



Genome-wide assessment of local Greek goats' diversity and comparison with global domestic breeds



Antonios Otapasidis¹, Valentina Tsartsianidou^{1,2}, Nikoleta Karaiskou^{1,2}, Georgios Arsenos¹, Alexandros Triantafyllidis^{1,2}

¹Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece ²Genomics and Epigenomics Translational Research (GENeTres), Center for Interdisciplinary Research and Innovation (CIRI-AUTH), Balkan Center, 57001, Thessaloniki, Greece

Introduction

Goat farming sector in Greece is one of the largest in Europe with over 3.8 million domestic goats and a main producer of goat dairy products. The national goat herd consists of 2 breeds: the Eghoria and the Skopelos. The Eghoria breed is reared in the whole country and the population exceeds ~2 million individuals. The Skopelos goat inhabits the Sporades island complex, and is considered endangered due to the total available number of female goats (7,250, 2010) (1, 3). Inbreeding estimation and characterization of homozygosity/heterozygosity status of the national goat flock may contribute to the future genetic improvement schemes and the respective conservation practices in Greece. The purpose of this study is the characterization of the genome-wide structure, the inbreeding and genetic diversity levels of the Greek national goat breeds.

Materials and methods

Sampling: Eghoria = 70 individuals from 2 farms in the North Greece, Skopelos = 289 samples from 2 farms in the Sporades island complex and 784 individuals from 21 global goat breeds from the AdaptMap project (2).

Genome-wide genotyping: GoatSNP50 BeadChip with 53,347 SNPs

Genetic diversity: inbreeding/heterozygosity levels (F_{ROH} , F_{IS} , H_e , H_o)

Principal Component Analysis: Greek goat breeds population structure assessment and genetic relatedness with other 21 breeds worldwide

Signatures of selection: ROH/ROHet identification using detectRUNS

Annotation: Gene annotation, Functional annotation, Functional Enrichment analysis, Goat related path identification using the KEGG database and DAVID software

Results & Discussion

Population structure and genetic relatedness

- The Greek national herd is placed between the Asian, African and the European breeds (Fig 1). This PCA provides information on the goat's expansion process beginning from Asia with direction to Europe (3).
- The Greek breeds show closer genetic relatedness mostly with the Italian goats (Garganica, Maltese), the American Kiko and the African Toggenburg breed (Fig 1, red circle).
- The Greek goats are also closely depicted with other European breeds, such as the French goats (except the Pyrenean) the Landrace breed, the Old Irish goat (Fig 1, blue circle).

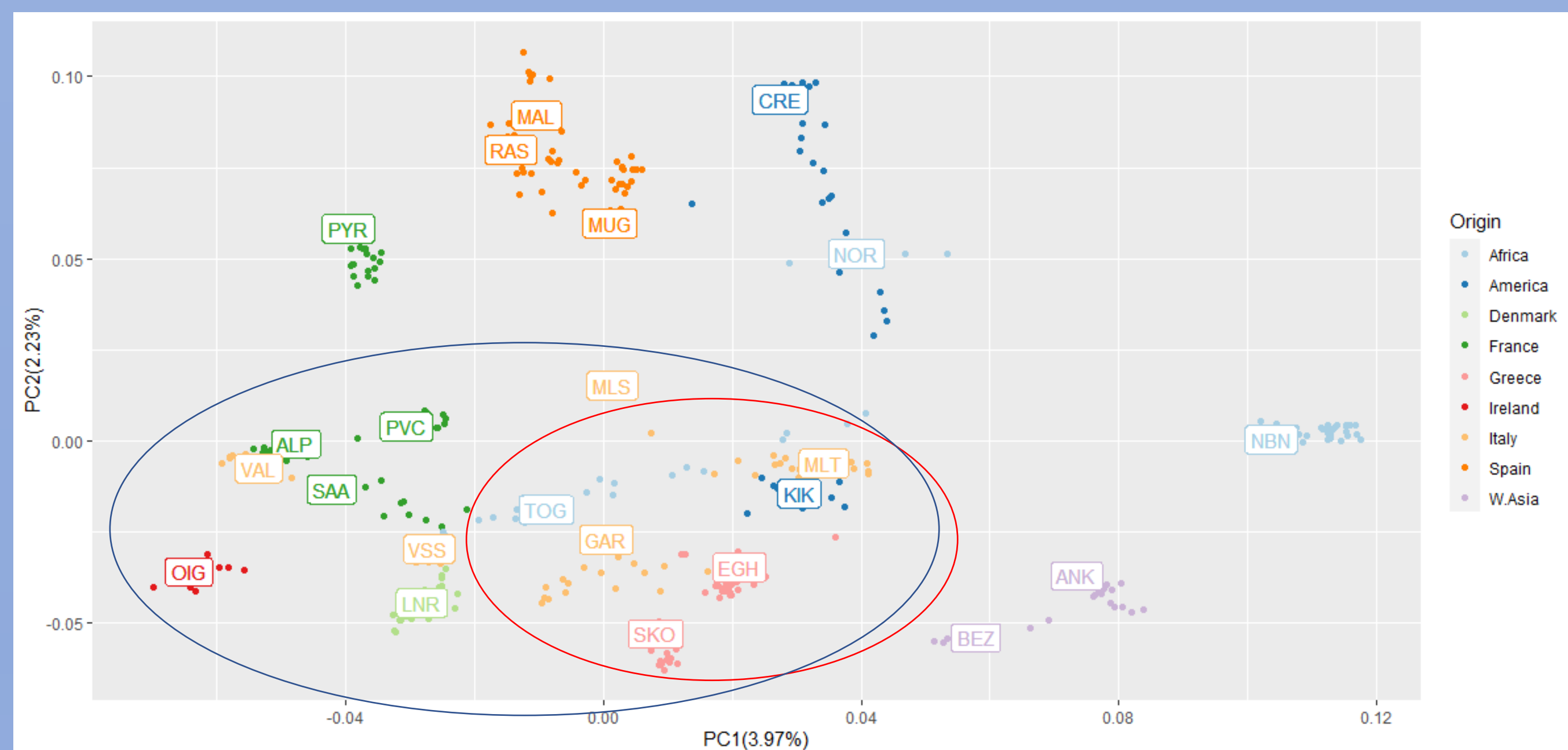


Fig 1. Principal Component analysis plot between the first and the second principal components (PC1 -PC2) for the sample of 496 goats from 23 breeds worldwide. EGH: Eghoria, SKO: Skopelos, ALP: Alpine (Camosciata), ANK: Ankara, BEZ: Bezoar, CRE: Creole, GAR: Garganica, KIK: Kiko, LNR: Landrace Goat, MAL: Mallorquina, MLS: Maltese Sarda, MLT: Maltese, MUG: Murciano - Granadina, NBN: Nubian, NOR: Nord, OIG: Old Irish Goat, PVC: Provencale, PYR: Pyrenean, RAS: Blanca de Rasquera, SAA: Saanen, TOG: Toggenburg, VAL: Valdostana, VSS: Valpessiera.

Genetic diversity of the Greek goat breeds

- High average levels of expected and observed heterozygosity and low inbreeding levels (F_{ROH} and F_{IS}) were estimated for Greek goat breeds (Table 1)

Table 1. Table illustrating the inbreeding index (F), inbreeding index based on regions of homozygosity (F_{ROH}), observed (H_o) and expected (H_e) heterozygosity in the Greek goat breeds, including standard error (SE) and standard deviation (sd).

Breed	F_{IS} (\pm SE)	F_{ROH}	H_e (\pm sd)	H_o (\pm sd)
Eghoria	0,0068 (\pm 0,0034)	0,02215	0,423 (\pm 0,088)	0,4203 (\pm 0,103)
Skopelos	0,01 (\pm 0,002)	0,022072	0,395 (\pm 0,114)	0,391 (\pm 0,116)

Runs of Heterozygosity

- ROH islands were not detected in the Greek goat breeds genome (Fig 2, Fig3). Instead, 6 Homozygosity rich regions in the Eghoria goat breed genome whereas 5 regions with relatively Homozygosity in the Skopelos breed, were identified.

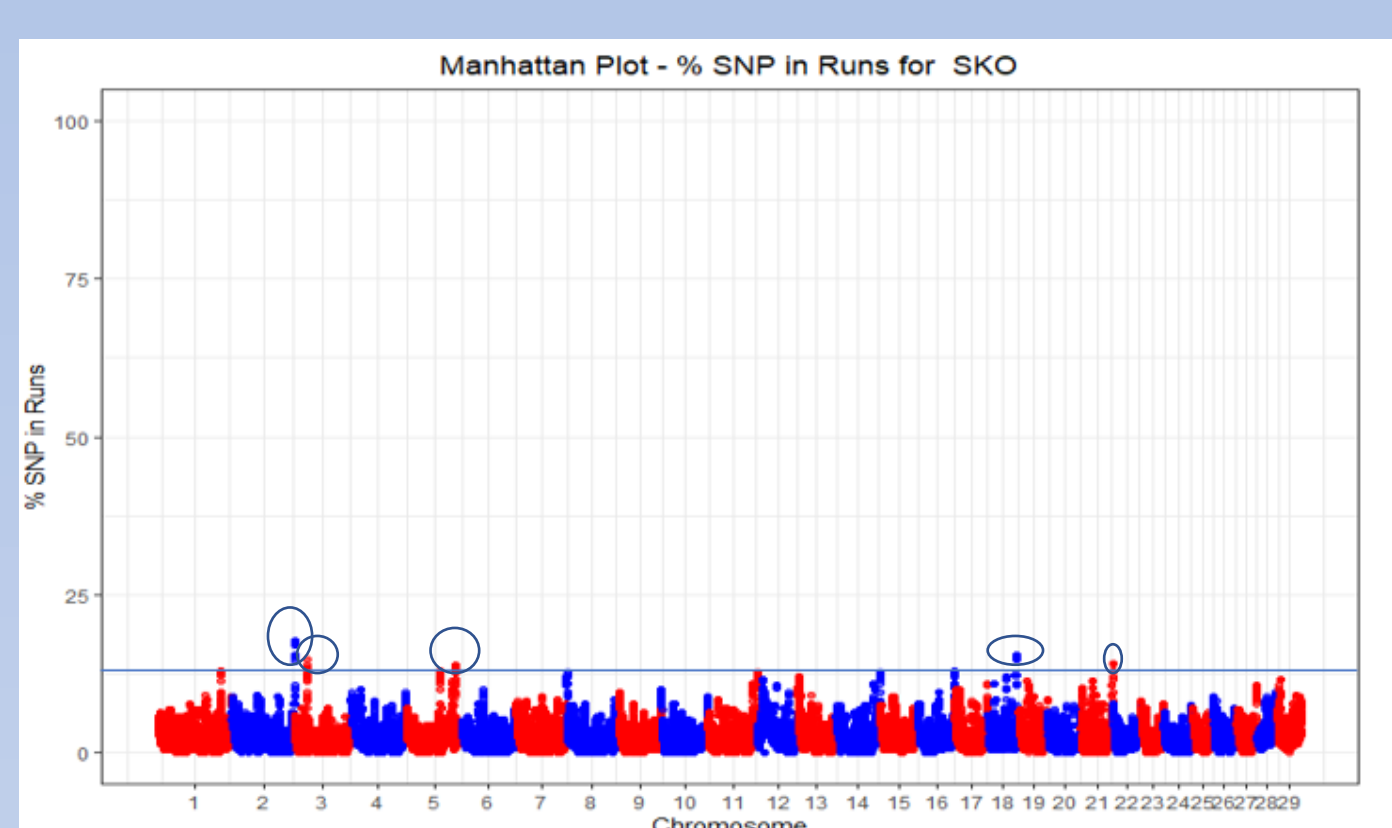


Fig 2. Manhattan plot showing the frequency of a SNP within homozygous segments in Skopelos goat breed. Runs of homozygosity (ROH). The blue line indicates the 99.9 quantile threshold, defining the ROH islands.

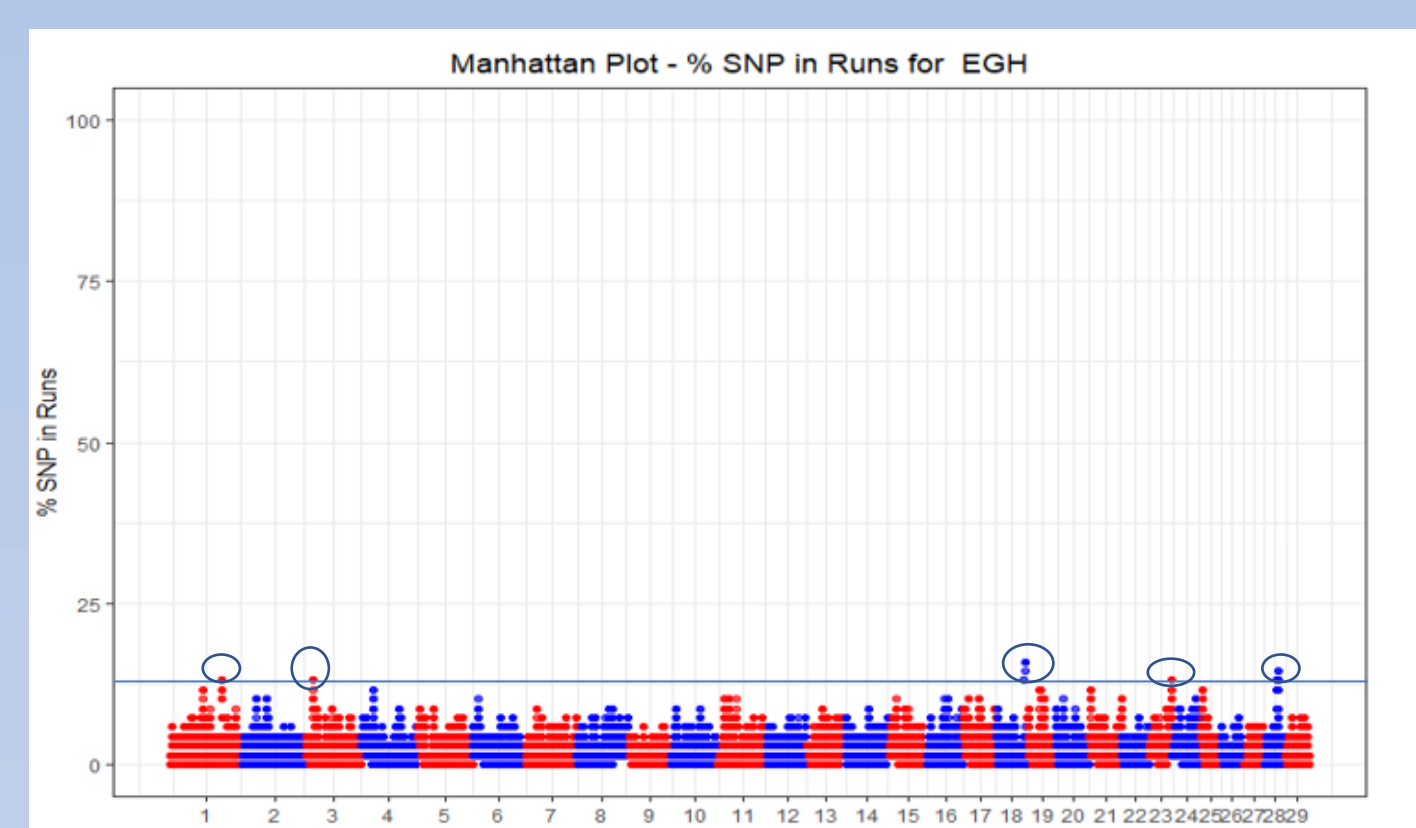


Fig 3. Manhattan plot showing the frequency of a SNP within homozygous segments in Eghoria goat breed. Runs of homozygosity (ROH). The blue line indicates the 99.9 quantile threshold, defining the respective ROH islands.

- A common ROH region (~0.7 Mb) in Greek goats on chromosome 18 (Table 2) associated with HSV-1 biological pathways (9 enriched genes for Eghoria & 7 enriched genes for Skopelos breed) was detected.

Table 2. Table illustrating the common ROH region between the Greek goat breeds.

Common ROH region	Chromosome	Region start (bp)	Region end (bp)	SNP number
Region A	18	60467072	61158064	8

Runs of Heterozygosity

- Runs of Heterozygosity Islands in the genome of both the Eghoria and Skopelos goat breed were discovered (Fig 3, Fig 4).

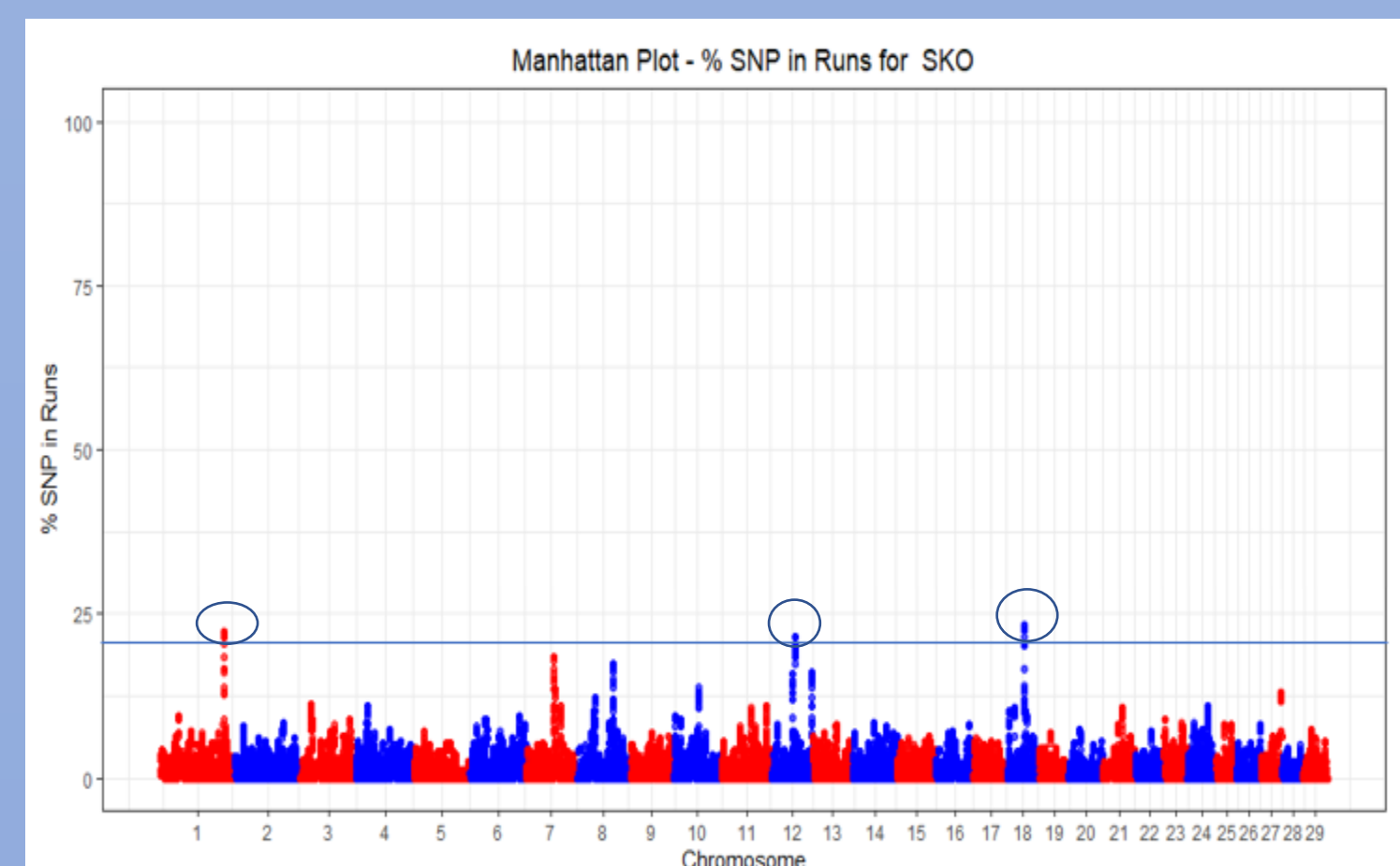


Fig 3. Manhattan plots showing the frequency of a SNP within heterozygous segments in Skopelos goat breed. Runs of heterozygosity (ROHet). The blue line indicates the 99.9 quantile threshold, defining the ROHet islands.

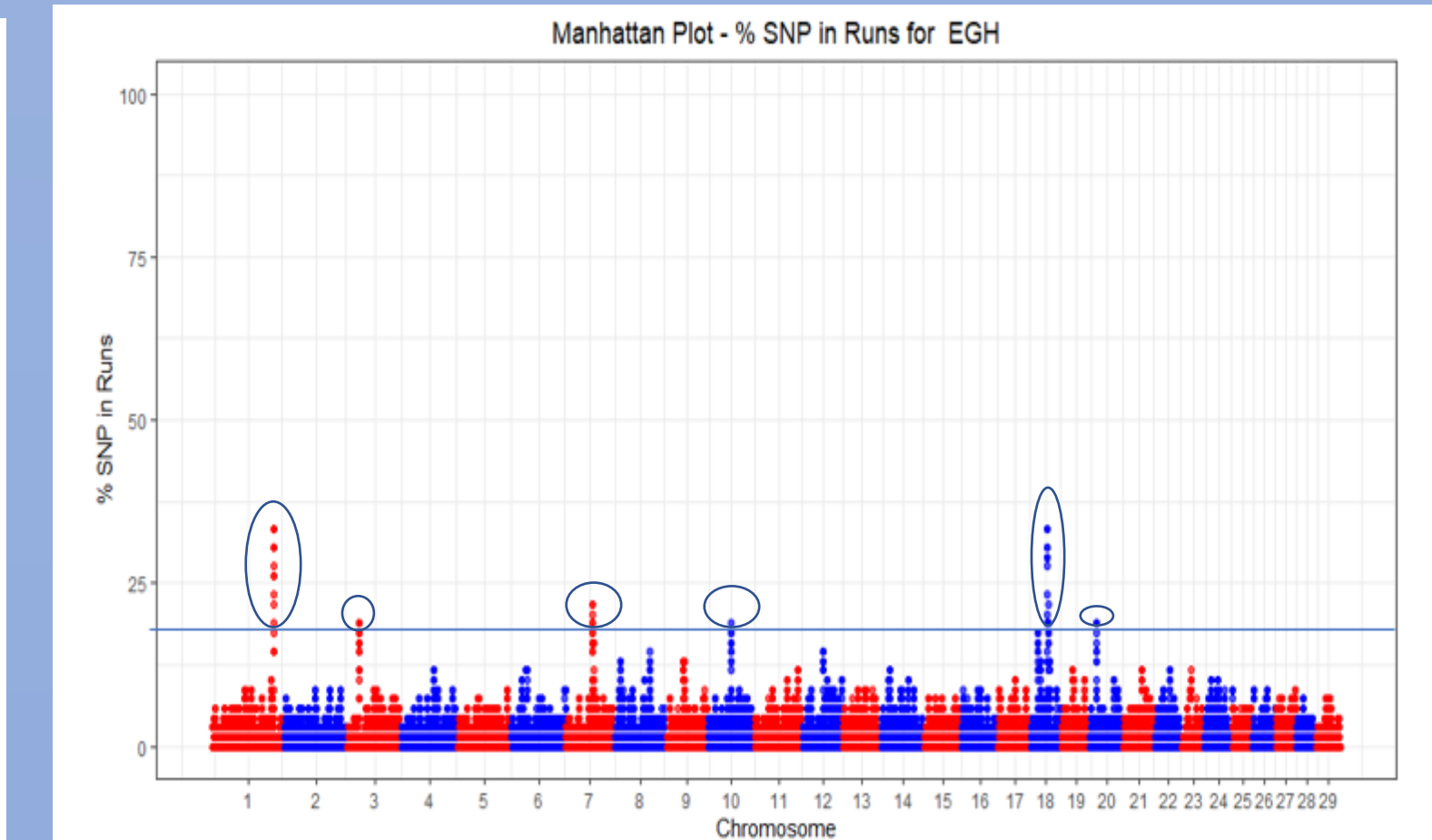


Fig 4. Manhattan plots showing the frequency of a SNP within heterozygous segments in Eghoria goat breed. Runs of heterozygosity (ROHet). The blue line indicates the 99.9 quantile threshold, defining the ROHet islands.

- 4 ROHet islands were identified for Skopelos and 6 for Eghoria goat (Table 3, Table 4).

Table 3. Table illustrating the homozygosity rich regions in the Eghoria goat breed.

Breed	Island	Chromosome	Island start (bp)	Island end (bp)	SNP number
Eghoria	Island A	18	36283603	37295931	18
	Island B	18	39636695	39926799	7
	Island C	1	131931194	132736269	17
	Island D	20	13847556	14250543	9
	Island E	3	24908534	25076519	5
	Island F	7	60015530	60137051	5

Table 4. Table illustrating the homozygosity rich regions in the Eghoria goat breed.

Breed	Island	Chromosome	Island start (bp)	Island end (bp)	SNP number
Skopelos	Island A	12	49884737	50328003	10
	Island B	12	50451655	50631677	5
	Island C	18	36360921	37006578	12
	Island D	1	131970293	132501737	11

- 2 common ROHet regions in Greek goats located on chromosomes i) 1 (~0,65 Mb) with 2 candidate genes associated to the embryonic development (STAG1, PCCB) (4) and ii) 18 (~0,5 Mb) with 34 candidate genes from which 2 genes were enriched (DPEP2, DPEP3).

Gene and functional annotation

- 80 and 103 genes candidate genes were identified for the Eghoria & the Skopelos breed respectively, located at the ROHet islands
- 41 and 39 candidate genes were found for the Eghoria & the Skopelos breed respectively, located at the Homozygosity rich regions.
- The functional annotation analysis revealed several biological pathways related to metabolism, immune response and response to the arid and semi arid environment (6).

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