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Poster session A

Monday, August 10th 1740-1940

Génopode 2000

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Symposium 28. Variation in natural selection: patterns, causes, and consequences

Searching for signatures of selection in Iberian honey bee (*Apis mellifera iberiensis*) using whole genome sequences

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Introduction

The Iberian Peninsula comprises a diverse set of habitats. It was an important glacial refugium during the Pleistocene and has served as a bridge for populations migrating between Africa and Europe, resulting in a complex mix of ancestry and diversity. The Iberian honey bee (*A. m. iberiensis)* is no exception and has been the subject of numerous incongruent population genetic surveys. Recent mtDNA and SNP analyses indicate a steep northeastern-southwestern cline of African-European ancestry along the peninsula, which has been explained by selection and demo.

Advances in DNA sequencing technology and computational tools provide unprecedented opportunities to study demography, search for signatures of selection across the genome and illuminate its role in shaping genomic diversity. We used Illumina technology to sequence the whole genomes of 86 Iberian honeybees. To verify whether there are signals of genetic adaptation we have done simple pairwise comparisons between populations located in extremes of environmental variables, like precipitation and temperature (Fig.1).

Methods

Genome sequencing

Illumina technology was used to sequence the whole genomes of 86 Iberian honeybees collected across three north-south transects in the Iberian Peninsula (Fig.1).
The reads were mapped against the reference genome Amel_4.5 using BWA.

Several steps of quality control were done:

- Identify and marked PCR duplicates: Picard;
- Identify regions of poorly and inconsistently mapped reads: GATK45;
- SNP calling: FreeBayes.

In order to reduce the number of false SNPs, we removed the following SNPs:

- Quality below 50;
- Depth below 123 or higher then 3000;
- With less then 37 alleles.

- In total the dataset is composed by 1289449 SNPs.

The environmental data were obtained from the Climatic Research Unit (www.cru.uea.ac.uk) and ArcGIS was used to manipulate the data

Searching for signals of selection:

In order to confirm whether there are signals of genetic adaptation to different environments we selected the populations that are in the extremes of environmental variables (precipitation and temperature) (Fig.1) and we performed a pairwise F_{ST} at all SNPs between these populations using plink (Fig.2).

The database DAVID Bioinformatics Resources 6.7 (https://david.ncifcrf.gov/) were used to check whether SNPs with extreme differences in frequency between populations from different environments are enriched in genes

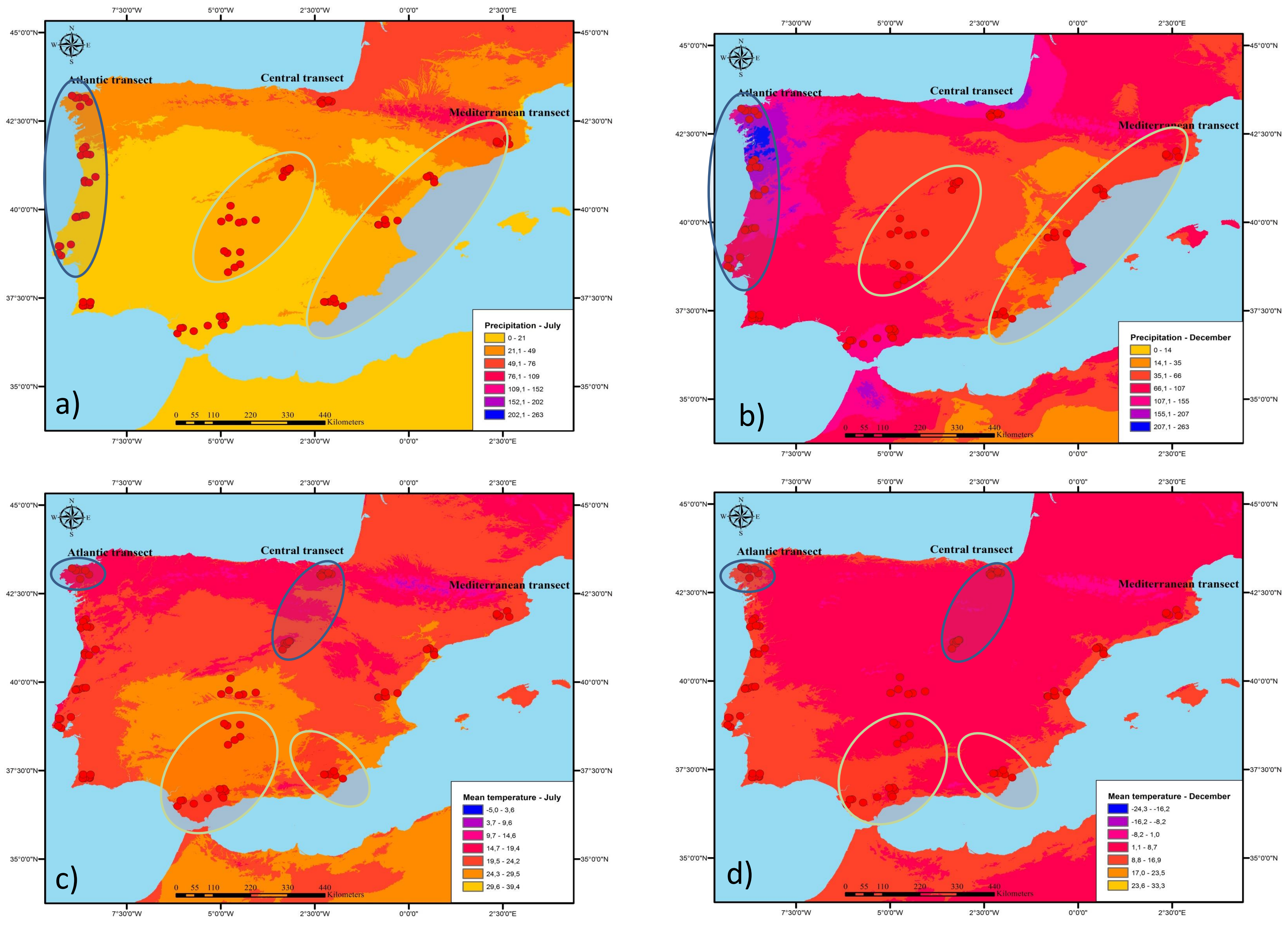
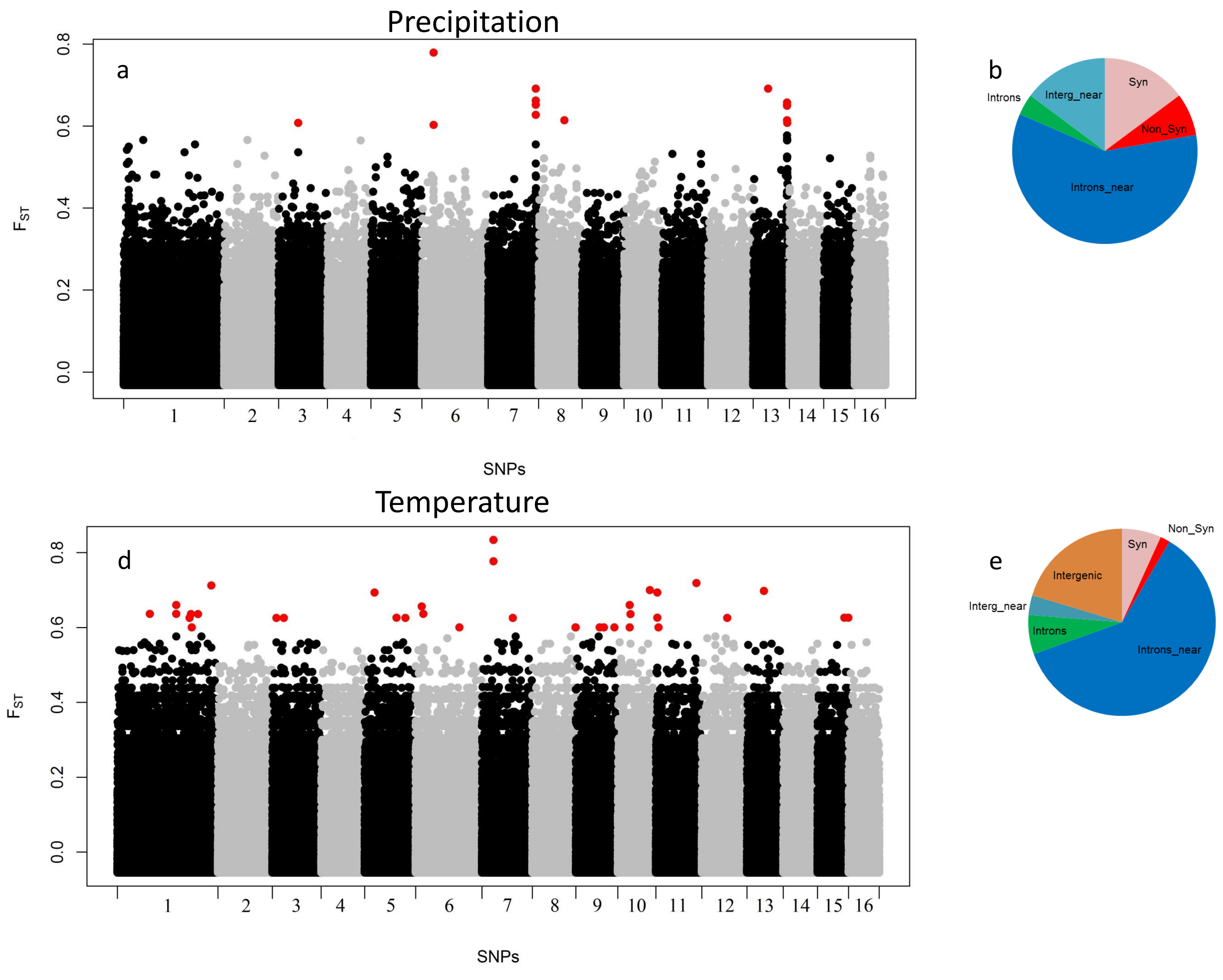


Figure 1: Map representing the distribution of precipitation in July (a) and December (b) and temperature in July (c) and December (d). The circles in different colors represent the two groups used to perform the analysis. The red spots are the samples position of the individuals used in this study.

Results and Discussion

Precipitation

A total of 29 SNPs have a F_{ST} higher than 0.60 (represented in red) and are mainly in chromosomes 7 and 13 (Fig.2,a), most of these SNPs are near or inside genes (a total of 9 genes) in this case just one SNP are in one intron with a distance from the nearest gene higher than 1000pb (Fig.2,1b). A total of 6 outlier SNPs are near to the gene GB48105, that is a probable cytochrome P450. The gene GB49882 also show 6 outlier SNPs, and this are responsible for potassium voltage channel. The functionally related groups obtained using the genes that have outlier SNPS have functions related to metabolism and cell structure (Fig2, 1c).



c

Nº of genes	Putative Function	Term	P-value
2	Metabolism	Response to insecticide	4.5 E-3
2	Metabolism	Response to toxin	6.0 E-3
2	Structural	Transmembrane protein	3.3 E-2
3	Structural	Membrane	9.1 E-2

f

Nº of genes	Putative Function	Term	P-value
3	Development	peripheral nervous system development	1.30E-02
4	Development	imaginal disc development	4.70E-02
3	Morphogenesis	imaginal disc morphogenesis	1.10E-01
3	Morphogenesis	post-embryonic organ morphogenesis	1.10E-01
3	Development	post-embryonic organ development	1.20E-01
3	Morphogenesis	instar larval or pupal morphogenesis	1.50E-01
3	Morphogenesis	post-embryonic morphogenesis	1.60E-01
3	Morphogenesis	metamorphosis	1.60E-01
3	Development	instar larval or pupal development	2.00E-01
3	Development	post-embryonic development	2.20E-01
3	Morphogenesis	pattern specification process	2.50E-01

Figure 2: Distribution of pairwise F_{ST} at all SNPs along the genome for the precipitation (a) and temperature(d). The red plots represent SNPs with a F_{ST} higher than 0.60. The distribution of the SNP outliers in different categories (b,e). Introns : SNPs located in introns at more than 1000pb from a gene; Introns_near: SNPS located at less than 1000pb from a gene; Syn:synonymous; Non_Syn: Non synonymous; Intergenic: SNPS located in intergenic region at more than 1000pb from a gene; Interg_near: SNPS located at less than 1000pb from a gene. In d is represent a table with functinally related groups for precipitation (c) and temperature (f).

Temperature

A total of 60 SNPs have a F_{ST} higher than 0.60 (represented in red) (Fig2,d). Most of the SNPs are located in introns but near to genes (less than 1000pb) (Fig2,e). The gene GB46620 has 15 outlier SNPs that is an uncharacterized protein homolog with C1orf11.

Two functionally related groups were obtained with the temperatures outliers, the group with higher enrichment score (0.95) it is in figure 2,f . It is possible to observe that the genes are very related with different phases of bee development and morphogenesis .

Conclusion

These results confirm the influence of environmental variables in the local adaptation of Iberian honey bee. Precipitation was associated a genes related to detoxidication mechanisms and membrane transports. It is known that temperature is important in the life cycle of insects, and this study confirm that, since temperature is associated with several genes related to development and morphogenesis.

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