Small Ruminant Research 251 (2025) 107570



Contents lists available at ScienceDirect

Small Ruminant Research

journal homepage: www.elsevier.com/locate/smallrumres





Unveiling genomic diversity and population structure in three Anatolian goats at genome-wide level

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ARTICLE INFO

Keywords:
Genetic diversity
Indigenous breed
Animal genetic resources
NGS
Turkish goats

ABSTRACT

Goats, domesticated around 10,000 years ago, have played a vital role in human civilization, particularly in developing regions where they are valued for their economic and ecological versatility. Türkiye, acting as a geographic bridge between Asia and Europe, is home to significant native goat populations, including Hair (HAI), Honamlı (HNM), and Kabakulak (KBK) goats. Despite their importance, limited genetic research has been conducted, especially on KBK, which has historically been classified as a sub-variety of HAI. This study is the first to apply double-digest restriction-site associated DNA sequencing (ddRADseq) to explore the genomic diversity and population structure of these breeds, analyzing 309,342 single nucleotide polymorphisms (SNPs) from 481 individuals. Genetic diversity parameters highlighted notable differences among the breeds. HAI exhibited the highest observed heterozygosity ($H_O = 0.307$) and nucleotide diversity ($\pi = 0.281$), whereas KBK displayed the lowest values ($H_O = 0.301$ and $\pi = 0.278$). Principal component analysis (PCA) and ADMIXTURE algorithm identified KBK as a separate genetic cluster, distinct from the overlapping HAI and HNM populations. Pairwise genetic differentiation (F_{ST}) values confirmed KBK's genetic distinctiveness. Effective population size (Ne) estimates showed a historical decline across all breeds, with HAI maintaining the largest values. Linkage disequilibrium (LD) decay patterns further highlighted higher genetic diversity in HAI. These findings underline the need for conservation strategies to preserve KBK's genetic composition. This study provides insights into the genetic diversity of Turkish native goats, offering a foundation for sustainable breeding programs and contributing important data to livestock genetics research.

1. Introduction

Domesticated in the Early Civilization Zone around 10,000 years ago, marking the beginning of animal husbandry, goats have been fundamental to the survival of mankind, especially in resource-limited regions where their adaptability ensures sustainability by supplying animal-derived products such as meat, milk, fiber, and hides (Zeder and Hesse, 2000). Today, over 1 billion goats are predominantly located in Asia and Africa, which highlight their crucial role in supporting economic stability and rural livelihoods. Türkiye, located between Asia and Europe, not only bridges continents but also harbors unique genetic diversity within its native goat populations, contributing critically to their global distribution.

The native goat population in Türkiye is estimated at approximately 10.3 million, with HAI goats comprising over 90 % of this population (Anonymous, 2023). HAI goats are extensively bred throughout the country, with a particularly high population density in the Taurus Mountains and the Mediterranean region. Other native goat breeds, such as HNM and KBK, are also reared in these regions. HNM goats are primarily raised in Antalya, Burdur, and Isparta provinces, while KBK goats are exclusively bred in geographic zones distributed between Antalya and Muğla provinces (Karsli et al., 2020; Aktaş et al., 2024). Historically,

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https://doi.org/10.1016/j.smallrumres.2025.107570
Received 17 February 2025; Received in revised form 21 June 2025; Accepted 8 July 2025
Available online 14 July 2025
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the KBK goat has been regarded as a sub-variety of the HAI goat, largely due to the limited availability of molecular and bioinformatics studies on native Turkish goat breeds. However, a study by Karsli et al. (2020), which utilized 20 microsatellite loci in 30 KBK goats, identified clear genetic differences between KBK and HAI goats. Moreover, a recent study has focused on revealing migration events among HAI, HNM, KBK, and Norduz breeds via 28 microsatellite loci in which no migration events were declared between HAI and KBK while migration edges have been identified between HAI and HNM breeds by the TreeMix algorithm (Demir, 2024). Despite these findings, KBK goats continue to be classified as a sub-variety of HAI. Consequently, within the framework of national goat breeding projects conducted by the Turkish Ministry of Agriculture, KBK goats are not evaluated as a separate breed but are included under HAI breeding programs. This approach results in the joint rearing of HAI and KBK goats, which inadvertently facilitates unintentional crossbreeding. Uncontrolled crossbreeding leads to the erosion of genetic diversity among these populations, thereby undermining conservation efforts.

Grasping the mechanisms by which natural selection alters genetic variation within populations is a key focus in evolutionary genetics (Brito et al., 2015). Genetic variability parameters offer crucial insights regarding population structure, adaptability, and evolutionary potential. Recent advancements in next-generation sequencing (NGS) technologies have modernized genetic research, allowing for high-resolution analyses of genomic diversity. Among these methods, ddRADseq has emerged as a highly efficient technique for yielding high-density SNP datasets, facilitating detailed evaluations of genomic variability and population stratification. This approach has proven invaluable in conservation genetics and breeding programs. Its utility has been demonstrated in various livestock species, including cattle (Demir et al., 2023), sheep (Karsli, 2024), chickens (Doublet et al., 2024), and pigs (Vani et al., 2024), as well as in aquaculture species such as fish (Magris et al., 2022), highlighting its broad applicability in genetic research.

Compared to SNP arrays, which are primarily developed for commercial breeds and thus have limited efficiency in detecting population-specific variants, ddRADseq provides significant advantages, particularly for native breeds. Unlike SNP chips, which rely on predefined markers and may fail to capture novel polymorphisms of local breeds, ddRADseq enables the identification of a wider range of genetic diversity and novel SNPs. By employing two restriction enzymes instead of one, as in RADseq, ddRADseq enhances locus coverage, optimizes fragment size selection, and improves SNP detection accuracy while remaining cost-effective for large-scale studies. This is further exemplified by Chen et al. (2022), who applied RADseq in 60 Yunling and Nubian goats and identified 151,071 SNPs, whereas the present study, utilizing ddRADseq, detected 309,342 SNPs across 481 Turkish goats, demonstrating its superior resolution and efficiency in SNP discovery.

This study aims to investigate the genetic variability and population structure of HAI, HNM, and KBK goats using approximately 310 K SNPs generated through the ddRADseq method. Specifically, the study focuses on elucidating the genetic differences between KBK and HAI goats to support the conservation and sustainable management of these breeds. Considering that KBK goats have long been classified as a sub-variety of HAI despite limited genomic evidence, this study seeks to test the hypothesis that KBK represents a genetically distinct population. Furthermore, the research aims to provide critical insights into the preservation of local genetic resources and inform future breeding programs. To the best of our knowledge, no previous studies have utilized NGS platforms or high-density SNP arrays to explore genetic diversity and population structure in Anatolian goat breeds, making this study a novel contribution to the field.

2. Material and method

2.1. Blood collection and DNA extraction

A total of 576 blood samples were collected from HAI, HNM, and KBK goats. The morphological characteristics of the goat breeds are illustrated in Fig. 1. The sample distribution included 200 HAI (180 females, 20 males), 184 HNM (164 females, 20 males), and 192 KBK (172 females, 20 males). HAI and HNM samples were collected from Elmalı and Korkuteli districts, but KBK samples were obtained from Kaş and western Elmalı regions of Antalya province (Fig. 2). Although Kaş is geographically a coastal district, goat breeding is almost exclusively conducted in its highland plateaus. These areas, located at high elevations, host the primary goat herding zones. Elmalı and Korkuteli lie in inner highland regions (approximately 1000–1500 m above sea level), characterized by a continental Mediterranean climate. The upland zones of Kaş used for goat husbandry share similar altitudinal and climatic features, enabling meaningful comparisons among geographically separate but ecologically comparable regions.

To ensure the selection of unrelated individuals, at least five herds were sampled per district, with animal selection guided by flock owners' information and the typical phenotypic traits of each breed. All sampled herds were part of the national conservation program. Approximately 3 mL blood was drawn from the jugular vein of each animal by a licensed veterinarian and transferred into 5 mL EDTA vacutainer tubes to prevent coagulation. Genomic DNA was extracted from blood samples using the GeneJET Genomic DNA Purification Kit (Thermo K0721), following the manufacturer's instructions. DNA concentrations were quantified using a Qubit 4™ fluorometer (Thermo Fisher Scientific) with a double-stranded DNA high-sensitivity assay kit (Invitrogen) to ensure consistency across all samples.

2.2. ddRADseq-based library preparation and sequencing

For ddRADseq library preparation, a modified version of the protocol described by Peterson et al. (2012) was implemented. A total of six genomic libraries were prepared, each containing DNA from 96 individuals. Genomic DNA was digested with *EcoR*-I and *Msp*-I restriction enzymes (New England Biolabs) and purified using AmPureXP beads (Beckman Coulter). Digested DNA fragments were ligated with P1 and P2 adapters, which contained unique five-base barcodes for sample identification, using T4 DNA ligase. After ligation, DNA fragments were cleaned again using AmPureXP beads, and reverse indexes were incorporated during PCR amplification for library enrichment. Following PCR, the libraries underwent a final purification step with AmPureXP beads to remove non-specific fragments and ensure high library quality. Sequencing was carried out on the Illumina HiSeq X Ten platform using paired-end reads (2×150 bp), producing high-quality SNP data for downstream analyses.

2.3. Data filtering and variant calling

Raw sequencing reads were demultiplexed and assigned to individual samples based on their unique barcode sequences using the *process_radtags* module in Stacks 2 software (Rochette et al., 2019). Reads containing unassigned or low-quality barcodes were discarded. Adapter sequences and low-quality bases were trimmed using the fastp program (Chen et al., 2018) under default parameters to ensure high-quality clean reads for subsequent analyses. Cleaned reads were then aligned to the *Capra hircus* reference genome assembly (ARS1.2) using the maximal exact match (MEM) algorithm implemented in the Burrows-Wheeler Aligner (BWA-MEM) software (Li and Durbin, 2009). The resulting alignments were stored in Binary Alignment Map (BAM) format, and further processing, such as sorting and indexing, was performed using SAMtools (Danecek et al., 2021). Variant discovery was conducted using the BCFtools pipeline (Danecek et al., 2021). Only

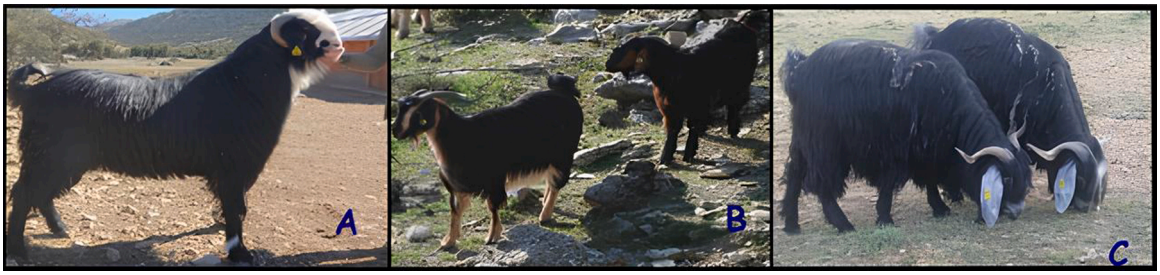

Fig. 1. Representative images for (A) HNM, (B) HAI, and KBK (C) goat breeds included in the study.

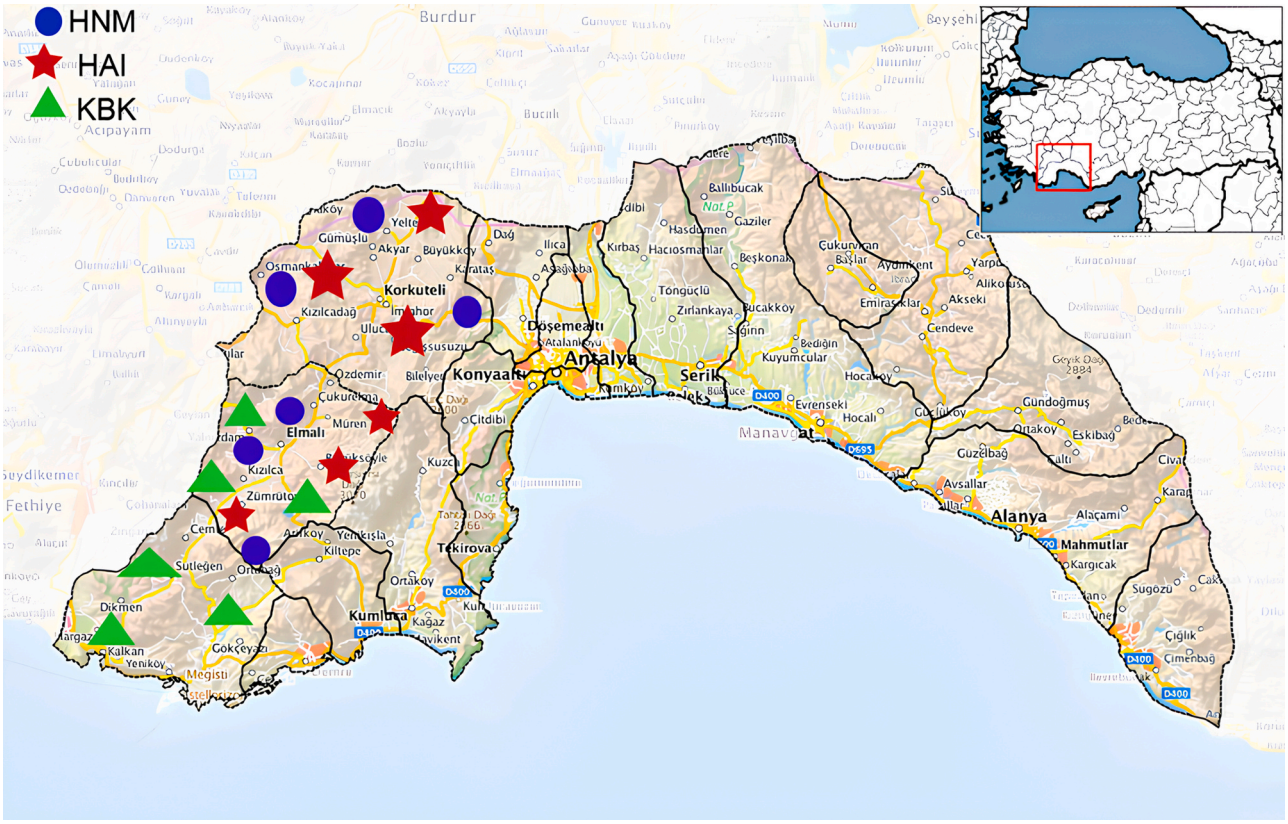

Fig. 2. A geographical representation of sampling strategy.

bi-allelic SNPs meeting the pre-set quality criteria were retained, which included a read depth between 20 and 500 ($20 \leq D \leq 500$) and a base quality score of $Q \geq 20$. Insertions, deletions, and SNPs located on non-autosomal chromosomes were excluded to refine the dataset to include only reliable genetic variants. Further filtering of the detected SNPs was performed using PLINK 1.9 (Chang et al., 2015) to ensure the inclusion of only high-quality variants. Filtering criteria included a minor allele frequency (MAF $\geq$ 0.01), marker missingness (–geno $\leq$ 0.1), and individual missingness (–mind $\leq$ 0.1), ensuring the retention of informative SNPs suitable for follow-up analysis of genetic variability and population structure in the HAI, HNM, and KBK goat breeds.

2.4. Bioinformatics analysis

Genetic variability was assessed through parameters including MAF, nucleotide diversity (π), observed heterozygosity (H_O), expected heterozygosity (H_E), and inbreeding coefficient (F_{IS}), calculated using VCFtools (Danecek et al., 2011). Pairwise genetic differentiation (F_{ST}) values were also computed with VCFtools (Danecek et al., 2011). Phylogenetic relationships were inferred by constructing an unweighted pair-group method with arithmetic average (UPGMA) dendrogram in the R environment (https://www.r-project.org) using the *dendextend* package (Galili, 2015) based on pairwise F_{ST} values. A heatmap based on genetic distance was generated using the *pheatmap* package (Kolde and Kolde, 2015) in the R environment (https://www.r-project.org). Principal component analysis (PCA) was performed using PLINK 1.9 (Chang et al., 2015), and the first two principal components were visualized using the *ggplot2* package (Wickham et al., 2016) in the R environment (https://www.r-project.org).

Effective population size (*Ne*) was estimated using SNeP v1.1 (Barbato et al., 2015) by analyzing linkage disequilibrium (LD) patterns between SNPs. LD was calculated as Pearson's squared correlation coefficient (r^2) for SNP pairs within a minimum distance of 10 kilobases (kb) and a maximum distance of 500 kb. A minor allele frequency (MAF) threshold of 0.05 was applied to exclude low-frequency alleles (Sved et al., 2008). *Ne* was estimated using the equation given below

$$E(r^2) = (1 + 4Nec)^{-1},$$

where *Ne* denotes effective population size and *c* represents the

recombination rate. Following the estimation of effective population size, population structure was analyzed using the ADMIXTURE v1.3.0 program (Alexander et al., 2009). Ancestral populations were evaluated for K values ranging from 2 to 6 while the optimal number of ancestral populations was identified via cross-entropy criterion.

3. Results

3.1. Sequencing data and SNP calling

Approximately 8.68 billion (8,680,962,327) short reads at 150 bp length were obtained using the Illumina HiSeq X Ten platform. Following the demultiplexing step, the average number of clean reads per individual was 241 million, ranging from 550.52 million (KBK) to 855.57 million (HAI) across the studied breeds. A significant proportion of the clean reads (82.33–98.12 %) aligned successfully with the goat reference genome. The highest number of aligned reads (855.57 million) and the highest alignment rate (98.12 %) were observed in the HNM breed. The variant calling process identified 309,342 high-quality bi-allelic SNPs. After applying filtering criteria and data curation steps, 481 individuals belonging to HAI (n = 156), HNM (n = 168), and KBK (n = 157) were retained for downstream analyses.

3.2. Estimation of genetic diversity

Genetic diversity parameters, including MAF, nucleotide diversity (π), H_O, H_E, and F_{IS} were calculated for HAI, HNM, and KBK populations (Table 1). The MAF values were consistent across the populations, with HAI and HNM exhibiting a mean of 0.322 and KBK slightly lower at 0.320. Nucleotide diversity showed minimal variation, ranging from 0.278 in KBK to 0.281 in HAI. H_O and H_E values were similar among the populations, with HAI exhibiting both metrics at 0.307, while HNM showed H_O at 0.305 and H_E at 0.306. KBK had the lowest H_O (0.301) and H_E at 0.305. The F_{IS} values were low in all populations, with HAI at 0.003, HNM at 0.011, and KBK at 0.015, indicating limited deviation from Hardy-Weinberg equilibrium.

3.3. Genetic relationships and population structure

Based on autosomal SNPs, PCA was conducted to examine genetic variation among HAI, HNM, and KBK populations. The first two principal components (PCA1 and PCA2) explained 18.75 % and 12.13 % of the total genetic variation, respectively. The PCA plot revealed that KBK formed a distinct cluster, clearly separated from HAI and HNM populations. While some individuals from HNM clustered separately, the majority overlapped with HAI, indicating partial genetic similarity between these populations (Fig. 3a). Pairwise F_{ST} values provided further insights into genetic differentiation among the populations. The lowest genetic differentiation was observed between HAI and HNM (F_{ST} = 0.0072), indicating a close genetic relationship, whereas the differentiation between HAI and KBK (F_{ST} = 0.0134) was slightly higher. The highest genetic differentiation was found between HNM and KBK (F_{ST} = 0.0184), highlighting the genetic distinctiveness of KBK. The UPGMA dendrogram constructed using pairwise F_{ST} values further illustrated these genetic relationships, showing that KBK was genetically distinct from other breeds while HAI and HNM were more closely related, reflecting a shared genetic background (Fig. 3b).

ADMIXTURE analysis was performed to assess population structure, with K values ranging from 2 to 6. Cross-entropy error values identified K = 2 as the optimal number of genetic clusters, with the lowest cross-validation error observed at 0.3374 (Fig. 3c). At K = 2, KBK was significantly distinct from HAI and HNM, while HAI and HNM displayed genetic admixture. At higher K values (K = 3 and K = 4), additional sub-structuring emerged, with unique ancestry components identified in some individuals from HNM (Fig. 3d).

A heatmap was constructed to illustrate pairwise genetic distances among 481 individuals from HAI, HNM, and KBK goat populations. KBK individuals (green labels) formed a cohesive genetic cluster with predominantly low pairwise distances (blue regions), indicating genetic homogeneity within this breed. In contrast, genetic distances between KBK and both HAI and HNM individuals were substantially higher (yellow to red regions), confirming the genetic distinctiveness of KBK. HAI and HNM individuals showed intermediate genetic distances with each other, consistent with their closer phylogenetic relationship observed in the UPGMA analysis. The diagonal pattern of low genetic distances (dark blue) represents self-comparisons, while the off-diagonal patterns reveal the hierarchical genetic structure among the three breeds (Fig. 4).

Table 1
Genetic diversity metrics for the studied populations.

Population	*MAF*	*π*	H_O	H_E	F_{IS}
HAI	0.322	0.281	0.307	0.307	0.003
HNM	0.322	0.280	0.305	0.306	0.011
KBK	0.320	0.278	0.301	0.305	0.015

MAF=Minor allele frequency; **π**=Nucleotide diversity; $\mathbf{H_O}$=Observed heterozygosity; $\mathbf{H_E}$=Expected heterozygosity; **FIS**=Inbreeding coefficient.

3.4. Effective population size and LD decay

The effective population size (*Ne*) was estimated over the past 1000 generations using LD-based methods. The *Ne* trends revealed consistent declines across all populations, reflecting a reduction in genetic variation over time. Among the studied populations, HAI exhibited the largest *Ne* values across generations, followed by HNM and KBK. At 400 generations ago, *Ne* was estimated to be approximately 11,250 for HAI, 8750 for HNM, and 9250 for KBK. This trend persisted in more recent generations, with HAI maintaining the largest *Ne* values, indicating distinct historical population dynamics (Fig. 5a).

The extent of LD was assessed for HAI, HNM, and KBK populations using pairwise r^2 values over SNP distances ranging from 0 bp to 1 Mb. The average LD (r^2) values showed a decreasing trend as the physical distance between SNPs increased. Among the populations, HAI exhibited the fastest LD decay, with consistently lower r^2 values across all distances, reflecting reduced levels of genetic linkage in this population. In general, HNM had slightly higher LD values than KBK across most distances; however, at approximately 1 Mb, KBK surpassed HNM, indicating a slower decay rate at longer distances (Fig. 5b).

4. Discussion

Although no previous studies have focused on genetic diversity in native Turkish goat breeds using SNP chips or NGS-based approaches, microsatellite-based studies have provided valuable insights into their genetic relationships. However, it is noteworthy that their resolution is limited compared to the approximately 310,000 high-quality SNPs generated in this study. Using 20 microsatellite loci, (Ağaoğlu and Ertuğrul, 2012) reported that HNM and HAI goats exhibited the closest genetic relationship among the breeds, with a low genetic distance (0.0587) which was explained by the geographic proximity of the rearing zones of these breeds. Similarly, Bulut et al. (2016) observed a genetic relationship between these two breeds, as indicated by a minimal level of genetic differentiation (FST = 0.005), supporting the possibility of gene flow due to overlapping geographical distributions. Karsli et al. (2020) carried out research derived from a genetic marker set of 20 microsatellites and reported low genetic divergence between HNM and HAI goats (FST = 0.012), whereas the differentiation between KBK and HAI goats (FST = 0.040), and between KBK and HNM goats (FST = 0.013), was slightly higher. In the present study, based on SNP data, the genetic differentiation between KBK and HAI goats was revealed to be relatively low (FST = 0.0134), though slightly higher

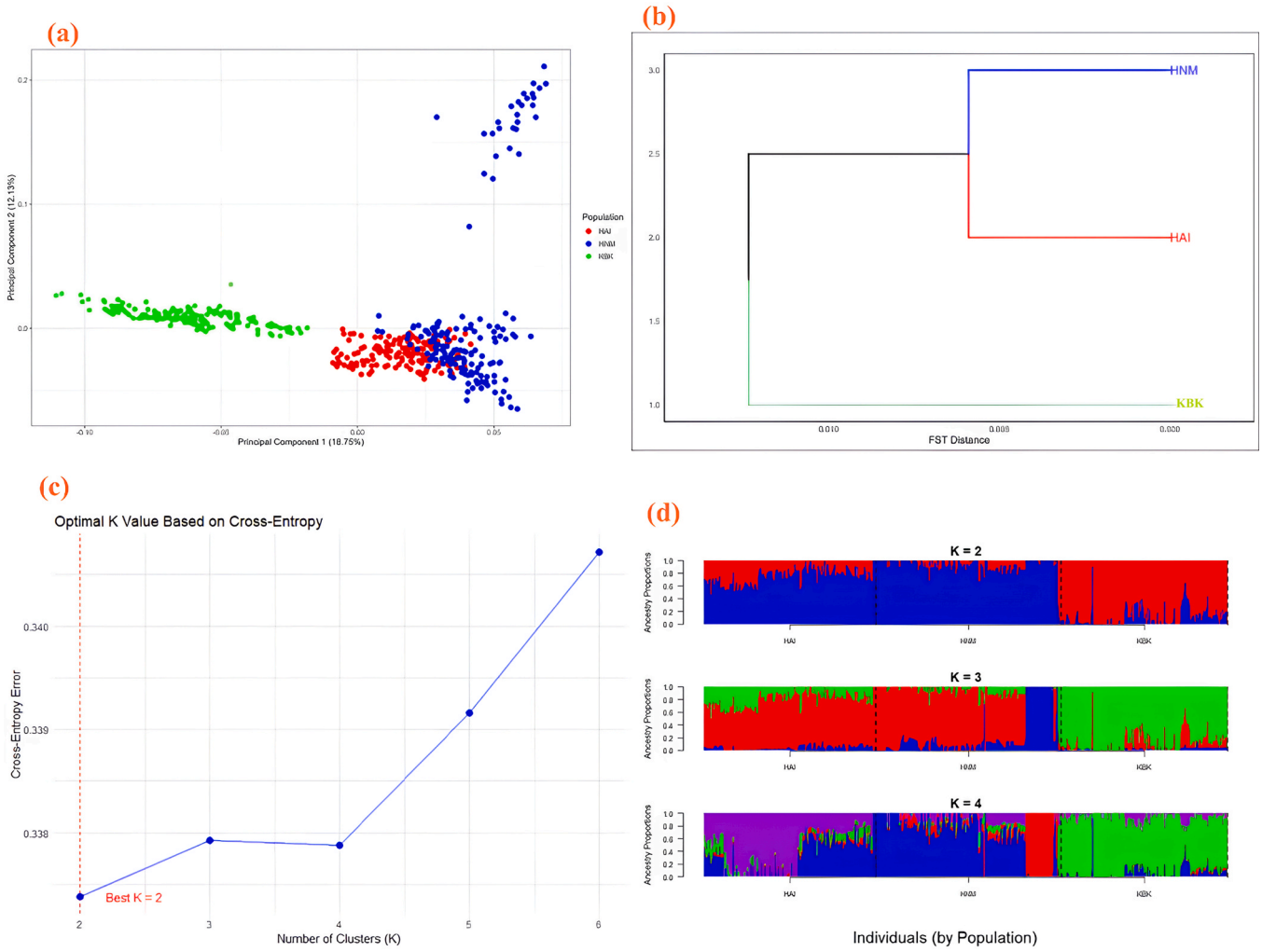

Fig. 3. Genetic relationships and population structure among goat populations. (a)PCA plot showing genetic variation, with PCA1 and PCA2 explaining 18.75 % and 12.13 % of the total variation, respectively. (b) UPGMA dendrogram based on pairwise FST values illustrating genetic differentiation among the populations. (c) Cross-entropy error values from ADMIXTURE analysis indicating K = 2 as the optimal number of genetic clusters. (d) ADMIXTURE analysis results for K = 2, K = 3, and K = 4 showing ancestry proportions for individuals in HAI, HNM, and KBK populations.

differentiation was observed between KBK and HNM goats (FST = 0.0184). These findings emphasize the improved resolution provided by the SNP dataset, which enables the detection of subtle genetic structures that microsatellite markers might not capture. Additionally, Karsli et al. (2020) noted that KBK goats, historically considered a variety of HAI goats, clustered more closely with Norduz which is a different Turkish local goat. However, the SNP-based analyses in this study identified KBK goats as a distinct genetic cluster, separate from both HNM and HAI goats. These results underline the value of high-resolution SNP data in characterizing genetic differentiation with greater precision and highlight the importance of treating KBK goats as a distinct genetic resource in conservation strategies.

In this study, the MAF values for HAI, HNM, and KBK were relatively uniform, ranging from 0.320 to 0.322, aligning with the 0.320 reported by Chokoe et al. (2020) in communal South African goats. This range is higher than the 0.154 observed in Hainan black goats by An et al. (2024) and the 0.253 recorded for Inner Mongolia Cashmere goats as noted by Wang et al. (2024). H_O was 0.307 for HAI, 0.305 for HNM, and 0.301 for KBK, which are slightly lower than Inner Mongolia Cashmere goats (0.355) (Wang et al., 2024) and Greek goats (0.395) (Michailidou et al., 2019), but higher than Moroccan domestic goats (0.223) (Benjelloun et al., 2019) and Hainan black goats (0.20) (An et al., 2024). The H_E values observed in this study were 0.307, 0.306, and 0.305 for HAI, HNM, and KBK, respectively. The values were lower than 0.412 reported for Iranian goats (Taheri et al., 2022) and 0.351 for Inner Mongolia Cashmere goats (Wang et al., 2024). In contrast, higher values were observed compared to 0.220 recorded for Hainan black goats (An et al., 2024) and 0.223 for Moroccan domestic goats (Benjelloun et al., 2019). The lowest F_{ST} value, 0.0072, was observed between HAI and HNM, showing a close genetic relationship. This result aligns with findings in Ethiopian Aberegalle and Central Highlands goats, where F_{ST} was reported as 0.003 by Gebreselase et al. (2024). In contrast, the highest value, 0.0184, was observed between HNM and KBK, suggesting greater genetic differentiation. This level of differentiation is comparable to that observed in Greek goat breeds, where F_{ST} was 0.0436, as noted by Michailidou et al. (2019).

The population structure analyses, including PCA, ADMIXTURE, and UPGMA dendrogram, revealed distinct genetic clusters among HAI, HNM, and KBK goat populations. PCA results demonstrated a clear separation between KBK and the other populations, while HAI and HNM exhibited partial overlap, suggesting closer genetic relationships. The first two principal components (PC1 and PC2) contributed 18.75 % and 12.13 % of the total genetic variance, respectively. ADMIXTURE analysis confirmed these findings, with K = 2 offering the best fit and

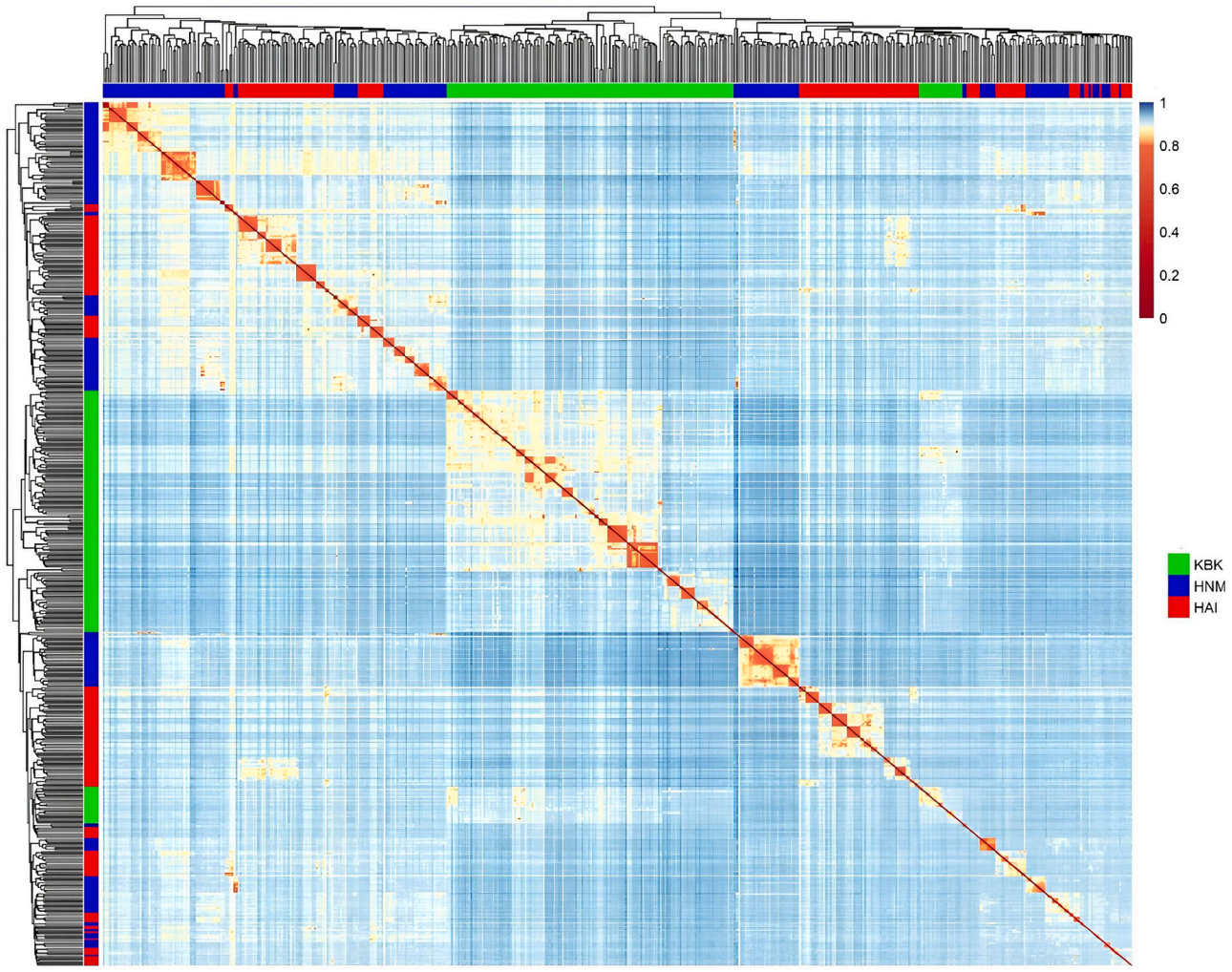

Fig. 4. Heatmap demonstrating pairwise genetic distances among individuals from three goat populations.

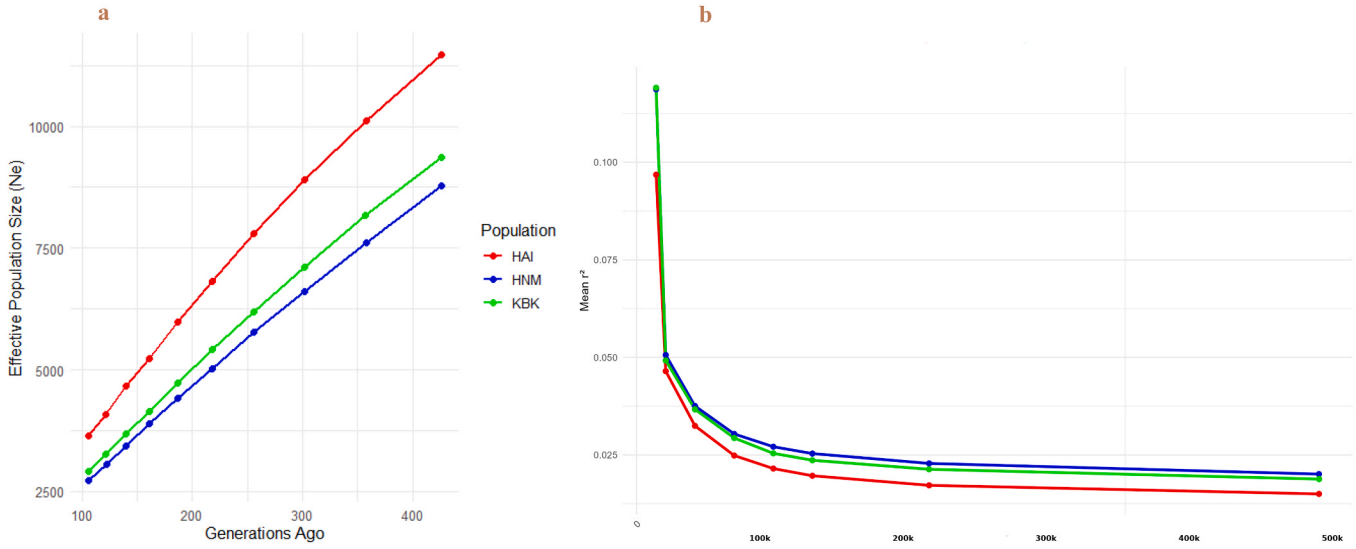

Fig. 5. LD decay and effective population size (*Ne*) for HAI, HNM, and KBK populations. (a) Effective population size (Ne) across the past 1000 generations. (b) LD decay measured as r^2 over increasing SNP distances.

demonstrating significant genetic differentiation between KBK and the other two populations. At K = 3 and K = 4, some samples belonging to HNM were assigned to another cluster, which is relevant to past breeding practices. Indeed, HNM and HAI were subjected to nonsystematic crossbreeding in the past, while KBK has been conserved by smallholder farmers via pure breeding. This information is also supported by the findings of F_{ST}-based dendrogram analysis, in which the lowest (0.0072) and highest (0.0184) values were calculated between HAI-HNM and KBK-HNM, respectively. Population structure analyses indicate that, despite being defined as a subtype of HAI, KBK goats have undergone genetic differentiation from both HAI and HNM populations. This differentiation observed in KBK goats may reflect the influence of localized breeding practices in relatively distinct zones, where breeder selection has maintained genetic separation through the years. Over time, this process may have facilitated local adaptation, further distinguishing KBK from HAI and HNM populations. It is noteworthy that animals were sampled from a narrow breeding site covering Kaş, Elmalı, and Korkuteli districts of Antalya, in which KBK is reared in mountainous areas of Kaş with high altitudes. However, the geographic distribution of this population could reach beyond Elmalı, especially during the summer season, leading to no clear geographic isolation between the studied goat populations. Compared to HNM and HAI breeds, which are reared in almost all districts of Antalya, KBK are kept by smallholder farmers in a limited geographic region without crossbreeding practices due to possessing unique characteristics such as walking ability over long distances. This breeding preference is believed to be the main reason for the genetic distinctiveness of KBK from HAI and HNM breeds rather than geographic distribution. In contrast, HNM was subjected to crossbreeding practices with HAI to improve the body weight trait.

Ne estimates over the past 400 generations depicted a gradual decline in all three populations, with HAI maintaining the largest *Ne* across time. This trend implies that HAI has a more stable population history, whereas smaller *Ne* in HNM and KBK populations points to stronger genetic drift and potential bottleneck events. This kind of declining trend in effective population size was also reported in several native small ruminant herds reared in Türkiye (Demir, 2024; Karsli, 2024). Comparable patterns have been reported in Ethiopian goat breeds, where Ne trends reflected historical demographic pressures (Gebreselase et al., 2024). The declining trend in effective population size in the studied goat populations is not surprising since goat breeding in Antalya was practiced in a nomadic style in the past with a higher population size. As of urbanization, negatively destroying breeding zones, the number of herds kept by smallholder farmers has reduced from one generation to another.

LD decay analyses revealed significant differences among populations. HAI exhibited the fastest LD decay, indicative of higher genetic diversity and less historical inbreeding. In contrast, KBK and HNM showed slower LD decay, reflecting more extensive genetic linkage and possibly reduced genetic variation due to geographic or demographic constraints. These results are consistent with findings in other livestock species, where slower LD decay is linked to smaller population sizes or limited gene flow Chokoe et al. (2020).

The observed *Ne* decline likely reflects domestication bottlenecks, as goats originate from a limited number of bezoars domesticated approximately 10,000 years ago (Amills et al., 2017), restricted gene flow in geographically isolated breeding zones, and crossbreeding with cosmopolitan breeds threatening local genetic resources (Ajmone-Marsan et al., 2014). Historical *Ne* studies confirm declining trends in goat populations due to long-term selection pressures (Berihulay et al., 2019). For KBK, joint conservation programs may have unintentionally reduced genetic distinctiveness through crossbreeding.

5. Conclusion

In this study, genetic diversity and population structure of three Anatolian goat populations were assessed for the first time by high-density SNP data obtained from the ddRADseq technique. A sufficient genomic variability and low inbreeding values were identified in all populations, which is promising for sustainable animal production in the future. Besides, the pattern of the past crossbreeding practices between HAI and HNM was validated by high-throughput genetic data. Population structure analyses demonstrated that KBK has become genetically different from HAI and HNM. This finding is expected to make a significant contribution to the recognition of KBK as a breed and to reshaping conservation programs. Further studies may focus on identifying genetic variants in KBK via genome-wide association analysis to design comprehensive selection programs for diverse purposes, including adaptability and economically important traits. Still, in this study, some samples belonging to HNM were genetically different from HAI samples. Future studies could also utilize a better sampling strategy to obtain deeper knowledge of genetic structure in HNM breeds.

CRediT authorship contribution statement

Kaya Sarp: Software, Investigation, Data curation. **Meydan Hasan:** Writing – review & editing, Resources, Investigation. **Yağci Serdar:** Resources. **Karacaören Burak:** Writing – review & editing, Formal analysis. **Karslı Taki:** Writing – review & editing, Writing – original draft, Supervision, Project administration, Methodology, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Bilginer Umit:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Demir Eymen:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation, Conceptualization. **Argun Karsli Bahar:** Methodology, Investigation. **Doğru Huriye:** Methodology, Investigation.

Ethical approval

Blood sample collection from animals for genetic studies was approved by the Local Ethics Committee of Animal Experiments at Eskişehir Osmangazi University (Protocol No: 1419/2002.03.001). All procedures complied with the ethical guidelines of the committee and adhered to ARRIVE guidelines. Blood sampling was performed exclusively by qualified veterinarians without the use of chemical agents.

Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the author(s) used ChatGPT (OpenAI) to improve the readability and language of the manuscript. After using this tool, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the published article.

Funding

This study was financially supported by The Scientific and Technological Research Council of Türkiye (TÜBİTAK) under Project No: 122O019.

Declaration of Competing Interest

No conflict of interest.

Acknowledgments

This study was conducted using goats from three sub-projects carried out in Antalya under the framework of national and conservation programs, coordinated by the General Directorate of Agricultural Research and Policies (TAGEM) under the Turkish Ministry of Agriculture and Forestry. We extend our sincere gratitude to TAGEM for their support.

We also express our appreciation to the Antalya Sheep and Goat Breeders' Association, including its management and technical staff, for their invaluable assistance in collecting phenotypic data and blood samples. Additionally, the numerical calculations presented in this paper were fully/partially performed using the High-Performance and Grid Computing Center (TRUBA resources) at TÜBİTAK ULAKBİM.

Data availability

The datasets analyzed during the current study are available for academic purposes upon signing a Material Transfer Agreement with the corresponding author (takikarsli@ogu.edu.tr).

References

Ağaoğlu, Ö.K., Ertuğrul, O., 2012. Assessment of genetic diversity, genetic relationship and bottleneck using microsatellites in some native Turkish goat breeds. Small Rumin. Res. 105, 53–60. https://doi.org/10.1016/j.smallrumres.2011.12.005.

Ajmone-Marsan, P., Colli, L., Han, J.L., Achilli, A., Lancioni, H., Joost, S., Globaldiv, Consortium, 2014. The characterization of goat genetic diversity: towards a genomic approach. Small Rumin. Res. 121 (1), 58–72. https://doi.org/10.1016/j.smallrumres.2014.06.010.

Amills, M., Capote, J., Tosser-Klopp, G., 2017. Goat domestication and breeding: a jigsaw of historical, biological and molecular data with missing pieces. Anim. Genet. 48 (6), 631–644. https://doi.org/10.1111/age.12598.

Berihulay, H., Islam, R., Jiang, L., Ma, Y., 2019. Genome-wide linkage disequilibrium and the extent of effective population sizes in six Chinese goat populations using a 50K single nucleotide polymorphism panel. Animals 9 (6), 350. https://doi.org/10.3390/ani9060350.

Aktaş, Y., Atmaca, V., Demir, E., Karslı, T., 2024. Assessment of InDel variations in CPT1A and PRDM6 genes across three Anatolian goat populations. J. Agric. Fac. Gaziosmanpaşa Univ. 41, 265–270. https://doi.org/10.55507/gopzfd.1568134.

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

An, Z.X., Shi, L.G., Hou, G.Y., Zhou, H.L., Xun, W.J., 2024. Genetic diversity and selection signatures in Hainan Black goats revealed by whole-genome sequencing data. Animal 18, 101147. https://doi.org/10.1016/j.animal.2024.101147.

Anonymous, 2023. Turkish Statistical Institute, Statistical Tables, Number of Animals by Type and Race, Number of Sheep and Goat. Available from: ⟨https://data.tuik.gov.tr/Kategori/GetKategori?p=tarim-111&dil=1⟩ (Accessed 15 February 2025).

Barbato, M., Orozco-TerWengel, P., Tapio, M., Bruford, M.W., 2015. SNeP: a tool to estimate trends in recent effective population size trajectories using genome-wide SNP data. Front. Genet. 6, 109. https://doi.org/10.3389/fgene.2015.00109.

Benjelloun, B., Boyer, F., Streeter, I., Zamani, W., Engelen, S., Alberti, A., Alberto, F.J., BenBati, M., Ibnelbachyr, M., Chentouf, M., Bechchari, A., Rezaei, H.R., Naderi, S., Stella, A., Chikhi, A., Clarke, L., Kijas, J., Flicek, P., Taberlet, P., Pompanon, F., 2019. An evaluation of sequencing coverage and genotyping strategies to assess neutral and adaptive diversity. Mol. Ecol. Resour. 19, 1497–1515. https://doi.org/10.1111/1755-0998.13070.

Brito, L.F., Jafarikia, M., Grossi, D.A., Kijas, J.W., Porto-Neto, L.R., Ventura, R.V., Salgorzaei, M., Schenkel, F.S., 2015. Characterization of linkage disequilibrium, consistency of gametic phase and admixture in Australian and Canadian goats. BMC Genet. 16, 1–15. https://doi.org/10.1186/s12863-015-0203-0.

Bulut, Z., Kurar, E., Ozsensoy, Y., Altunok, V., Nizamlioglu, M., 2016. Genetic diversity of eight domestic goat populations raised in Turkey. BioMed Res. Int. 2016, 2830394. https://doi.org/10.1155/2016/2830394.

Chang, C.C., Chow, C.C., Tellier, L.C.A.M., Vattikuti, S., Purcell, S.M., Lee, J.J., 2015. Second-generation PLINK: rising to the challenge of larger and richer datasets. Gigascience 4, s13742–015. https://doi.org/10.1186/s13742-015-0047-8.

Chen, S., Zhou, Y., Chen, Y., Gu, J., 2018. Fastp: an ultra-fast all-in-one FASTQ preprocessor. Bioinformatics 34, i884–i890. https://doi.org/10.1093/bioinformatics/bty560.

Chen, Y., Li, R., Sun, J., Li, C., Xiao, H., Chen, S., 2022. Genome-wide population structure and selection signatures of Yunling goat based on RAD-Seq. Animals 12, 2401. https://doi.org/10.3390/ani12182401.

Chokoe, T.C., Mdladla-Hadebe, K., Muchadeyi, F., Dzomba, E., Matelele, T., Mphahlele, T., Mpofu, T.J., Nephawe, K., Mtileni, B., 2020. Genetic diversity of South African indigenous goat population from four provinces using genome-wide SNP data. Sustainability 12, 10361. https://doi.org/10.3390/su122410361.

Danecek, P., Auton, A., Abecasis, G., Albers, C.A., Banks, E., DePristo, M.A., Handsaker, R.E., Lunter, G., Marth, G.T., Sherry, S.T., McVean, G., Durbin, R., 1000 Genomes Project Analysis Group, 2011. The variant call format and VCFtools. Bioinformatics 27, 2156–2158. https://doi.org/10.1093/bioinformatics/btr330.

Danecek, P., Bonfield, J.K., Liddle, J., Marshall, J., Ohan, V., Pollard, M.O., Whitwham, A., Keane, T., McCarthy, S.A., Davies, R.M., Li, H., 2021. Twelve years of SAMtools and BCFtools. Gigascience 10, giab008. https://doi.org/10.1093/gigascience/giab008.

Demir, E., 2024. Microsatellite-based bottleneck analysis and migration events among four native Turkish goat breeds. Arch. Anim. Breed. 67, 353–360. https://doi.org/10.5194/aab-67-353-2024.

Demir, E., Moravčíková, N., Kaya, S., Kasarda, R., Doğru, H., Bilginer, Ü., Balcioglu, M.S., Karsli, T., 2023. Genome-wide genetic variation and population structure of native and cosmopolitan cattle breeds reared in Türkiye. Anim. Biotechnol. 34, 3877–3886. https://doi.org/10.1080/10495398.2023.2235600.

Doublet, M., Degalez, F., Lagarrigue, S., Lagoutte, L., Gueret, E., Allais, S., Lecerf, F., 2024. Variant calling and genotyping accuracy of ddRAD-Seq: comparison with 20X WGS in layers. PLoS One 19, e0298565. https://doi.org/10.1371/journal.pone.0298565.

Galili, T., 2015. dendextend: an R package for visualizing, adjusting and comparing trees of hierarchical clustering. Bioinformatics 31, 3718–3720. https://doi.org/10.1093/bioinformatics/btv428.

Gebreselase, H.B., Ngussie, H., Wang, C., Luo, C., 2024. Genetic diversity, population structure and selection signature in Begait goats revealed by whole-genome sequencing. Animals 14, 307. https://doi.org/10.3390/ani14020307.

Karsli, B.A., 2024. Genetic diversity and population structure of four Anatolian sheep revealed by genome-wide ddRADseq data. Small Rumin. Res. 235, 107285. https://doi.org/10.1016/j.smallrumres.2024.107285.

Karsli, T., Demir, E., Fidan, H.G., Aslan, M., Argun Karsli, B., Arik, I.Z., Sahin Semerci, E., Karabag, K., Balcioglu, M.S., 2020. Determination of genetic variability, population structure and genetic differentiation of indigenous Turkish goat breeds based on SSR loci. Small Rumin. Res. 190, 106147. https://doi.org/10.1016/j.smallrumres.2020.106147.

Kolde, R., Kolde, M.R., 2015. Package 'Pheatmap. R Package, Version 1.7. ⟨https://cran.r-project.org/web/packages/pheatmap/⟩.

Li, H., Durbin, R., 2009. Fast and accurate short read alignment with Burrows–Wheeler transform. Bioinformatics 25, 1754–1760. https://doi.org/10.1093/bioinformatics/btp324.

Magris, G., Marroni, F., D'Agaro, E., Vischi, M., Chiabà, C., Scaglione, D., Kijas, J., Messina, M., Tibaldi, E., Morgante, M., 2022. ddRAD-Seq reveals the genetic structure and detects signals of selection in Italian Brown Trout. Genet. Sel. Evol. 54, 8. https://doi.org/10.1186/s12711-022-00698-7.

Michailidou, S., Tsangaris, G.T., Tzora, A., Skoufos, I., Banos, G., Argiriou, A., Arsenos, G., 2019. Analysis of genome-wide DNA arrays reveals the genomic population structure and diversity in autochthonous Greek goat breeds. PLoS One 14, e0226179. https://doi.org/10.1371/journal.pone.0226179.

Peterson, B.K., Weber, J.N., Kay, E.H., Fisher, H.S., Hoekstra, H.E., 2012. Double digest RADseq: an inexpensive method for de novo SNP discovery and genotyping in model and non-model species. PLoS One 7, e37135. https://doi.org/10.1371/journal.pone.0037135.

Rochette, N.C., Rivera-Colón, A.G., Catchen, J.M., 2019. Stacks 2: analytical methods for paired-end sequencing improve RADseq-based population genomics. Mol. Ecol. 28, 4737–4754. https://doi.org/10.1111/mec.15253.

Sved, J.A., McRae, A.F., Visscher, P.M., 2008. Divergence between human populations estimated from linkage disequilibrium. Am. J. Hum. Genet. 83, 737–743. https://doi.org/10.1016/j.ajhg.2008.10.019.

Taheri, S., Zerehdaran, S., Javadmanesh, A., 2022. Genetic diversity in some domestic and wild sheep and goats in Iran. Small Rumin. Res. 209, 106641. https://doi.org/10.1016/j.smallrumres.2022.106641.

Vani, A., Kumar, A., Mahala, S., Janga, S.C., Chauhan, A., Mehrotra, A., De, A.K., Sahu, A.R., Ahmad, S.F., Vempadapu, V., Dutt, T., 2024. Revelation of genetic diversity and genomic footprints of adaptation in Indian pig breeds. Gene 893, 147950. https://doi.org/10.1016/j.gene.2023.147950.

Wang, R., Wang, X., Qi, Y., Li, Y., Na, Q., Yuan, H., Rong, Y., Ao, X., Guo, F., Zhang, L., Liu, Y., Shang, F., Zhang, Y., Wang, Y., 2024. Genetic diversity analysis of Inner Mongolia Cashmere goats (Erlangshan subtype) based on whole genome re-sequencing. BMC Genom. 25, 698. https://doi.org/10.1186/s12864-024-10485-x.

Wickham, H., Chang, W., Wickham, M.H., 2016. ggplot2: Create Elegant Data Visualisations Using the Grammar of Graphics. R Package Version 2.1. ⟨https://ggplot2.tidyverse.org⟩.

Zeder, M.A., Hesse, B., 2000. The initial domestication of goats (Capra hircus) in the Zagros Mountains 10,000 years ago. Science 287, 2254–2257. https://doi.org/10.1126/science.287.5461.2254.