA	00	2.00		
	AC	08	4.00	0.03	*		AC	07	3.50	0.04	*		AC	07	3.50	0.04	*		AC	08	4.00	0.03	*		AC	08	4.00	0.03	*
	BC	00	2.00				CC	00	1.75				CC	00	1.75				CC	00	2.00				CC	00	2.00		
Tr.	BB	03	3.78			Tf	BB	03	3.78			Tf	DD	00	1.75			Tf	DD	02	3.13			Tf	AA	02	2.04		
	BD	05	3.44	0.49	N		BD	05	3.44	0.49	N		DB	07	3.50	0.04	*		DA	06	3.75	0.44	N		AD	03	2.92	1.00	N
	DD	00	0.78				DD	00	0.78				AB	00	1.75				AA	00	1.13				DD	01	1.04		
Ca.	BB	04	2.5			Ca.	AA	00	2.0			a.	AA	00	2.0			Ca.	AA	04	4.5			Ca.	AA	04	4.5		
	AB	01	3.94	0.05	*		AB	08	4.00	0.03	*		AC	08	4.00	0.03	*		AC	04	3.00	1.00	N		AC	04	3.00	1.00	N
	AC	03	1.53				BC	00	2.00				CC	00	2.00				CC	00	0.50				CC	00	0.50		

Hb = haemoglobin, Al = albumin, Tf = transferrin, Ca = carbonic anhydrase, L = loci, Ge = genotype, Ho = observed, He = expected, S = level of significant, H = hardy Weinberg equilibrium, * = significant at 5% probability and N = not significant, Tiv ecotype: location 1-5 = Uikpan, Daudu, Kadarko, Yelwata and Cohor

Table 2: Hardy -Weinberg Equilibrium for the Fulani Local Chicken Ecotype base on Location

Location 1						Location 2						Location 3						Location 4						Location 5					
L	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**	**L**	**Ge**	**H_o**	**H_e**	**H**	**S**
Hb.	AA	06	6.13			Hb.	AA	07	7.00			Hb.	AA	01	2.53			Hb.	AA	00	2.00			Hb.	AA	07	7.03		
	AB	02	1.75	1.00	N		AB	00	0.00	ND	-		AB	07	3.94	0.14	N		AB	08	4.00	0.03	*		AB	01	0.94	1.00	N
	BB	00	0.13				BB	00	0.00				BB	00	1.53				BB	00	2.00				BB	00	0.03		
Al.	AA	00	2.00			Al.	AA	01	2.53			Al.	AA	02	3.13			Al.	AA	00	1.75			Al.	AA	00	2.00		
	AC	08	4.0	0.0	*		AC	07	3.9	0.1	N		AC	06	3.7	0.4	N		AC	07	3.5	0.0	*		AC	08	4.0	0.0	*
	CC	00	2.00				CC	00	1.53				CC	00	1.13				CC	00	1.75				CC	00	2.00		
Tf	AA	02	2.45			Tf.	AA	05	5.28			Tf.	AA	08	8.00			Tf.	AA	05	5.28			Tf	DD	07	6.13		
	AD	03	2.10	1.00	N		AD	03	2.44	1.00	N		AD	00	0.00	ND	-		AD	03	2.44	1.00	N		DA	00	1.75	0.07	N
	DD	00	0.45				DD	00	0.28				DD	00	0.00				DD	00	0.28				AA	01	0.13		
Ca.	AA	02	3.13			Ca.	AA	05	5.28			Ca.	AA	03	3.57			Ca.	AA	00	1.75			Ca.	AA	00	2.00		
	AC	06	3.75	0.44	N		AC	03	2.44	1.00	N		AC	04	2.86	1.00	N		AC	07	3.50	0.04	*		AC	08	4.00	0.03	*
	CC	00	1.13				CC	00	0.28				CC	00	0.57				CC	00	1.75				CC	00	2.00		

Hb = haemoglobin, Al = albumin, Tf = transferrin, Ca = carbonic anhydrase, L = loci, Ge = genotype, Ho = observed, He = expected, S = level of significant, H = hardy Weinberg equilibrium, * = significant at 5% probability and N = non-significant, Fulani ecotype: location 1-5 = Lafia, Akurba, Adogi, Asakio and Namu.

DISCUSSION

The study reveals a mostly significant (P<0.05) deviations from Hardy-Weinberg expectations in both the Tiv and the Fulani chicken ecotypes across locations. This finding strongly agree with the report of Habimana *et al.,* (2020) who reported significant deviations from Hardy-Weinberg equilibrium Rwandan local chicken. Similarly, this study agrees with the earlier report of Ojo (2014) for four breeds of Nigerian goats. Deviation from Hardy-Weinberg equilibrium at microsatellite level has been reported in various studies (Barker *et al*., 2001; Hassan *et al*., 2003; Laval *et al*., 2000; Luikart *et al*., 1999). The significant difference of the allelic frequencies from the Hardy-Weinberg equilibrium implies that mating in these populations was not random. Inbreeding, selective mating, migration and selection of individuals may have occurred that had altered the allelic frequencies of the haemoglobin, albumin, transferen and carbonic anhydrase at their respective loci. Generally, these findings provide the fundamental step in the direction of judicious decision-making before the development of genetic enhancement and preservation programmes without interfering with the uniqueness of the Tiv and Fulani chicken ecotypes in Nigeria.

CONCLUSION

The finding of this research has revealed that the population of the Tiv and Fulani chicken ecotypes deviated from Hardy-Weinberg expectations across locations due probably to migration, mutation and selection. The deviation from Hardy-Weinberg expectations at the four blood proteins loci of the two ecotypes indicated that the Tiv and the Fulani chicken 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 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. Generally, these findings provide the fundamental step in the direction of judicious decision-making before the development of genetic enhancement and preservation programmes without interfering with the uniqueness of the Tiv and Fulani chicken ecotypes in Nigeria.

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