Short Communication

Hemoglobin polymorphism in Nigerian indigenous small ruminant populations - preliminary investigation

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Hemoglobin polymorphism was studied in the Nigerian indigenous small ruminants. The populations studied comprised of 50 West African Dwarf (WAD) sheep and 150 red sokoto (RS) goats. Blood samples collected from the animals by jugular venipuncture were subjected to starch gel electrophoresis to reveal the activities of hemoglobin (HB). The acetate strip exposed only two co-dominant alleles (A and B) that gave rise to three phenotypes (AA, AB and BB) in the sheep population. Three co-dominant alleles producing four phenotypes (AA, AB, BB and AC) were detected among red Sokoto individuals. The frequencies of allele A and B were 0.61 and 0.40 respectively in the WAD and 0.27, 0.72 and 0.003 corresponding to allele A, B and C, respectively, in red Sokoto goats. In the later, 58% and 42% of the individuals were heterozygote and homozygote, respectively. Genotypic frequencies for AA, BB and AB in rams were 0.40, 0.25 and 0.35 respectively and 0.40, 0.10 and 0.50, respectively, in the ewes. Estimated heterozygosities that represent the proportion of the population that is heterozygote were 0.47 and 0.48 in the red Sokoto goats and WAD sheep population, respectively. The observed gene frequencies deviated significantly from the theoretical Hardy-Weinberg's proportion.

Key words: Hemoglobin, polymorphism, small ruminants, Nigeria indigenous.

INTRODUCTION

Genetic variations among the small ruminant varieties that are indigenous to Nigeria have been a point of concern to geneticists. This is because the amount of genetic variation detectable in an animal population is related to the magnitude of genetic improvement achievable within the species. Thus genetic variation has become the objective tool traditionally used for improving animal species.

The small ruminants of Nigeria have been variously evaluated for genetic variation based on morphological and productive characters/data (Adu and Ngere, 1979). However, morphological variations have been documented to underestimate true levels of genetic variations. In the recent decades, advances in the field of biotechnology has opened up a completely new area at molecular levels with the introduction of techniques such as routine electrophoresis employed for the detection of polymorphi-

sm at protein and enzyme loci as well as other serological and immunogenetic procedures for the measurement of variation.

Data obtained from this type of study could be useful as genetic markers for important economic characters and diseases and could aid significantly in selection of superior animals for breeding purposes. Reports and documentations on these animals from the molecular standpoint are scarce. One of the important erythrocyte proteins that have attracted attention because of its relevance to the selection process is the hemoglobin, an alpha and beta containing polypeptide chain. For example, FAO (1988) reported that the presence of allele A in an individual heep confers upon it a characteristic helminth resistance. Tella (2000) reported that after electrophoresis on cellulose acetate, the Hb genotype that migrated faster from point of application at pH of 8.5-9.0 was labeled HbAA, the slow moving fraction was identified as HbB while the heterozygote was HbAB.

In this study, the small ruminant population of Nigeria precisely the WAD sheep and goats were investigated for

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Table 1. Genotype and gene frequencies of the sheep and goat population.

Species	Sex	No	Genotype frequency				Gene frequency		
			AA	AB	BB	AC	A	B	C
Sheep	F	30	12(0.4)	3 (0.1)	15 (0.5)	-	0.45	0.55	-
	M	20	8 (0.4)	5 (0.25)	7 (0.3)	-	0.28	0.72	-
	Total	50	20 (0.4)	8 (0.17)	22 (0.4)	-	0.48	0.52	-
Goats	Total	150	20 (13.0)	54 (36)	75 (50)	1 (0.01)	0.32	0.68	-

Heterozygosity: sheep = 0.483; Goats = 0.435.

genetic variation at this locus. This documentation will be useful in providing objective basis for planning future selection programmes towards genetic improvement in the species as well as keep pace with the FAO global mandate and strategy for livestock genetic improvement.

MATERIALS AND METHODS

10 ml of blood was drawn from each of 150 adult red Sokoto goats and 40 West African Dwarf (30 ewes and 20 rams) by jugular venipuncture, using needle and syringe into heparinized vacutainer tubes. The blood samples were then washed with normal saline and then hemolysed with distilled water to release the hemoglobin. The supernatant was removed after centrifuging at 3000 rpm for 5 min and sample hemoglobin stored until ready for electrophoresis. Starch gel electrophoresis was employed to separate the globin fractions (Hb and Hb). On electrophoresis, HbA was faster than HbB while the intermediate band was labeled HbAB (hetero-zygote) and the slow band was labeled HbBB. There was another band HbC found in just one of the red Sokoto sample.

Statistical analysis

Estimations

1. Genotype frequency was obtained for each population using the following:

AA = no. of AA x 100/total no. of individual sheep or goat. AB = no. of AB x 100/ total no. of individual sheep or goat.
BB = no. of BB x 100/total no. of individual sheep or goat.
AC = no. AC x 100/ total no. of goats sampled.

2. Gene frequencies for the respective alleles in the populations were obtained using the following:

In case of the sheep:

A = Total number of individuals carrying AA x 2 + ½ no of individuals with AB
B = Total number of individuals carrying BB x 2 + ½ no of individuals with AB

In the case of goats:

A= (Total number of individuals carrying with AA x 2) + (no. of individuals with
    AB) + (no. of individuals with AC).
B= (Total no of individuals carrying BB x 2) + (no. of individuals carrying AB).
C= No. of animals carrying AC.

3. Heterozygosity

Heterozygosity (h) = 1- $_i$

Where = the gene frequency of the ith allele in the ith locus, and i = the no. of loci.

Hardy-Weinberg's equilibrium used for testing the significance of genotypic ratios was based on the expansion of the binomial $(p + q)^2 = p^2 + 2pq + q^2$.

RESULTS AND DISCUSSION

Table 1 presents the genotype and allelic frequencies of individual animals of the sheep and goats. The genotype frequency of HbAA, HbAB and HbBB were 0.4, 0.1 and 0.5 for the ewes and 0.4, .025 and 0.35 for the rams. When sex is ignored, the respective genotypic frequen-cies were 0.4, 0.17 and 0.42 corresponding to 20, 9 and 21 animals. For the goat population genotype frequencies were 13.0, 36.0, 50.0 and 0 corresponding to 20, 54, 75 and 1 animal for allele AA, AB BB and AC. Gene frequen-cies are as presented in Table 1.

Allele B had the highest frequency in both populations: 0.72 in the rams, 0.55 in the ewes and 0.52 when sex was ignored and for the goat populations 0.68. Allele B generally followed with 0.45, 0.28 and 0.48 for the ewes, rams and total, respectively. This did not follow the same trend as with B allele because the frequency for the ewe was higher than that of rams (Table 1). In the red Sokoto goat sample, the same trend was followed, as figure for allele A was lower than that for B, although in all the cases the significance of the difference was not tested. The allele C found only in the red Sokoto popula-tion was associated with the lowest frequency (0.003). This allele, also called a switch gene, whenever found in small ruminants has always been associated with incidence of anemia due to illness and environmental stress.

Evans and Warren (1958) reported that HbA has a selective advantage in sheep at higher altitudes because it constitutes the most common allele in highland breeds of English and Scottish sheep. The population sampled here appeared to have higher HbB frequency suggesting their fit for lower altitude. This agrees with the geographi-cal location from where the samples were derived.

The HbA gene has been reported to confer helminth resistance on the carriers such that the degree of helminth resistance is directly related to the number of doses of the A allele in the locus (FAO, 1988). It is therefore not surprising that the WAD sheep of Nigeria do not demonstrate considerable resistance against helminth infection.

Results from this study present the possibility for increasing helminth/disease resistance (for diseases that are associated with the hemoglobin genetic types) through selection against the HbB allele. The low frequency of HbC emanating from only one animal could either be associated with stress/illness condition of the individual or error in reading the bands due to poor resolution normally experienced for certain experimental protocols in electrophoresis. However, it agrees with the report of Deza et al. (2000) in the goats of Colon and Ischlin who reported 0.02 and 0.04 as the allele frequency for HbC allele respectively. The goat populations of Central Argentina have been reported to have high frequency of allele B while in Japanese stock HbA frequency is higher.

From this study, heterozygotes with intermediate heredity are more than either homozygote. Heterozygosities were also estimated to be 0.483 and 0.435 for the sheep and goat samples, respectively. This value usually indicates the proportion of heterozygotes in a random mating population. The similarity in the estimated heterozygosities therefore suggests that proportions of heterozygotes in the samples were similar although the significance of the difference was not tested. It has a clear bio- logical meaning as it can be regarded as the measure of genetic diversity at the hemoglobin locus in the investigated populations and that both of them have similar flock structure and mating pattern.

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