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**Review Article** 

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# Usefulness of short sequence repeat markers in goat genetic diversity studies on the Asian and African continents

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**Abstract:** Goat genetic diversity studies are very important since extinction of germ plasma is increasing very rapidly. The African and Asian continents are the hotspots for indigenous animals with fewer genetic manipulations. Conservation studies using molecular markers have the capability to validate the real status of the animals. The use of molecular markers has revolutionized studies of genetic diversity. Even though a number of markers are used for these types of studies, short sequence repeats are in the forefront due to their superior features such as high variability, high mutation rate, large number, distribution throughout the genome, codominant inheritance, and neutrality with respect to selection. The aim of this review is to emphasize the importance of microsatellite markers for studies of genetic diversity in the goat and their use in conservation strategies in Asia and Africa.

Key words: Molecular markers, conservation, breeding, dominant, variability

# 1. Introduction

Domestication of animal species probably occurred during the Mesolithic period around 8000–7000 BC (1,2). Later, evolutionary forces of migration, mutation, selection, genetic drift, and creative human activity jointly contributed to the origin of numerous identifiable morphological characteristics and a colossal amount of variability in production performance. Hence, a vast array of landraces, populations, and breeds constitute domestic animal diversity. Interestingly, it was only in the 17th century that Robert Bakewell from Dishley, England, categorized animals with similar morphological characteristics into a population and developed the foundation of pedigree breeding based on the concept of "like begets like" and "breed the best to the best" (3).

In the 18th and 19th centuries, raising sheep was more or less traditional and under conditions of sedentary, nomadic, and seminomadic management. A number of sheep breeds evolved in the desert, tropical, temperate, and mountainous regions of the world where rainfall, wind, temperature, solar radiation, and vegetation varied. During this period, increased emphasis was placed on conformation, hardiness, and productivity, and this emphasis invigorated interest in the development of new sheep breeds derived from a combination of 2 or more breeds (4).

#### 2. Historical background

History sometimes takes ironic twists, and the history of science is no exception. Microsatellites have been detected in eukaryote genomes for over 30 years, though they were regarded as sequences of no particular interest. With the rise of polymerase chain reaction (PCR), it was realized in the late 1980s that microsatellites may be the most powerful Mendelian markers ever found (see Table 1). They have since been widely studied in conjunction with some genetic diseases. They also have been used in mapping programs and by population biologists for kinship investigations and for more classical studies of population genetic structure (5–9).

Worldwide, recognition exists for the need for conservation of livestock diversity (10) and for characterization of breeds and populations including their genetic differentiation and relationships. These unique characteristics are the result of evolutionary forces and their interactions over long periods of time. However, the adaptations and unique characteristics might have been diluted due to intermixing, substructuring, and/ or consequent genetic drift in the population over time. Moreover, the small population of microsatellites, approximately less than 5000 (11,12), makes them further vulnerable to the various forces of genetic change, thus

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modifying the foundation genetic structure of the breed (13). Therefore, an investigation of genetic variation within the breed and its body structure may help to evaluate these factors and provide genetic information to be used for conservation and improvement of goats of Asia and Africa (14,15).

The International Goat Genome Consortium (IGCC) is a very good initiative to increase the genomic tools and knowledge dissemination in the public sphere on goat species. The current projects of the IGCC are to produce a goat whole-genome reference sequence that will be initiated via de novo assembly from an expressed sequence tag-based virtual goat genome and bacterial artificial chromosome clones. Discovery of single nucleotide polymorphism (SNP) chips using next-generation sequencing techniques and mapping of markers for RH map and Hap Map developments are some of the other projects of the IGCC (16).

# 2.1. Comparison of goat genetic diversity

Methodology for research in population genetic diversity has improved tremendously over the past 2 decades since the application of advanced molecular techniques (17). Genetic characterization studies also showed a steep increase. Genetic characterization is carried out in livestock using various molecular biology techniques such as allozymes, restriction fragment length polymorphism (RFLP), protein polymorphism, randomly amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), mitochondrial DNA (mtDNA), short tandem repeat (STR or microsatellites), and SNPs (18-23). Comparisons of the attributes of some of these important molecular markers are listed in Table 1. Attempts have been made by using microsatellite (24,25) and mitochondrial (26) markers to establish breed characteristics and to determine relationships among indigenous goat breeds. Out of many genetic markers now available, microsatellite loci are best suited for answering some of these questions (27) because of their high variability, high mutation rate, large number, distribution throughout the genome, codominant inheritance, and neutrality with respect to selection (28). They are very

useful to analyze the degree and pattern of genetic variability within and differences between populations.

Once extracted from the chosen matrix (animal tissue, blood, muscle, hair, sperm, feces, or even processed food such as cheese or canned meat), the DNA is analyzed by molecular markers to obtain a fingerprint or specific allelic frequencies allowing for individual, breed, or species identification. Since the introduction of PCR in 1989, many different markers have been studied. Presently the most widely used are microsatellites, which are also known as STRs and SNPs (29). Although DNA analysis furnishes different levels of identification, the individual one is of great interest for the verification of a meat cut for food safety purposes, while breed and species discrimination are of interest to detect fraud and to protect and validate typical productions. The use of these technologies in animals and their products is just an extension of techniques already in use for human testing and routinely applied in forensic casework (30). The most widely used markers are microsatellites (31-37) and, most recently, SNPs (35,38,39). The results from this research in goats, the type of markers utilized, and the breeds studied are shown in Table 1.

It is worth mentioning an important aspect when choosing the markers and the breeds to be analyzed (37): in a study on 4 cattle breeds, the informative content of each microsatellite varied among breeds depending on the breeds' allelic frequencies (alleles always present in one breed and always absent in the others), especially in genetic characterization studies. When implementing a genetic trace-back system it would be interesting to choose different panels for each breed to achieve good efficacy in all breeds. In both cases, preliminary analyses of all breeds are needed to determine the genetic structure of each population.

Another interesting trait of microsatellites is that we can relatively easily gain information on their molecular structure and mutation rate as well. This has not escaped the attention of population geneticists. Recent work at the population level may also shed light on the molecular forces acting on microsatellites. Microsatellites have

Characters	RFLP	RAPD	SSR
Genomic abundance	High	Very high	Medium
Level of polymorphism	Medium	Medium	High
Dominance	Codominance	Dominance	Codominance
Quantity of DNA needed	2–10 µg	10–20 µg	50–100 μg
Sequence information requires	None	None	Yes
Null alleles	Rare	Not applicable	Occasional
Automation or multiplexing	Difficult	Possible	Possible
Radioactive probes	Yes/no	No	No

Table 1. Comparison of some of the important DNA markers.

become widely applied for several types of studies, due to their advantages over other markers, such as the relative ease in obtaining markers, high polymorphism rates, neutrality, and easy automation of analytical procedure (40,41). Additionally, variation in simple nucleotide repeats, random and abundant distribution across the genome, and codominance can be determined (42).

Due to their close chromosomal resemblance, microsatellites developed for cattle and sheep normally

work well in goats (43). The International Society of Animal Genetics described more than 1400 microsatellite markers that have been listed in cattle and around 40% of those markers can be amplified efficiently in goat (44).

Microsatellites are the marker of choice in animal genetic studies due to the above mentioned advantages, and these markers have been used in numerous studies all over the world. The use in goats is listed for the Asian and African continents in Table 2. At the turn of the century,

**Table 2.** Overview of goat genetic diversity studies in Asia and Africa, microsatellites used, observed sizes of their alleles, number of alleles  $(n_a)$ , observed heterozygosity (Ho), and expected heterozygosity (He).

Breed/origin	n	H	H	Reference
South Asian breeds (n)	u	0		
Jamunapari	4.91	0.42	0.54	(45)
Kutchi	12	0.59	0.80	(46)
Gohilwari	10.12	0.51	0.69	(47)
Marwari	5.8	0.45	0.63	(48)
Barbari	6.33	0.85-1.0	0.62-0.85	(49)
Zalawadi	7	0.6	0.58	(50)
Gohilwadi	7.82	0.63	0.67	(50)
Surti	7.06	0.58	0.64	(50)
Mehsana	12.28	0.65	0.77	(51)
Indian domestic goats (7)	8.1-9.7	0.37-0.43	0.74 - 0.78	(52)
Kanniadu	5-14	0.71-0.98	0.64-0.87	(53)
Sirohi	5-25	0.5	0.79	(54)
Chegu	6-11	0.66	0.81	(25)
South Indian goats (5)	7-31	0.11-0.81	0.51-0.92	(55)
Bangladesh goats (5)	5.23-6.08	0.51-0.56	0.53-0.59	(56)
East Asian breeds				
East Asian goats (18)	5.8	0.31-0.71	0.30-0.72	(57)
Chinese goats (12)	5.24-7.77	0.60-0.78	0.61-0.78	(18)
Korean goats	3.4	0.36	0.38	(58)
Western Asian breeds				
Markhoz	8.1	-	0.80	(59)
Tali	7.4	-	0.74	(60)
Lori	7	-	0.78	(43)
Raeini	7.8	-	0.81	(60)
Taleshi	6.7	0.42	0.74	(61)
Native breeds (3)	7.3–11	-	0.74-0.8	(43)
African goats				
Namibia (4)	4.67-6	-	0.6-0.71	(62)
Burkina Faso (3)	4-33	0.02-0.86	0.02-0.93	(63)
Kalahari Red	7.77	-	0.63	(64)
South Africa (3)	9-10	0.49-0.69	0.46-0.67	(65)
Sub-Saharan Africa (19)	3.82-5.91	0.44-0.56	0.45-0.54	(66)
Egyptian goats (3)	5.3-7.6	0.61-0.66	0.67-0.79	(67)
West African local (9)	11.7	0.60-0.73	-	(66)

African and Asian researchers have concentrated on genetic diversity studies using microsatellites. From these studies, we can conclude that goat genetic diversity studies using microsatellites markers have been extensively conducted on the African and Asian continents. These studies have paved the way for future genetic and conservation studies. As an overall observation, the admixtures of local and foreign breeds have shown greater genetic diversity than single breed structures (16).

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### 3. Conclusion

At present, DNA-based techniques seem to be the appropriate tool for the verification of the origin of animal breeds. In conclusion, microsatellite markers are a useful and trusted tool for identification of goat breeds and their usage could be the solution to conservation with high confidence. However, to be really applicable, more cooperation among researchers and people involved in conservation is necessary.

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