



ARE PATHOGENS AND PARASITES SHAPING THE GENETIC DIVERSITY OF THE HONEY BEES?: A CASE STUDY IN THE IBERIAN PENINSULA

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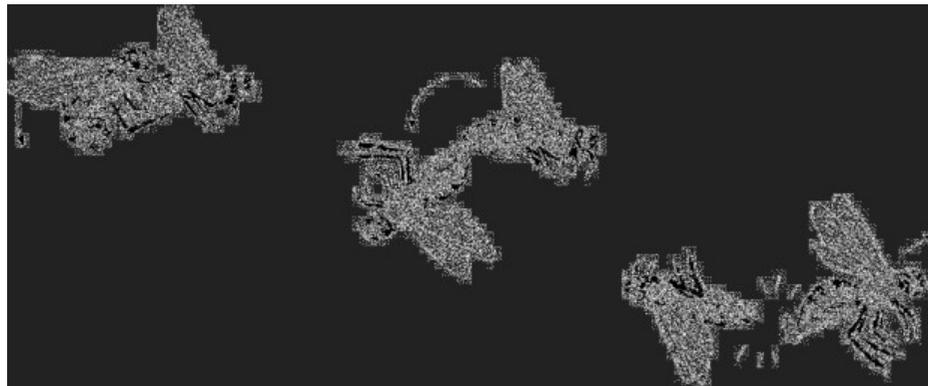
Genetic diversity

- Maintaining the genetic diversity is crucial for populations since it allows withstanding potential pathogenic perturbations
- Reduction of genetic diversity may lead to homogenization and impoverishment of the population:
 - Loss of local ecotypes
 - Loss of alleles (some of them related to pathogen tolerance)



Genetic diversity

- Human management (domestication) may reduce the population effective size and decrease the genetic diversity... but honeybees are not fully domesticated due to the mating system of honeybee queens with unselected drones



Colony losses

- Elevated and sometimes unexplained winter losses in Europe (from 21 millions to ~15.5 in 1970- 2007) and also in North America have been repeatedly detected
- This phenomenon known as Colony Collapse Disorder (CCD in the U.S.) is characterized by a series of specific symptoms

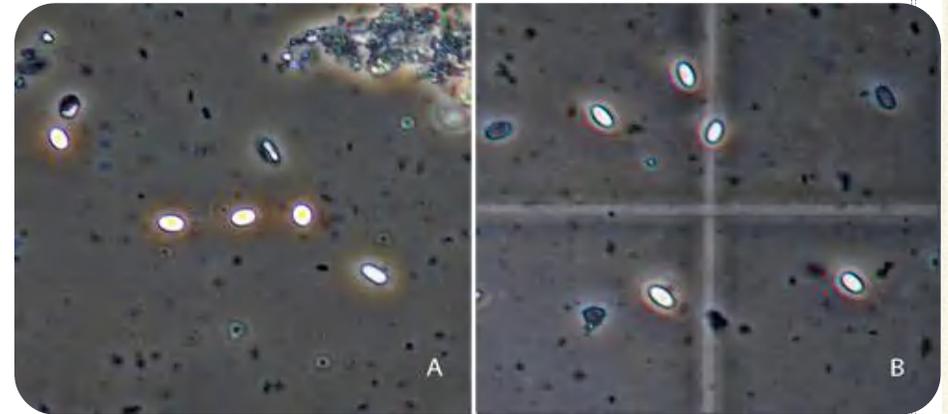


Colony losses

- A multitude of factors have been analyzed and discussed →to date, the most significant variable related to the population decline is the increased incidence of parasites and pathogens in honeybees



Varroa destructor and *Nosema spp.*



Parasitic mite from Asia (adapted to *A. cerana*)

Causing varroosis

Detected in *A. mellifera* in 1963.

Spread in Spain since 80s → loss of ~40% of the colonies

Two species causing nosemosis: *N. apis* and *N. ceranae* (type C)

N. apis: traditionally infected *A. mellifera*

N. ceranae: recently detected in *A. mellifera* and world-wide spread → related to the massive loss of colonies in temperate climate

Host-parasite relationships

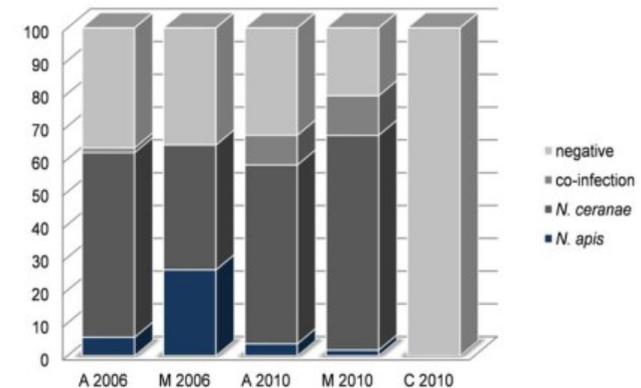
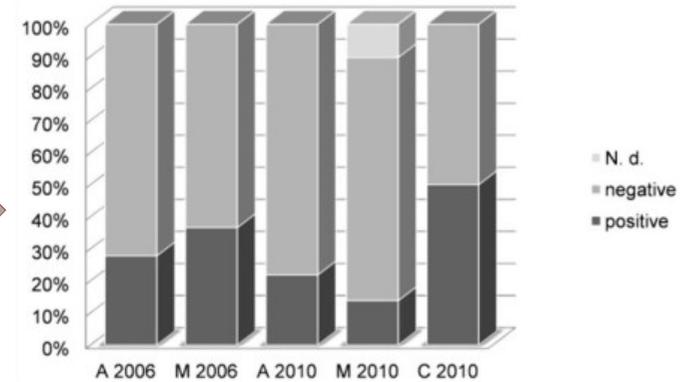
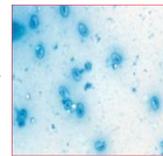
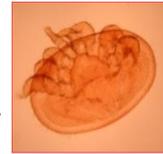
- Genetic variation can influence host susceptibility to pathogens
- The Iberian Peninsula provides an interesting scenario to study this correlation: A and M evolutionary lineages are dispersed naturally throughout
- **mtDNA** data show a clinal distribution in *A. m. iberiensis* as a consequence of the secondary contact of both lineages



Host-parasite relationships



Jara et al. (2012)
Journal of Invertebrate Pathology



- ▶ Evolutionary lineage and pathogen prevalence were determined in Iberian honey bees in 2006 and 2010
- ▶ In 2006 *N. apis* was found in a significant higher frequency in M than in A honey bees
- ▶ In 2010 *N. ceranae* was found in a high frequency in both M and A honey bees
- ▶ In both years no significant mite-evolutionary lineage relation was observed

Aims



To study the correlation of the incidence of *V. destructor* and *N. ceranae* and *N. apis* and the genetic diversity of *A. m. iberiensis*

We analyzed first the genetic diversity (H_e) of *A. m. iberiensis* in 2006 and 2010s.

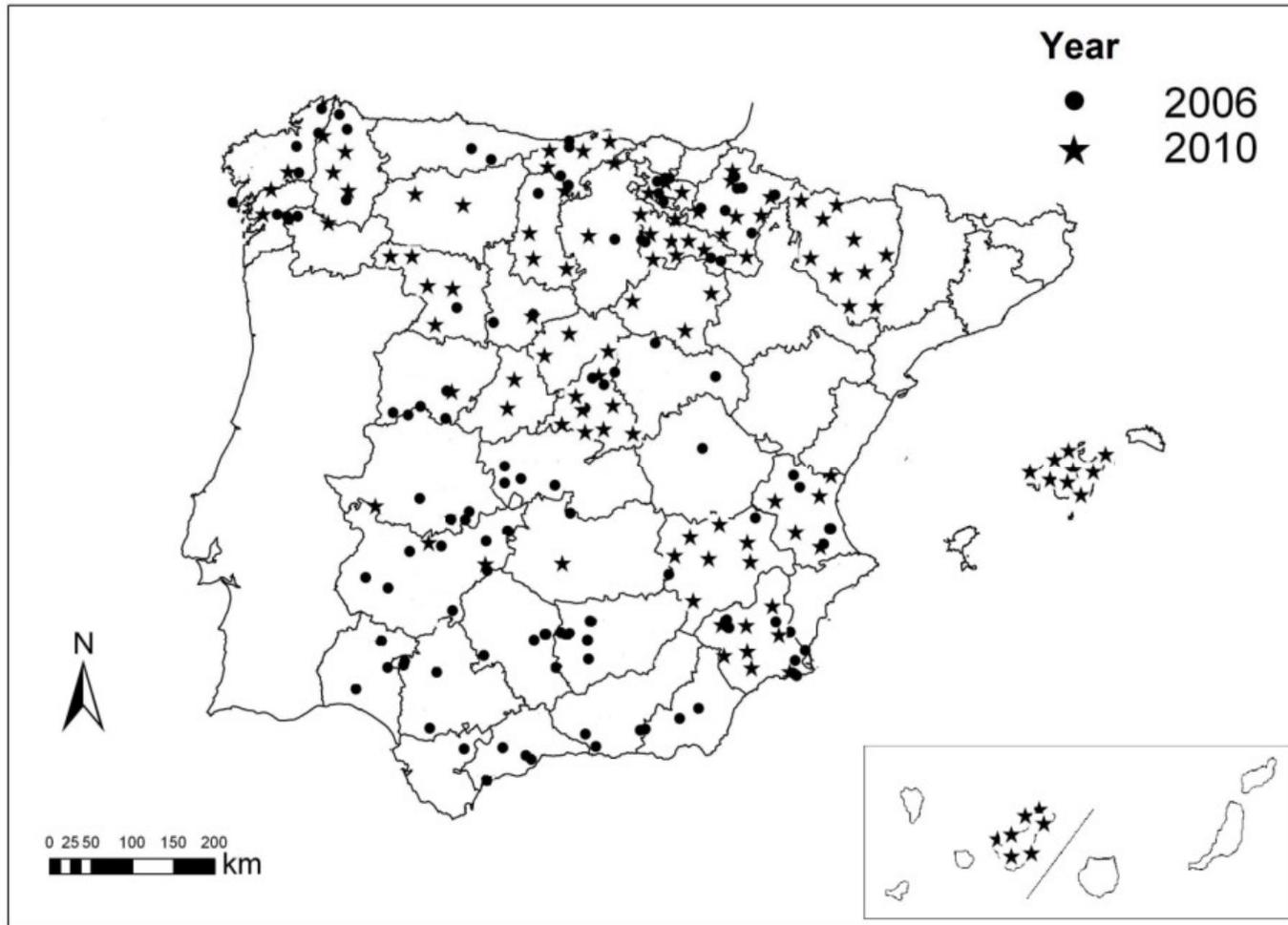
We analyzed the correlation of H_e with the prevalence of *V. destructor* and *Nosema spp.*

To study whether any of the analyzed microsatellite loci was subjected to selection

We detected differences in allele frequencies among different populations (F_{st})

Outlier loci were submitted to sequence similarity search and functional annotation

Sampling and Analysis



2006 → 113 colonies

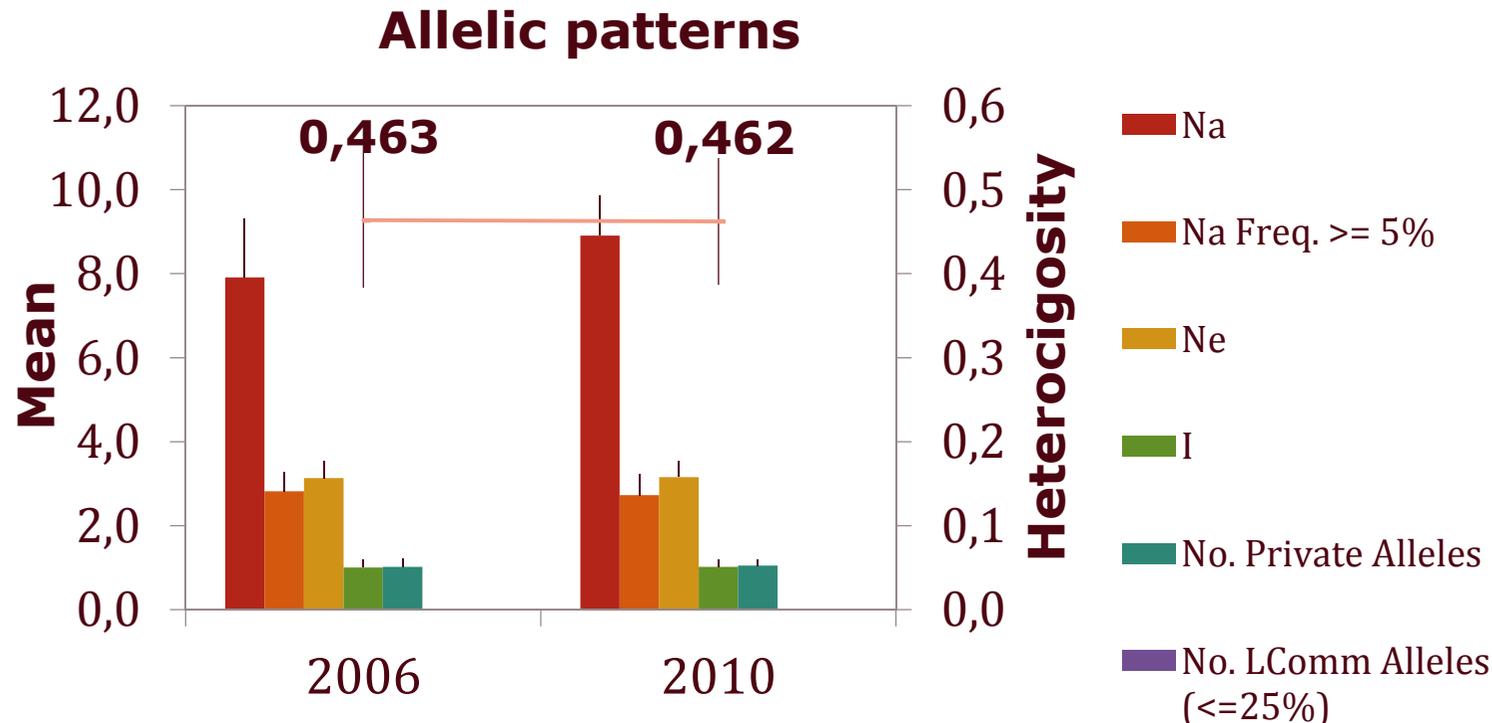
2010 → 115 colonies

Pathogen detection:

Varroa → direct visualization

N. apis and *N. ceranae* → specific PCR reactions

Genetic diversity in 2006 and 2010



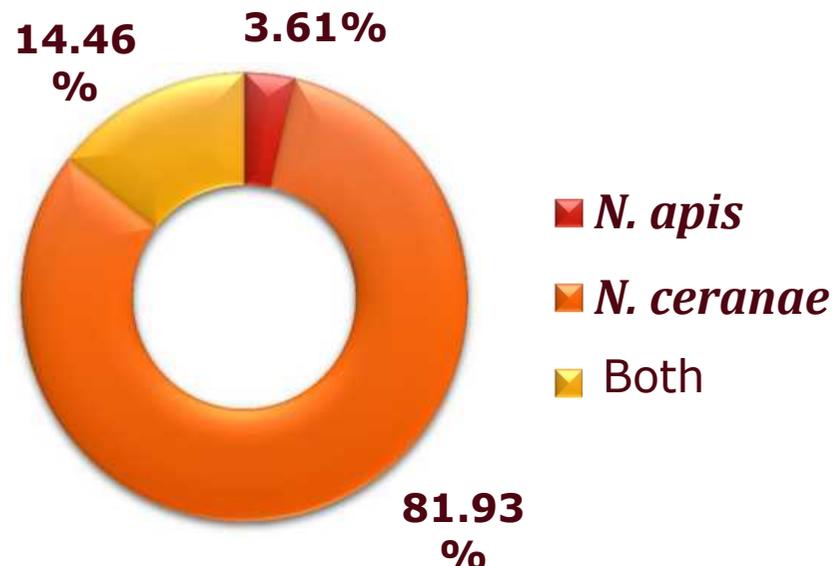
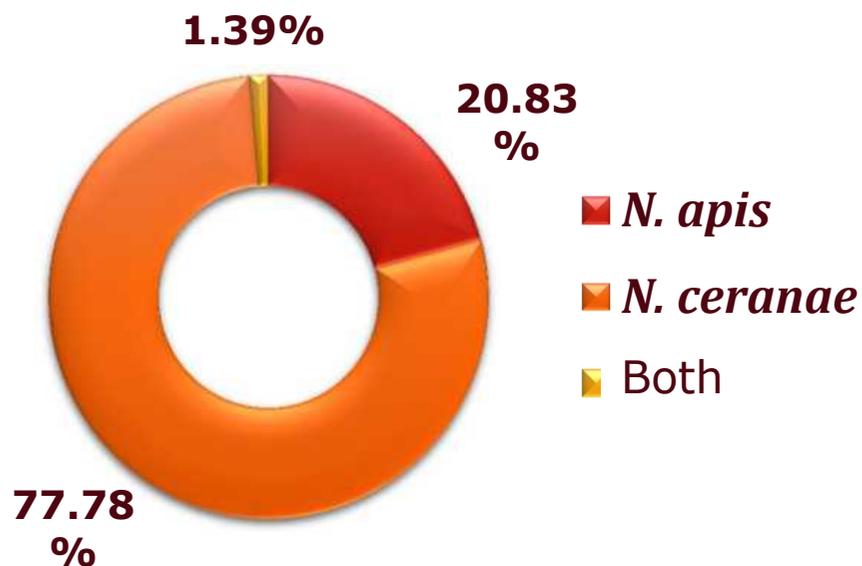
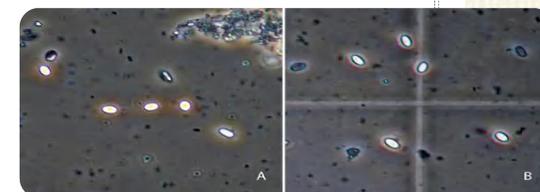
- H_e no significantly different from 2006 to 2010 (t-Student= 0.012; $p= 0.495$)
- Mean $H_e = \mathbf{0.437}$ ($\approx H_e$ to our sampling from 2000)

V. destructor, *N. apis* and *N. ceranae* prevalence

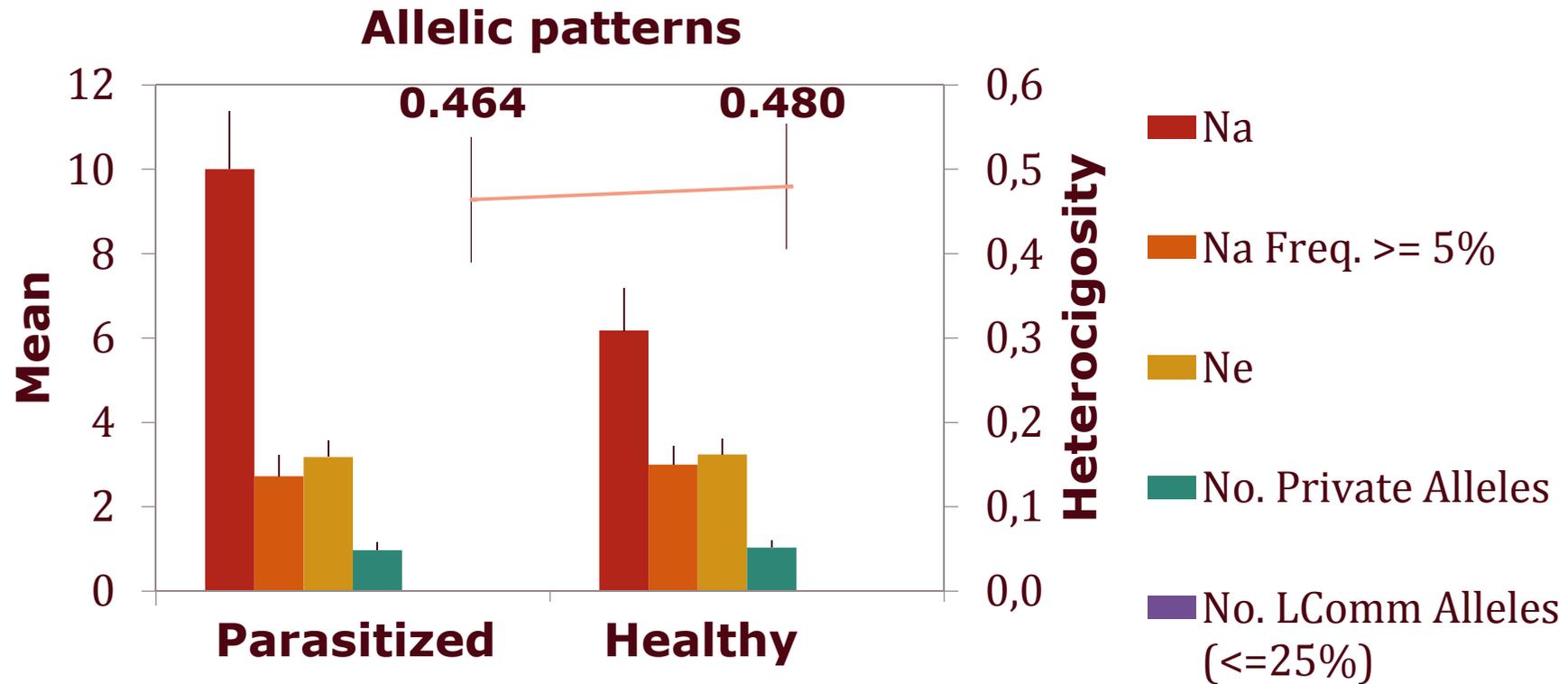
2006: 31% parasitized > 2010: 19.3% parasitized



2006: 62.5% infected < 2010: 72.2% infected

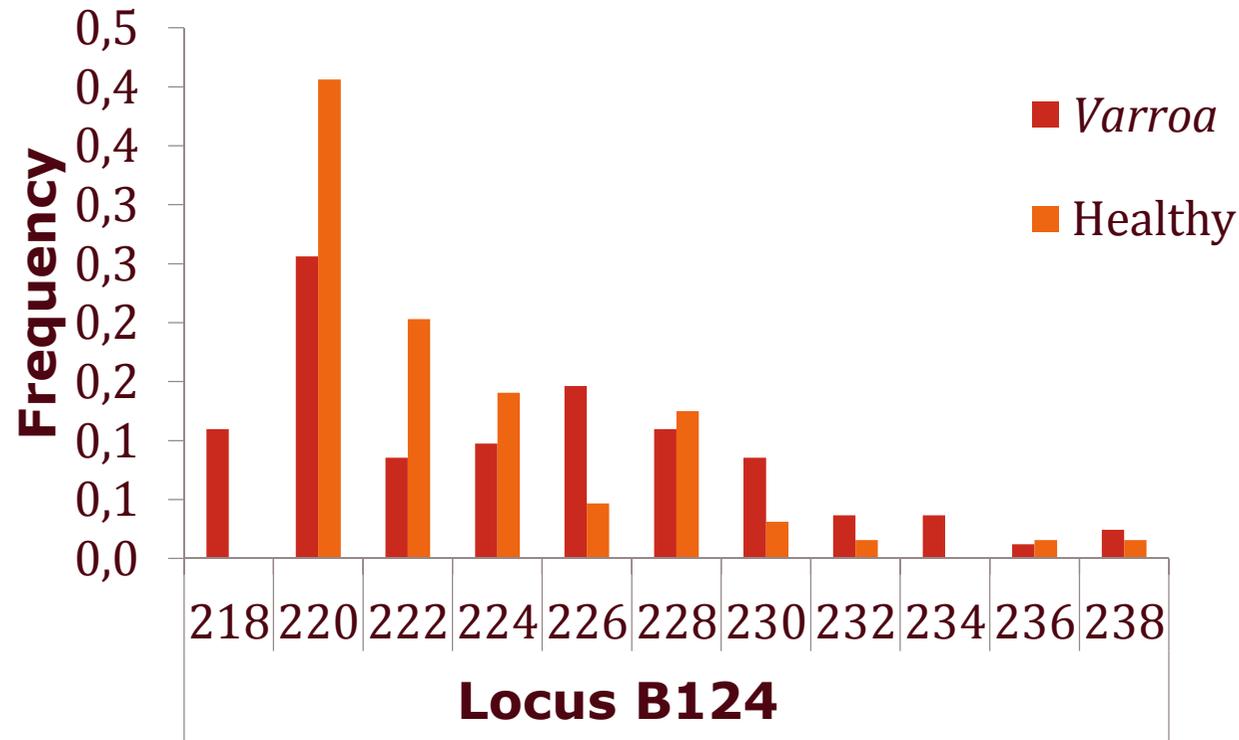


Genetic diversity in parasitized and healthy



- H_e greater in healthy than in parasitized honeybees but no significantly different (t-Student = -0.149, $p = 0.441$)

Loci under selection



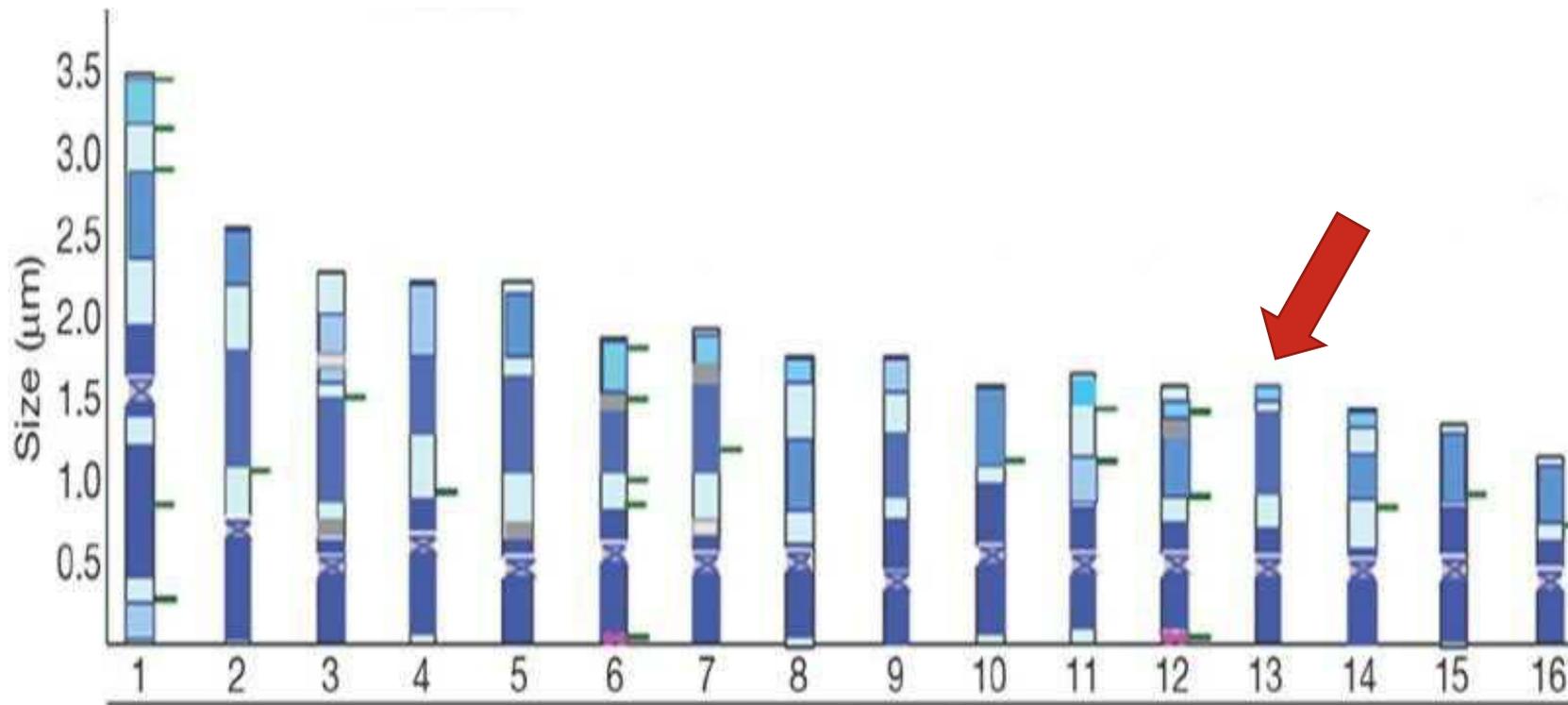
- candidate for positive directional selection with a specific Fst value higher than that of the rest loci

Locus	Het	Fst	P(Simul Fst < sample Fst)
B124	0.866319	0.047409	0.985782



Loci under selection

- B124 marker appears located on chromosome LG13 of *A. mellifera* and is associated with the *dpr7* gene, that encodes to immunoglobulin proteins.



Conclusions

Despite the observed variation of *Varroa* and *Nosema* prevalence, the genetic diversity of the Iberian honeybee populations has not changed over the last 12 years → influenced by factors:

- ✓ uncontrolled breeding of queen bees with drones from different colonies (polyandry),
- ✓ anthropogenic factors such as transhumance and swarm catching.



Conclusions

The genetic diversity decrease (although not significantly) in colonies with presence of the studied pathogenic elements compared to healthy ones → consistent with the theory stating that populations with lower genetic diversity suffer higher prevalence of parasites and pathogens



Conclusions

One microsatellite locus appear to be subject to selective forces with alleles showing different patterns of frequency between parasitized and no-parasitized honeybees → locus associated with genes involved in immune response, may be linked to traits of tolerance to pests in honeybees

