Genetic diversity of the Northern Morocco goat population assessed with microsatellite markers

Najat El Moutchou1,2, Ana M. González-Martínez2, Mouad Chentouf2, Khalid Lairini1 and Evangelina Rodero2

1University Abdelmalek Essaâdi, Fac. Science and Technology, Km 10 Zieten, BP 416, Tangier, Morocco 2University of Cordoba, Department of Animal Production, Campus of Rabanales, c/eA3. 14071, Córdoba, Spain. 1National Institute of Agricultural Research, Regional Centre of Tangier, 78 Avenue Sidi Mohamed Ben Abdellah, 90010 Tangier, Morocco.

Abstract

The main goal of this work was to study the genetic diversity of the Northern Morocco goat population through the analysis of 19 microsatellites in 144 animals from 61 herds. To detect a possible population structure, three distinct geographic subpopulations were characterized as a function of climate and environmental influences. Most of the markers were highly polymorphic, and the results revealed considerable genetic variation across the studied loci. A total of 204 alleles were detected, with an average number of 10.7 per locus. The PIC average was 0.728, and four microsatellites showed a significant deviation ($p < 0.05$) from Hardy-Weinberg Equilibrium. Analysis of molecular variance (AMOVA) indicated that only 0.5% of the variation corresponded to differences among subpopulations, and 99.5% corresponded to differences among individuals. Factorial correspondence analysis showed intense admixtures across the putative subpopulations, and the subdivision related to geographical or environmental adaptation was undetectable. The Northern Morocco goat population presented high genetic diversity and a lack of population structure. The main reason for these findings is the absence of the breed concept (reproductively closed population), resulting in uncontrolled crossbreeding with exotic breeds and other local goats.

Additional keywords: animal genetic resources; local population; sustainable development.

Abbreviations used: AFC (Factorial correspondence analysis); AMOVA (Analysis of Molecular Variance); An (Number of alleles per locus); FIS (Inbreeding coefficient of an individual relative to the subpopulation); FIT (Inbreeding coefficient of an individual relative to the total population); FST (Effect of subpopulations compared to the total population); He (Expected heterozygosity); Ho (Observed heterozygosity); PIC (Polymorphic Information Content).

Authors’ contributions: Conceived, designed, performed the experiments, analyzed the data and wrote the paper: NEM, AMGM, and ER. Contributed reagents/materials/analysis tools: ER and MC. All authors read and approved the final manuscript.

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Correspondence should be addressed to Najat El Moutchou: najatemoutchou@gmail.com

Introduction

Molecular genetic characterization is the first approach for the sustainable use of animal genetic resources. In the absence of genetic information, development of local populations is often ignored in favour of the introduction of exotic breeds. Therefore, characterization of breeds is essential at both the level of animal phenotypes and genetic variability (FAO, 2007, 2011, 2013). Microsatellite marker analysis is very indicative of the historical progression of breeds by showing the evolution and differentiation of animal populations; this technique has been used in genetic diversity studies of European, African and American goats (Ajmone-Marsan et al., 2014).

In Morocco, three major goat populations have been identified according to their geographic location or production system: the Black population (with three subpopulations: Atlas, Barcha, and Ghazalia), the Draa population and the Northern population (Ibnelbachyr et al., 2015). The genetic characterization of Moroccan goats using microsatellites has been limited to studies of the Draa (Tadlaoui Ouafi et al., 2002), Black Rahalli breeds (Ouragh et al., 2012) and the Hamra population (Hilal et al., 2016). Previous studies reporting the genetic structure and diversity of Moroccan goat
populations have been achieved using mitochondrial DNA markers (Benjelloun et al., 2011) and WGS data (Benjelloun et al., 2015).

In Northern Morocco, the goat herding sector plays a vital role in the socioeconomic development of the region, providing more than 70% of the revenue in rural communities (Chentouf et al., 2011) and includes approximately 788,000 goats; these goats represent 13% of the total census in Morocco and 43% of the total small ruminants in the region (Chentouf, 2014). In a previous study, morphological differentiation was conducted in this goat population based on measurable and qualitative morphological traits, considering that this population was likely subdivided into three subpopulations according to geographic location, origin and breed influences (El Moutchou et al., 2014, 2017). The results showed high phenotypic variability, very heterogeneous subpopulations and undetectable differences among the groups. In the literature, the goats of Northern Morocco are related to Spanish and French breeds such as Murciana, Malagueña and Serrana Andaluza (Benlekhal & Tazi, 1996; Analla & Serradilla, 1997). However, management practice improvement and breeding plans in this unstudied goat population are very challenging and difficult to achieve due to a lack of genetic information.

The aim of the present research was to study the genetic diversity of the Northern Morocco goat population using microsatellite markers. To detect a possible population structure, three distinct geographic subpopulations were characterized as a function of climate and environmental influences because the genetic diversity among breeds or populations has been shown to be related to geographical location (Iamartino et al., 2005). This work was developed as part of a comprehensive international project oriented towards cataloguing and conserving animal genetic resources as a basis for rural development in Morocco according to FAO directives (El Moutchou, 2016). The results obtained from this characterization provided useful information that may contribute to the improvement of management practices and productivity in this population.

Material and methods

DNA sampling

Blood samples were collected from 144 unrelated goats in the north and northwest of Morocco (Fig. 1). Animals were selected according to the following criteria. First, individuals with unique phenotypes that clearly resembled well-known breeds were excluded. Second, based on the geographic distribution of the goats, the sampled territory was divided into three main geographical zones according to climate conditions and environmental influences: Dual (42 goats from 15 herds), Mediterranean (55 goats from 22 herds) and Atlantic (47 goats from 24 herds). To make the geographical sampling more representative and to avoid possible herd bias, the terrain was divided into cells; in each cell, 1 to 4 herds were selected. From each herd, 1 to 3 animals were sampled. Blood samples were collected by jugular venepuncture in Vacutainer EDTA-containing tubes. A commercial extraction kit (Qiagen® Mini Blood) was used to isolate DNA from whole blood according to the manufacturer's protocol.

Microsatellite amplification and analysis

A panel of nineteen microsatellites recommended by ISAG/FAO (FAO, 2011) for the genetic analysis of goats was used, including BM1258, BM1329, CSRD247, ETH10, FCB20, HSC, ILSTS11, ILSTS19, ILSTS030, ILSTS87, INRA005, INRA006, INRA023, INRA063, INRA172, MAF65, SRCRSP5, SRCRSP8, and TGLA53 (Table 1). Genotypes for all 19 microsatellites were determined via four multiplex fluorescent PCR reactions, and fragment lengths were determined in a single semi-automated multiplex electrophoresis run in an ABI Prism® 3130xl Genetic Analyzer using Gene Mapper ™ software from Applied Biosystems. Each reaction was performed in a total volume of 20 μL containing 50 ng of template DNA, 1× Qiagen Multiplex PCR Master Mix, 1× PCR Master Mix, primer mix, and nuclease-free water. The PCR programme consisted of an initial denaturation at 95°C for 15 min, followed by 32 cycles of 45 s at 95°C, 1 min 50 s at 58°C, and 1
min 20 s at 72°C, with a final extension at 60°C for 30 min. The analysis of the amplified fragments was carried out by regression analysis with GeneScan v.3.7 software using standard size fragments and presented with GeneMapper 3.7 software.

### Statistical analysis

The Cervus 3.0 program (Kalinowski et al., 2007) was used to calculate the number of alleles (An), the allele frequencies, the polymorphic information content

| Table 1. Description of amplification conditions used in this study. |
|---------------------------------------------------------------|
| **Microsatellites** |
| **Primer sequence** (forward and reverse) | **Chromosome** | **Size range (bp)** | **Annealing temperature (°C)** |
| Multiplex 1 |
| BM1258 | GTATGTAATTTTCCCCACCTGC GAGTCAGACATGACTGAGCCTG | OAR 23 | 110-120 | 50 |
| BM1329 | TTTTTAGCAAGTCCAAAGTC AACACCGAGCTTCATCC | OAR 6 | 145-161 | 50 |
| Multiplex 2 |
| HSC | CTGCAAATGCAGGACAACAAGAGTC TGTCTCCTGTCTCGTAC | 271 - 304 | 55 |
| ILSTS19 | CTGCAGGTTCTGCATATGTGG CTTAGACAACAGAAGGGTGTGGG | 2q(2) | 144-158 | 55 |
| INRA005 | CAATCAGCATGAGTAAATAAT CTTTCAGCAATACCTACAAC | 12 | 118-126 | 55 |
| INRA063 | GACCAAAAGGATTGCGAAGAGC AAACGAGAAATGCTCGAGAG | CHI18 | 171-181 | 55 |
| SRCRSP5 | GAATCTCAACCTAGCTGACAACGTGAAAGCTAAAGACTGC | CHI21 | 158-180 | 55 |
| SRCRSP8 | TGCGTCTGGTTCTGATTCAC GCTCTCTTCTGACATGAGAAGTCGATGCTTAG | Unknown | 209-235 | 55 |
| INRA023 | GAGTAGAGCTACAAGATAAACCTTC TAACTAGGGGTGTAGATGAAC | BTA3 | 197-215 | 55 |
| Multiplex 3 |
| ETH10 | GTTCAGAGCTGGGCTGCATACCA CTTCCAGCCCCTCCTCCCT | CHI5 | 200-210 | 55 |
| ILSTS030 | CTGCAAGTTCTGCATAGTGG CTTAGACAACAGAAGGGTGTGGG | 2q(2) | 146-158 | 55 |
| INRA006 | AGGAATCTGATGCTACGCATAC | 3 | 109-123 | 55 |
| TGLA53 | GCTTTCAGAAATAGTTGGCATTC ATCTTCACATGATACATACGC | BTA16 | 126-160 | 55 |
| Multiplex 4 |
| CSRD247 | GGACTTTGCGCAGAATCTCAGAAT CACTGTGTTTGTATATATGCAG | OAR14 | 220-247 | 58 |
| FCB20 | AAGTTGGTTAAAGTCTACATACGTG GAAGAAACCCCCACATACACACTCAC | OAR 2 | 93-117 | 58 |
| ILSTS87 | AGCAGGACATGATGACTGAC CTTGCCCTTTTCTGAGAG | BTA6 | 137-155 | 58 |
| ILSTS11 | GCTGTCTACATGGAAGATGTC CTAIAATGCAGAGGCCCTTACC | BTA14 | 230-300 | 58 |
| INRA172 | CGGAGGTGCTGGAATTTGCGG TGGTCGCCCTAGGTGAGAC | BTA26 | 135-153 | 58 |
| MAF65 | AAAGGCCAGAAGTATGCAATTAGGAG CCACCTCCTGTGAAATACATG | OAR15 | 116-158 | 58 |
Results

Nineteen microsatellite markers were successfully amplified, and a total of 204 alleles were observed from the 144 samples analysed in the studied population (Table 2). The number of alleles per locus (An) ranged from 5 (ETH10, INRA005) to 23 (HSC), with a global average of 10.7 alleles per locus. Most of the markers were highly polymorphic: 8 of the 19 loci analysed exhibited more than 10 alleles. The PIC values ranged from 0.457 (ILSTS19) to 0.867 (HSC), and the average value was 0.728. Four microsatellites (ETH10, ILSTS11, INRA023, and MAF65) showed significant ($p < 0.05$) deviations from Hardy-Weinberg Equilibrium.

Genetic variability within the three geographical groups was relatively high and very similar, as indicated in Table 3. The mean values over all loci for allelic richness, allele number, private alleles, and observed and expected heterozygosity were 9.97, 0.717, and 0.694, respectively, for the Mediterranean group; 9.85, 0.712, and 0.685, respectively, for the Atlantic group; and 9.85, 0.712, and 0.685, respectively, for the Dual group. The first three components explained only 50% of the total variation. The analysis of molecular variance (AMOVA) (Table 4) indicated 50% of the genetic variability within subpopulations and 98% of the genetic variability between subpopulations.

Discussion

Genetic diversity findings concur with the high variation previously observed in phenotypic traits of the studied population (El Moutchou et al., 2014, 2017). The genetic variability estimated by the various studied parameters was very high. The An (10.7) and He (0.758) values obtained here were higher than those reported by other authors examining other Moroccan goats. Tadlaoui et al. (2002), with five microsatellite markers, obtained mean values for An (7.83 and 8.33) and He (0.673 and 0.670) for Black Rahali and Draa goats, respectively. Using 12 microsatellite markers and a highly polymorphic milk protein gene, Ouragh et al. (2012) studied three Moroccan goat populations and reported An mean values of 8.53, 8.23, and 7.92, and He values of 0.746, 0.783 and 0.726 for the Black, Draa, and Northern goat populations, respectively. In their work, Ouragh et al. used a small sample size located only in the Dual area of our study. Hilal et al. (2016) obtained an An value of 8.67 for Beni Arouss and a value of 8.07 for Rommani. Our results are in agreement with those obtained by Benjelloun et al. (2011) in the analysis of mitochondrial DNA. These authors attributed the high level of variability observed in Moroccan goats to the high heterogeneity of the founder population and the influence of Spanish breeds across the Strait of Gibraltar. Genetic differentiation (FST) among the populations was not detected (2%), and 98% of the genetic variability of the studied subpopulations was due to differences among individuals of the total population and not to differences among the geographical subpopulations. Our FST values are notably inferior to those reported by Ouragh et al. (2012) but are similar to those obtained with SNPs for other Moroccan goats (Black of the Atlas, Draa and Northern populations) by Benjelloun et al. (2015).

The Dual goats were different from the other goats and had greater variability and introgression from the Mediterranean than the Atlantic goats, probably because they are not affected by the north-south movements following the Rif valleys and the Atlas Mountains. Furthermore, due to the geographic
Table 2. Diversity parameter estimates in the goat population of Northern Morocco. Total number of alleles detected per locus (An), total number of private alleles (PA), index of polymorphic information content (PIC), observed heterozygosity (Ho), expected heterozygosity (He), significance of Hardy-Weinberg Equilibrium test (HWE) and F-statistics (FIS, FST, FIT) according to Weir and Cockerham (1984) values for 19 microsatellites markers analysed in the goat population of Northern Morocco (N=144).

| Locus     | An | PA | PIC   | Ho   | He   | HWE  | FIT  | FST  | FIS  |
|-----------|----|----|-------|------|------|------|------|------|------|
| BM1258    | 12 | 1  | 0.820 | 0.847| 0.841| n.s  | -0.006| 0.004| -0.010|
| BM1329    | 10 | 1  | 0.794 | 0.785| 0.820| n.s  | 0.042 | -0.003| 0.045|
| CSRD247   | 10 | 2  | 0.838 | 0.875| 0.858| n.s  | -0.018| 0.006| -0.025|
| ETH10     | 5  | 0  | 0.601 | 0.618| 0.665| *    | 0.079 | 0.028| 0.053|
| FCB20     | 9  | 2  | 0.736 | 0.757| 0.767| n.s  | 0.012 | -0.004| 0.016|
| HSC       | 23 | 7  | 0.876 | 0.875| 0.888| n.s  | 0.017 | 0.005| 0.012|
| ILSTS11   | 8  | 0  | 0.680 | 0.688| 0.709| *    | 0.031 | 0.002| 0.029|
| ILSTS19   | 7  | 1  | 0.457 | 0.479| 0.482| n.s  | 0.007 | 0.002| 0.004|
| ILSTS30   | 17 | 4  | 0.836 | 0.889| 0.855| n.s  | -0.041| -0.002| -0.038|
| ILSTS87   | 10 | 1  | 0.572 | 0.604| 0.598| n.s  | -0.010| 0.002| -0.012|
| INRA005   | 5  | 1  | 0.585 | 0.604| 0.633| n.s  | 0.047 | 0.004| 0.044|
| INRA006   | 13 | 4  | 0.849 | 0.819| 0.867| n.s  | 0.055 | -0.001| 0.055|
| INRA023   | 12 | 1  | 0.800 | 0.792| 0.823| *    | 0.037 | -0.002| 0.040|
| INRA063   | 6  | 1  | 0.599 | 0.618| 0.664| n.s  | 0.068 | -0.006| 0.073|
| INRA172   | 9  | 2  | 0.756 | 0.750| 0.788| n.s  | 0.057 | 0.025| 0.032|
| MAF65     | 14 | 2  | 0.817 | 0.792| 0.838| *    | 0.058 | 0.007| 0.051|
| SRCRSP5   | 10 | 3  | 0.755 | 0.813| 0.787| n.s  | -0.029| 0.011| -0.040|
| SRCRSP8   | 12 | 2  | 0.767 | 0.771| 0.799| n.s  | 0.034 | -0.002| 0.036|
| TGLA53    | 12 | 3  | 0.698 | 0.708| 0.725| n.s  | 0.029 | 0.018| 0.011|
| Mean      | 10.7|   | 0.728 | 0.741| 0.758|      | 0.024 | 0.005| 0.019|
| Total     | 204| 38 |      |      |      |      |      |      |      |

**a*p≤0.05, **p≤0.01, ***p≤0.001, n.s>0.05; bJackknifing estimate over the loci.

Table 3. Mean values over all loci for diversity parameter estimates in Northern Morocco goats. Total number of alleles (An), number of private alleles (PA), allelic richness (AR), polymorphic information content (PIC), observed (Ho) and expected (He) heterozygosity.

| Source of variation | N  | An  | PA  | AR  | PIC | Ho  | He  |
|---------------------|----|-----|-----|-----|-----|-----|-----|
| Dual                | 42 | 8.842| 12 | 8.842| 0.729| 0.773| 0.767|
| Mediterranean       | 55 | 8.789| 15 | 8.456| 0.718| 0.737| 0.753|
| Atlantic            | 47 | 9.053| 11 | 8.893| 0.710| 0.718| 0.748|
| Population total    | 144| 10.7| 38 | 8.043| 0.728| 0.741| 0.758|

Table 4. Partitioning of genetic variation by the fixation indices (FST, FIS, FIT) and the analysis of molecular variance (AMOVA) based on 19 microsatellite loci for the Northern Morocco goat populations.

| Source of variation | Degrees of freedom | Variance components | Percentage of variation | Fixation indices | p value |
|---------------------|--------------------|---------------------|------------------------|-----------------|---------|
| Among subpopulations| 2                  | 0.03502             | 0.49                   | FST = 0.005      | 0.650   |
| Among individuals within subpopulations | 141 | 0.13904 | 1.93 | FIS = 0.019 | 0.042   |
| Among individuals within total population | 144 | 7.04167 | 97.59 | FIT = 0.024 | 0.020   |
| Total               | 287                | 7.21573             |                        |                 |         |
proximity, it is more probable that the influence of local Black goat breeds occurs more on goats from the Mediterranean and Atlantic areas than on Dual goats. The Draa breed in southern Morocco could have influenced the Atlantic goats more than the goats in the other two areas. Additional studies are necessary for a better understanding of Moroccan genetic resources.

The AFC analysis showed a lack of population structure in the 144 studied goats, and the high admixture in the three groups confirms their high similarity, as indicated by genetic parameters. This could be a result of high goat mobility across different regions, providing the opportunity for introgression and resulting in reduced differentiation (Luikart et al., 2001; Naderi et al., 2007).

These findings are very similar to those examining other Moroccan goat populations, i.e., Black, Draa and Northern (Benjelloun et al., 2015), for which three principal components explained only 5.8% of the variance among the three populations. Hilal et al. (2016) reported weak differentiation in the Hamra goat population in two different locations of Morocco. In general, this result could be due to the absence of a breed concept in North Africa (reproductively closed population), resulting in uncontrolled crossbreeding with exotic breeds and other local goats.

In summary, the Northern Morocco goat population presented high genetic diversity and an absence of population structure. This population may be considered valuable in order to meet current production needs while preserving its purity and avoiding uncontrolled crossbreeding. These findings reinforce the need for improved management practices and implemented breeding plans based on genetic data, to avoid inbreeding and preserve genetic and allelic diversity. We recommend adopting a goat sector approach and orienting the traditional production system towards labelling using local skills, which would allow these genetic resources to be integrated into the economic and social development of Morocco.

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References

Ajmone-Marsan P, Colli L, Han JL, Achilli A, Lancioni H, Joost S, Crepaldi P, Pilla F, Stella A, Taberlet P, et al., 2014. The characterization of goat genetic
Genetic diversity of the Northern Morocco goat population

FAO, 2007. Plan d'action mondial pour les ressources zoogénétiques et la déclaration d'Interlaken. FAO, 36 pp. http://www.fao.org/3/a-i3405e.pdf

FAO, 2011. Molecular genetic characterization of animal genetic resources. FAO Anim Prod Health Guidelines 9. http://www.fao.org/docrep/014/i2413e/i2413e000.htm

FAO, 2013. In vivo conservation of animal genetic resources. FAO Anim Prod Health, Guidelines 14. http://www.fao.org/3/a-i3327e.pdf

Goutet J, 1995. FSTAT (Version 1.2): A Computer Program to Calculate F-Statistics. J Hered 86: 485-486. https://doi.org/10.1093/oxfordjournals.jhered.a111627

Hilal B, Boujenane I, El Otmani S, Chentouf M, Piro M, 2016. Genetic characterization of Hamra goat population in two different locations of Morocco using microsatellite markers. IR J App Ani Sci 4(6): 901-907.

Iamartino D, Bruzzone A, Lanza A, Blasi M, Pilla F, 2005. Genetic diversity of Southern Italian goat populations assessed by microsatellite markers. Small Rum Res 57: 249-255. https://doi.org/10.1016/j.smallrumres.2004.08.003

Ibnelbachy M, Boujenane I, Chikhi A, 2015. Morphometric differentiation of Moroccan indigenous Draa goat based on multivariate analysis. J Anim Genet Res 57 : 81-87. https://doi.org/10.1017/S207863615000296

Kalinowski ST, Taper ML, Marshall TC, 2007. Revising how the computer program CERVUS accommodates genotyping error increases success in paternity assignment. Mol Ecol 16: 1099-1106. https://doi.org/10.1111/j.1365-294X.2007.03089.x

Lebart L, Morineau A, Warwick K, 1984. Multivariate descriptive statistical analysis. John Wiley & Sons.

Luikart G, Gielly L, Excoffier L, Vigne JD, Bouvet J, Taberlet P, 2001. Multiple maternal origins and weak phylogeographic structure in domestic goats. Proc Natl Acad Sci USA 98: 5927-5932. https://doi.org/10.1073/pnas.091591198

Maderi S, Rezaei HR, Taberlet P, Zundel S, Rafat SA, Naghash HR, El-Barody MAA, Ertugrul O, Pompanon F, Econogene Consortium, 2007. Large-scale mitochondrial DNA analysis of the domestic goat reveals six haplogroups with high diversity. PLoS ONE 2: e1012. https://doi.org/10.1371/journal.pone.0001012

Nei M, 1978. Estimation of average heterozygosity and genetic distance from a small number of individuals. Genetics 99: 583-590.

Ouralgh L, Pantano T, El Fadili M, Fagouri S, Babillot M, Hossaini-Hilali J, 2012. Analyse génétique des populations caprines marocaines. ABARK: 107-120. http://www.agrimaroc.net/seminaire_caprin/ouragh_genetique_caprins.html

Raymond M, Rouset F, 1995. GENEPOP (Version 1.2): Population Genetics Software for Exact Tests
and Ecumenicism. J Hered 86: 248-249. https://doi.org/10.1093/oxfordjournals.jhered.a111573
Saitbekova N, Gaillard C, Obexer-Ruff G, Dolf G, 1999. Genetic diversity in Swiss goat breeds based on microsatellite analysis. Ani Genet 30: 36-41. https://doi.org/10.1046/j.1365-2052.1999.00429.x
Tadlaoui Ouafi A, Babilliot JM, Leroux C, Martin P, 2002. Genetic diversity of the two main Moroccan goat breeds: phylogenetic relationships with four breeds reared in France. Small Rum Res 45: 225-233. https://doi.org/10.1016/S0921-4488(02)00111-6

Weir BS, Cockerman C, 1984. Estimating F-statistics for the analysis of population structure. Evolution 38: 1358-1370. https://doi.org/10.1111/j.1558-5646.1984.tb05657.x
Whitlock MC, McCauley DE, 1999. Indirect measures of gene flow and migration: FST not equal to 1/(4Nm + 1). Hered 82: 117-125. https://doi.org/10.1038/sj.hdy.6884960
Wright S, 1965. The interpretation of population structure by F-statistics with special regard to systems of mating. Evolution 19: 395-420. https://doi.org/10.1111/j.1558-5646.1965.tb01731.x