Preliminary investigation of haemoglobin polymorphism among Indigenous cattle in north western Nigeria.

Abstract

The aim of this study was to characterize the genetic pool of the Bunaji and Sokoto-Gudali cows using haemoglobin (Hb) polymorphism. Blood samples were collected from a total of 150 mature Bunaji, Sokoto-Gudali and HolsteinXBunaji cows in north western Nigeria. The red cell lysates were subjected to cellulose acetate electrophoresis and specific staining procedure to reveal the band patterns of haemoglobin. Three co-dominant alleles, causing the presence of three genotypes (AA, AB and AC) were detected among individual cattle. The gene frequencies of the A and B alleles were 0.62 and 0.38, respectively. The corresponding genotype frequencies for AA, AB and AC in the cattle population were 0.34, 0.57 and 0.10, respectively. The discrepancy between the observed and the expected genotype number was significant ($P<0.05$) thereby violating the Hardy-Weinberg frequencies. There is a need for further study to determine the association between haemoglobin variants and production traits in cattle.

Key words: haemoglobin, polymorphism, morphometric traits, cattle, Nigeria

Introduction

One key component of a strategy to expand food security is genetic characterisation of indigenous breeds and their biochemical properties [1]. In Nigeria, several studies on diverse livestock species have been published [3]. Due to low production, local indigenous breeds have gotten less attention, causing breeders to transfer their focus to temperate cattle breeds in order to improve their local genetic resources. Much genetic research is currently focused on determining the link between physiological, biochemical, and metabolic products/markers and farm animal productivity efficiency [2]. The physiological basis of performance qualities and the influence of heterosis have been investigated using biochemical traits such as blood types, blood proteins, and enzymes [4]. The traditional approach to better animal breeding is based on phenotypic differences found within and between related groups of individuals, although they are only partly attributable to genetic differences and partly owing to environmental factors. Haemoglobin is a blood protein that is made up of four globin chains (two each of alpha and beta) plus a prosthetic group called haem [3]. Human haemoglobin was the first protein to be investigated, and it continues to be the most studied blood protein [13]. Several studies in cattle and sheep have already connected these markers to production qualities and environmental adaptation, in addition to several critical activities of blood proteins [5]. [10] found that Romanian Brown cows in Moldavia with haemoglobin type AB had higher milk yield (2271–2900 vs 1900–2471 litres) and milk fat yield (84.6–105.0 vs 67.5–103.7 Kg) than other kinds (AA, BB). In terms of milk yield and fat percentage, BB cows were shown to be inferior to the other two categories (AB, AA). According to [10] Romanian Brown Cows of Hb types A, AB, and B had an average first calving age of 45.14, 40.52, and 36.66 months, average milk yields of 2478, 2578, and 2277kg, and average fat yields of 95.78, 95.78, and 95.78, respectively. [11] found that Hb types significantly age at first calving in all groups except Holstein crossbreds, service period in Brown Swiss crossbreds, and milk yield in Brown Swiss crossbreds and Hariana purebreds in crossbreds (Holstein X Hariana, Brown Swiss X Hariana, Jersey X Hariana) and Haria Cattle. Although morphological classification of cattle has been attempted in Nigeria [14]), there is a scarcity of information on their molecular characterization. Therefore, the present investigation aimed at describing the genetic structure from the level of the determinant locus of haemoglobin variants and its association with milk and morphological traits in Bunaji, Sokoto-Gudali and FriesianXBunaji cattle.

Materials and Methods

Location of the experiment

The research was conducted at the Dairy Breeding Unit of National Animal Production Research Institute (NAPRI), Shika, Zaria, Kaduna State. NAPRI is geographically located between latitude 11^0 and 12^0N and longitude 7^0 and 8^0E at an altitude of 640m above sea level.

Animals and Management

Animals used for this research were sourced in National Animal Production Research Institute (NAPRI). They were raised under semi intensive system of management. A total of 150 cattle comprising of equal number of Friesian X Bunaji cross, Sokoto Gudali and White Fulani were used for the study.

Blood Collection

The red cells were separated, washed in saline solution and lysed with distilled water. Haemoglobin was typed using cellulose acetate electrophoresis as described by [6] with a slight modification [13]. The identification of the haemoglobin types in cattle was achieved in accordance with the migration speed of the light spots on the electrophoretical substratum, detected from the start line towards the cathodal zone. The direct gene counting method was used to score Hb bands based on the separation of Hb variants.

Statistical Analysis

Genotype and gene frequencies of Haemoglobin (Hb) alleles were estimated according to Hrinca (2008). Data on Hb alleles and of genotype frequencies were subjected to chi-square analysis to test for goodness-of-fit for observed and expected frequencies under Hardy-Weinberg equilibrium (HWE). Heterozygosity (H) was estimated as the expected proportion of heterozygotes under HWE using J.M.P genomics software.

Result and Discussion

Genotype and Gene Frequencies of Haemoglobin among Breeds

Table 1 presents the gene and genotypic frequencies of haemoglobin variants. The observed and expected numbers were also indicated. The HbAA overall count was 28 with a frequency of 0.34, HbAB was 47 and 0.57 and HbBB was 8 and 0.10. Allelic count and frequency were (51.50 and 0.62) and (31.50 and 0.38) for the A and B variants. In the Bunaji the count and frequency were HbAA (5.00 and 0.24), HbAB (12.00 and 0.57), HbBB (4.00 and 0.19) while allelic frequency indicated similar count and frequency of HbA (11.00 and 0.52) and HbB (10.00 and 0.48). The Friesian X Bunaji cross indicated (3.00 and 0.25), (7.00 and 0.58) and (2.00 and 0.17) respectively for genotypes observed; (6.50 and 0.54) and (5.50 and 0.46) for allelic counts and frequencies. In the Gudali the following were observed for the genotype; (20.00 and 0.40), (28.00 and 0.56) and (2.00 and 0.04) while the allelic counts and frequencies were (34.00 and 0.68) and (16.00 and 0.32) respectively. The χ^2 value 1.27 was not significant ($p>0.05$) indicating conformity to Hardy-Weinberg equilibrium for the haemoglobin loci across all the breeds. Due to their polymorphic nature, large number and simple Mendelian pattern of inheritance, the biochemical systems are used in the study of breed origin, determining structure and relations between breeds, and for the correlations that can be established with several traits such as production, resistance to diseases [10]. Though after the widespread use of molecular characterization techniques, the enthusiasm of biochemical polymorphism studies has gone down considerably, however, it is still popular especially in the developing world where sophisticated lab facilities are not readily available. The frequencies 0.62 (HbA) and 0.48 (HbB) obtained for the pooled Bunaji, Friesian X Bunaji and Gudali cattle populations were comparable to the report of 0.64 and 0.36 respectively reported by [1] for Zebu cattle and their crosses in Zaria. The higher frequencies of HbA (0.52, 0.54 and 0.68) obtained respectively for the Bunaji, Friesian X Bunaji and Gudali cattle were consistent with the observation of [10] that most of the animals studied revealed a high genotypic frequency of 0.518 for HbAA in haemoglobin types in Bunaji cattle and their Friesian crosses in Shika, Zaria-Nigeria. The Preponderance of the HbAB over the HbAA and HbBB across all the breeds and in the pooled information were consistent with the report that Zebu cattle and their crosses with HbAB show a high degree of dominance over HbAA and HbBB and this was a result of natural selection [1]. Also the lack of significant ($p>0.05$) variation among the frequencies observed also agreed with the findings of [5]) and may perhaps, apart from pointing to the stability of the population with regards to these genes, indicate that the Zaria environment may be favouring the HBAA genotype by natural selection, this observation confirms the conclusion of [5] that Hb type AA in the Nigerian Friesian-Bunaji crosses of cattle of Zaria environment was favoured by natural selection, possibly for adaptation against the hot tropical weather and tolerance of tick and helminthic infections which was not achievable with exotic cattle breed. Also [1] in reporting the effect of age, sex and haemoglobin type on adaptive and blood biochemical characteristics of Red Sokoto goats have stated that the variation of Hb type with adaptive and blood biochemical characteristics was significant ($P<0.05$) and that the relationship between Hb types and HR can be linked to the different oxygen carrying capacity of the Hb types. In their study, higher Heart Rate was observed in goats with HbAA and HbAB, and HbA is known to be the haemoglobin allele with highest affinity for oxygen. This is in line with the earlier report of [13] who related the preponderance of HbA to greater affinity for oxygen, thus explaining the high adaptive coefficient they observed on goats with Hb types AA and AB since adaptive coefficient is a function of Heart Rate and Rectal Temperature.

Table 1: Genotype and Gene Frequencies of Haemoglobin among Breeds of Indigenous Cattle

Variable	Genotype			Gene		χ2 df=4	LOS
	AA	AB	BB	A	B	1.27	NS
Overall							
Observed	28.00	47.00	8.00	51.50	31.50		
Expected	27.70	27.70	27.70	41.50	41.50		
Frequencies	0.34	0.57	0.10	0.62	0.38		
Bunaji							
Observed	5.00	12.00	4.00	11.00	10.00		
Expected	9.00	9.00	9.00	10.50	10.50		
Frequencies	0.24	0.57	0.19	0.52	0.48		
Friesian X Bunaji							
Observed	3.00	7.00	2.00	6.50	5.50		
Expected	4.00	4.00	4.00	6.00	6.00		
Frequencies	0.25	0.58	0.17	0.54	0.46		
Gudali							
Observed	20.00	28.00	2.00	34.00	16.00		
Expected	16.70	16.70	16.70	25.00	25.00		
Frequencies	0.40	0.56	0.04	0.68	0.32		

NS-Not significant

The effect of the haemoglobin polymorphism on growth and milk traits of the breeds of cattle are presented in Table 2: Polymorphic forms of haemoglobin only significantly ($p<0.05$) influenced BW, BL and CW. While the other traits indicated non-significant variations due to haemoglobin types. The highest BW and BL was observed with the AB genotype (391.15 and 178.95) while the BB and AA homozygote individuals exhibited statistically similar ($p>0.05$) magnitude for these traits. CW was also larger with the homozygote individual and differed from the BB genotype, while the BB genotype differed from the AA genotype. It was also noted that for most traits studied which did not show significant variation, the heterozygote had higher mean values for most of these traits. It was observed that BW, BL and CW were observed to be significantly ($p<0.05$) influenced by Hb types. There exists a deficit of literature reports on the impact of Hb types on growth and production traits. However, [8] reported that haemoglobin types had no significant influence on LL. No direct evidence exists of differences among the three Hb phenotypes (AA, AB and BB) for fitness in cattle [4]. [2] reported that Hb type did not significantly affect milk yield and butterfat percentage in dairy cattle. The influence of Hb variants on BL and CW were comparable to the observation of [13] in goats. There exist variations in literature reports on the effect of polymorphic forms of Hb on body traits; [2] had reported that haemoglobin type had influence on performance of sheep and goats. While [3] found no significant relationship between Hb type and body weight, body length, heart girth and height at withers in Garole sheep. It should be noted that goats exhibit a very complex Hb polymorphism due to the presence of a number of allelic and non-allelic chains both in the alpha and beta globin systems [9]. This may be responsible for the lack of clear pattern and accord in obtained results and literature reports on their impact on morphological traits. However, the significant effect of the interaction between breed and Hb types on growth and milk production traits in this study may point to the impact of Hb on fitness which may in turn influence growth and productivity. However, the lack of preceding literatures makes it difficult to compare and contrast. It may be posited that breed and Hb interaction may be a good source of variation in adaptability and productivity.

[abc:] Means with different superscript across rows differ significantly ($p<0.05$

Table 2: Effect of Haemoglobin Types on Morphology and Milk Production

Haemoglobin	AA	AB	BB	SEM
BW	383.83[b]	394.15[a]	387.92[b]	3.35
BL	176.99[b]	180.95[a]	178.59[b]	1.01
HW	174.17	174.87	173.88	0.78
CW	24.98[c]	28.58[a]	25.75[b]	1.13
HG	126.5	125.63	125.26	1.20
Rumwidth	45.29	46.43	44.80	3.08
TL	4.66	4.83	4.72	0.36
RUH	20.42	20.59	20.72	1.95
UC	41.52	42.04	41.99	1.56
TY	1187.81	1187.81	1172.24	47.71
ADY	4.49	5.45	5.37	0.76
LL	233.34	244.82	241.64	9.61

Conclusion

The study revealed overall frequencies of 0.62 (HbA) and 0.48 (HbB). Higher frequencies of the A allele (0.52, 0.54 and 0.68) were obtained respectively for the Bunaji, Friesian X Bunaji and Gudali cattle in the Haemoglobin locus. Study of blood protein polymorphism and productivity indicated significant ($p<0.05$) influence of the haemoglobin, on both body and milk production traits with no definite patterns, but rather it was more of the bye product of the fitness and adaptability they confer on the animals. However, it was noted that the heterozygote genotype for most of the locus and breed were superior to the homozygote.

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