Supporting Information

In situ genetic association for serotiny, a fire-related trait, in Mediterranean maritime pine (*Pinus pinaster* Aiton)

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Table S1 Range-wide SNP genotyping to test for accuracy outside the focal population of the phenotypic model for serotiny constructed in the eastern Iberian Peninsula (see also Figure 1): populations sampled, geographical region, mtDNA lineage (i.e., maternal genetic lineage; Burban & Petit, 2003) and sample size (*N*); the mean observed percentage of serotiny is also provided (retrieved from Tapias *et al*., 2004; see *Material and Methods*).

Population	**Geographical region**	**mtDNA lineage**	***N***	**Serotiny (%)**
Mimizan	Atlantic France	western	19	9.03
Galicia	Atlantic Spain	western	21	0.00
Leiria	Atlantic Portugal	western	24	0.00
Tabuyo del Monte	Central Spain	western	20	43.30
Arenas de S. Pedro	Central Spain	western	27	29.73
Cuéllar	Central Spain	western	28	1.07
Coca	Central Spain	western	18	0.66
San Leonardo de Yagüe	Central Spain	western	20	3.64
Boniches	Eastern Spain	western	11	17.31
Olba	Eastern Spain	western	21	1.19
Cazorla	Southern Spain	western	15	11.11
Cómpeta	Southern Spain	western	4	3.31
Corsica	Corsica (France)	eastern	37	2.61
Tabarka	North Africa (Tunisia)	eastern	21	62.76
Tamrabta	North Africa (Morocco)	Moroccan	24	26.52

Table S2 Illumina VeraCode OPA design file and designability scores provided as Excel file (‘GGT_Sequence_v1_Score251109.xls’).

Table S3 Marker effects of 26 SNPs with $P<0.05$ in the mixed-effects linear models (MLMs) for the additive and allele-dominance genetic models (no significant SNPs were found for the over-dominance model). SNP refers to SNP marker names reported by Chancerel *et al.* (2011) except for *myb2-337*; ns: not significant.

SNP	Genetic effects					
	Additive model			Allele dominance model		
	F	P	R^2	F	P	R^2
m15	6.0523	0.0028	0.0620	6.4522	0.0119	0.0330
m39	3.5752	0.0299	0.4529	ns	ns	ns
m79	5.8002	0.0036	0.0588	11.5088	0.0008	0.0583
m237	5.0627	0.0072	0.0542	9.9162	0.0019	0.0531
m493	7.2695	0.0076	0.0371	ns	ns	ns
m526	ns	ns	ns	3.9169	0.0492	0.3276
m594	3.4602	0.0334	0.0350	ns	ns	ns
m646	4.6751	0.0104	0.0489	9.3322	0.0026	0.0488
m663	4.3030	0.0148	0.0435	8.5825	0.0038	0.0434
m692	6.6319	0.0016	0.0686	ns	ns	ns
m696	ns	ns	ns	5.7223	0.0177	0.0294
m698	ns	ns	ns	4.9335	0.0275	0.0249
m705	3.3688	0.0365	0.0344	ns	ns	ns
m716	3.6902	0.0267	0.4529	ns	ns	ns
m757	3.5286	0.0313	0.0360	ns	ns	ns
m816	ns	ns	ns	5.3404	0.0219	0.0277
m817	3.2994	0.0390	0.4629	ns	ns	ns
m912	3.8011	0.0240	0.0409	5.7912	0.017	0.0312
m942	ns	ns	ns	4.6263	0.0327	0.0234
m955	3.0488	0.0497	0.0310	4.8997	0.028	0.0249
m965	3.5331	0.0311	0.0357	5.7636	0.0173	0.0292
m974	ns	ns	ns	5.3869	0.0213	0.0272
m983	ns	ns	ns	5.4546	0.0205	0.0278
m1194	3.7141	0.0261	0.0376	6.9170	0.0092	0.0350
m1196	ns	ns	ns	3.9690	0.0477	0.0200
myb2-337	3.1416	0.0454	0.0319	ns	ns	ns

Table S4 Gene function, site annotation, and mapping information for the 17 SNPs potentially involved in associations with serotiny.

SNP	Annotation	Putative function (UniProtKB/Swiss-Prot)	Site annotation[c]	Linkage Group
m15	defectively organized tributaries 2 (DOT2)	root, shoot, and flower development	nc	
m289	calcium-dependent protein kinase (CDPK)	signalling, stress response	syn	1[a,b]
m594	pyrophosphate-energized vacuolar membrane proton (AVP)	membrane transport	syn	8[a,b]
m692	unknown	unknown	unk	3[a]
m696	arabinogalactan-like protein (AGP)	cell walls, plant defense, stress response	nc	3[a,b]
m698	nascent polypeptide-associated complex subunit alpha-like protein (NAC-alpha)	protein transport, response to salt stress	syn	
m705	carotenoid cleavage dioxygenase (CCD)	stress response, signalling	nc	
m816	receptor protein kinase clavata1 (CLV1)	cell differentiation	syn	1[a,b]
m817	receptor protein kinase clavata1 (CLV1)	cell differentiation	nc	1[b]
m912	peroxidase 72-like (PER72)	stress response	non-syn	
m955	unknown	unknown	unk	3[a,b]

m959	unknown	unknown	unk	5[b]
m974	1-aminocyclopropane-1-carboxylate synthase (ACC)	ethylene metabolism, stress response	syn	11[b]
m1194	cell division related protein (DnaJ and myb-like DNA-binding domain-containing protein)	cell division	syn	
m1196	peptidyl-prolyl cis-trans isomerase (PPI)	protein folding	syn	
m1211	unknown	unknown	unk	1[b]
m1414	auxin response factor 4 (ARF4)	auxin metabolism, plant growth and development	nc	

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[a] LG from Chancerel *et al*. (2011)

[b] LG from De Miguel *et al*. (2012)

[c] Site annotation: syn, synonymous; non-syn, non-synonymous; nc, non-coding (UTRs or introns); unk, unknown

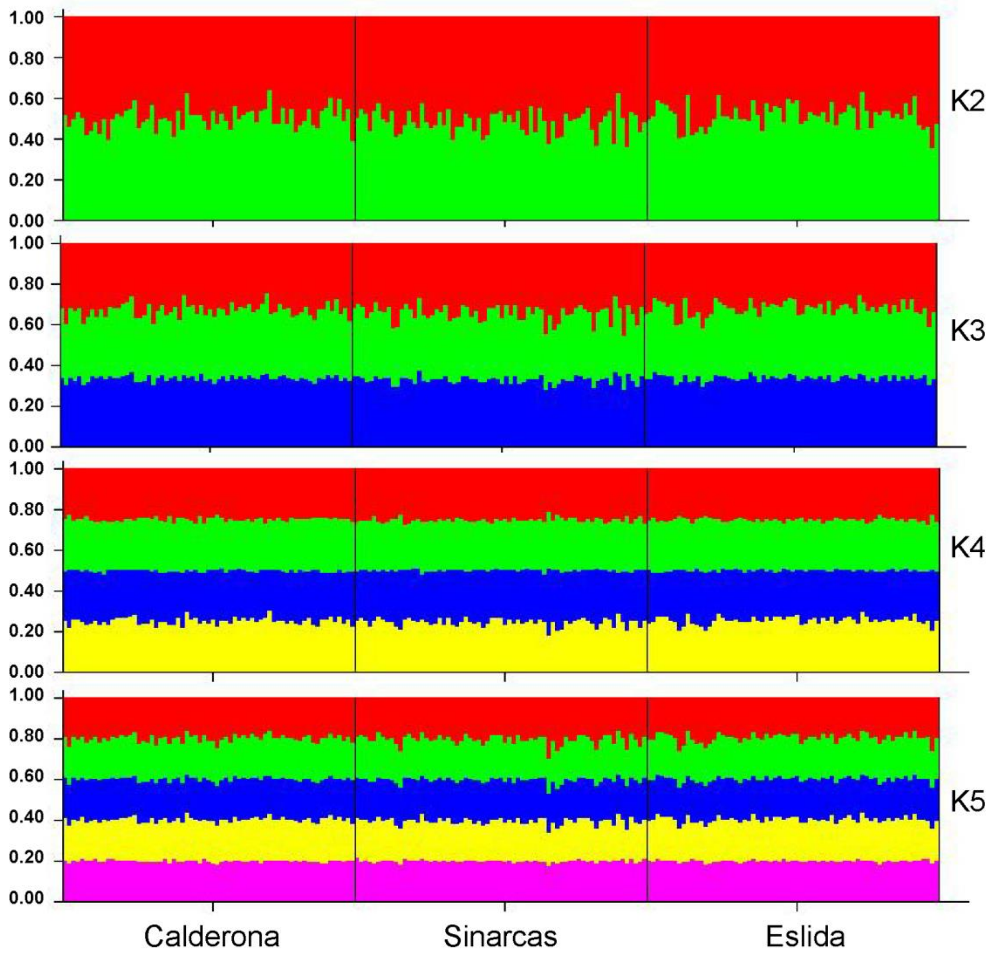

Figure S1 Bar plots showing ancestry proportions for *K*=2 to *K*=5 clusters (i.e., gene pools) as produced by STRUCTURE 2.2 software. Each individual is represented as a line segment which is vertically partitioned into *K* coloured components representing the individual's estimated proportions of ancestry in the *K* clusters. Approximately equal ancestry proportion for each gene pool in all individuals indicates lack of genetic structure.

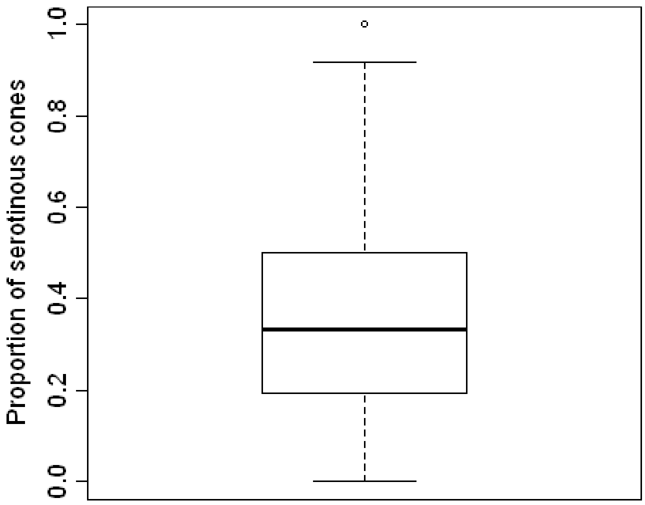

Figure S2 Phenotypic variability (box plot) for serotiny in eastern Iberian Peninsula (mean serotiny of 36.29% and standard deviation of 23.36%) based on 199 trees genotyped with SNP markers.

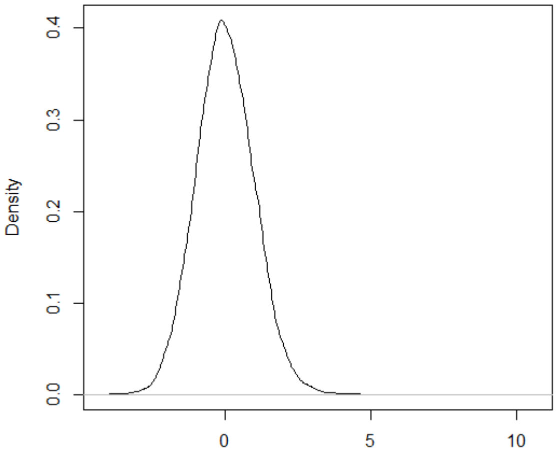

Figure S3 Density plot for pairwise kinship (as estimated by the Loiselle *et al*. [1995] kinship estimator implemented in SPAGeDi 1.3). Notice the positive skewness (D'Agostino's skewness test: skew = 0.208, z = 7.777, *P*-value = 7.387E-15) indicating some relatedness among trees within the studied stands.

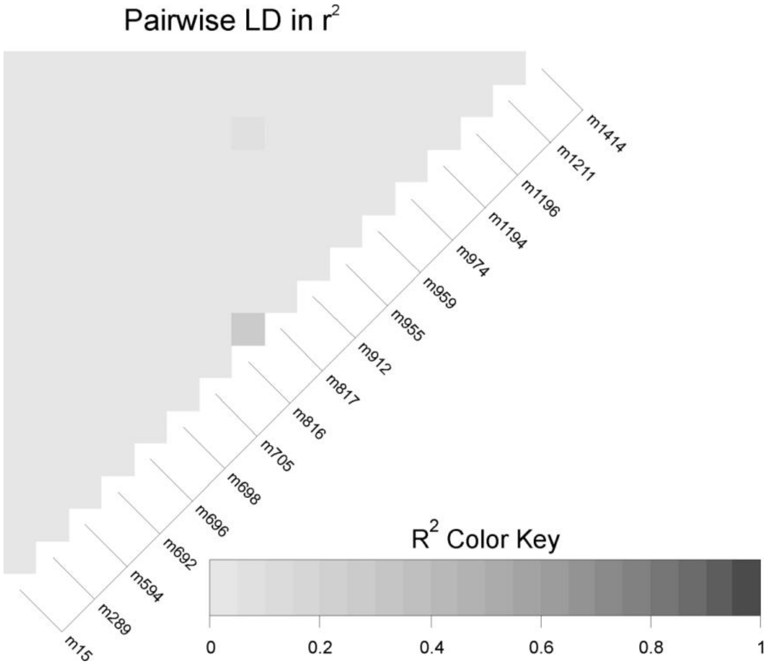

Figure S4 Linkage disequilibrium (LD) heatmap (LDheatmap, R software package) for the 17 SNPs potentially involved in associations with serotiny. Low, but significant, LD was detected between *m816* and *m817* (r^2=0.262), two SNPs from the same gene.

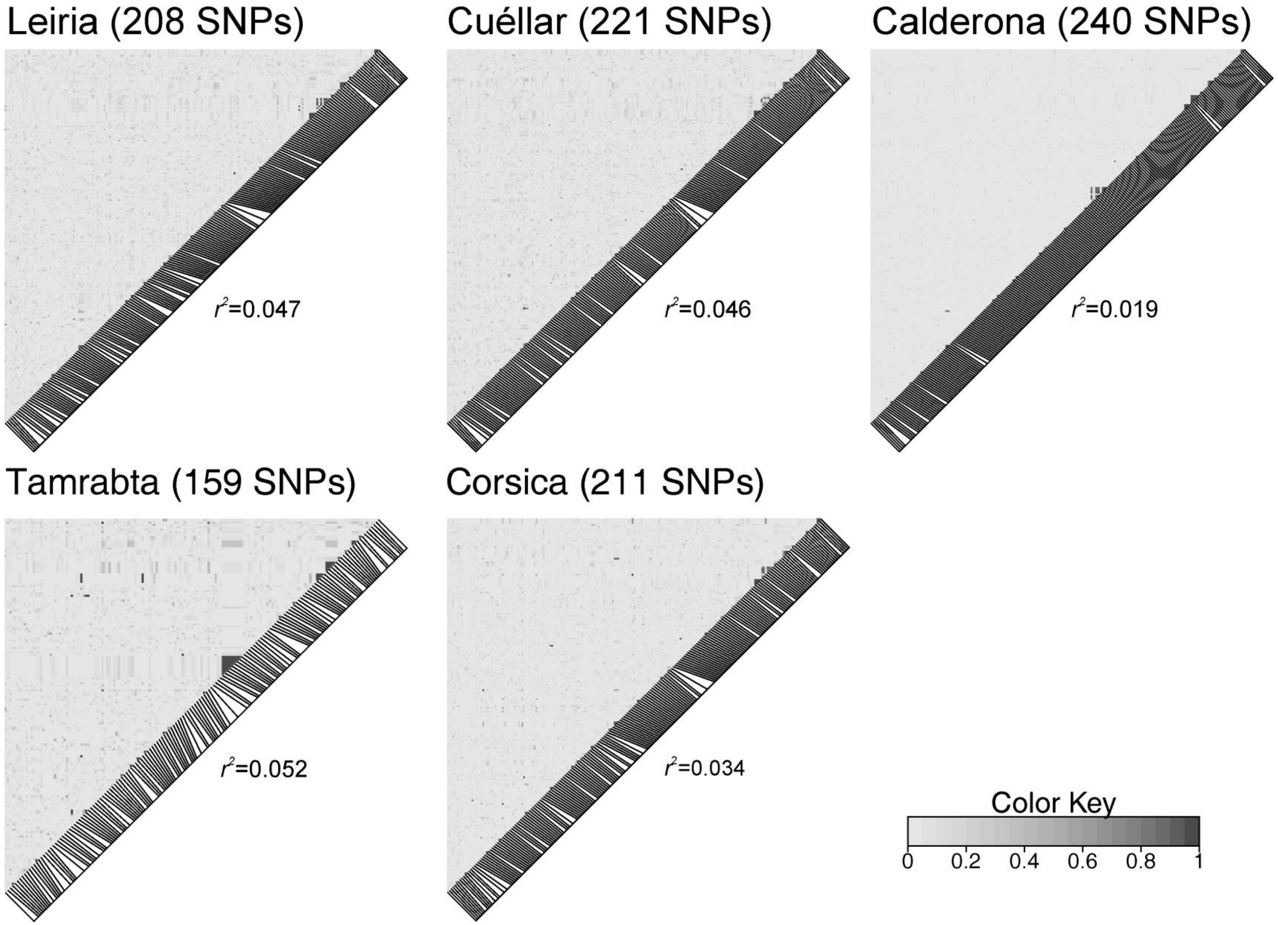

Figure S5 Linkage disequilibrium (LD) heatmaps (LDheatmap, R software package) for all polymorphic SNPs in four range-wide populations (Leiria: Portugal, Cuéllar: central Spain, Tamrabta: Morocco, and Corsica) and one stand (Calderona) from the focal population in the east of the Iberian Peninsula. For each population, the mean r^2 is given.