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GENETIC DIVERSITY OF SELECTED CHICKEN BREEDS IN NIGERIA, SEGREGATING AT THE CHICKEN GROWTH HORMONE GENE LOCUS

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ABSTRACT

The study was carried out to evaluate genetic diversity of selected chicken breeds in Nigeria focussing on the chicken growth hormone gene locus. The chickens studied were: Fulani ecotype, FUNAAB Alpha, frizzled feathered, Noiler and Cobb 500 broilers chicken. Genomic DNA was extracted from blood samples collected from 50 chickens (10/chicken breed). The extracted DNA was amplified using PCR and the gene sequenced. The sequenced data was used to evaluate genetic diversity and to carry out phylogenetic analysis. Results showed that the mean number of alleles (Na), mean number of effective alleles (Ne), mean Shannon information index (I), mean observed heterozygosity (Ho), mean expected heterozygosity (He), mean unbiased expected heterozygosity (uHe), mean fixation index (F) and % polymorphic locus were 2.000±0.000, 1.956±0.014, 0.682±0.004, 0.449±0.067, 0.489±0.004, 0.516±0.004, 0.083±0.138 and 100%, respectively. The gene flow (Nm) for all the population was 13.141. Results of Analysis of Molecular Variance (AMOVA) showed that the percentage of molecular variance among and within individuals was 13 and 87%, respectively. Phylogenetic analysis divided the five chickens into two broad claves; Noiler and the frizzled feathered occupied one while, the Fulani ecotype, FUNAAB Alpha and Cobb 500 occupied the other. It was concluded that great genetic diversity exists in the chicken breeds which could be exploited for further improvement especially in the more native breeds.

Key words: Genetic diversity, chickens, growth, hormone, gene locus.

INTRODUCTION

Growth performance and associated traits such as carcass traits are very important economic characters in broiler chickens production, and they are controlled by sets of multifarious genes. Growth is a very complicated procedure which is synchronized by an extensive network of neuroendocrine pathways. It is therefore very difficult to make rapid progress using more conventional methods of genetic selection within breeds (Zhang *et al.*, 2008). Recent advances in molecular technology have however provided new lee ways to evaluating genetic variability existing at the DNA level of animals (Kaya and Yildiz, 2008). The candidate gene approach has become a very powerful technique for genetic improvement in chicken breeding programmes. Applying this approach (the candidate gene approach), will likely result in higher efficiency in the detection of desired traits necessary to improve productive performances. The chicken growth hormone (*cGH*) gene located on chromosome 27, is one of the most promising candidate genes for improving growth performance and carcass quality traits in chickens (Anh *et al.*, 2015). Growth hormone and the transforming growth factor-β subfamily are the most important groups of hormones that play key roles in many physiological functions such as growth and reproduction. The *cGH* is a 22-kDa protein, consisting of 191 amino acid residues (Hrabia *et al.*, 2008). The *cGH* gene contains four exons and five introns with an overall length of 4.1kb (Kansaku *et al.*, 2008). A cursory look at the growth performance of the various chickens reared in Nigeria will show that they do not grow at the same rate. Could it be that differences exist in the *cGH* of the chickens manifesting in differential performances of the birds especially between the exotic and the more indigenous ones? It is with this in hindsight that the current study was conceived to evaluate genetic diversity at the *cGH* gene locus focussing on selected chicken breeds.

MATERIALS AND METHODS

Blood samples were collected from 50 chickens, 10 each of Fulani ecotype, FUNAAB Alpha, frizzled feathered, Noiler and Cobb 500 broilers chicken. The blood samples kept on ice packs were transported to the laboratory (African Biosciences, Ibadan, Oyo State, Nigeria) where genomic DNA was extracted using the protocols of gSYNC™ DNA extraction kit (Geneaid). The extracted DNA was checked for quality and then PCRed. The PCR products were sequenced and then aligned against the chicken genome using the BLAST programme to verify their identity. Prior to sequencing, the DNA were cleaned to ensure that the gene is not contaminated with impurities. This was done using DNA clean and concentrator kit according to the manufacturers protocol (ZYMO Research). The measurement of genetic diversity including Na, Ne, Ho, He, uHe, I, % polymorphic locus, F and AMOVA were estimated using GenAlEx 6.2 software (Peakall and Smouse, 2012). Dendrogram based on Nei's unbiased genetic distances, using the unweighted pair group method with arithmetic mean (UPGMA), was generated to show the genetic distances of the populations or subpopulations was generated using MEGA X.

RESULTS AND DISCUSSION

The genetic differentiation at the *cGH* gene locus of the chicken breeds is presented in Table 1. The FUNAAB Alpha and Cobb 500 had lower Ne (1.923), I (0.673), He (0.480) and uHe (0.505). The lowest Ho (0.200) was observed in the Cobb 500 while the highest F (0.583) was observed in the Cobb 500 broilers chicken. The entire gene locus was found to be polymorphic (100%) and the Nm over all populations for each locus was 13.141. The mean Na over the loci for each population (2.000) is much lower than the 5.10-6.28 and 3.52-6.62 reported by Lyimo *et al.* (2013) and Mtileni *et al.* (2010) in Tanzanian and South African chickens, respectively. The lower Na in the current study as compared to these other populations is indicative of the presence of a relatively limited sample of gene pool. The mean expected heterozygosity (0.464) concurs with the 0.351-0.434 reported by Marle-Koster and Nel (2000). Mtileni *et al.* (2010) reported expected heterozygosity of 0.67-0.69 among South African free range chickens while Lyimo *et al.*(2013) reported expected heterozygosity values of 0.58-0.67 in Tanzanian chicken populations. The difference observed in heterozygosity values may be due to variation in geography, chicken types, sample sizes, laboratory and sources of microsatellites used. Expected heterozygosity value of <0.5 may be due to inbreeding and admixture associated with population constraints and bottlenecks (Fariba, 2008).

Table 1. Genetic differentiation at the *cGH* gene locus of five chicken breeds

Population	N	Na	Ne	I	Ho	He	uHe	F	%P	Nm
Noiler	10	2.000	1.980	0.688	0.500	0.495	0.521	-0.010	100	
Fulani ecotype	9	2.000	1.976	0.687	0.444	0.494	0.523	0.100	100	
FUNAAB Alpha	10	2.000	1.923	0.673	0.600	0.480	0.505	-0.250	100	
Frizzled feather	10	2.000	1.980	0.688	0.500	0.495	0.521	-0.010	100	
Cobb 500	10	2.000	1.923	0.673	0.200	0.480	0.505	0.583	100	
Mean	**9.800**	**2.000**	**1.956**	**0.656**	**0.516**	**0.464**	**0.477**	**-0.075**	**100**	13.141
SE	0.200	0.000	0.014	0.031	0.126	0.030	0.031	0.225	0.000	

SE=standard error, Na=number of alleles, Ne=number of effective alleles, I=Shannon's information index, Ho=observed heterozygosity, He=expected heterozygosity, uHe=unbiased expected heterozygosity, F=fixation index, %P=percentage of polymorphic locus, Nm=gene flow.

The negative fixation index (F, an index of inbreeding) suggests out-breeding is still occurring in the Noiler, FUNAAB Alpha and frizzled feathered chickens. The reverse is the case in the Fulani ecotype and Cobb 500. The high values could be due to high level of selection involved in the creation of Cobb 500; the close population, in which Fulani ecotype chickens are reared probably, explains the positive nature of the fixation index. Negative fixation index in a population indicates that homozygous deficiencies may have arisen from population sub divisions owing to null alleles, genetic drift and selection against inbreeding (Pemberton *et al.*, 1995). The % polymorphic loci observed (100%) showed that markers used in this study were highly informative in showing genetic diversity. Halima *et al.* (2007) used polymorphic information content to assess how informative markers were and got an average value of 0.71. The main effect of gene flow (Nm) is the homogenization of allele frequencies between populations. The more flow of a particular gene between populations, the more their similarity (El Hentati *et al.*, 2012). The estimated Nm in the studied chicken's populations (13.141) is high; this is evidence of the popular use of commercial chicken lines to improve indigenous ones. The impact of such introgressions is rather limited, possibly due to poor adaptation of exotic birds to village conditions/consumers' preference for local chickens. Diffusive gene flow prevents substantial genetic differentiation due to genetic drift, if gene flow is greater than unity (Slatkin, 1985 cited by Udeh, 2015).

Variation within and between populations of chicken breeds estimated using AMOVA (Table 2) revealed that a large proportion (87%) of the observed variance occurred within the breeds and 13% of the variance was contributed due to differences among individuals within populations. Variation among populations was 0%.

Table 2: AMOVA table showing variation within and between chicken populations

Source of variation	df	SS	MS	Estimate of variance component	% variation
Among population	4	0.464	0.116	0.000	0
Among individual	44	12.944	0.294	0.035	13
Within animal	49	11.000	0.224	0.224	87
Total	97	24.408		0.259	100

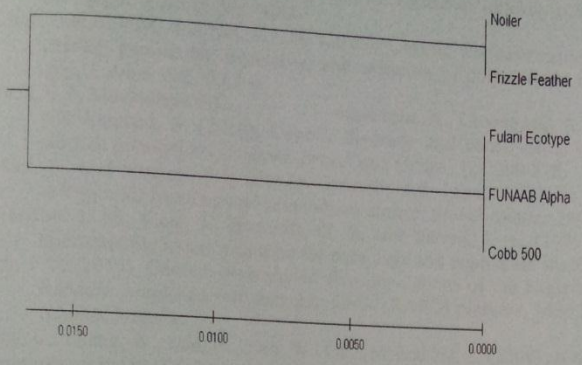

Figure 1: Phylogeny showing genetic relationships between chicken breeds at the *cGH* gene locus

A cluster analysis generated based on Nei's standard distance matrix (Figure 1) showed that Noiler and frizzled feathered chickens were in the first cluster, while Fulani ecotype, FUNAAB Alpha and Cobb 500 were in the second cluster. The closeness of the Fulani ecotype chicken to FUNAAB Alpha and Cobb 500 might not be too surprising because it has some exotic bloodline. The Fulani ecotype chicken could have been developed from crosses between exotic cockerels and indigenous hens. Halima *et al.* (2007) clustered Ethiopian indigenous chickens into two (similar to this result), showing the presence of two major breeds. A cluster shows the level of inbreeding and populations in it could be having coancestry (Halima *et al.*, 2007).

CONCLUSION

Results of the study showed the existence of great genetic diversity in the chicken breeds which could be exploited for the genetic improvement especially of the more native breeds.

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