Sanna Olsson, David Macaya-Sanz, Delphine Grivet, Carlos Guadaño, David Leon Carbonero, Sara Pinosio, Francesca Bagnoli, Camilla Avanzi, Giovanni G. Vendramin, Santiago C. González-Martínez, Ricardo Alía, Sven Mutke

Utility of the 4TREE SNP array in clonal identification of Mediterranean stone pine (*Pinus pinea* L.)

B4EST International Conference *Managing Forest Genetic Resources (FGR) for an uncertain future*
Lisboa 20-23 June, 2022

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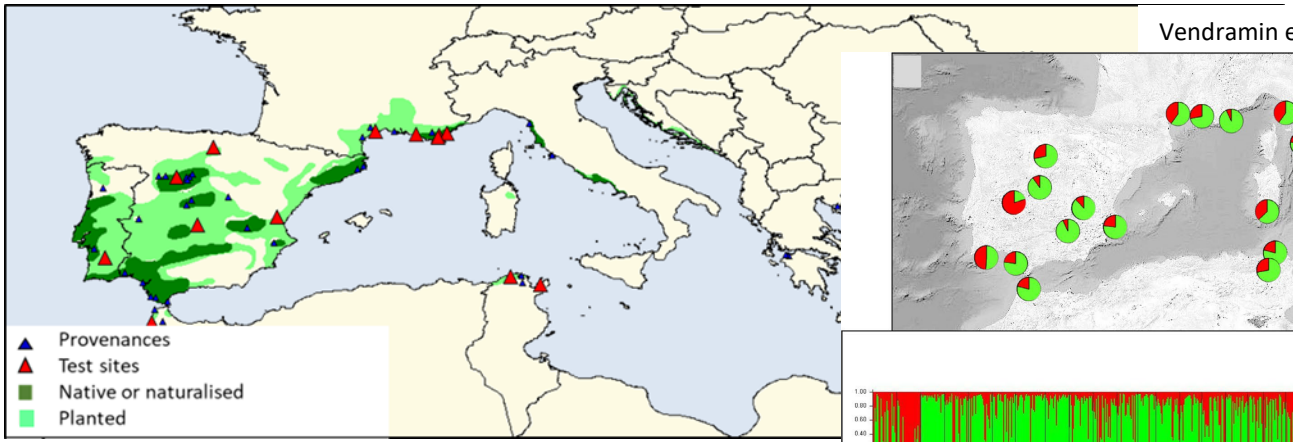
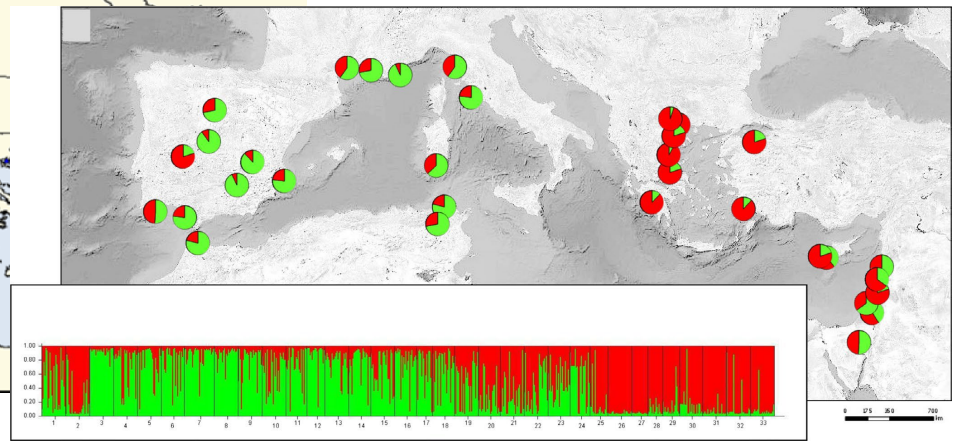
Pinus pinea

- Very low molecular and quantitative genetic variation
- 5,671 new SNP markers from the 4TREE array
- Breeding program established to target edible nuts

FAO provenance test sites *Silva Mediterranea,* 1994 (Mutke et al. 2010, 2013; Vizcaíno-Palomar et al. 2019, 2020)



Vendramin et al. 2008; Jaramillo-Correa et al. 2019, 2021



Clone	Proportion genotyped	Relative genetic distance*	SNP mismatches
CL_1.011	0.970	0.009	35
CL_1.012	0.959	0.014	53
CL_1.073	0.965	0.032	117
CL_1.123	0.959	0.017	66
CL_1.201	0.958	0.031	113
CL_2.004	0.948	0.048	174
CL_2.048	0.960	0.033	118
CL_3.029	0.952	0.047	173
CL_3.029	0.962	0.037	139
CL_3.048	0.955	0.018	68
CL_3.057	0.955	0.020	68
CL_3.063	0.961	0.016	58
CL_6.010	0.960	0.037	139
CL_6.015	0.957	0.032	119
CL_6.053	0.967	0.014	54

* Manhattan method for pairwise similarity

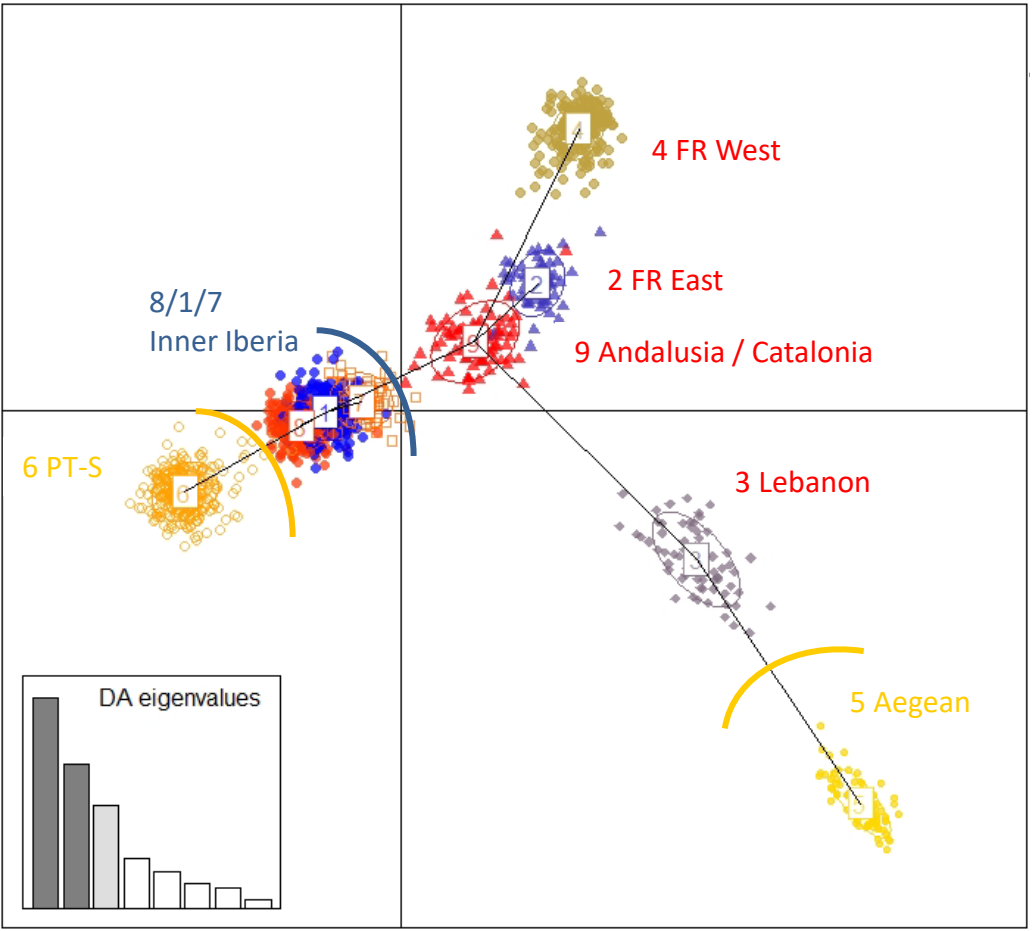
Basic materials (clones) s. 2015, 7 SSR (Pinzauti et al., 2012):

- Low allelic polymorphism
- Erroneous genotypes in 16% of grafted trees?
- Inconsistent results

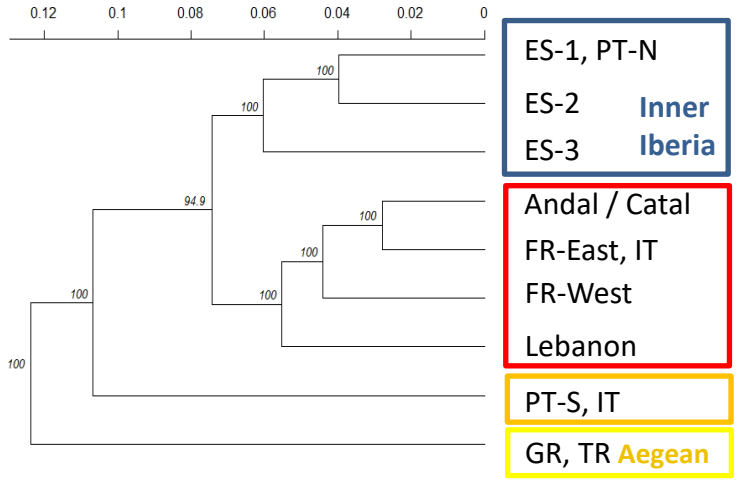
Axiom 50K array (TF) 4TREE:

- High proportion of genotyped SNPs
- Much lower reported error rate than SSRs!
- Unambiguous interpretation

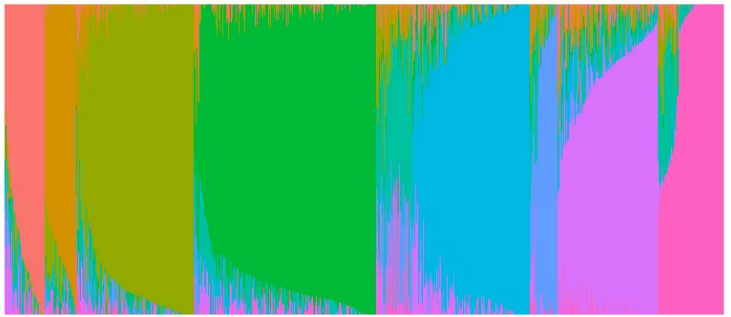
- Practical application of results: exclusion of 14 ramets from their corresponding genets (10%)



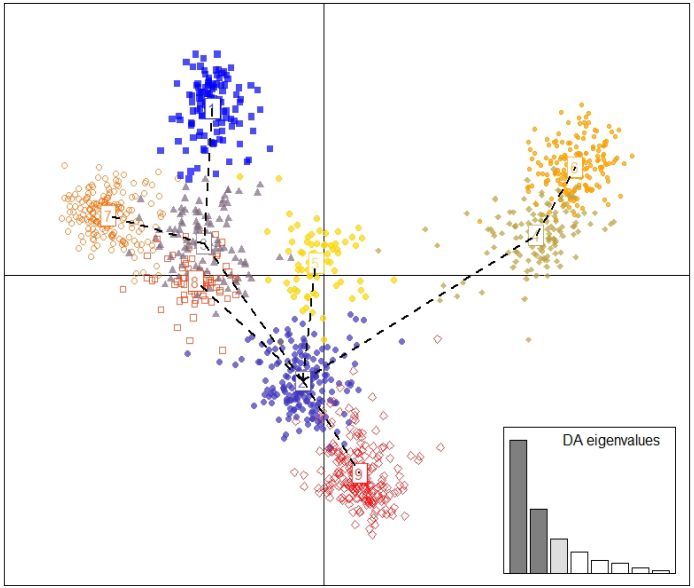
DAPC, with LD-pruned SNPs

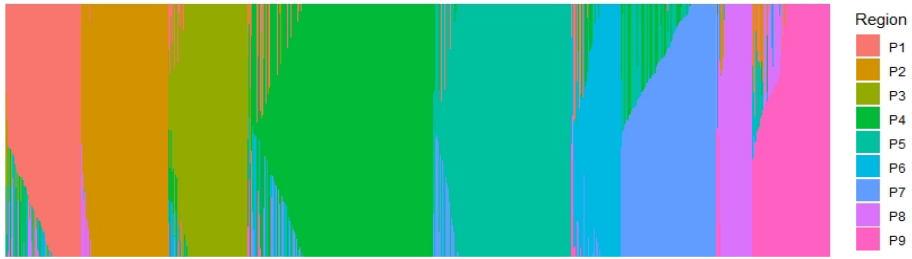


Unprecedented resolution of population structure!



sNMF (individual ancestry coefficient)





Subset of 200 most informative SNPs with high F_{ST} across training sets (assignPOP)

- Intermediate-priced identification of clones
- Similar structure but less definition
- **Will they distinguish between clones in same region?**

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sanna.olsson@inia.csic.es