Genetics & Biodiversity Journal

Journal homepage: https://journals.univ tlemcen.dz/GABJ/index.php/GABJ

ISSN: 2588-185X

OPEN ACCESS

Original Research Paper

Identification of selection signatures in Algero-Tunisian sheep breeds using medium-density SNP chips

Ghazi YAHYAOUI[1], Slim BEN-JEMAA[2*], Samia KDIDI[3], Semir B. S GAOUAR[4], Mohamed H. YAHYAOUI[3]

[1] Département de Biologie, Faculté des Sciences de Tunis, Université Tunis El Manar, 2092 Tunis, Tunisie.
[2] Laboratoire des Productions Animales et Fourragères, Institut National de la Recherche Agronomique de Tunisie, Université de Carthage, 2049, Ariana, Tunisia.
[3] Laboratoire d'Elevage et Faune Sauvage, Institut des Régions Arides, Medenine, Tunisia.
[4] Applied genetic in agriculture, ecology and public health laboratory, university of Tlemcen

***Corresponding Author**: Slim Ben-Jemaa, Laboratoire des Productions Animales et Fourragères, Institut National de la Recherche Agronomique de Tunisie, Université de Carthage, 2049, Ariana, Tunisia; **Email**: benjemaaslim@gmail.com

Article history: Received: 5 February 2024; Revised: 26 February 2024; Accepted: 27 February 2024

Abstract

North Africa has a wide range of great diversity of indigenous sheep breeds whose origin is linked to its environmental characteristics but also to certain historical events that took place in the region. Knowledge of population structure and identification of genomic regions that have been targets of selection are key steps for genetic resource conservation and breeding strategies designing. This work explores the population structure of two Tunisian and four Algerian sheep breeds and identifies genomic regions under selection in four of them. Leveraging high-throughput genomic data from Illumina Ovine SNP50K array from 59 animals belonging to the six breeds, we conducted a comprehensive analysis to unravel their population structure. Our results indicate a clear genomic admixture between the fat-tailed Tunisian and Algerian Barbarine with thin-tailed breeds raised in each of the two countries. Additionally, through the detection of genomic regions under selection and candidate gene identification, we provide insights into some putative genetic mechanisms that would have contributed to the adaptation and evolution of these breeds in response to local environmental pressures. We also highlight several genes related to economical traits. This study provides valuable information for the conservation and sustainable management of the studied Tunisian and Algerian sheep breeds and offers opportunities for future breeding and genomic improvement programs.

Keywords: North African sheep, Single Nucleotide Polymorphism, Selection signature, Extended haplotype homozygosity, genetic structure, Ovine SNP50K.

الملخص

تتمتع شمال أفريقيا بتنوع كبير في سلالات الأغنام الأصلية التي يرتبط أصلها بخصائصها البيئية وبعض الأحداث التاريخية التي وقعت في المنطقة، وتعد معرفة التركيبة السكانية وتحديد المناطق الجينومية التي تخضع للانتقاء، خطوات أساسية للحفاظ على هاته الموارد الوراثية وتصميم استراتيجيات تهجين تتماشى معها. تستكشف هذه الورقة العلمية التركيبة الجينية لسلالتين من الأغنام التونسية وأربعة جزائرية وتحدد المناطق الجينومية قيد الانتقاء في أربعة منها، وذلك بالاستفادة من البيانات الجينومية عالية الإنتاجية المستوردة من رقاقة "Illumina Ovine SNP50K" لـ59 حيوانًا ينتمون الى الستة سلالات المذكورة. بإجراء تحليل شامل للبيانات تمكنا من كشف التنوع الوراثي والتركيبة الجينية لهذه المجموعة، إذ كشفت النتائج التي توصلنا إليها عن اختلاط جيني بين البربري التونسي والجزائري ذو الليّة وسلالات أخرى ذات ذيول رقيقة. بالإضافة إلى ذلك، ومن خلال تحديد المناطق الجينومية قيد الانتقاء وكشف الجينات التي تحتويها، فإننا نقدم نظرة ثاقبة عن الآليات الوراثية التي ساهمت في تكيف وتطور هذه السلالات استجابتا للضغوط البيئية المحلية، كما نسلط الضوء على العديد من الجينات المرتبطة بصفات اقتصادية معينة لكل سلالة. تساهم هذه الدراسة بمعلومات قيّمة للحفاظ على سلالات الأغنام التونسية والجزائرية وإدارتها المستدامة وتوفر معلومات مرجعية أساسية لبرامج التربية والتحسين الجيني في المستقبل.

كلمات مفتاحية: أغنام شمال أفريقيا، تعدد أشكال النوكليوتيد المفرد، بصمات الانتقاء، التركيبة الجينية.

Introduction

Sheep farming is a national capital resource with great economic value in the Maghreb region. This part of North Africa is endowed with many diverse indigenous sheep breeds. In the Maghreb countries sheep population is composed of one or two major breeds and several other minor populations. Many of these

breeds are common to at least two countries. For instance, the Barbarine, the main sheep breed in Tunisia is also raised in Libya and Algeria. Also, the D'men, a highly prolific breed, is common to both Algeria and Morocco and, more recently, introduced in Tunisia. Several of these breeds thrive well in challenging local agro-climatic conditions characterized by excessive heat load, poor nutrition and disease-infested areas (Ben Salem et al., 2011; Djaout et al., 2017). This resulted in a wide variety of genetic adaptations to both environmental conditions and production systems. The genetic basis of adaptation is detectable by genome-wide scanning technology using SNP arrays using specific statistics based on contrasting haplotype homozygosity (EHH) either within a population between a pool of ancestral and derived haplotypes (Voight et al., 2006) or between populations (Sabeti et al., 2007). Various studies have been performed within or across sheep breeds in order to determine regions harboring adaptive genes under potential selection for resilience traits (e.g. Cao et al., 2021; Lv et al., 2014; Mastrangelo et al., 2019; Rochus et al., 2018). Most of sheep in Algeria and Tunisia are raised in extensive production systems where cross-breeding is the dominant mode of genetic improvement. Despite the fact that such a practice may ensure a faster impact on performance than long selection schemes, repeated cross-breeding will lead to the disappearance of local genetic diversity through genetic erosion of minority breeds. Yet, small breeds are the custodians of rare allelic variants of genes responsible for adaptation to specific environmental conditions such as higher feed conversion ratio of local feed resources of low nutrient density and parasite resistance. Therefore, minority breeds can be regarded as an important genetic reservoir of variants conferring adaptation to multiple environmental stressors triggered by climate change. Previous genetic studies highlighted the effects of the widespread of cross-breeding on the population structure of sheep breeds from Algeria (Gaouar et al., 2017, 2015) and Tunisia (Jemaa et al., 2019; Kdidi et al., 2015). All these studies highlighted a significant change in the genomic structure of minority breeds.

Belabdi et al., (2019), highlighted a strong homogenization of the Maghreb sheep stock affecting the major part of Morocco and most of Northern Algeria. Paradoxically, this might offer a unique opportunity to map loci underlying some morphologic traits between breeds with a similar genomic structure but differing in one or more morphologic features. Furthermore, the relatively recent crossbreeding of Maghreb sheep can provide an excellent proxy to investigate footprints of ongoing selection left by recent events of hybridization. Recent advancements in genomic technology, allow us to uncover candidate genetic regions through genome scans using appropriate statistical approaches. The aim of our study is to detect potential signatures of ongoing selection in Algerian and Tunisian local sheep populations. We also leveraged the genomic similarity between the Tunisian Barbarine fat-tailed breed and the thin-tailed Algerian Hamra and Ouled-Djellal populations to identify genomic regions under differential selection pressure between the two breeds.

Materials and methods

Breeds and genotyping data

Our work focused on two Tunisian sheep breeds (the Tunisian Barbarine (BART) and Black Thibar, (BT)) and four Algerian breeds (The Algerian Barbarine (BRBA), Ouled-Djellal (OLDA), Hamra (HAMA) and D'men (DMNA)). We used already available genotypes for 59 individuals from the six aforementioned breeds (Belabdi et al., 2019; Gaouar et al., 2017; Jemaa et al., 2019). Table 1 provides more details on the six populations. We used PLINK v.1.09 software (Purcell et al. 2007) to perform a quality control of the dataset i) SNPs with call rates <90% or minor allele frequency (MAF) <0.01 and SNPs in linkage disequilibrium as revealed by PLINK default parameters (SNP window size:50, step 5 SNPs, r2: 0.5) were removed; ii) individuals with call rates <90% of markers were also discarded from further analysis. After merging genotypes from various datasets and applying the aforementioned quality filters, a total of 39954 SNPs mapping to the 26 sheep autosomes on the Oar_v4.0 ovine (Ovis aries) genome sequence assembly were used in this study.

Population structure and genetic relationship analyses

Various methods were used to provide a fine scale assessment of the genetic structure of the studied populations. First, a Principal Component Analysis (PCA) implemented in the adegenet R package (Jombart, 2008) was performed to visualize the major axes of genetic variation. Then, we performed a Discriminant Analysis of Principal Components (DAPC) of the same package to identify an optimal

number of genetic clusters that best describe the data by using the "find.clusters" function implemented in adegenet. Following this analysis each individual was assigned to a genetic cluster without providing any a priori population assignment.

Table 1. The six sheep breeds analysed in this study.

Breed	Code	Origin	N	Tail	Reference
Tunisian Barbarine	BART	Tunisia	15	Fat	Ben Jemaa et al., 2019
Black Thibar	BT	Tunisia	16	Thin	Ben Jemaa et al., 2019
Algerian Barbarine	BRBA	Algeria	5	Fat	Belabdi et al., 2019
D'men	DMNA	Algeria	5	Thin	Gaouar et al., 2017
Hamra	HAMA	Algeria	12	Thin	Belabdi et al., 2019
Ouled-Djellal	OLDA	Algeria	6	Thin	Belabdi et al., 2019

N. number of individuals

Unsupervised model-based clustering was undertaken using ADMIXTURE 1.23 software (Alexander et al., 2009) with K values as the number of populations varying from 3 to 6, results were then graphically displayed using DISTRUCT (Rosenberg, 2003). Genetic differentiation between populations was estimated with Genepop 4.0 software (Rousset, 2008) using all available SNPs. Finally, we computed the genetic distance for each pair of individuals using the ape R package (Paradis et al., 2004). The distance between two individuals was defined as the number of loci for which they differ. A neighbor-joining tree was then computed based on the resulting distance matrix using the phyclust R package (Chen and Dorman, 2013).

Identification of selection signatures

To identify selection signatures, we investigated haplotype extended patterns within populations using the *iHS* test (Voight et al., 2006). For this test, information on the ancestral and derived allele state is needed for each SNP because it is based on the ratio of the extended haplotype homozygosity (EHH) associated with each allele. In our analysis, the ancestral allele was inferred as the most common allele within a panel of 34 worldwide sheep breeds. We also contrasted haplotype patterns between the Tunisian Barbarine (fat-tailed) and the two Algerian Hamra and Ouled-Djellal using the *Rsb* test (Tang et al., 2007). Haplotypes were reconstructed using fastPHASE 1.4 software (Scheet & Stephens, 2006). For each SNP, EHH tests are constructed to have an approximately standard Gaussian distribution (Tang et al., 2007; Voight et al., 2006). Both tests were computed within nonoverlapping 1-Mb sliding windows using the rehh package v. 3.2.2 in R (Gautier and Vitalis, 2012). The scores, calculated for each SNP, were transformed into p-values. A window is classified as putatively under selection when it contains at least 3 markers exceeding the significance threshold of $-\log10$ (p-value) = 3.

Gene identification and functional enrichment analysis

Genes in candidate genomic regions were identified using the Oar_v4.0 assembly annotation. Functional enrichment analysis was performed for the genes overlapping the candidate regions using the online tool DAVID (Database for Annotation, Visualization and Integrated Discovery https://david.ncifcrf.gov/), version 6.8. DAVID uses thousands of annotation terms in several annotation categories, such as Gene Ontology (GO), Biological Process, GO Molecular Function and InterPro Domains. An adjusted Benjamini-corrected p-value of 0.05 was used as the criterion for statistical significance of over-enrichment of genes in one of the categories.

Results

Population structure and phylogenetic analyses

In PCA analysis, the first two principal components (PC) 1 and 2 accounted for 2.73 % and 2.24 % of the total variation, respectively and showed a genetic proximity between the Algerian Ouled-Djellal

(OLDA), Hamra (HAMA) and the Tunisian Barbarine individuals (BART) (Figure 1a). The latter showed a low dispersion around their centre of gravity unlike the Algerian Barbarine (BRBA) whose individuals are more scattered reflecting a high genetic heterogeneity. Overall, the genetic diversity of the studied breeds is represented by a triangle with the Tunisian Black Thibar (BT) and the Algerian Barbarine (BRBA) and D'men (DMNA) forming the apexes. In order to have an unbiased interpretation of population structure, we performed a DAPC in which each individual was assigned to a cluster without providing any a priori breed assignment (Figure 1b). The analysis showed that the individuals of Black Thibar population were assigned mostly to one cluster (14 of 16, cluster 3). Similarly, all Tunisian Barbarine belong to a unique cluster (cluster 4) and are positioned closely to genetic cluster 2 composed of all OLDA and HAMA animals in addition to 2 BT, 1 BRBA and 1 DMNA individuals. Four of the 5 DMNA individuals were assigned to cluster 6. It's worth to note that clusters 3 (BT), 6 (DMNA), 1 and 5 (BRBA) are positioned far from clusters 2 and 4 mostly including BART, HAMA and OLDA. Pairwise Fst values (Table 2) revealed the absence of genetic differentiation between HAMA and both OLDA and the Tunisian Barbarine (Fst almost null). By contrast, BT, DMNA an BRBA showed higher pairwise Fst values which agrees with PCA and DAPC results.

Model based hierarchical clustering with varying K of predefined genetic clusters implemented in the ADMIXTURE software (Figure 1c), essentially confirmed the pattern of the structuring of genetic diversity revealed by PCA and DAPC where BT, DMNA and BRBA breeds separated as a distinct cluster. The genetic heterogeneity of BRBA revealed by both PCA and DAPC was also confirmed by ADMIXTURE analysis. By contrast, HAMA, OLDA and BART breeds showed a similar genetic structure through all K values.

Table 2. Pairwise Fst values between the studied sheep populations.

	BART	BRBA	BT	DMNA	HAMA	OLDA
BART						
BRBA	0.032					
BT	0.026	0.047				
DMNA	0.042	0.055	0.053			
HAMA	0.008	0.028	0.021	0.032		
OLDA	0.014	0.024	0.024	0.035	0.006	

The neighbor-joining tree constructed based on a distance matrix between pairs of individuals (Figure 2) showed BART and BT as distinct breeds. In contrast, no clear separation is observed between HAMA and OLDA which agrees with previous population structure analyses (PCA, DAPC and Fst). DMNA branched in an intermediate position between individuals from these two last breeds.

Selective sweeps detection

Because it has been shown that it is better to have populations with at least 15 diploid individuals to get reasonable power to detect signatures of ongoing selection (Ma et al., 2015), intra-population *iHS* test was performed only on populations composed of at least 15 animals. This led to the exclusion of DMNA and BRBA. It is worth noting that HAMA and OLDA were considered as a single population because of their similar genomic structure highlighted in all the previous analyses.

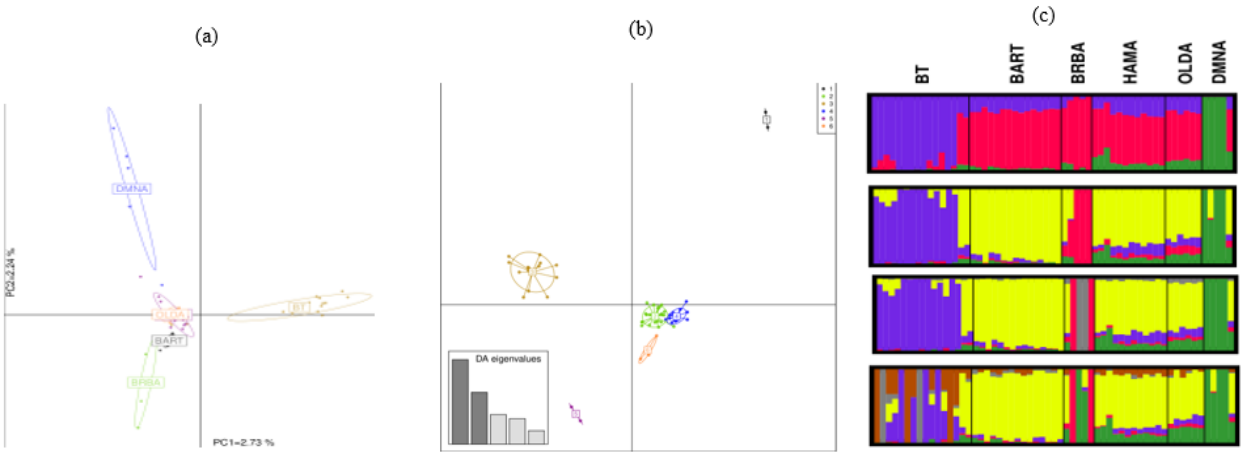

Figure 1. Population structure analyses. (a) PCA results. (b) DAPC results obtained when 5 genetic clusters are defined (optimal number of clusters). Genetic cluster 1 corresponds to 2 BRBA individuals. Cluster 2 includes Hamra (HAMA) and Ouled-Djellal (OLDA) in addition to 1 DMNA, 1 BRBA and 2 BT individuals. Cluster 3 consists of the remaining 14 BT individuals. Cluster 4 includes all BART animals. Clusters 5 and 6 consist of 2 BRBA and 4 DMNA individuals, respectively. (c) ADMIXTURE results (K=3 to 6).

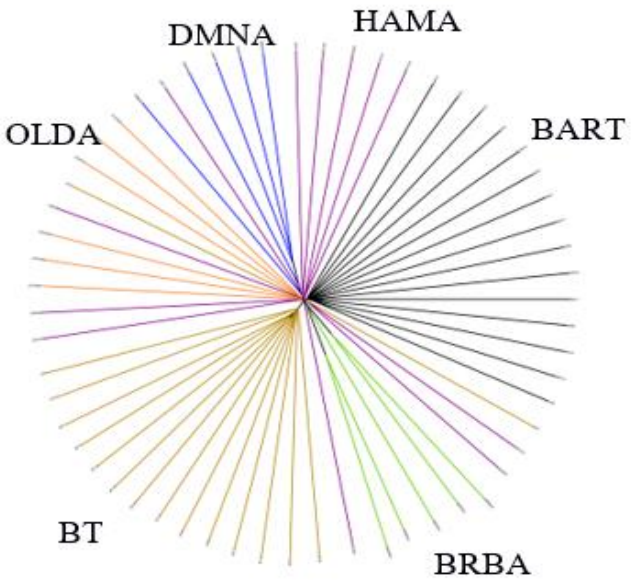

Figure 2. Neighbor-Joining tree relating the 36 Algerian and the 33 Tunisian sheep individuals

The *iHS* test revealed 3, 18 and 5 regions putatively under selection in BART, BT and the mixture (HAMA, OLDA), respectively (Table 3). The 2 Mb region on OAR13 detected within the Tunisian Barbarine showed the strongest selection signal with 11 SNPs (52% of the total SNPs in the window) exceeding the significance threshold (Figure 3a). For Black Thibar, a 9 Mb interval on OAR12 extending from 37 to 49 Megabases (Mb), is the strongest selection signal (52 SNPs exceeding the significance threshold; Figure 3b). For *iHS* test within HAMA and OLDA, the region on OAR10 (at position: 42-43 Mb) could be considered as the candidate region with the most consistent selection signal with 4 SNPs (33% of the total SNPs in the region) exceeding the significance threshold (Figure 3c). Contrasting BART and HAMA_OLDA haplotypes revealed 13 regions under differential selection between the two breeds (Figure 3d) most of them (10) were shown to be under selection in the HAMA_OLDA genome (Table 4). The highest scores were found in OAR04 where 4 SNPs passed the threshold of −log10(p-value) equal to five within a region of 268 Kb long

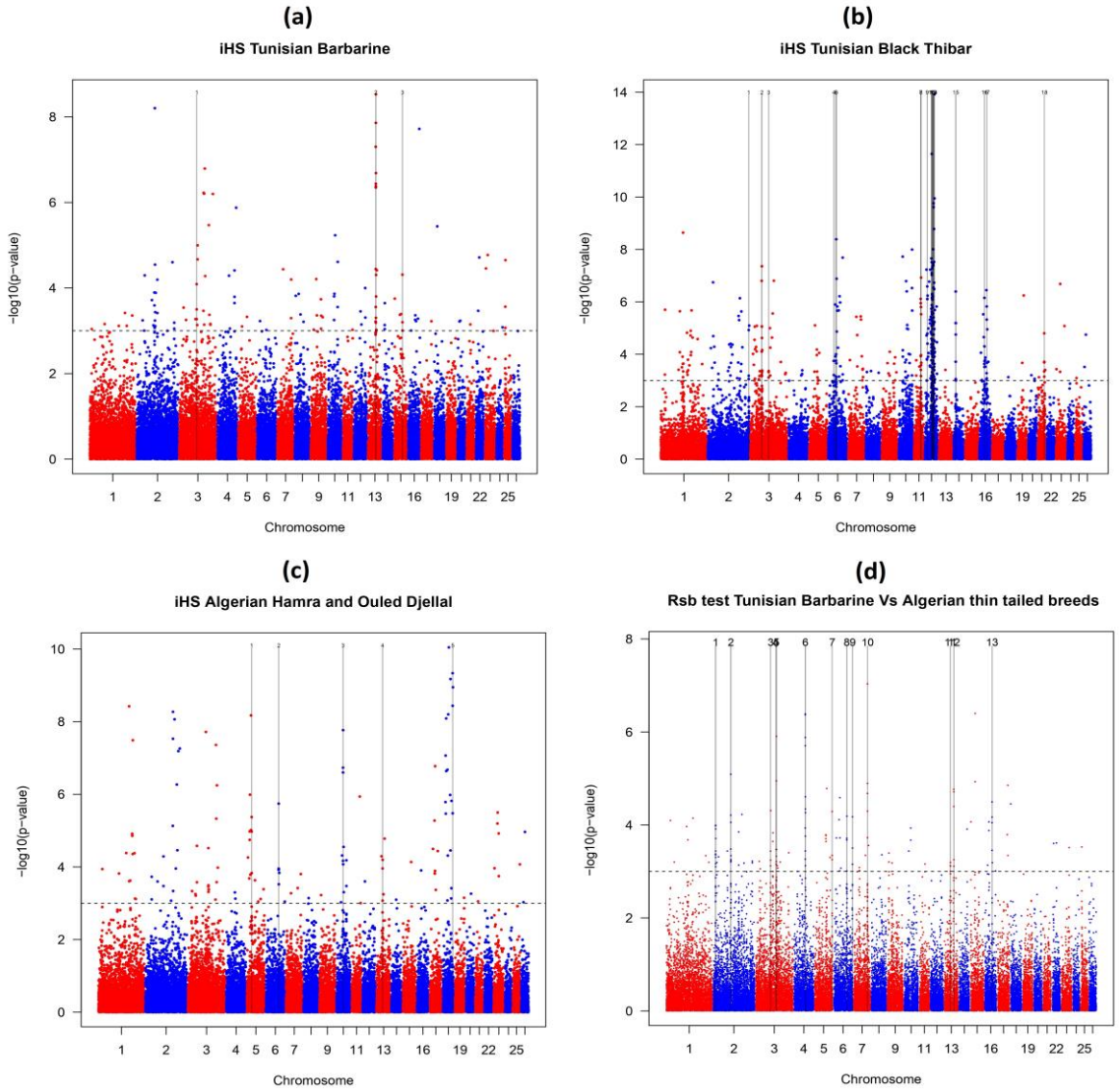

Figure3. Manhattan plots showing results of Extended Haplotype Homozygosity-based tests (a) *iHS* test computed on Tunisian Barbarine. (b) *iHS* test computed on Tunisian Black Thibar. (c) *iHS* test computed on Hamra and Ouled-Djellal population. (d) *Rsb* test contrasting haplotypes of fat-tailed Tunisian Barbarine vs Algerian thin-tailed Hamra and Ouled-Djellal.

Table 3. Genomic regions putatively under selection identified using the *iHS* statistic and gene content of these regions. Genes in bold are located near the selection signals. Underlined genes are strong candidates related to important traits.

Breed	OAR	Start (Mb)	End (Mb)	Maximum Peak (Mb)	Genes
BART	3	104	105	104.567	GPAT2, FAHD2A, KCNIP3, PROM2, LOC106990992, ZNF2, ZNF514, LOC105611091, MRPS5, LOC101117493, LOC101115527, MALL, LOC105614684, NPHP1, **LOC101117149**, LOC101117749, BUB1, ACOXL
	13	46	48	47.527	ZMYND11, PRNP, LOC105609932, PRND, PRNT, RASSF2, SLC23A2, LOC105609938, TMEM230, PCNA, CDS2, LOC101109379, PROKR2, GPCPD1, LOC101109635, C13H20orf196, TRNAF-GAA, CHGB, TRMT6, MCM8, LOC105609936, CRLS1, LRRN4, **FERMT1**, LOC106991507, LOC101117437
	15	50	51	50.361	CLPB, PDE2A, **ARAP1**, STARD10, LOC105602286, ATG16L2, FCHSD2, LOC105602287, LOC106991638, P2RY2, P2RY6,

					ARHGEF17, RELT, FAM168A
BT	2	245	246	245.044	MUL1, **CAMK2N1**, VWA5B1, UBXN10, PLA2G2C, PLA2G2F, PLA2G2D, PLA2G5, LOC105611501, LOC101107171, LOC101107420, LOC101107675, PLA2G2E, OTUD3, LOC105611511, LOC105613466, RNF186, LOC105611518, TMCO4, HTR6, NBL1, MINOS1
	3	68	70	69.813	EFEMP1, LOC105613776, PNPT1, PPP4R3B, CFAP36, CCDC88A, LOC101122262, MTIF2, LOC106991051, RPS27A, CLHC1, TRNAE-UUC, LOC106990985, RTN4, EML6, SPTBN1, C3H2orf73, LOC106991052, ACYP2, TSPYL6, LOC105613853, **PSME4**, GPR75
	3	110	111	110.523	LOC101120997, ATXN7L3B, **KCNC2**, LOC101123110, CAPS2
	6	34	35	34.821	CCSER1, MMRN1, **SNCA**
	6	47	48	47.882	LOC101107099, **LOC105612659**
	6	49	50	49.096	**PCDH7**
	11	41	42	41.03	LOC100526782, **KRT34**, V15, K38, KRT32, KRT35, KRT36, LOC101117431, KRT15, LOC101117946, KRT9, LOC101118459, LOC101108147, KRT17, LOC101118712, EIF1, GAST, HAP1, JUP, P3H4, FKBP10, LOC105611406, NT5C3B, KLHL10, KLHL11, ACLY, TTC25, CNP, DNAJC7, NKIRAS2, ZNF385C, LOC106991400, DHX58, KAT2A, HSPB9, RAB5C, LOC106991401, LOC101110253, HCRT, GHDC, STAT5B, STAT5A, STAT3, PTRF, ATP6V0A1
	11	43	44	43.880	SOST, DUSP3, C11H17orf105, MPP3, CD300LG, MPP2, PPY, PYY, LOC101114885, NAGS, TMEM101, LSM12, G6PC3, HDAC5, LOC105616384, C11H17orf53, LOC105616385, ASB16, TMUB2, ATXN7L3, UBTF, LOC105612015, SLC4A1, RUNDC3A, SLC25A39, GRN, FAM171A2, ITGA2B, GPATCH8, LOC105612017, FZD2, LOC105612018, LOC101116922, MEIOC, CCDC43, LOC106991432, DBF4B, ADAM11, **GJC1**, HIGD1B, EFTUD2, CCDC103, LOC101118203, GFAP, KIF18B
	12	13	14	13.27	LOC106990525, **BRINP3**, LOC105613926
	12	37	41	40.223	LOC105606472, PRRC2C, TRNAS-AGA, MYOC, LOC105616523, VAMP4, METTL13, DNM3, MIR199A, C12H1orf105, PIGC, LOC106991483, SUCO, FASLG, TNFSF18, LOC105606465, TNFSF4, LOC106991453, LOC101110527, AADACL4, LOC106991466, DHRS3, VPS13D, LOC105606462, TNFRSF1B, TRNAS-GGA, TNFRSF8, MIIP, MFN2, LOC105606461, PLOD1, KIAA2013, NPPB, NPPA, CLCN6, MTHFR, C12H1orf167, AGTRAP, LOC105606456, LOC105606457, DRAXIN, LOC105606457, MAD2L2, FBXO6, LOC101112585, FBXO2, **PTCHD2**, LOC105606453, UBIAD1, MTOR, ANGPTL7, EXOSC10, SRM, MASP2, LOC106991484, TARDBP, LOC105616526, CASZ1
	12	42	46	44.196	SLC25A33, LOC101113870, LOC105606443, LOC105606444, LOC106991486, SPSB1, LOC105616528, H6PD, LOC105606441, LOC105606442, GPR157, SLC2A5, CA6, ENO1, RERE, TRNAE-CUC, LOC101118715, SLC45A1, LOC101118971, LOC101119233, LOC105609137, ERRFI1, PARK7, TNFRSF9, UTS2, PER3, VAMP3, CAMTA1, **TRNAG-UCC**, DNAJC11, THAP3, LOC106991487, KLHL21, ZBTB48, LOC101116416, NOL9, PLEKHG5, TNFRSF25, ESPN, HES2, ACOT7, TRNAE-UUC, GPR153, HES3, LOC101117436, ICMT, RNF207, RPL22, CHD5, KCNAB2, NPHP4, TRNAS-GCU
	12	47	49	48.003	C12H1orf174, DFFB, CEP104, LRRC47, LOC105609712, SMIM1, CCDC27, TP73, WRAP73, TPRG1L, MEGF6,

					ARHGEF16, PRDM16, LOC105616533, **ACTRT2**, TRNAC-GCA, LOC101103167, TTC34, MMEL1, FAM213B, LOC105609715, HES5, PANK4, PLCH2, PEX10, RER1, MORN1, LOC105609719, SKI, LOC105616534, FAAP20, PRKCZ, GABRD, LOC106991455, LOC106991467, CFAP74, LOC101123057, TMEM52, CALML6, GNB1, NADK, LOC106991489, LOC101104843, MMP23B, MIB2
	12	50	52	50.784	CASP9, LOC101111210, CTRC, EFHD2, FHAD1, TMEM51, LOC105616540, KAZN, **LOC105610224**, LOC105616544, LOC106991445, PRDM2, LOC106991490, PDPN
	12	54	57	56.148	RABGAP1L, LOC106991446, MRPS14, CACYBP, TNN, KIAA0040, TNR, RFWD2, LOC105612974, LOC101117093, LOC101117349, TRNAW-CCA, PAPPA2, **ASTN1**, LOC101117862, BRINP2, LOC101118972
	14	11	13	11.814	GSE1, GINS2, C14H16orf74, EMC8, LOC101105179, LOC106991584, IRF8, LOC105616838, FOXF1, **MTHFSD**, FOXC2, FOXL1, LOC105616839, C14H16orf95, LOC106990561, FBXO31, MAP1LC3B, ZCCHC14, JPH3, KLHDC4, LOC105612266, LOC106991585, SLC7A5, CA5A, BANP, LOC105616840, ZNF469
	16	30	31	30.581	MRPS30, **FGF10**
	16	44	45	44.099	**LOC101116348**, LOC101116603, LOC101116863, LOC101120194, LOC101120441
	21	40	41	40.432	LOC101110371, ASRGL1, SCGB1A1, AHNAK, EEF1G, TUT1, MTA2, EML3, ROM1, B3GAT3, GANAB, INTS5, C21H11orf98, METTL12, LBHD1, LOC101112013, UBXN1, BSCL2, LOC101114066, HNRNPUL2, TTC9C, LOC101114319, ZBTB3, POLR2G, TAF6L, TMEM179B, TMEM223, NXF1, STX5, **WDR74**, SLC3A2, CHRM1, LOC101112776, TRNAE-UUC, LOC101117286, SLC22A8, LOC101117802, LOC101118064, LOC101113025, LOC101118318, LOC101118571, LOC101118829, LOC105604101, LOC101113546, LOC101119087, TRNAC-GCA, LOC101119346
HAMA & OLDA	5	28	29	28.261	PRDM6, LOC105611875, PPIC, **SNX24**, SNX2, SNCAIP
	6	77	78	77.814	ADGRL3, LOC101114018, **LOC101119476**
	10	41	42	41.404	**LOC101121273**
	13	35	36	35.9645	BAMBI, WAC, LOC105609169, LOC105609165, LOC101119745, MPP7, ARMC4, LOC101118374, LOC105609170, **MKX**
	18	59	60	59.121	LOC105603234, LOC105603235, GLRX5, **TCL1B**, LOC105603238, TCL1A, C18H14orf132, LOC105603241, BDKRB2, BDKRB1, ATG2B, GSKIP, AK7, LOC101121806, LOC105603242, PAPOLA

Genes overlapping these regions were retrieved. A total of 125 genes for BART, 416 genes for BT and 107 genes for HAMA and OLDA were identified, of which 82, 250 and 55 genes were mapped for BART, BT and HAMA_OLDA, respectively using the DAVID database. Only the genes retrieved from *iHS* BT have significantly enriched functional classes (Benjamini-corrected p-value <0,05) (Additional File 1). The first cluster presented Keratin type I associated genes according to Interpro domain, these genes are involved in *Staphylococcus aureus* infection and Estrogen signaling pathway according to KEGG pathway annotation, the second cluster represented Phospholipase A2 domain related genes, involved in a variety of functions related to lipid metabolism by controlling fatty acids digestion and absorption

Table 4. Genomic regions putatively under selection identified using the *Rsb* BART Vs HAMA_OLDA statistic. Genes in bold are located near the selection signals. Underlined genes are strong candidates related to important traits.

CHR	Start (Mb)	End (Mb)	Max peak	Selected breed	Genes
4	68	70	68.381	BART	JAZF1, TAX1BP1, LOC106991102, **LOC101104741**, HIBADH, LOC101104988, EVX1, HOXA13, LOC105606592, HOXA11, HOXA10, HOXA9, LOC105606599, HOXA3, HOXA5, HOXA4, HOXA2, LOC105606593, HOXA1, SKAP2, TRNAW-CCA, TRNAS-GGA, LOC106991133, SNX10, CBX3, HNRNPA2B1, NFE2L3, MIR148A, LOC101107436
7	82	83	82.587	BART	ELMSAN1, PTGR2, ZNF410, FAM161B, COQ6, ENTPD5, BBOF1, ALDH6A1, LIN52, VSX2, ABCD4, VRTN, **SYNDIG1L**, NPC2, ISCA2, LTBP2, AREL1, LOC105606507, LOC106991293, FCF1, YLPM1, PROX2, DLST
13	56	57	56.037	BART	**PHACTR3**, EDN3, LOC105616728, ZNF831, PRELID3B, LOC105606223, TUBB1, CTSZ, NELFCD, LOC101115640, LOC105616729, LOC101102411, LOC105606221, NPEPL1, STX16, APCDD1L
2	14	16	15.261	HAMA & OLDA	EPB41L4B, FRRS1L, TMEM245, CTNNAL1, FAM206A, IKBKAP, ACTL7A, ACTL7B, LOC105606795, LOC105606800, **LOC105606808**, TRNAE-CUC, LOC105606814, KLF4, TRNAW-CCA, RAD23B
2	105	107	105.883	HAMA & OLDA	GLRA3, LOC106990533, HPGD, LOC101113210, CEP44, FBXO8, LOC106990444, **LOC105608584**, HAND2, LOC105608588, SAP30, LOC105608589, HMGB2, GALNT7, LOC105608592, GALNTL6, TRNAW-CCA
3	89	90	89.530	HAMA & OLDA	**LOC105614903**, LOC105614904
3	122	123	122.792	HAMA & OLDA	MGAT4C, TRNAG-CCC, **TRNAC-GCA**
3	124	125	124.942	HAMA & OLDA	C3H12orf50, C3H12orf29, CEP290, TMTC3, KITLG, **TRNAG-CCC**
5	104	105	104.333	HAMA & OLDA	**FBXL17**, FER
6	80	81	80.531	HAMA & OLDA	LOC106991226, **LOC101119727**, EPHA5
6	114	115	114.475	HAMA & OLDA	LOC105608268, TRMT44, CPZ, HMX1, **ADRA2C**, LOC105615584, LOC106991218, LRPAP1, LOC106991219, LOC106991220, HGFAC, DOK7, RGS12, MSANTD1, HTT
13	35	36	35.917	HAMA & OLDA	BAMBI, WAC, LOC105609169, LOC105609165, LOC101119745, MPP7, ARMC4, LOC101118374, LOC105609170, **MKX**
16	44	45	44.188	HAMA & OLDA	**LOC101116348**, LOC101116603, LOC101116863, LOC101120194, LOC101120441

Discussion

In the present study, we investigated the population structure of two Tunisian (BART and BT) and four Algerian sheep breeds (BRBA, DMNA, HAMA and OLDA). All the analyses done (Fst, ADMIXTURE, DAPC and PCA) point to a genomic similarity between the fat-tailed Tunisian Barbarine and the thin-

tailed Algerian breeds HAMA and OLDA. This can be mainly explained by the widespread of cross-breeding of BART with the Tunisian Queue fine de l'ouest, a thin-tailed breed closely related to the before-mentioned Algerian populations. The extensive use of such cross-breeding is dictated by a change in consumers preference who became more inclined towards low-fat meat (Kdidi et al., 2015). The Algerian Barbarine is found to be more heterogeneous than its Tunisian counterpart. This is clearly demonstrated in ADMIXTURE and DAPC analyses with the latter showing the five BRBA individuals assigned to three different genetic clusters: 1, 2 and 5 (Figure 1b). The BRBA individual assigned to cluster 2 consisting mainly of HAMA and OLDA and having the same genetic structure as these two breeds in ADMIXTURE is in line with previous studies indicating that the Algerian Barbarine is threatened by dilution with the Ouled-Djellal population (Gaouar et al., 2017). However, conclusions regarding this breed should be drawn with caution because of the low number of BRBA individuals in this study. Further studies must be conducted including additional individuals sampled across the whole country to quantify more precisely the dilution phenomenon. On the other hand, our results corroborate the study of (Belabdi et al., 2019) who reported a significant intensification of crossing practices between OLDA and HAMA over the past 30 years leading to a dilution of the latter breed. All the population structure analyses revealed a clear separation of Black Thibar which reflects the fact that this breed benefits from intra-breed selection program. This latter is favored by the distinctive black coat color and a restricted geographical location. Indeed, the majority (99%) of Black Thibar flocks is located in North Tunisia (GIVLAIT, 2007).

We identified several regions putatively under ongoing selection for the three populations. The Black Thibar breed showed the highest number and the most obvious signals of selection. Indeed, five candidate genomic regions located on OAR12 containing more than 10 SNPs exceeding the significance threshold were identified for this breed. This often represents more than 30% of the total number of markers located within each genomic region. The high number of candidate regions detected within BT could be explained by the young age of this breed created during the early 20th century by the White Fathers with a reciprocal cross between the Queue fine de l'ouest and the imported Arles Merino, the latter originating from local ewes crossed with Spanish Merino rams (Mason, 1969).

The *iHS* test is known to be highly efficient in detecting ongoing selective sweeps (Voight et al., 2006). The selection signature on chromosome 12 spanning the interval [39.778 Mb – 40.383 Mb] may be the most biologically relevant due to the presence of 4 highly significant SNPs (7.02 <P-value<11.64). Furthermore, *iHS* values within this interval are largely negative indicating that selection is occurring on the derived allele (data not shown). The SNP with the highest P-value (position: 40,223,491 bp) is located within the *PTCHD2* gene (OAR12: 40,205,947 – 40,246,125 Mb). *PTCHD2* (called also *DISP3*) encodes a protein involved in neuronal proliferation and differentiation (Zíková et al., 2014). *PTCHD2* is also a tumor suppressor gene. Mutations in this gene predispose patients to Basal Cell Carcinoma (BCC), a type of skin cancers that most often develops on areas of skin exposed to the sun (Fujii et al., 2013; Smyth et al., 1999). Bearing in mind that Black Thibar was created and selected to fix the black color in the skin in order to tackle the issue of photosensitization of light-skinned animals engendered by the consumption of the St. John's wort (*Hypericum perforatum*) weed (Abdellatif, 1968), we can hypothesize that the selection signal within *PTCHD2* could be considered as another adaptive feature to counter the adverse effects of photosensitization on skin in Black Thibar. Further functional studies are needed to set whether or not there is a relationship between this gene and photosensitization. Another relevant candidate region on OAR12 putatively under ongoing selection in BT is located between 42,297 Mb and 44,841 Mb. This interval includes 8 highly significative SNPs (6.08 <P-value< 7.34) and overlaps with a previously reported genomic region potentially associated with nematode resistance in autochthonous Tunisian sheep (Ahbara et al., 2021). Likewise, our candidate region overlaps with selection signature identified in the Italian Laticauda and the Libyan Barbary (Mastrangelo et al., 2019). Within this interval, we selected one candidate gene, *SLC45A1* (Solute Carrier Family 45 Member 1) which was shown to be associated with wool production traits (more precisely with staple length) in Turkish sheep (Arzik et al., 2023). Our choice is motivated by two main reasons: i) *SLC45A1* is only 210 kb downstream the selection signal peak. ii) *SLC45A2*, a paralog to *SLC45A1*, is associated with

variation in coat color in several species such as sheep (Wang et al., 2016), horses (Sevane et al., 2019), dog (Wijesena and Schmutz, 2015) and mice (Le et al., 2020). Black Thibar was primarily selected for wool quality (Chalh et al., 2007). Therefore, we do not exclude an influence of *SLC45A1* on coat color in Black Thibar given that paralogous proteins have the same biochemical function even though they may act on different targets. Our candidate regions harbour two other genes influencing wool quality, *Keratin 34* (*KRT34*, OAR11: 41-42 Mb) and *WD Repeat Domain 74* (*WDR74*, OAR21:40-41 Mb). *KRT34* is among the keratin intermediate filaments (KIF) genes involved in wool formation in sheep (Yu et al., 2010). WDR74 is associated with hair follicle development in goat (Jin et al., 2020). In addition to wool, BT was selected since the 1960s for growth and reproduction traits. Under intensive breeding conditions, this breed has growth and reproductive performances significantly higher than that of the other Tunisian meat sheep breeds, namely the Barbarine and the Queue fine de l'ouest (Ben Salem et al., 2009). Accordingly, we expect that some of the genes located within the candidate regions are associated with both types of traits. On OAR12, we identified *ACTRT2* (*Actin Related Protein T2*), a cytoskeletal gene encoding a protein that belongs to the actin family which plays a crucial role in cell and tissue growth (García-González and van Gelderen, 2021). The selection signal peak is only 17 kb upstream *ACTRT2*. Importantly, this gene is also associated with male fertility and spermatogenesis (Chen et al., 2023; Louvandini et al., 2020). Another gene with pleiotropic effects on growth and reproduction traits is *PSME4*, a gene located 65 Kb upstream the selection signal peak on OAR03 (the SNP with the highest P-value in this region is located at 69,813,088 bp). *PSME4*, also called *PA200* (*Proteasome Activator PA200)*, has its highest RNA expression in skeletal muscle and testes (www.proteinatlas.org). PA200-/- mice has a faster decline in Gastrocnemius muscle fibre diameter than that in the wild-type mice (Jiang et al., 2021). CircPSME4 (circular RNA transcribed from PSME4 gene) was demonstrated to be a novel regulator of skeletal muscle development in pig (Zeng et al., 2023). PA200 has also an important non-redundant function during spermatogenesis. Indeed, loss of this gene led to a marked reduction in male fertility in mice owing to defects in spermatogenesis observed in meiotic spermatocytes and during the maturation of postmeiotic haploid spermatids (Khor et al., 2006).

Regarding the Tunisian Barbarine, we identified a clear signal, revealed by iHS test, on chromosome 13 spanning 2 Mb [46 – 48 Mb]. The candidate region on this chromosome includes 11 significant SNP (52% of the total number of SNP within this window). Examining the genes of this region, we identified an interesting candidate, CDS2, which is involved in phospholipid biosynthetic process and has been shown to be an important novel regulator of lipid storage in mammals (Qi et al., 2016). Therefore, this gene could be involved in the fat-tail phenotype of BART. A closer look to the region allowed us to identify a well-known gene: Bone morphogenetic protein 2 (BMP2, OAR13: 48,387,181 – 48,400,67) located less than 1Mb of our peak signal. This gene, identified in several studies (Ahbara et al., 2019; Lu et al., 2020; Mastrangelo et al., 2019; Zhao et al., 2020), as one of the major genes linked to fat-tail phenotype in sheep.

By contrasting haplotypes of fat-tailed Tunisian Barbarine and thin-tailed Algerian Hamra and Ouled-Djellal we retained another genomic region located [68 – 70 Mb] under selection in BART. This interval included 12 SNPs (38% of the total number of SNPs within the window) passing the threshold of −log10(p-value) equal to five. Nearby the peak signal, we found HIBADH (OAR04: 68,423,031 – 68,542,169), a gene that is shown to be involved in mitochondrial function of spermatozoa and the maintain of sperm motility (Tasi et al., 2013). Several studies highlighted the effect of heat stress on sheep fertility, as temperatures above 32 to 35°C decreases considerably sperm motility (Barragán Sierra et al., 2021; Mieusset et al., 1992; van Wettere et al., 2021). We speculate that the mutation observed in *HIBADH* allows BART to maintain good fertility rates under hot climate. We identified a second relevant region on OAR7 [82 – 83 Mb] under selection in BART spanning 1 Mb [82 – 83 Mb] with a peak signal at 82.587 Mb. The nearest gene to the peak is *SYNDIG1L*. This gene is known to be involved in body development of numerous livestock species. It was reported to be a factor affecting the final body weight and back-fat thickness in Landrace pigs (Lee et al., 2018). *SYNDIG1L* also affects the formation of bovine body shape in cattle (An et al., 2020) and regulates thoracic vertebral numbers in sheep (Zhong et al., 2021). From either side of this gene, we identified two other additional potentially

candidate genes: *VRTN* (OAR07: 82,533,315 – 82,535,355) and *NPC2* (OAR07: 82,634,501 – 82,644,085). *VRTN* is a major selection locus for the number of thoracic vertebrae in sheep (Li et al., 2019) while *NPC2* plays an important role in cholesterol hemostasis. Its recombinant protein increases triglyceride level in body fat (Adachi et al., 2014). *NPC2* was presented as a potential candidate gene for lipid metabolism in Asian sheep (Zhao et al., 2017).

The strongest selection signal detected within the thin-tailed Algerian populations, Hamra and Ouled-Djellal, was located on OAR13 [35-36 Mb]. This region is jointly identified by *iHS* and *Rsb* tests, with a peak signal (35.9645 Mb) found within *Mohawk homeobox* (*MKX*, OAR13: 35,886,096 – 35,953,956), a gene which regulates the number of vertebrae in pigs (Duijvesteijn et al., 2014). Furthermore, *MKX* negatively regulates MyoD expression and myoblast differentiation (Chuang et al., 2014). Importantly, reduced expression of MyoD gene in mice muscle might be associated with meat tenderness (Lian et al., 2013). Hence, *MKX* gene would be one of the genes under selection explaining differences in meat quality between the fat-tailed Barbarine and the thin-tailed Algerian breeds. Another, relevant region identified with *Rsb* BART Vs HAMA_OLDA, was located on OAR06 (114 – 115 Mb). In total, 25% of the SNPs in this window exceeded the significance threshold. *Adrenergic α2C receptor (ADRA2C)* (OAR06: 114,453,422 – 114,455,456) coincides with the selection peak in this region. *ADRA2C* is an inhibitory modulator of the sympathetic nervous system. Knockout mice for this gene show physiological and behavioral alterations such as enhanced startle response, shortened attack latency, and diminished acoustic prepulse inhibition (Sallinen et al., 1998). *ADRA2C* is located near to one of the strongest selection signatures identified in the context of chicken domestication (Rubin et al., 2010). Studies about differences in behavior between sheep populations from North Africa are almost inexistant. Hence, for now we are not able to find an explanation for the reason of this gene being under divergent selection between BART and thin-tailed Algerian breeds.

In the present study, the majority of the genes putatively under selection in fat-tailed sheep or under divergent selection between fat- and thin-tailed breeds are associated either with lipid metabolism or with vertebral development. This is in agreement with the review made by (Kalds et al., 2021) in which they observed that differences between fat- and thin-tailed sheep was not only perceptible from fat deposition level but also from caudal vertebrae number and tail length.

Conclusion

Our study highlighted several genes related to important traits playing a key role in the continuity and the survive of Tunisian and Algerian sheep breeds granting their thrive in the North African environment with a firm performance. We also shed light on the importance of preserving these genetically unique resources. Additionally, recognizing the impact of hybridization on the genomic architecture of these breeds would emphasize the need for suitable management strategies that balance the preservation of genetic diversity with targeted breeding goals.

Conflict of interest

The authors declare that they have no conflict of interest.

References

Abdellatif, K., 1968. Le mouton Noir de Thibar. Thèse pour le Doctorat Vétérinaire, Ecole Nationale Vétérinaire de Toulouse, N°32, 62pp.

Adachi, T., Ishii, K., Matsumoto, Y., Hayashi, Y., Hamamoto, H., Sekimizu, K., 2014. Niemann-Pick disease type C2 protein induces triglyceride accumulation in silkworm and mammalian cell lines. Biochem J 459, 137–147. DOI: 10.1042/BJ20130876

Ahbara, A., Bahbahani, H., Almathen, F., Al Abri, M., Agoub, M.O., Abeba, A., Kebede, A., Musa, H.H., Mastrangelo, S., Pilla, F., Ciani, E., Hanotte, O., Mwacharo, J.M., 2019. Genome-Wide Variation, Candidate Regions and Genes Associated with Fat Deposition and Tail Morphology in Ethiopian Indigenous Sheep. Frontiers in Genetics 9. DOI: 10.3389/fgene.2018.00699

Ahbara, A.M., Rouatbi, M., Gharbi, M., Rekik, M., Haile, A., Rischkowsky, B., Mwacharo, J.M., 2021. Genome-wide insights on gastrointestinal nematode resistance in autochthonous Tunisian sheep. Sci Rep 11, 9250. DOI: 10.1038/s41598-021-88501-3

Alexander, D.H., Novembre, J., Lange, K., 2009. Fast model-based estimation of ancestry in unrelated individuals. Genome Res 19, 1655–1664. DOI: 10.1101/gr.094052.109

An, B., Xu, Lei, Xia, J., Wang, X., Miao, J., Chang, T., Song, M., Ni, J., Xu, Lingyang, Zhang, L., Li, J., Gao, H., 2020. Multiple association analysis of loci and candidate genes that regulate body size at three growth stages in Simmental beef cattle. BMC Genetics 21, 32.
DOI: 10.1186/s12863-020-0837-6

Arzik, Y., Kizilaslan, M., Behrem, S., White, S.N., Piel, L.M.W., Cinar, M.U., 2023. Genome-Wide Scan of Wool Production Traits in Akkaraman Sheep. Genes 14, 713. DOI: 10.3390/genes14030713

Barragán Sierra, A., Avendaño-Reyes, L., Hernández Rivera, J.A., Vicente-Pérez, R., Correa-Calderón, A., Mellado, M., Meza-Herrera, C.A., Macías-Cruz, U., Barragán Sierra, A., Avendaño-Reyes, L., Hernández Rivera, J.A., Vicente-Pérez, R., Correa-Calderón, A., Mellado, M., Meza-Herrera, C.A., Macías-Cruz, U., 2021. Thermoregulation and reproductive responses of rams under heat stress. Review. Revista mexicana de ciencias pecuarias 12, 910–931.
DOI: 10.22319/rmcp.v12i3.5624

Belabdi, I., Ouhrouch, A., Lafri, M., Gaouar, S.B.S., Ciani, E., Benali, A.R., Ould Ouelhadj, H., Haddioui, A., Pompanon, F., Blanquet, V., Taurisson-Mouret, D., Harkat, S., Lenstra, J.A., Benjelloun, B., Da Silva, A., 2019. Genetic homogenization of indigenous sheep breeds in Northwest Africa. Sci Rep 9, 7920. DOI: 10.1038/s41598-019-44137-y

Ben Salem, H., Lassoued, N., Rekik, M., 2011. Merits of the fat-tailed Barbarine sheep raised in different production systems in Tunisia: digestive, productive and reproductive characteristics. Trop Anim Health Prod 43, 1357–1370. DOI: 10.1007/s11250-011-9863-8

Ben Salem, I., Rekik, M., Hammami, H., Ben Hamouda, M., Aloulou, R., Sâadoun, L., 2009. Facteurs de variation non génétique de la productivité des brebis de race Noire de Thibar. Rev. Elev. Med. Vet. Pays Trop. 62, 59. DOI: 10.19182/remvt.10095

Cao, Y.-H., Xu, S.-S., Shen, M., Chen, Z.-H., Gao, L., Lv, F.-H., Xie, X.-L., Wang, X.-H., Yang, H., Liu, C.-B., Zhou, P., Wan, P.-C., Zhang, Y.-S., Yang, J.-Q., Pi, W.-H., Hehua, Ee., Berry, D.P., Barbato, M., Esmailizadeh, A., Nosrati, M., Salehian-Dehkordi, H., Dehghani-Qanatqestani, M., Dotsev, A.V., Deniskova, T.E., Zinovieva, N.A., Brem, G., Štěpánek, O., Ciani, E., Weimann, C., Erhardt, G., Mwacharo, J.M., Ahbara, A., Han, J.-L., Hanotte, O., Miller, J.M., Sim, Z., Coltman, D., Kantanen, J., Bruford, M.W., Lenstra, J.A., Kijas, J., Li, M.-H., 2021. Historical Introgression from Wild Relatives Enhanced Climatic Adaptation and Resistance to Pneumonia in Sheep. Molecular Biology and Evolution 38, 838–855.
DOI: 10.1093/molbev/msaa236

Chalh, A., Gazzah, M., Djemali, M., Noureddine, C., 2007. Genetic and Phenotypic Characterization of the Tunisian Noire De Thibar Lambs on Their Growth Traits. Journal of Biological Sciences 7.
DOI: 10.3923/jbs.2007.1347.1353

Chen, H., Li, Y., Deng, C., Liang, X., Liu, G., 2023. O-254 ACTRT2 deficiency increases spermatogonia vulnerability to ferroptosis. Human Reproduction 38, dead093.308.
DOI: 10.1093/humrep/dead093.308

Chen, W.C., Dorman, K., 2013. Phyclust: Phylogenetic Clustering (Phyloclustering). http://cran.r-project.org/package=phyclust

Chuang, H.-N., Hsiao, K.-M., Chang, H.-Y., Wu, C.-C., Pan, H., 2014. The homeobox transcription factor Irxl1 negatively regulates MyoD expression and myoblast differentiation. FEBS J 281, 2990–3003. DOI: 10.1111/febs.12837

Djaout, A., Farida, A.-B., Chekal, F., El-Bouyahiaoui, R., Rabhi, A., Boubekeur, A., Benidir, M., Chekal, F., Ameur, A.A., Gaouar, S.B.S., 2017. Biodiversity state of Algerian sheep breeds. Genetics & Biodiversity Journal 1, 1–18. DOI: 10.46325/gabj.v1i1.74

Duijvesteijn, N., Veltmaat, J.M., Knol, E.F., Harlizius, B., 2014. High-resolution association mapping of number of teats in pigs reveals regions controlling vertebral development. BMC Genomics 15, 542. DOI: 10.1186/1471-2164-15-542

Fujii, K., Ohashi, H., Suzuki, M., Hatsuse, H., Shiohama, T., Uchikawa, H., Miyashita, T., 2013. Frameshift mutation in the PTCH2 gene can cause nevoid basal cell carcinoma syndrome. Familial Cancer 12, 611–614. DOI: 10.1007/s10689-013-9623-1

Gaouar, S.B.S., Lafri, M., Djaout, A., El-Bouyahiaoui, R., Bouri, A., Bouchatal, A., Maftah, A., Ciani, E., Da Silva, A.B., 2017. Genome-wide analysis highlights genetic dilution in Algerian sheep. Heredity 118, 293–301. DOI: 10.1038/hdy.2016.86

Gaouar, S.B.S., Silva, A.D., Ciani, E., Kdidi, S., Aouissat, M., Dhimi, L., Lafri, M., Maftah, A., Mehtar, N., 2015. Admixture and Local Breed Marginalization Threaten Algerian Sheep Diversity. PLOS ONE 10, e0122667. DOI: 10.1371/journal.pone.0122667

García-González, J., van Gelderen, K., 2021. Bundling up the Role of the Actin Cytoskeleton in Primary Root Growth. Frontiers in Plant Science 12. DOI: 10.3389%2Ffpls.2021.777119

Gautier, M., Vitalis, R., 2012. rehh: an R package to detect footprints of selection in genome-wide SNP data from haplotype structure. Bioinformatics 28, 1176–1177. DOI: 10.1093/bioinformatics/bts115

GIVLAIT, 2007. Faisabilité d'un signe de qualité pour la viande d'agneau de race Noire de Thibar. http://www.givlait.com.tn/medias/files/Noire%20de%20thibar(1)_compressed.pdf

Jemaa, S., Kdidi, S., Gdura, A., Dayhum, A., Eldaghayes, I., Boussaha, M., Rebours, E., Yahyaoui, M., 2019. Inferring the population structure of the Maghreb sheep breeds using a medium-density SNP chip. Animal Genetics 50. DOI: 10.1111/age.12831

Jiang, T.-X., Ma, S., Han, X., Luo, Z.-Y., Zhu, Q.-Q., Chiba, T., Xie, W., Lin, K., Qiu, X.-B., 2021. Proteasome activator PA200 maintains stability of histone marks during transcription and aging. Theranostics 11, 1458–1472. DOI: 10.7150/thno.48744

Jin, M., Lu, J., Fei, X., Lu, Z., Quan, K., Liu, Y., Chu, M., Di, R., Wang, H., Wei, C., 2020. Genetic Signatures of Selection for Cashmere Traits in Chinese Goats. Animals (Basel) 10, 1905. DOI: 10.3390/ani10101905

Jombart, T., 2008. adegenet: a R package for the multivariate analysis of genetic markers. Bioinformatics 24, 1403–1405. DOI: 10.1093/bioinformatics/btn129

Kalds, P., Luo, Q., Sun, K., Zhou, S., Chen, Y., Wang, X., 2021. Trends towards revealing the genetic architecture of sheep tail patterning: Promising genes and investigatory pathways. Animal Genetics 52, 799–812. DOI: 10.1111/age.13133

Kdidi, S., Calvo, J.H., González-Calvo, L., Ben Sassi, M., Khorchani, T., Yahyaoui, M.H., 2015. Genetic relationship and admixture in four Tunisian sheep breeds revealed by microsatellite markers. Small Ruminant Research 131, 64–69. DOI: 10.1016/j.smallrumres.2015.08.012

Khor, B., Bredemeyer, A.L., Huang, C.-Y., Turnbull, I.R., Evans, R., Maggi, L.B., White, J.M., Walker, L.M., Carnes, K., Hess, R.A., Sleckman, B.P., 2006. Proteasome activator PA200 is required for normal spermatogenesis. Mol Cell Biol 26, 2999–3007. DOI: 10.3390/biom13040586

Le, L., Escobar, I.E., Ho, T., Lefkovith, A.J., Latteri, E., Haltaufderhyde, K.D., Dennis, M.K., Plowright, L., Sviderskaya, E.V., Bennett, D.C., Oancea, E., Marks, M.S., 2020. SLC45A2 protein stability and regulation of melanosome pH determine melanocyte pigmentation. Mol Biol Cell 31, 2687–2702. DOI: 10.1091/mbc.E20-03-0200

Lee, Y.-S., Shin, D., Song, K.-D., 2018. Dominance effects of ion transport and ion transport regulator genes on the final weight and backfat thickness of Landrace pigs by dominance deviation analysis. Genes Genomics 40, 1331–1338. DOI: 10.1007/s13258-018-0728-7

Li, C., Li, M., Li, X., Ni, W., Xu, Y., Yao, R., Wei, B., Zhang, M., Li, H., Zhao, Y., Liu, L., Ullah, Y., Jiang, Y., Hu, S., 2019. Whole-Genome Resequencing Reveals Loci Associated With Thoracic Vertebrae Number in Sheep. Front Genet 10, 674. DOI: 10.3389/fgene.2019.00674

Lian, T., Wang, L., Liu, Y., 2013. A New Insight into the Role of Calpains in Post-mortem Meat Tenderization in Domestic Animals: A review. Asian-Australas J Anim Sci 26, 443–454.
DOI: 10.5713/ajas.2012.12365

Louvandini, H., Corrêa, P.S., Amorín, R., Liu, L., Ieda, E.H., Jimenez, C.R., Tsai, S.M., McManus, C.M., Peñagaricano, F., 2020. Gestational and lactational exposure to gossypol alters the testis transcriptome. BMC Genomics 21, 59. DOI: 10.1186/s12864-020-6487-2

Lu, Z., Liu, J., Han, J., Yang, B., 2020. Association Between BMP2 Functional Polymorphisms and Sheep Tail Type. Animals (Basel) 10, 739. DOI: 10.3390/ani10040739

Lv, F.-H., Agha, S., Kantanen, J., Colli, L., Stucki, S., Kijas, J.W., Joost, S., Li, M.-H., Ajmone Marsan, P., 2014. Adaptations to climate-mediated selective pressures in sheep. Mol Biol Evol 31, 3324–3343. DOI: 10.1093/molbev/msu264

Ma,Y., Ding, X., Qanbari, S., Weigend, S., Zhang, Q. & Simianer, H., 2015. Properties of different selection signature statistics and a new strategy for combining them. Heredity 115, 426–436. https://doi.org/10.1038/hdy.2015.42

Mason, I.L., 1969. A world dictionary of livestock breeds, types and varieties (4th Edition).

Mastrangelo, S., Bahbahani, H., Moioli, B., Ahbara, A., Abri, M.A., Almathen, F., Silva, A. da, Belabdi, I., Portolano, B., Mwacharo, J.M., Hanotte, O., Pilla, F., Ciani, E., 2019. Novel and known signals of selection for fat deposition in domestic sheep breeds from Africa and Eurasia. PLOS ONE 14, e0209632. DOI: 10.1371/journal.pone.0209632

Mieusset, R., Quintana Casares, P., Sanchez Partida, L.G., Sowerbutts, S.F., Zupp, J.L., Setchell, B.P., 1992. Effects of heating the testes and epididymides of rams by scrotal insulation on fertility and embryonic mortality in ewes inseminated with frozen semen. J Reprod Fertil 94, 337–343.
DOI: 10.1530/jrf.0.0940337

Paradis, E., Claude, J., Strimmer, K., 2004. APE: Analyses of Phylogenetics and Evolution in R language. Bioinformatics 20, 289–290. DOI: 10.1093/bioinformatics/btg412

Proteinatlas.org - The Human Protein Atlas PSME4 Protein Expression Summary. Available online: https://www.proteinatlas.org/ENSG00000068878-PSME4 (Accessed on February 02, 2024)

Qi, Y., Kapterian, T.S., Du, X., Ma, Q., Fei, W., Zhang, Y., Huang, X., Dawes, I.W., Yang, H., 2016. CDP-diacylglycerol synthases regulate the growth of lipid droplets and adipocyte development [S]. Journal of Lipid Research 57, 767–780. DOI: 10.1194/jlr.M060574

Rochus, C.M., Tortereau, F., Plisson-Petit, F., Restoux, G., Moreno-Romieux, C., Tosser-Klopp, G., Servin, B., 2018. Revealing the selection history of adaptive loci using genome-wide scans for selection: an example from domestic sheep. BMC Genomics 19, 71. DOI: 10.1186/s12864-018-4447-x

Rosenberg, N.A., 2003. distruct: a program for the graphical display of population structure: PROGRAM NOTE. Molecular Ecology Notes 4, 137–138. DOI: 10.1046/j.1471-8286.2003.00566.x

Rousset, F., 2008. genepop'007: a complete re-implementation of the genepop software for Windows and Linux. Molecular Ecology Resources 8, 103–106. DOI: 10.1111/j.1471-8286.2007.01931.x

Rubin, C.-J., Zody, M.C., Eriksson, J., Meadows, J.R.S., Sherwood, E., Webster, M.T., Jiang, L., Ingman, M., Sharpe, T., Ka, S., Hallböök, F., Besnier, F., Carlborg, Ö., Bed'hom, B., Tixier-Boichard, M., Jensen, P., Siegel, P., Lindblad-Toh, K., Andersson, L., 2010. Whole-genome resequencing reveals loci under selection during chicken domestication. Nature 464, 587–591. DOI: 10.1038/nature08832

Sabeti, P.C., Varilly, P., Fry, B., Lohmueller, J., Hostetter, E., Cotsapas, C., Xie, X., Byrne, E.H., McCarroll, S.A., Gaudet, R., Schaffner, S.F., Lander, E.S., 2007. Genome-wide detection and characterization of positive selection in human populations. Nature 449, 913–918.
DOI: 10.1038/nature06250

Sallinen, J., Haapalinna, A., Viitamaa, T., Kobilka, B.K., Scheinin, M., 1998. Adrenergic alpha2C-receptors modulate the acoustic startle reflex, prepulse inhibition, and aggression in mice. J Neurosci 18, 3035–3042. DOI: 10.1523/JNEUROSCI.18-08-03035.1998

Sevane, N., Sanz, C.R., Dunner, S., 2019. Explicit evidence for a missense mutation in exon 4 of SLC45A2 gene causing the pearl coat dilution in horses. Anim Genet 50, 275–278. DOI: 10.1111/age.12784

Smyth, I., Narang, M.A., Evans, T., Heimann, C., Nakamura, Y., Chenevix-Trench, G., Pietsch, T., Wicking, C., Wainwright, B.J., 1999. Isolation and Characterization of Human Patched 2 (PTCH2), a Putative Tumour Suppressor Gene in Basal Cell Carcinoma and Medulloblastoma on Chromosome 1p32. Human Molecular Genetics 8, 291–297. DOI: 10.1093/hmg/8.2.291

Tang, K., Thornton, K.R., Stoneking, M., 2007. A New Approach for Using Genome Scans to Detect Recent Positive Selection in the Human Genome. PLOS Biology 5, e171.
DOI: 10.1371/journal.pbio.0050171

Tasi, Y.-C., Chao, H.-C.A., Chung, C.-L., Liu, X.-Y., Lin, Y.-M., Liao, P.-C., Pan, H.-A., Chiang, H.-S., Kuo, P.-L., Lin, Y.-H., 2013. Characterization of 3-hydroxyisobutyrate dehydrogenase, HIBADH, as a sperm-motility marker. J Assist Reprod Genet 30, 505–512. DOI: 10.1007/s10815-013-9954-8

Van Wettere, W.H.E.J., Kind, K.L., Gatford, K.L., Swinbourne, A.M., Leu, S.T., Hayman, P.T., Kelly, J.M., Weaver, A.C., Kleemann, D.O., Walker, S.K., 2021. Review of the impact of heat stress on reproductive performance of sheep. Journal of Animal Science and Biotechnology 12, 26.
DOI: 10.1186/s40104-020-00537-z

Voight, B.F., Kudaravalli, S., Wen, X., Pritchard, J.K., 2006. A Map of Recent Positive Selection in the Human Genome. PLOS Biology 4, e72. DOI: 10.1371/journal.pbio.0040072

Wang, H., Xue, L., Li, Y., Zhao, B., Chen, T., Liu, Y., Chang, L., Wang, J., 2016. Distribution and expression of SLC45A2 in the skin of sheep with different coat colors. Folia Histochemica et Cytobiologica 54, 143–150. DOI: 10.5603/FHC.a2016.0015

Wijesena, H.R., Schmutz, S.M., 2015. A Missense Mutation in SLC45A2 Is Associated with Albinism in Several Small Long Haired Dog Breeds. Journal of Heredity 106, 285–288.
DOI: 10.1093/jhered/esv008

Yu, Z.D., Sw, G., Je, W., Oam, W., Aj, N., Aj, P., 2010. Identification of novel wool keratin intermediate filament genes in sheep skin. Proceedings of the New Zealand Society of Animal Production 70, 222–228. https://www.nzsap.org/system/files/proceedings/2010/ab10052.pdf

Zeng, M., Yan, S., Yang, P., Li, Q., Li, J., Fan, X., Liu, X., Yao, Y., Wang, W., Chen, R., Han, G., Yang, Y., Tang, Z., 2023. Circular RNA transcriptome across multiple tissues reveal skeletal muscle-specific circPSME4 regulating myogenesis. International Journal of Biological Macromolecules 251, 126322. DOI: 10.1016/j.ijbiomac.2023.126322

Zhao, F., Deng, T., Shi, L., Wang, W., Zhang, Q., Du, L., Wang, L., 2020. Genomic Scan for Selection Signature Reveals Fat Deposition in Chinese Indigenous Sheep with Extreme Tail Types. Animals (Basel) 10, 773. DOI: 10.3390/ani10050773

Zhao, Y.-X., Yang, J., Lv, F.-H., Hu, X.-J., Xie, X.-L., Zhang, M., Li, W.-R., Liu, M.-J., Wang, Y.-T., Li, J.-Q., Liu, Y.-G., Ren, Y.-L., Wang, F., Hehua, Ee., Kantanen, J., Arjen Lenstra, J., Han, J.-L., Li, M.-H., 2017. Genomic Reconstruction of the History of Native Sheep Reveals the Peopling Patterns of Nomads and the Expansion of Early Pastoralism in East Asia. Mol Biol Evol 34, 2380–2395. DOI: 10.1093/molbev/msx181

Zhong, Y.-J., Yang, Y., Wang, X.-Y., Di, R., Chu, M.-X., Liu, Q.-Y., 2021. Expression analysis and single-nucleotide polymorphisms of SYNDIG1L and UNC13C genes associated with thoracic vertebral numbers in sheep (Ovis aries). Arch Anim Breed 64, 131–138. DOI: 10.5194/aab-64-131-2021

Zíková, M., Konířová, J., Ditrychová, K., Corlett, A., Kolář, M., Bartůněk, P., 2014. DISP3 promotes proliferation and delays differentiation of neural progenitor cells. FEBS Letters 588, 4071–4077. DOI: 10.1016/j.febslet.2014.09.03