

# Utilization of microsatellite markers in rubber tree for genetic diversity analysis and genetic mapping of agronomic traits.

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418 microsatellite markers already developed on rubber tree by CIRAD geneticists (*Seguin et al 2008*)

Most of them from enriched libraries

Dinucleotides: majority of AG/CT, some AC/GT

Non coding regions of genomic DNA

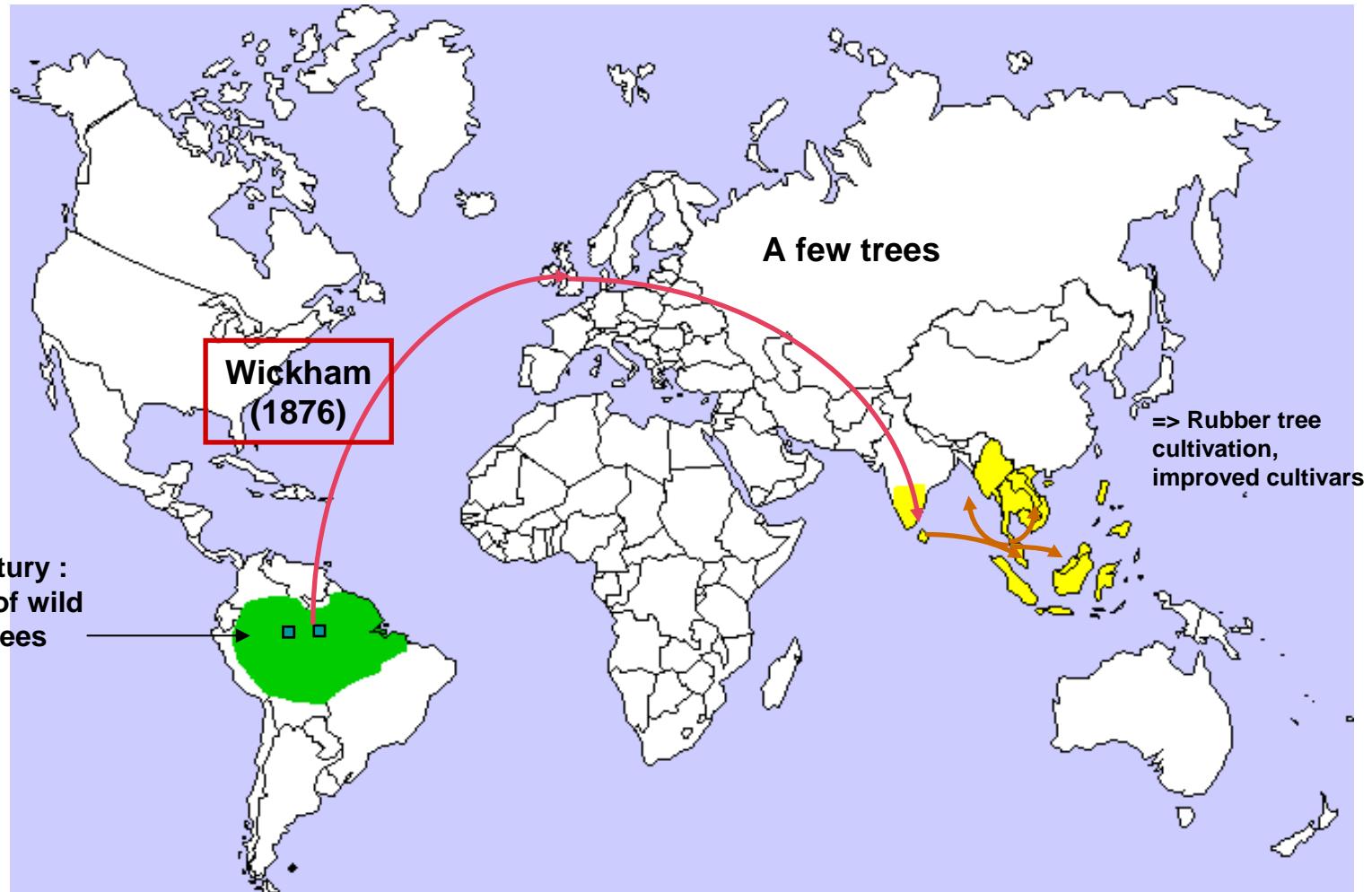
365 polymorphic microsatellite markers



Two examples of their use in rubber tree genetics studies

- ✓ Genetic diversity
- ✓ Linkage mapping

# *Analysis of genetic diversity among wild Amazonian rubber tree populations*



Rubber tree : a trans-domestication



*Hevea brasiliensis* (Willd. ex Adr. de Juss.) Muell.-Arg.

(Priyadarshan & Gonçalves, 2003)

# Genetic diversity of wild rubber tree populations

Sample under study comes from an *ex situ* collection located in French Guiana

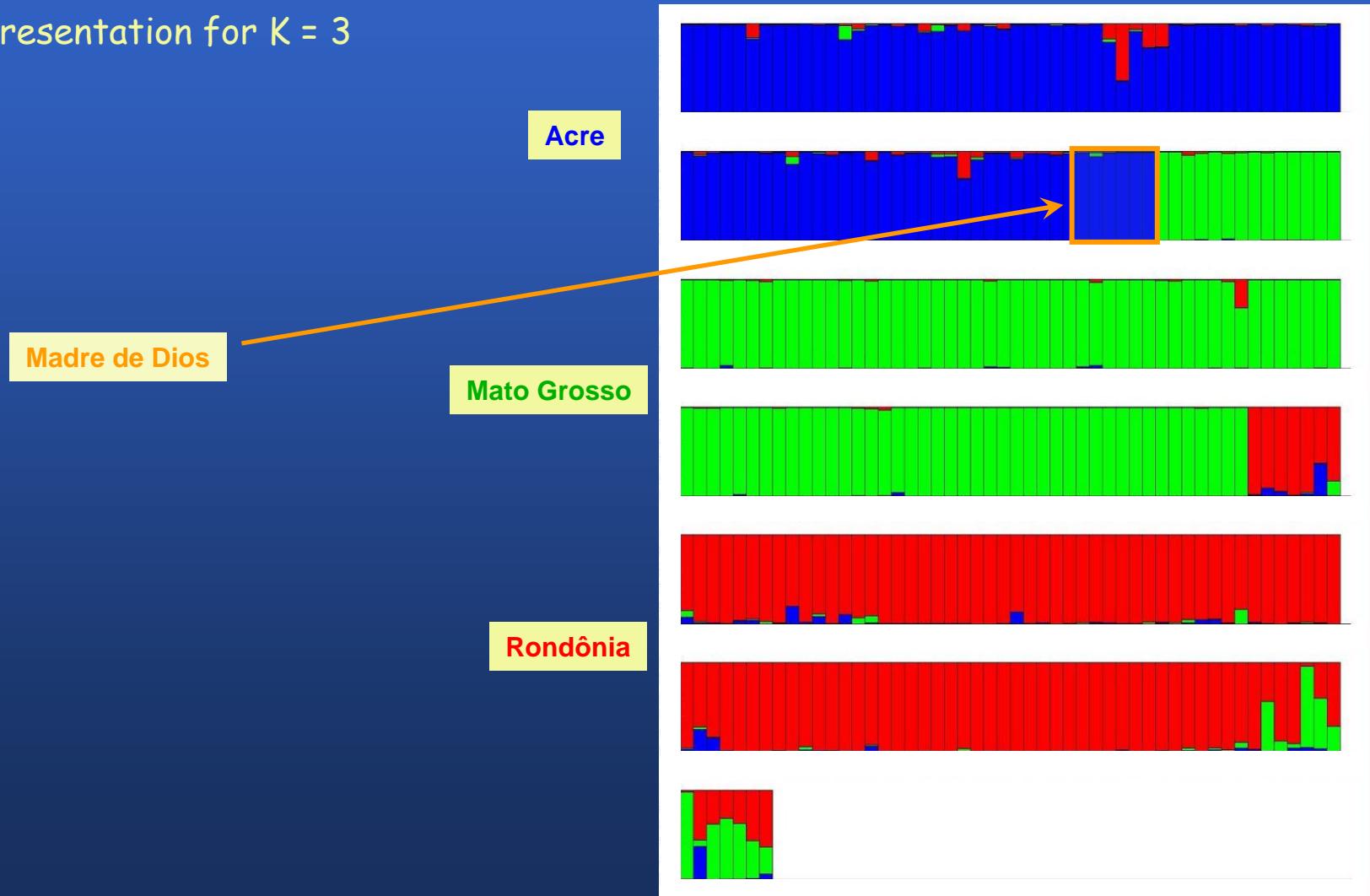
Initial survey	Area of origin	Number of locations	Number of genotypes
Peruvian Minist. Agric.(1948)	Madre de Dios (Peru)	1	6
EMBRAPA/IRCA (1974)	Acre (Brazil)	2	8
	Rondônia (Brazil)	2	7
IRRDB (1981)	Acre (Brazil)	5	72
	Mato Grosso (Brazil)	4	108
	Rondônia (Brazil)	7	106
<i>Total number of clones in sample:</i>			307

15 microsatellite markers, each on a different linkage group

# Genetic diversity of wild rubber tree populations

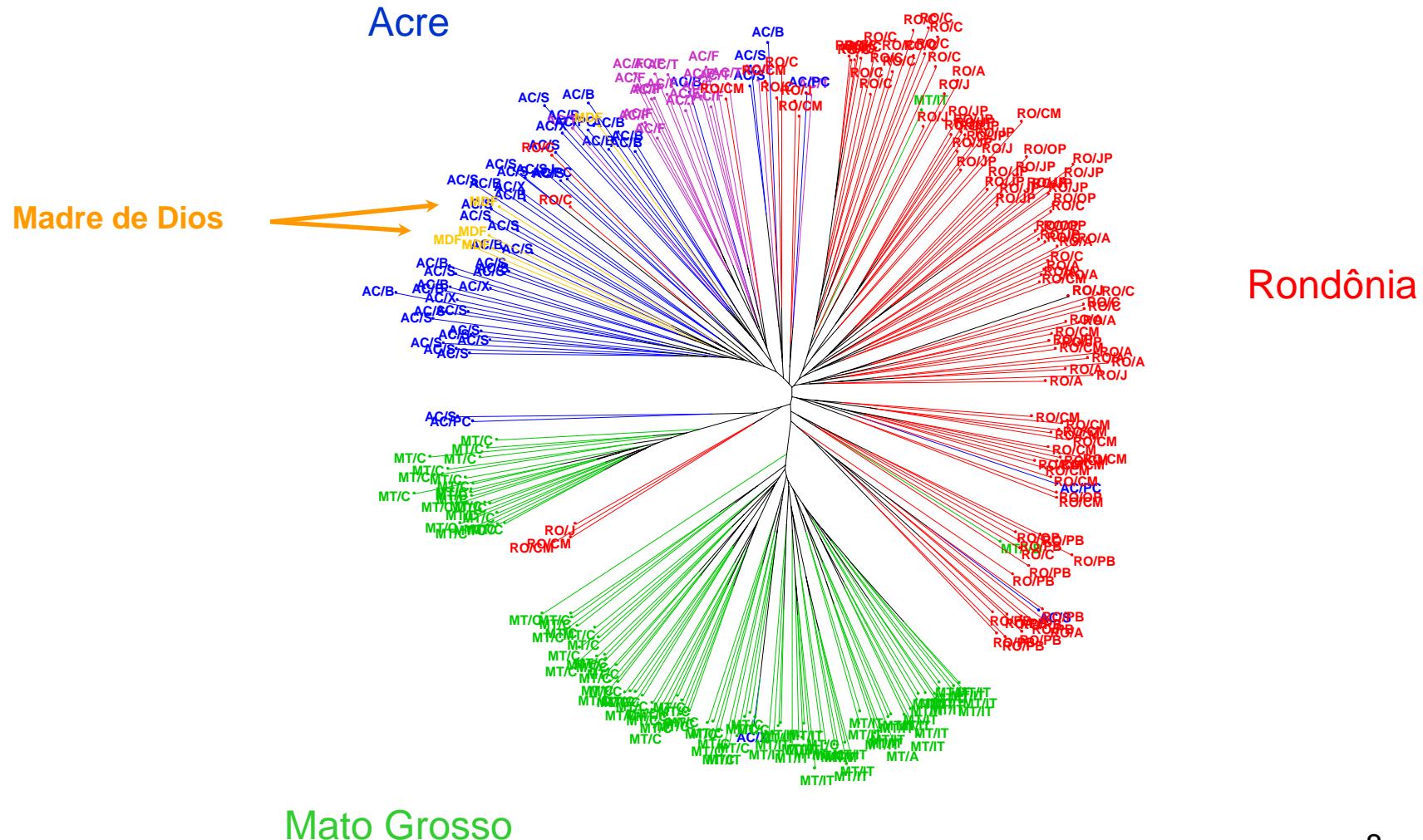
Structure software: a clustering algorithm to infer the structure of populations

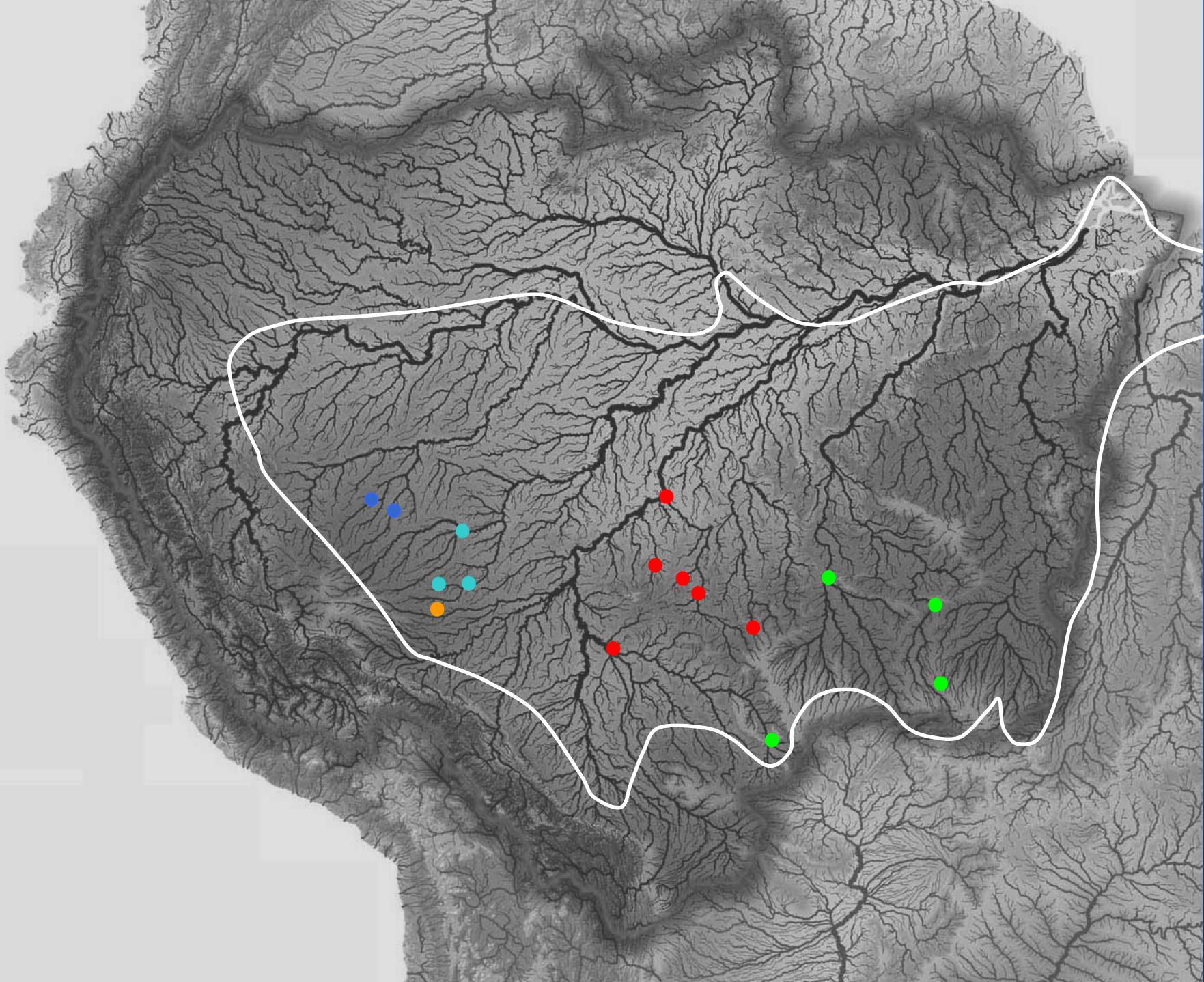
Best representation for K = 3 clusters

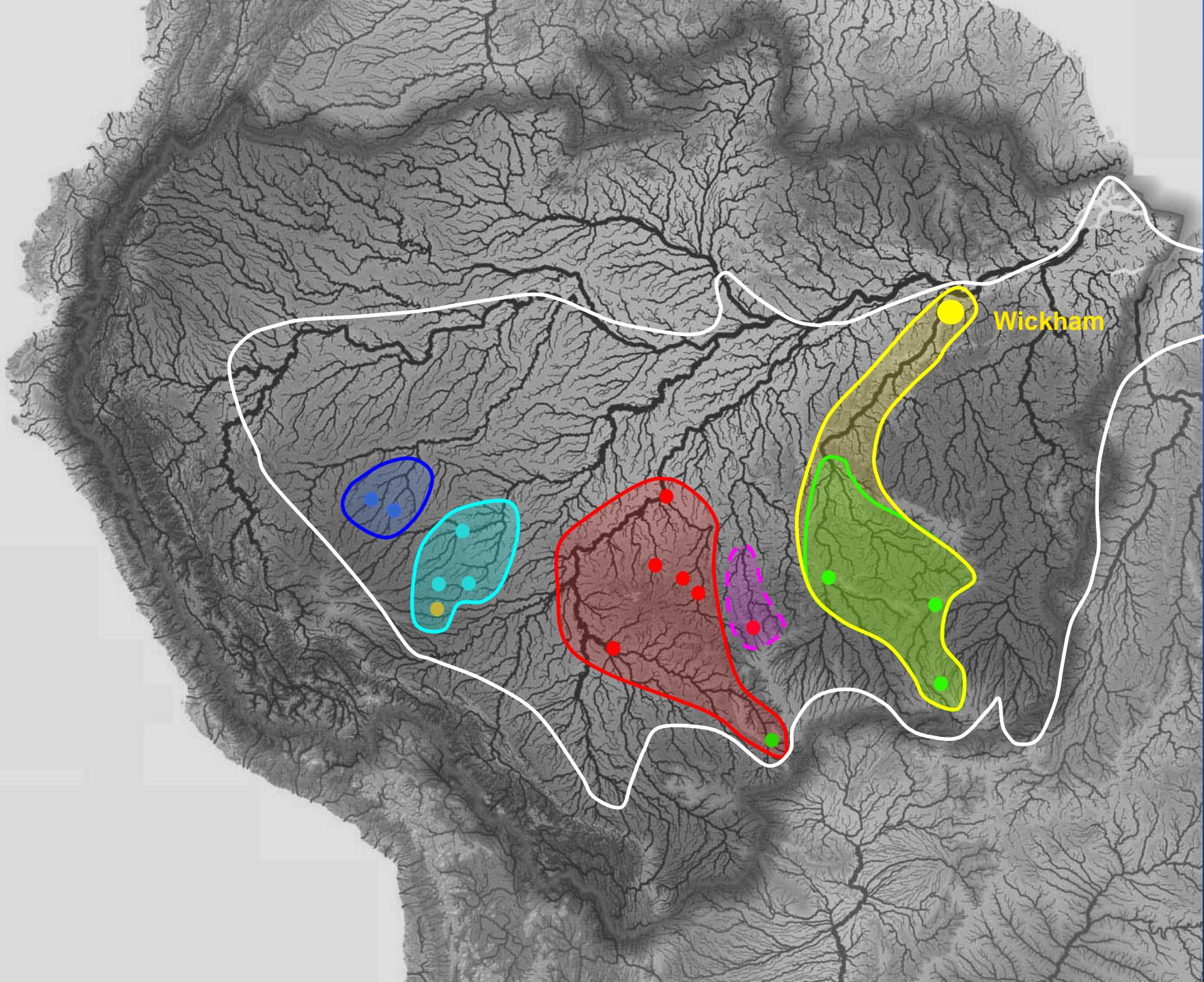


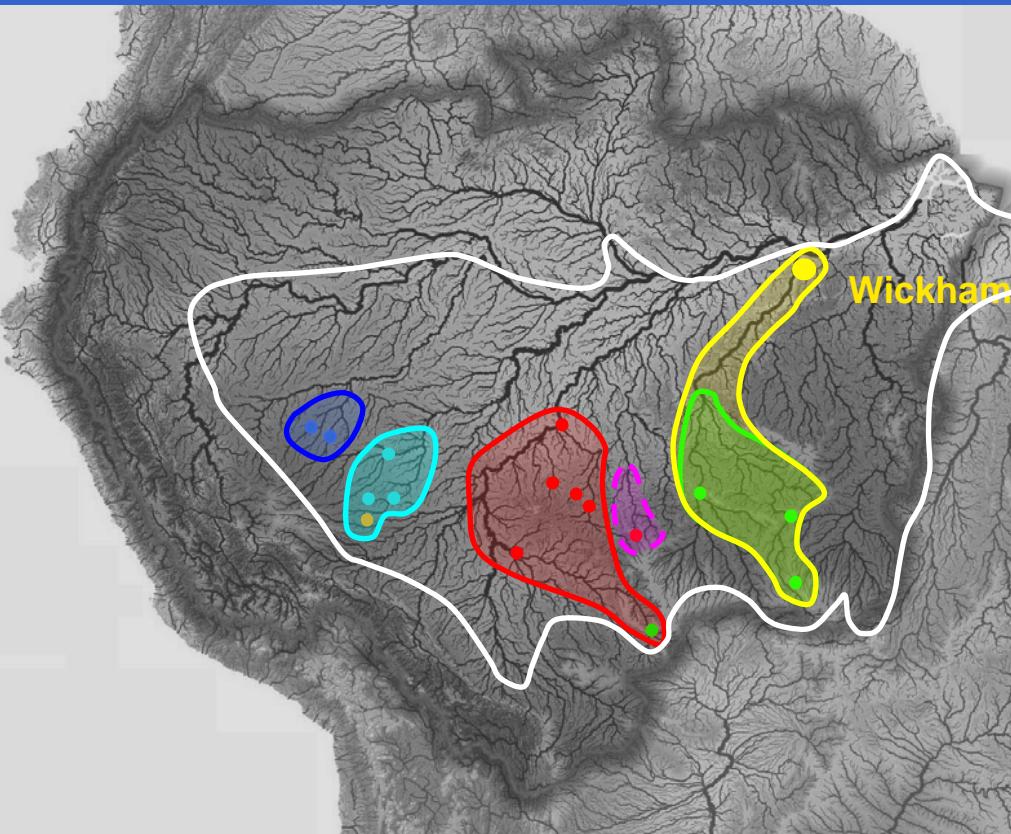
# Genetic diversity of wild rubber tree populations

Darwin software: construction of a dissimilarity measure to draw Neighbor-Joining trees









- Hydrographical network and isolation by distance
- Some populations may not exist any more
- Necessity of additionnal surveys
- Highly polymorphic markers (21.7 alleles/locus)
- Not suitable for association genetics studies

# Genetic linkage mapping for the detection of quantitative trait loci

# Three progeny populations already mapped at Cirad

- PB260 x FX3899
  - Detection of loci for SALB resistance  
(Lespinasse et al 2000a; Lespinasse et al 2000b; Le Guen et al 2003; Le Guen et al 2007)
- RRIM600 x PB217
  - Detection of QTLs for growth and latex production  
(Prapan et al 2006; Rattanawong et al 2008)
- PB260 x MDF180
  - Identification of loci for durable resistance to SALB

# South American Leaf Blight due to *Microcyclus ulei*

Asiatic clones highly susceptible



MDF180 is a resistant cultivar:

High level of partial resistance  
Durable resistance

(Le Guen et al 2008)



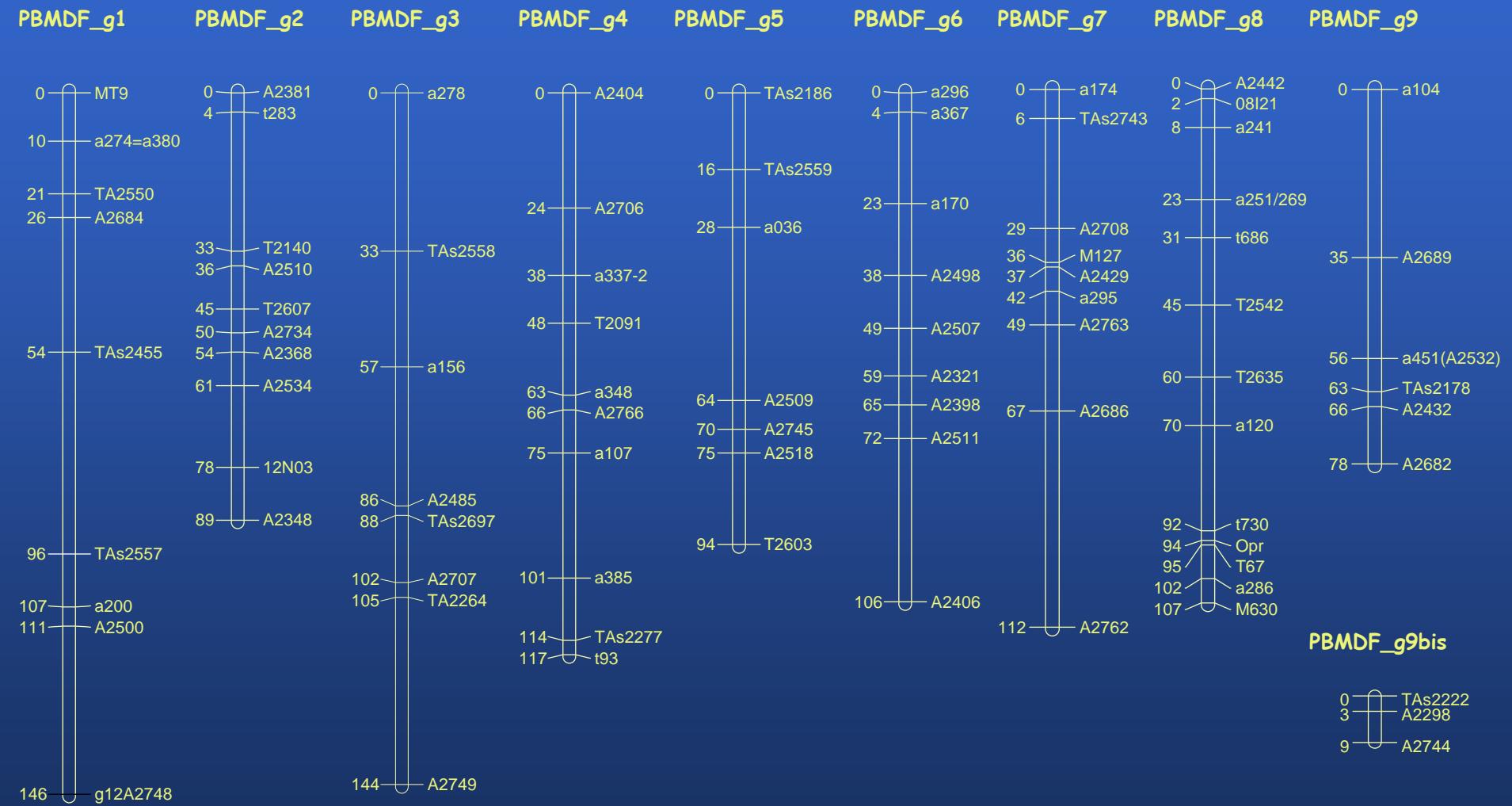
298 clones from PB260 x MDF180 cross were genotyped with 177 microsatellites

Disease observation under controlled conditions and in the field

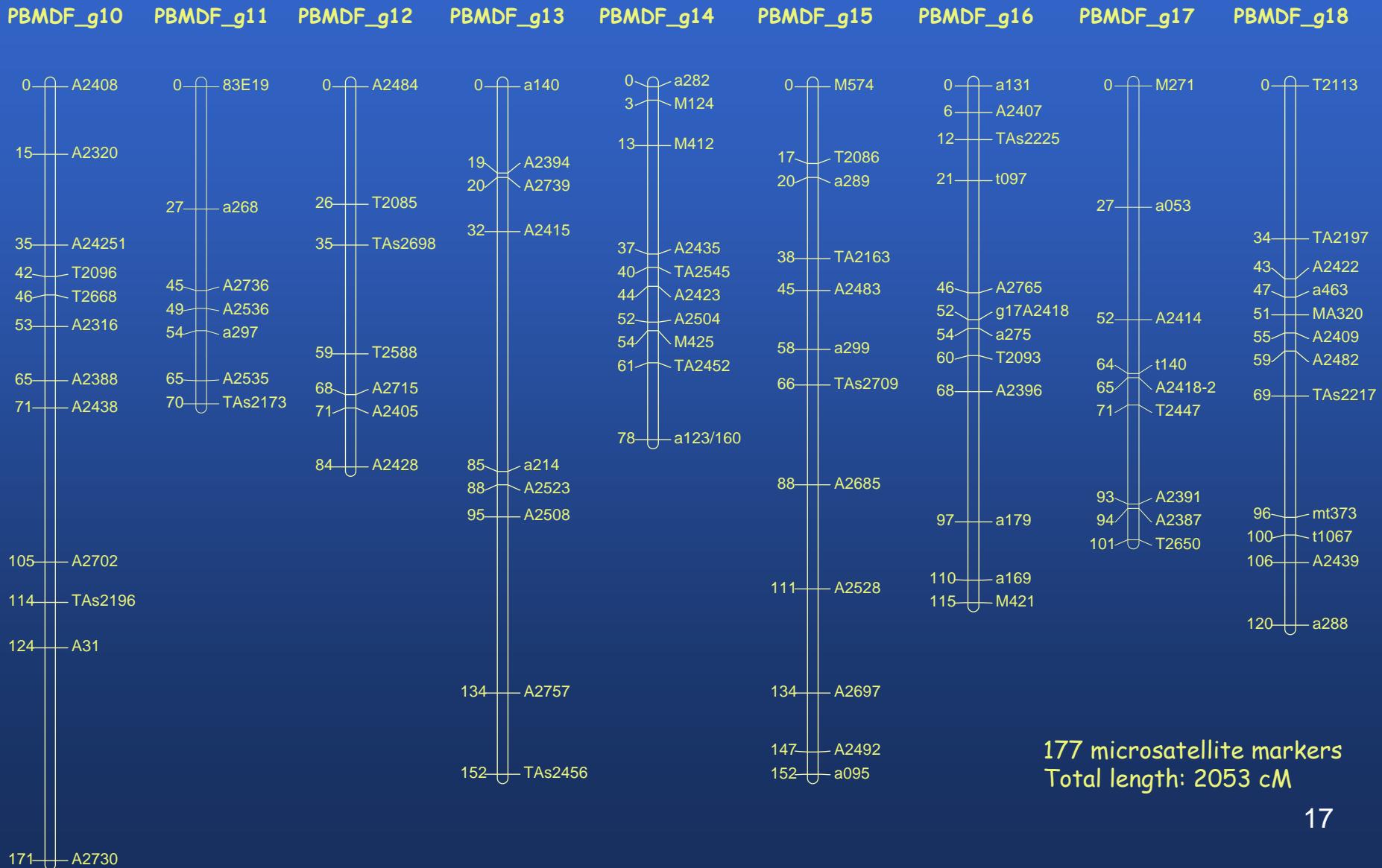
Construction of a linkage map (JoinMap)

Location of markers linked to the expression of resistance (MapQTL)

## Genetic linkage map of PB260 x MDF180 (JoinMap 3.0)

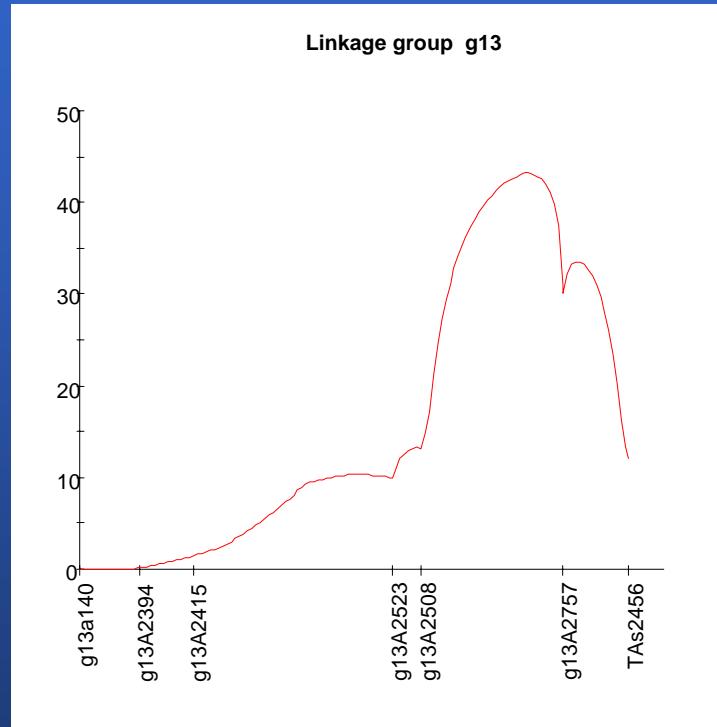


## Genetic linkage map of PB260 x MDF180 (JoinMap 3.0)

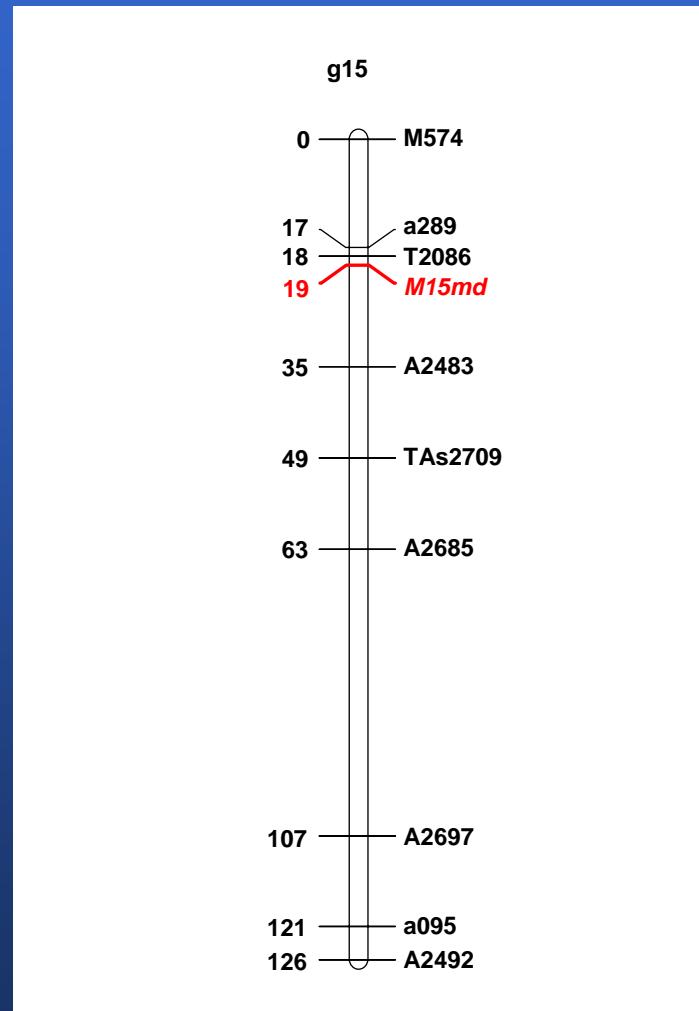


177 microsatellite markers  
Total length: 2053 cM

## Lod score value for an important resistance QTL on linkage group 13



## Location of the major resistance gene M15md on linkage group 15



Four minor QTLs were also detected on other linkage groups

Microsatellite-based linkage map allowed identification of a major gene and QTLs for SALB resistance

However they are some large gaps in the linkage map

There is a need of additional locus-specific markers for Marker Assisted Selection

# Conclusion

- Advantages of microsatellite markers in non-coding DNA:  
Well adapted to diversity analysis and genetic mapping:  
Neutrality, locus specificity, multi-allelism, co-dominant  
segregation pattern, ease of use
- Drawbacks:  
Multi-allelism is a flaw for linkage disequilibrium assessment  
Their number is not sufficient for precise QTL location
- Sequencing rubber tree (either whole genome, or expressed  
genes or BACs) would be very useful for geneticists to:
  - ✓ Identify new and less variable microsatellite markers
  - ✓ Identify SNPs (the most useful type of marker for association  
genetics studies)
  - ✓ Combine « candidate gene » and « QTL detection » approaches

# References

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