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Genetic diversity and relationship of indigenous goats of Sub-saharan Africa using microsatellite DNA markers

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Abstract

Sub-Saharan African goats with an estimated population of 180 millions are major asset for farmer communities in a range of agro-ecological zones. This study was undertaken to determine the genetic diversity in and differentiation of relationships among 18 populations of goats from Uganda (4), Tanzania (5), Kenya (2), Mozambique (2), Nigeria (3), Mali (1) and Guinea Bissau (1). Heterozygosity, estimates of F_{ST}, genetic diversity and distances were performed using data from 11 microsatellite DNA loci.

Expected heterozygosity ranged from 0.450 in Guinea Bissau population to 0.541 in Mbeya population (Tanzania), while the observed heterozygosity ranged from 0.441 in Pafuri population (Mozambique) to 0.560 in Sebei population (Uganda). Mean number of alleles (*MNA*) per population ranged from 3.82 to 5.91. Gene differentiation (F_{ST}) among populations was low (5.3%), a result confirmed by genetic distances (D_A).

Our results reveal that genetic relationships between populations reflect their geographical proximity rather than morphological classification.

Key Words: DNA markers, gene diversity, selection

Introduction

Africa comprises diverse agricultural environment determined by climate, natural resource and human population density. The economic importance of livestock in African farming systems increases with decreasing rainfall. Livestock production is vital to subsistence and economic development (Winrock International 1992). The ever increasing demand for livestock production to cater for the nutritional needs of rapidly growing human population has led to indiscriminate crossbreeding and replacement of indigenous goats with exotic breeds in an effort to improve productivity. These goats are endowed with unique qualities such as water economy, heat tolerance, disease resistance, mothering and walking abilities, and the ability to efficiently metabolize low quality feeds (Trail and Gregory 1984).

If genetic diversity is very low, none of the individuals in a population may have the characteristics needed to cope with the new environmental conditions or challenges. Such a population could be suddenly wiped out. Low amounts of genetic diversity increase the vulnerability of populations to catastrophic events such as disease outbreaks. Low genetic diversity may also indicate high levels of inbreeding with its associated problems of expression of deleterious alleles or loss of over-dominance. Change in the distribution of the pattern of genetic diversity can destroy local adaptations and break up co-adapted gene complexes. These problems combine to lead to a poorer 'match' of the population to its habitat increasing and eventually leading to the probability of population or species extinction.

Additionally introgression from other populations, uncontrolled interbreeding among indigenous goat breeds, absence of breed improvement programme and political instability in some countries further endanger them. As a result some indigenous African goat breeds are at a risk of extinction, while others might be losing their unique genetic adaptation to local production systems. It is therefore urgent to understand the genetic diversity of indigenous goats of Africa to implement steps to ensure their conservation and rational utilization for improvement of productivity for the benefit of the farmers**.**

Materials and methods

Sampling

A total of 749 blood samples were randomly collected in 19 populations (Table 1) including one population from Switzerland used as reference population. Genomic DNAs were extracted from the blood samples following the method of Sambrook et al (1989). Eleven microsatellite loci studied were: *MAF209*, *INRA132*, *BM1818*, *ILSTS011*, *INRA063*, *SRCRSP03*, *BMS1494*, *ILSTS044*, *ILSTS005*, *ILSTS087* and *MAF35*.

PCR amplification

PCR amplification was done on 20 ng template DNA in a 10 µl reaction volume. The 10 µl reaction volume contains 1 µl template DNA, 1 µl 10 X PCR buffer, 0.5 µl dNTPs, 0.1 µl forward primer, 0.1 µl reverse primer, 0.1 µl *Taq* polymerase and 7.2 µl of double distilled water. Cycling profile included an initial denaturation step at 95^{o}C for 5 minutes, followed by 35 cycles of 30 seconds at 95^{o}C, 1 minute at 55-58^{o}C depending on the primers used and 1 minute at 72^{o}C, and a final extension step at 72^{o}C for 10 minutes using a GeneAmp 9700 (Applied Biosystems) thermal cycler.

Table 1. Sample information

	Country	Population/breed	Sample size
1	Guinea Bissau	Gubu/Bissau	46
2	Kenya	Boran Galla	36
3	Kenya	Small East African	39
4	Mali	Maure	37
5	Mozambique	Pafuri	38
6	Mozambique	Landim	37
7	Nigeria	Red Sokoto	41
8	Nigeria	Born White	36
9	Nigeria	West African Dwarf	37
10	Switzerland	Grison Striped	31
11	Tanzania	Maasai	44
12	Tanzania	Small East African (Coat)	41
13	Tanzania	Small East Africa (Mbeya)	38
14	Tanzania	Ugogo	46
15	Tanzania	Ujiji	39
16	Uganda	Karamoja	36
17	Uganda	Teso	49
18	Uganda	Kigezi	38
19	Uganda	Sebei	40

Microsatellite genotyping

The resultant PCR products were electrophoresed for 2 hours through a 36 cm, 4.25% polyacrylamide denaturing gel on an ABI Prism™ 377 automated DNA sequencer to separate the alleles. The Genescan™ analysis software version 3.1.2 was used to size the resultant DNA fragments following electrophoresis. The Internal size standard used in this study was the Genescan™ 350-Tamra. The resultant data was imported into the Genotyper™ analysis Software version 2.0. The first step in Genotyper analysis was to check whether the internal size standard DNA fragments were accurately assigned (35, 50, 75, 100, 139, 150, 160, 200, 250, 300, 340 and 350 bp).

Results

Genetic variation

A total of 101 alleles were detected at the 11 microsatellite loci for all populations. The highest number of alleles observed was 14 at *BMS1494* while the lowest was five at *MAF35*. Table 2 shows the observed and expected heterozygosities and the mean number of alleles (MNA) per population averaged across the 11 microsatellite loci. The expected heterozygosity ranged from 0.450 in Guinea Bissau population to 0.541 in Mbeya population. The observed heterozygosity ranged from 0.441 in Pafuri population to 0.559 in Sebei population. The MNA ranged from 3.82 ± 1.89 in Ujiji and Guinea Bissau populations to 5.91 ± 2.88 in Ugogo population.

Table 2. Expected and observed heterozygosities and MNA with their standard errors within each population studied

Population	Sample size	He ± s.e.	Ho ± s.e.	MNA
Karamoja	36	0.525 ± 0.060	0.495 ± 0.025	4.64 ± 1.63
Teso	49	0.515 ± 0.061	0.464 ± 0.022	5.09 ± 2.07
Kigezi	38	0.509 ± 0.060	0.506 ± 0.025	4.55 ± 2.11
Sebei	40	0.470 ± 0.064	0.559 ± 0.024	4.18 ± 1.72
Mbeya	38	0.541 ± 0.051	0.530 ± 0.025	5.00 ± 1.84
Coat	41	0.510 ± 0.074	0.465 ± 0.024	5.55 ± 2.07
Ugogo	46	0.527 ± 0.067	0.466 ± 0.022	5.91 ± 2.88
Maasai	44	0.482 ± 0.066	0.527 ± 0.023	4.27 ± 2.20
Ujiji	39	0.473 ± 0.066	0.459 ± 0.024	3.82 ± 1.89
Grison Striped	31	0.460 ± 0.089	0.468 ± 0.027	4.45 ± 2.77
Pafuri	38	0.458 ± 0.071	0.441 ± 0.024	5.00 ± 2.45
Landim	37	0.453 ± 0.072	0.435 ± 0.025	4.18 ± 1.40
Red sokoto	41	0.527 ± 0.062	0.516 ± 0.024	4.82 ± 2.44
Born white	36	0.531 ± 0.064	0.489 ± 0.025	5.09 ± 1.76
Maure	37	0.526 ± 0.061	0.492 ± 0.025	5.18 ± 1.99
West African Dwarf	37	0.506 ± 0.070	0.548 ± 0.025	4.64 ± 2.54
Small East African	39	0.504 ± 0.068	0.477 ± 0.024	4.18 ± 1.60
Boran Galla	36	0.492 ± 0.063	0.485 ± 0.025	4.18 ± 2.23
Guinea Bissau	46	0.450 ± 0.067	0.465 ± 0.022	3.82 ± 1.83

Genetic distances and relationships between populations

The highest D_S value was observed between West African Dwarf (WAD) and Landim populations (0.111) and the lowest was between Karamoja and Teso populations (0.002) (Table 4). Large differences were observed between populations from different region: West Africa, East Africa and Southern Africa countries, but slight differences were observed between populations from the same country. The reference populations (Grison Striped and Guinea Bissau goats) had D_S values

significantly different from the rest of the populations. The largest D_A distance was observed between WAD, Ujiji and Landim populations (0.095) and the lowest between Karamoja and Teso populations (0.028). The distances between all populations were generally low, but significantly different from the reference populations (Guinea Bissau and Grison Striped goats).

Population differentiation

Population differentiation was estimated using F_{ST} and G_{ST} values. The mean genetic differentiation within population (H_S) was 0.498 and in the total populations (H_T) was 0.524. Both values were lowest at *MAF35* (0.146 and 0.149 respectively) and highest at *BM1818* (0.727 and 0.748 respectively). The genetic differentiation (G_{ST}) ranged from 0.024 at *MAF35* to 0.086 at *BMS1494*. The overall G_{ST} values for all markers across all populations was 0.05 indicating that 5% of the total genetic diversity was observed among populations, while 95% was within population.

In this study, a relatively low F_{ST} (5.3%) indicated that genetic differentiation among populations was limited. The F_{ST} estimator of Weir and Cockerham (1984) (5.3%) was close to the Nei's G_{ST} estimator (1978) (5%). They both indicated that the genetic differentiation among the sub-Saharan goat populations is very low. 95% of the total allelic variations account for genetic variation among individuals within populations and only 5% of the total allelic variation account for genetic variation among populations.

Table 3. Genetic diversity at the 11 microsatellite loci studied

Locus	H_O	H_S	H_T	G_{ST}	F_{ST}
ILSTS087	0.276	0.322	0.334	0.036	0.035
ILSTS005	0.143	0.160	0.167	0.037	0.038
ILSTS044	0.502	0.626	0.660	0.051	0.055
ILSTS011	0.667	0.641	0.688	0.068	0.071
INRA132	0.538	0.551	0.565	0.025	0.025
INRA063	0.702	0.632	0.656	0.037	0.039
BM1818	0.648	0.727	0.748	0.028	0.029
BMS1494	0.511	0.613	0.671	0.086	0.095
MAF35	0.125	0.146	0.149	0.024	0.025
MAF209	0.516	0.516	0.561	0.080	0.080
SRCRSP03	0.749	0.543	0.570	0.049	0.051
Overall	0.489	0.498	0.524	0.050	0.053

Phylogenetic analysis

The D_A NJ tree clustered the population into three major clusters mainly along the geographical locations. The first cluster consisted of 11 populations from East African countries. These populations included Coat, Ugogo, Maasai, Small East African, Boran Galla, Karamoja, Teso, Mbeya, Ujiji, Kigezi and Sebei populations. The second cluster comprised of Pafuri and Landim populations from Mozambique. The third cluster included four populations of WAD, Borno White, Red Sokoto and Maure from West Africa. The reference populations formed another group with the Grison Striped goats been closer to the West African populations compared to Guinea Bissau population.

Discussion

The microsatellite DNA loci have shown a high genetic polymorphism. The sub-Saharan goats had a considerable amount of within population variation based on analysis of molecular variance, heterozygosities and number of alleles. The number of alleles ranged from 5 to 14 which are lower than Ethiopian goats (4 to 23) (Tesfaye 2004), West African Dwarf goats (4 to 21) (Mujibi 2005) and Swiss goat breeds (3 to19) (Saitbekova et al 1999). The relatively low number of heterozygotes indicates excess of homozygotes which could be due to locus under selection, null alleles, inbreeding or presence of population of substructure (Wahlund effect).

All populations studied showed significant differentiation and structuring within themselves. The F_{ST} and G_{ST} values overall populations was 0.053 and 0.05 respectively. This indicates that about 5% of the total genetic diversity was observed among populations and 95% was observed within populations. The total genetic diversity observed between populations was similar to other studies done on African goat populations namely West African Dwarf goats was 5.4% (Mujibi 2005). But populations outside Africa showed slightly higher between population variation [17% among Swiss goats (Saitbekova et al 1999), 11% among Italian goats (Ajamone-Marsan et al 2001) and 10.5% among Chinese goats (Li et al 2002)]. When a population is divided into isolated subpopulations, there is less heterozygosity than there would be if the population was undivided. Founder effects acting on different demes generally lead to subpopulation with allele frequencies that are different from the larger population.

The NJ tree classified the populations into three major clusters mainly along the geographical locations. The reference breeds stood out distinctly different from the rest thus suggesting a different ancestry. The information obtained in this study will aid their rational development, utilization and conservation.

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