

GENETIC DIVERSITY OF THE CAPORE GOAT IN ALBANIA BASED ON 30 MICROSATELLITE MARKERS

Anila Hoda

*Department of Animal Production, Faculty of Agriculture and Environment,
Agricultural University of Tirana, Albania*
hodanila@yahoo.com

The Capore goat, is an important local goat breed in the South East of Albania especially for milk production. For the first time, we report the genetic diversity of this breed, by the use of 30 microsatellite markers. Heterozygosities and the gene diversity were estimated. A total of 233 alleles were distinguished for 30 microsatellite markers. Twenty nine of 30 markers showed more than 4 alleles. The average observed and expected heterozygosity values were 0.667 and 0.732 respectively. The mean polymorphism information content (PIC) value was 0.7, reflecting a high level of polymorphism across the loci. Within the population inbreeding estimate ($F_{IS}=10.6$), showed a moderate level of inbreeding. This can be explained with the small population size, with the limited geographical location of the breed and with the small number of the breeding males. The data suggest that the Capore breed has not encountered a recent genetic bottleneck. The high level of genetic variability indicates that this breed is a reservoir of genetic diversity that has to be conserved.

Key words: genetic variability; local goat breed; microsatellite markers; inbreeding; genetic bottleneck

ГЕНЕТСКА РАЗНОВИДНОСТ НА КОЗИТЕ КАПОРЕ ВО АЛБАНИЈА БАЗИРАНА НА 30 МИКРОСАТЕЛИТСКИ МАРКЕРИ

Козата капоре е важна локална раса кози во југоисточниот дел на Албанија, особено за производството на млеко. За прв пат ја претставуваме генетската разновидност на оваа раса, употребувајќи 30 микросателитски маркери. Беа проценувани хетерозиготите и различноста на гените. Беа одделени 233 аели за 30 микросателитски маркери. Дваесет и девет од 30 маркери покажаа повеќе од 4 аели. Просечните набљудувани и очекувани вредности на хетерозиготност беа 0,667 и 0,732, соодветно. Средната вредност на содржина на информациски полиморфизам (СИП) беше 0,7, одразувајќи високо ниво на полиморфизам на локусите. Во рамките на процената на инбридингот во популацијата ($F_{IS} = 10,6$) е утврдено дека неговото ниво е променето. Ова може да биде објаснето со мала големина на популацијата и ограничена географска локација на расата, како и со мал број приплодни машки грла. Податоците укажуваат дека козата капоре не била пресретната од генетска препрека. Високото ниво на генетска варијабилност покажува дека оваа раса е извор на генетска разновидност која мора да се зачува.

Клучни зборови: генетска варијабилност; локална раса кози; микросателитски маркери; инбридинг; генетска препрека

1. INTRODUCTION

The domestic goat is one of the most important livestock species in the mountainous area of Albania. Goat breeds are defined mainly by the

geographic position, morphological characteristics and production performance. In this study we intend to determine the levels of genetic variation of an important goat breed, namely Capore. The animals of this breed are well adapted to extensive

management conditions. There is a high risk for the extinction of native goat breeds, because there is a lack of herd book, breeding programmes are absent and native breeds are displacing with other breeds according to the preferences of the farmers. Therefore the genetic characterization of a breed is very important for the evaluation of genetic variability.

Polymorphic DNA markers are very useful in assessment of genetic diversity within and between breeds. Microsatellites are widely used as genetic markers for the analysis of genetic variability within and between breeds due to their high number, distribution throughout the genome and the efficacy of genotyping.

Microsatellite markers are used for the study of genetic diversity among livestock breeds. There are several studies on genetic diversity of goats, based on microsatellite markers, such as Swiss breeds (Saitbekova et al. 1999), Chinese indigenous populations (Li et al., 2002), goats from Europe (including also the breeds represented here) and Middle East (Canon et al., 2006), Mehsana goat (Aggarwal et al., 2007), Indian domestic goats (Rout et al., 2008), Barbari goats (Ramamoorthi et al., 2009). In this study we are using 30 microsatellite markers in order to characterize the genetic diversity of the Capore goat breed.

2. MATERIALS AND METHODS

Sample collection and microsatellite markers

A total of 31 randomly sampled animals of an Albania goat breed was analyzed. The individuals were selected unrelated individuals (two females and one male) per flock, based on the information provided by the farmer. Sampling was carried out from an average of 11 flocks. The breeds are marginally farmed and autochthones.

30 microsatellite markers were used: CSRD247, DRBP1, ILSTS011, ILSTS087, INRA023, INRA063, InraBern172, MAF65, McM527, OarAE54, OarFCB20, OarFCB48, SPS113, SRCRSP09, SRCRSP23, SRCRSP3, MAF70, SRCRSP5, ILSTS005, ETH10, TGLA53, SRCRSP8, BM6444, P19, MAF209, SRCRSP7, ILSTS029, SRCRSP15, TCRVB6, INRABERN185.

Observed and expected heterozygosity estimates, observed number of alleles, effective number of alleles, and departures from Hardy-Weinberg equilibrium were computed using the POP-

GENE software (Yeah et al., 1999). The polymorphic information content (PIC) index for each marker was calculated according to Botstein et al. (1980).

3. RESULTS AND DISCUSSION

The allelic and genotypic frequencies of 30 microsatellite markers were determined in the Capore goat breed of Albania. The mean number of alleles per locus and the observed and expected heterozygosities for all loci are presented in Table 1. A total of 233 alleles was detected across 30 microsatellite loci and the number of alleles per locus ranged from 3 (MAF 209) to 17 (BM6444). All the markers had more than 4 alleles, except MAF209. The effective number of alleles ranged from 1.89 (MAF209) to 11.174 (BM6444). Since all the markers had more than 4 alleles, they were appropriate to analyze diversity in the Capore breed.

The observed heterozygosity ranged from 0.323 (Inrabe) to 0.903 (MAF 65). The expected heterozygosity ranged from 0.471 (MAF 209) to 0.911 (BM6444), with an average of 0.732. A marker is considered to be useful for measuring genetic variation and should have an average heterozygosity ranging from 0.3 to 0.8 in the population (Takezaki and Nei, 1996). This again confirms that the markers used in this study were appropriate for measuring genetic variation.

The polymorphism information content (PIC) values for all 30 microsatellite loci ranged from 0.424 (MAF 209) to 0.904 (BM6444). PIC is a parameter indicative of the degree of informativeness of a marker. Except MAF209 and Inrabe, all the other markers had PIC values higher than 0.5, therefore these markers appeared to be highly informative (Botstein et al., 1980).

Eight loci showed significant heterozygote deficiency (Table 1). Within population inbreeding estimate (F_{IS}) for the investigated loci was rather high, 0.103. The estimates for each locus are presented in Table 1. The values ranged from -0.182 (SRCRSP) to 0.586 (BM6444). Eleven loci revealed negative F_{IS} values. The heterozygote deficiency may be a result of inbreeding. This breed is located in a small geographical area. It is characterized by a small population size, of 8800 individuals, and an insufficient number of breeding males (a total of 400 males). Another possible reason may be the Wahlund effect, since the sampling was carried out in an 11 goat flock.

Table 1

Measures of genetic variability in the Capore goat

Locus	N	Na	Ne	I	Ho	He	F	PIC	F _{IS}	P-value
CSRD24	31	7.000	2.843	1.381	0.548	0.648	0.154	0.616	0.17	0.005**
DRBP1	31	9.000	3.230	1.602	0.484	0.690	0.299	0.666	0.314	0.0181*
ILSTS0	31	6.000	3.540	1.448	0.710	0.717	0.011	0.673	0.027	0.6151
ILSTS0	30	9.000	5.325	1.863	0.733	0.812	0.097	0.801	0.114	0.0446*
INRA02	31	9.000	4.358	1.760	0.710	0.771	0.079	0.746	0.095	0.2306
INRA06	31	4.000	2.658	1.106	0.645	0.624	-0.034	0.549	-0.018	0.6779
InraBe	31	7.000	6.160	1.870	0.871	0.838	-0.040	0.817	-0.023	0.6415
MAF65	31	13.000	9.196	2.340	0.903	0.891	-0.013	0.881	0.003	0.4428
McM527	31	7.000	4.388	1.663	0.742	0.772	0.039	0.742	0.055	0.5657
OarAE5	31	9.000	5.824	1.914	0.871	0.828	-0.052	0.806	-0.035	0.8015
OarFCB	31	6.000	3.829	1.471	0.806	0.739	-0.092	0.696	-0.075	0.7891
OarFCB	31	8.000	5.539	1.844	0.774	0.819	0.055	0.796	0.072	0.0616
SPS113	31	11.000	6.493	2.071	0.871	0.846	-0.030	0.828	-0.013	0.645
SRCRSP	30	10.000	3.010	1.599	0.800	0.668	-0.198	0.671	-0.182	0.9964
SRCRSP	31	14.000	7.813	2.280	0.839	0.872	0.038	0.86	0.055	0.2781
SRCRSP	31	5.000	2.394	1.178	0.516	0.582	0.113	0.549	0.13	0.2331
MAF70	31	6.000	4.470	1.602	0.516	0.776	0.335	0.741	0.35	0.0046**
SRCRSP	31	9.000	4.398	1.768	0.774	0.773	-0.002	0.748	0.014	0.2968
ILSTS0	31	5.000	2.412	1.112	0.613	0.585	-0.047	0.532	-0.031	0.7649
ETH10	31	4.000	3.166	1.221	0.710	0.684	-0.037	0.622	-0.021	0.6182
TGLA53	31	7.000	3.710	1.478	0.645	0.730	0.117	0.684	0.133	0.2043
SRCRSP	31	10.000	6.407	2.041	0.871	0.844	-0.032	0.827	-0.016	0.5158
BM6444	31	17.000	11.174	2.581	0.387	0.911	0.575	0.904	0.586	NA
P19	31	7.000	5.339	1.786	0.452	0.813	0.444	0.787	0.457	NA
MAF209	31	3.000	1.890	0.825	0.516	0.471	-0.096	0.424	-0.08	0.7548
SRCRSP	31	5.000	2.474	1.101	0.387	0.596	0.350	0.526	0.365	0.0025**
ILSTS0	31	5.000	3.105	1.286	0.484	0.678	0.286	0.62	0.301	NA
SRCRSP	31	6.000	3.269	1.359	0.710	0.694	-0.022	0.644	-0.006	0.4994
TCRVB6	31	9.000	5.414	1.896	0.806	0.815	0.011	0.793	0.027	0.3838
INRABE	31	6.000	1.901	1.025	0.323	0.474	0.319	0.451	0.334	0.0772
Mean		7.767	4.524	1.616	0.667	0.732	0.088	0.700	0.103	

The sign test revealed differences between the observed and expected number of loci with heterozygosity excess under IAM. Twenty six of 30 loci had heterozygosity excess and only four loci had significant heterozygosity deficiency ($p \leq 0.01$). The dataset showed mutation-drift equilibrium under TPM and SMM. The standardized difference test revealed significant heterozygosity

excess under IAM, mutation drift equilibrium under TPM and significant heterozygote deficiency under SMM ($T_2 = -3.718$, $p \leq 0.01$). The Wilcoxon test revealed that the population had undergone recent bottleneck assuming IAM and TPM. The three tests revealed significant deficit of heterozygotes under SMM and a recent bottleneck under IAM and TPM.

Table 2

Test for mutation drift equilibrium at 30 microsatellite markers, under three mutation models in the Capore goat

Test	IAM		TPM		SMM	
	Expected	Observed	Expected	Observed	Expected	Observed
Sign test (Number of loci with heterozygosity excess)	17.78	26**	17.83	21	17.74	14
Standardized differences test (T_2 values)	3.878**		1.216		-3.718**	
Wiloxon test (Probability of heterozygosity excess)	0.00002**		0.01636*		0.959	

* $p \leq 0.05$; ** $p \leq 0.01$; IAM – Infinite allele model; TPM – Two phase mutation model; SMM – Strict one step mutation model

Also a mode shift test is carried out. The allele frequency spectrum is shown in Figure 1. The microsatellite alleles were organized into 10 frequency classes. The distribution followed the normal L-shaped form. The alleles with low frequencies (0.01–0.1) are the most numerous. The observed distribution suggests that the breed did not encounter a recent genetic bottleneck.

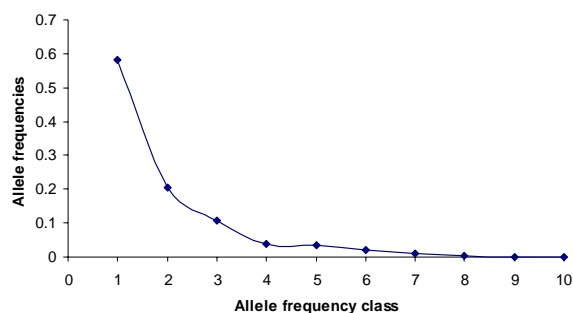


Fig. 1: L-shaped mode shift graph

The results of this study suggested that all the markers were highly polymorphic and useful for the molecular characterization of the Capore breed. Nevertheless of the small population size, the Capore breed displayed a high level of variability. That means that this breed is a reservoir of genetic diversity that is to be conserved. There was a moderate level of inbreeding. Efforts have to be made in order to avoid inbreeding in the population.

4. CONCLUSIONS

- Genetic diversity of the Capore goat breed is reported for the first time, using 30 microsatellite markers.
- A total of 233 alleles was distinguished for 30 microsatellite markers.
- Markers were highly polymorphic.

- There were high levels of observed and expected heterozygosity values.
- There was a moderate level of inbreeding.
- The Capore breed has not encountered a recent genetic bottleneck.
- The Capore breed is a reservoir of genetic diversity that has to be conserved.

Acknowledgements: This work has been supported by the ECONOGENE project, funded by the European Union (project QLK5-CT2001-02461).

REFERENCES

- Aggarwal R. A. K., S. P. Dixit, N. K. Verma, S. P. S Ahlawat, Y. Kumar, S. Kumar, R. Chander, K. P. Singh, Population genetic analysis of Mehsana goat based on microsatellite markers, *Current science*, **92** (8), 1133–1137 (2007).
- Botstein D., R. L. White, M. Skolnick, R. W. Davis, Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *A. J. Hum. Genet.*, **32**, 314–331 (1980).
- Li S-L, Valenti A., Genetic diversity of Chinese indigenous goat breeds based on microsatellite markers. *Journal of Animal Breeding and Genetics*, **121**, 350–55 (2004).
- Ramamoorthi J., Thilagam K., Sivaselvam S. N., Karthickeyan A. M. K., Genetic characterization of Barbari goats using microsatellite markers, *Journal of Veterinary Science*, **10** (1), 72–76 (2009).
- Rout P. K., Joshi M. B., Mandal A., Laloe D, Singh L., Thangaraj K., Microsatellite based phylogeny of Indian domestic goats. *BMC Genetics*, **9**, 11 (2008).
- Saitbekova N., Gaillard C., Obexer-Ruff G., Dolf G., Genetic diversity in Swiss goat breeds based on microsatellite analysis, *Animal Genetics*, **30**, 36–41 (1999).
- Takezaki N., Nei M., Genetic distances and reconstruction of phylogenetic trees from microsatellite DNA. *Genetics*, **144**, 389–3999 (1996).
- Yeh Francis C., Yang R.-C., Boyle Timothy B. J., Ye -H., Mao Judy X., Popgene version 1.32. Molecular Biology and Biotechnology Centre, University of Alberta Canada (<http://www.ualberta.ca/~fyeh/>) (1999).