Polygenic adaptation and negative selection across traits, years and environments in a long-lived plant species (*Pinus pinaster* Ait.)

Marina de Miguel[†], Isabel Rodríguez-Quilón[‡], Myriam Heuertz[†], Agathe Hurel[†], Delphine Grivet[‡], Juan-Pablo Jaramillo-Correa[*], Giovanni G. Vendramin[§], Christophe Plomion[†], Juan Majada[††], Ricardo Alía[‡], Andrew J. Eckert[‡‡], Santiago C. González-Martínez[†**]

[†]INRAE, Univ. Bordeaux, BIOGECO, F-33610 Cestas, France

[‡] Department of Forest Ecology and Genetics, Forest Research Centre, INIA, Carretera de la Coruña km 7.5, 28040 Madrid, Spain

[*]Department of Evolutionary Ecology, Institute of Ecology, Universidad Nacional Autónoma de México, AP 70-275 México City, CDMX 04510, Mexico

[§]Institute of Biosciences and Bioresources, Division of Florence, National Research Council, 50019 Sesto Fiorentino (FI), Italy

[††]Sección Forestal, SERIDA, Finca Experimental ‘‘La Mata’’, 33820 Grado Principado de Asturias, Spain

[‡‡]Department of Biology, Virginia Commonwealth University, Richmond, VA 23284, USA

**Corresponding author

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Corresponding author:

Santiago C. González-Martinez

UMR 1202 INRAE – Univ. Bordeaux

69 rout d'Arcachon, F-33610 Cestas, France

Tel: +33 (0) 5 35 38 53 20

santiago.gonzalez-martinez@inrae.fr

Abstract

Results from a decade of association studies in different organisms suggest that most complex traits are polygenic, that is, their genetic architectures are determined by numerous causal loci distributed across the genome each with small effect-size. Thus, determining the degree of polygenicity is a central goal to understand the genetic basis of phenotypic variation. Recently, multi-loci methods able to detect variants associated with a phenotype of interest despite the subtle allele frequency changes between populations usually observed have been developed. In this study, we applied two multi-loci methods to estimate the degree of polygenicity of fitness-related traits in a long-lived plant species (maritime pine) and to analyze how polygenicity changes across years and environments. For this purpose, we evaluated five categories of fitness related traits, such as, height, survival, phenology-related, biotic-stress resistance and functional traits in a clonal common garden network planted in contrasted environments. Most of the analyzed traits showed evidences of local adaptation. We observed a remarkably stable degree of polygenicity (average 6%) across traits, environments and years. Additionally, some of the measured traits showed evidences of negative selection that could explain the observed degree of polygenicity, as previously suggested in humans. The observed genetic architecture of fitness-related traits in maritime pine is in accordance with the polygenic adaptation model. Because polygenic adaptation can take place rapidly, our results can contribute to improve the predictions about the capacity of natural populations of forest trees to adapt to new environments, which is of special relevance in the current context of climate change.

Introduction

Adaptive responses by populations to environmental changes depend on the genetic architecture of traits related to fitness (Hayward and Sella 2019). Thus, the identification of genetic variants affecting fitness-related traits, their allele frequencies, the magnitudes of their effects, and their interactions with one another and the environment has been a major focus of evolutionary studies for humans (reviewed by Visscher *et al.* 2017), other animals (e.g. Sharma *et al.* 2015; Pitchers *et al.* 2019) and plants (González-Martínez *et al.* 2006; Huang and Han 2014; Alonso-Blanco *et al.* 2016). In this regard, the information provided by genome-wide association studies (GWAS) was essential in clarifying the genetic basis of complex traits.

Over the past decade of GWAS for humans, a substantial number of genetic variants associated with phenotypes of interest have been identified (Visscher *et al.* 2017). Surprisingly, however, they accounted for only small fractions of trait heritabilities, as estimated through pedigree analysis, causing the so-called 'missing heritability' paradox (Maher 2008). While the 'missing heritability' paradox continues to be debated for model plant species (Brachi *et al.* 2011; Hamblin *et al.* 2011), it is expected to also apply to forest trees because of their large effective population sizes and outcrossing mating systems, which create genomic patterns of diversity mirroring those of humans (Resende *et al.* 2012; Lind *et al.* 2018). Several explanations of missing heritability have been identified (Manolio *et al.* 2009; Brachi *et al.* 2011; Björkegren *et al.* 2015; Pallares 2019). For instance, rare variants, routinely excluded from GWAS studies because of the associated lack of statistical power, can account for a considerable fraction of heritability (Bloom *et al.* 2019; Fournier *et al.* 2019). Epistasis may also be an important component for understanding the genetic architecture of adaptive phenotypes, but its contribution to non-additive genetic variance seems low and recent work argues that it is not a likely source of 'missing heritability' (Mäki-

74 Tanila and Hill 2014; Sella and Barton 2019). Epigenetics has also been identified as a
75 measurable component of 'missing heritability' (Cortijo *et al.* 2014; Koch 2014). Finally,
76 several sources of evidence point to polygenicity –trait architectures determined by a large
77 number of variants located in a large number of genes, each with small effect size - as a
78 potential reason for the low levels of heritability explained by GWAS results. First, estimates
79 of heritability based on all or at least extremely large sets of the single nucleotide
80 polymorphisms (SNPs) used in GWAS, regardless of the statistical significance of their
81 association to the phenotype of interest, are consistent with an explanation that most of the
82 heritable variance is linked to loci that are difficult to detect with realistic sampling designs
83 (Yang *et al.* 2010; Shi *et al.* 2016; Boyle *et al.* 2017). In addition, the proportion of the
84 heritability contained within chromosomes is strongly correlated to their length for many
85 traits (Shi *et al.* 2016), suggesting that causal variants are abundant and scattered throughout
86 the genome.

87 The study of adaptation has traditionally been addressed from contrasting research paradigms
88 (Höllinger *et al.* 2019). While quantitative genetic approaches viewed adaptation as the result
89 of changes in allele frequencies at an idealized and infinite number of loci each with
90 infinitesimal effects on fitness (Fisher 1918), population genetic approaches make more
91 emphasis in the detection of selective sweeps where new beneficial mutations rapidly became
92 fixed at a small number of loci (Smith and Haigh 1974). It was Pritchard *et al.* (2010) who
93 first brought together population and quantitative genetic theory with the conclusions from
94 GWAS to formulate a new framework for the study of adaptation – the polygenic adaptation
95 model. The hypothesis articulated under this model states that natural selection (mostly) acts
96 through subtle allele frequency shifts on standing genetic variation at numerous loci
97 distributed across the genome. Some genes may harbor new mutations that have been fixed by
98 natural selection, but the most common pattern would be the genome-wide increase of

99 favored alleles, without fixation for most of the causative alleles. Thus, the expected genome-
100 wide footprint resulting from natural selection would not be that of classical hard sweeps, but
101 would rather involve a large number of causal variants each with subtle allele frequency
102 changes (Pritchard *et al.* 2010; Pritchard and Rienzo 2010; Hermisson and Pennings 2017b).
103 This new evolutionary context may explain why GWAS have failed to explain the bulk of
104 heritable variance using methods designed to identify signatures from hard selective sweeps
105 that drive new mutations to fixation (Pritchard and Rienzo 2010; Stephan 2016) .

106 From the formulation of the polygenic adaptation model, a lot of work has been devoted to
107 assess how much adaptation occurs by any of the different models and mechanisms proposed.
108 New methods have been developed to detect the genetic signatures of natural selection under
109 the polygenic adaptation framework (Guan and Stephens 2011; Daub *et al.* 2013; Field *et al.*
110 2016; Zeng *et al.* 2018; Edge and Coop 2019; Speidel *et al.* 2019). Some of these methods
111 were based on the identification of coordinated changes in allele frequency at many loci
112 associated to a trait of interest, as identified by GWAS (Turchin *et al.* 2012; Berg and Coop
113 2014). Results from these latter methods had to be modulated and contextualized because they
114 were biased by population stratification (Liu *et al.* 2018; Josephs *et al.* 2019; Rosenberg *et al.*
115 2019; Berg *et al.* 2019a; Sohail *et al.* 2019). Nonetheless, even considering the inflation of the
116 polygenic signal due to neutral genetic structure, cumulating evidence in last years, using a
117 variety of methodologies, supports the polygenic adaptation model, mainly in humans
118 (Hancock *et al.* 2010a; b; Daub *et al.* 2013; Shi *et al.* 2016; Zeng *et al.* 2018; Gnecchi-
119 Ruscone *et al.* 2018; Berg *et al.* 2019b), but also in model plants (He *et al.* 2016), crops
120 (Josephs *et al.* 2019; Wisser *et al.* 2019) and forest trees (Lind *et al.* 2017; De La Torre *et al.*
121 2019b).

122 Strikingly, heritability of complex traits is not only widespread across the genome, but also
123 comprised in genes and pathways that do not show a clear connection with the trait (Boyle *et*

124 *al.* 2017). The omnigenic model, formulated by Boyle *et al*. in 2017, provides a plausible
125 hypothesis to explain this observation: the interconnection of gene regulatory networks
126 implies that virtually all the expressed genes in a relevant tissue likely influence the functions
127 of core genes that are directly relate with the phenotype. In this regard, recent work with
128 expression networks revealed that genes with higher connectivity experienced higher
129 intensities of selection and consequently presented higher variation, increasing the probability
130 of being detected by traditional GWAS (Mähler *et al.* 2017; Chateigner *et al.* 2019). In
131 addition, the study of polygenic adaptation at pathway level revealed that gene sets related
132 with resistance to pathogens in humans showed enriched signals of selection (Daub *et al.*
133 2013), highlighting its importance in shaping human adaptation. Polygenic adaptation at
134 pathway level has been shown as a useful approach to detected evidences of selection that
135 were unnoticed when using traditional single-locus approaches in a non-model tree species
136 (Mayol *et al.* 2020). By this approach, Mayol et al. (2020) found that most selective
137 signatures in *Taxus baccata* were driven by the effect of negative selection.

138 Unraveling the genetic architecture of adaptive traits allow to draw conclusions on the kind of
139 natural selection acting on them. The studies that addressed this question from a polygenic
140 adaptation perspective concluded that negative selection drove the observed genetic
141 differences between individuals (Zeng *et al.* 2018; O'Connor *et al.* 2019). This conclusion
142 was based on modeling SNP effect size conditional to MAF, which allowed not only to test
143 for evidences of selection but also to infer the type of action accordingly with the sign of the
144 detected correlation (i.e. positive or negative). Accordingly, negative selection has been
145 suggested to favor polygenicity of complex traits by removing large-effect variants, because
146 of their deleterious effect, while small-effects variants would remain unaffected (O'Connor *et*
147 *al.* 2019). Furthermore, unraveling the genetic basis of adaptive traits in forest trees is not
148 only useful to understand past adaptive processes but also to predict the future of forests under

149 climate change (Aitken *et al.* 2008). Polygenic adaptation is expected to produce rapid
150 changes in phenotypic variation because it requires only small changes in allele frequencies at
151 the numerous loci contributing to genetic variation (Hermisson and Pennings 2017b; Dayan *et*
152 *al.* 2019; Wisser *et al.* 2019). Consequently, considering information about the genetic
153 architecture of fitness-related traits can significantly improve the predictions of species range
154 shifts under climate change and inform forest management for conservation and reforestation
155 purposes (Valladares *et al.* 2014; Fady *et al.* 2016). Ultimately, a better understanding of the
156 genetic architecture of polygenic productive traits could improve the predictive accuracy of
157 genomic-assisted breeding, allowing to build genomic selection models in accordance with
158 the underlying genetic architecture of the trait of interest (Grattapaglia *et al.* 2018).

159 Despite the advances in theory and the development of new methods to study polygenic
160 adaptation, there still remains multiple open questions regarding the degree of polygenicity
161 for adaptive traits, the effect size distribution of these loci, and how the genetic architecture
162 changes under varying selective forces, specially for non-model species (Lind *et al.* 2018). In
163 this work, we addressed these questions taking advantage of the opportunity that plants offer
164 to replicate the genotypes through clonal propagation and investigate selective forces under
165 contrasting environments. We, i) tested the hypothesis that most complex adaptive traits for a
166 long-lived plant species, maritime pine (*Pinus pinaster* Ait.), are polygenic and 2) that their
167 genetic architecture is determined by the effect of negative selection, as it has been suggested
168 in humans (Zeng *et al.* 2018; O'Connor *et al.* 2019). We then, 3) tested how these patterns
169 change across years and environments in this iconic long-lived forest tree. Maritime pine has
170 nearly undomesticated random-mating populations with high genetic diversity (González-
171 Martínez *et al.* 2002; Jaramillo-Correa *et al.* 2015). It expanded from isolated glacial refugia
172 and now can be found in scattered populations, living under contrasting environments across
173 the occidental Mediterranean Basin and the European Atlantic front (Bucci *et al.* 2007),

174 making it an ideal case study to analyze the modes of evolution across traits, years and
175 environments.

176 **Materials and Methods**

177 *CLONAPIN clonal common garden network*

178 The *P. pinaster* clonal common garden network (CLONAPIN) is formed by five common
179 gardens covering the range of environments where the species is present, from harsh
180 Mediterranean climates to mild Atlantic ones. Overall, it comprises trees from 36 populations,
181 sampled across the species natural distribution range and covering the six distinct gene pools
182 previously identified for this species (Jaramillo-Correa *et al.* 2015; Supplemental Figure S1).
183 Open-pollinated seeds were collected in natural stands, germinated in a nursery and one
184 seedling per open-pollinated family was vegetatively propagated by cuttings (following
185 Majada *et al.* 2011). A total of 535 genotypes (clones) belonging to 35 populations were used
186 to establish four of the clonal trials (in Asturias, Cáceres, Madrid, and Portugal) with eight
187 ramets per clone set in a randomized complete block design (N=4,273 trees). These clonal
188 trials were planted in 2010 and constitute identical copies. In 2011, a fifth trial was
189 established in Bordeaux, comprising 443 clones from 36 populations (N=3,434 trees). Trials
190 in Asturias, Portugal and Bordeaux are located in the Atlantic region, with high annual
191 rainfall and mild temperatures. Trials in Madrid and Cáceres are located in continental areas
192 under Mediterranean influence, with large seasonal temperature oscillations and a marked
193 summer drought (Table 1). In addition, clay soils in Cáceres prevented maritime pine survival
194 and growth in this site.

195 *Phenotypic evaluation*

196 A total of 29 phenotypic trait-environment combinations were evaluated in this study.
197 Phenotypic traits considered can be classified in five groups: survival, height, phenology-
198 related traits, functional traits and biotic-stress response (see Supplemental Table S1 for an
199 exhaustive list of the measured traits). In brief, survival and height were evaluated in the five
200 common garden sites (including different years in Bordeaux, with measurements in 2013,
201 2015 and 2018). Phenology-related traits were evaluated in the Atlantic sites only (Asturias,
202 Bordeaux and Portugal), including different years of evaluation in Bordeaux (2015 and 2017).
203 In Asturias and Portugal, growth phenology was estimated through the presence of
204 polycyclism and a phenology growth index (1):

205 $$\text{Phenology Growth Index} = \frac{spring\ \text{growth}}{total\ \text{growth}} = \frac{(tree\ \text{height}\ may_n - tree\ \text{height}\ dec_{n-1})}{(tree\ \text{height}\ dec_n - tree\ \text{height}\ dec_{n-1})} \quad (1)$$

206 where *may* and *dec* correspond to the months May and December of the year n and the year n-
207 1, respectively.

208 In Bordeaux, phenology of bud burst was estimated through a scale ranging from 0 to 5 (0:
209 bud without elongation during winter, 1: elongation of the bud, 2: emergence of brachyblasts,
210 3: brachyblast begins to space, 4: elongation of the needles, 5: total elongation of the needles)
211 (see Hurel *et al.* 2020). The Julian day of entry in each stage (S1 to S5) was scored for each
212 tree. Julian days were converted into accumulated degree-days (0°C basis) from the first day
213 of the year, to take into account the between year variability in temperature.

214 Functional traits, including nitrogen and carbon amount and isotopic composition, as well as
215 Specific Leaf Area (SLA), were evaluated in the common garden trial located in Portugal.
216 SLA, a measure of leaf area per unit dry mass, is an estimation of the compromise between
217 light-capture and CO_2 assimilation and the restrictions imposed by water loss through
218 transpiration. Therefore, it is a key leaf trait conditioning fitness components, such as survival

219 (Greenwood *et al.* 2017). Since the establishment of the direct positive relationship between
220 $\delta^{13}C$ and water use efficiency (Farquhar and Richards 1984), $\delta^{13}C$ has been widely used as a
221 surrogate to study tree adaptation to water limiting environments (e.g. Aranda *et al.* 2010;
222 Walker *et al.* 2015). Complementary, $\delta^{15}N$ is an indirect indices of different nitrogen cycle
223 process (Craine *et al.* 2015). In this study, nitrogen and carbon amount did not show genetic
224 variation (i.e. heritability) and polygenic association methods failed to converge for these
225 traits. Thus, they were excluded for further analyses.

226 Biotic-stress response was evaluated based on susceptibility to two major pine pathogens,
227 *Diplodia sapinea* and *Armillaria Ostoyae*, based on an inoculation experiment at controlled
228 conditions and the incidence of a defoliator pest, the pine processionary moth (*Thaumetopoea*
229 *pityocampa*) (see Hurel *et al.* 2020 for details on biotic-stress response evaluation). *D.*
230 *sapinea* causes several diseases in conifers that may be exacerbated in the context of climate
231 change compromising the health of maritime pine forests (Desprez-Loustau *et al.* 2006). In
232 this study, susceptibility to *D. sapinea* was evaluated through two parameters: length of the
233 lesion around the inoculation (hereafter referred as necrosis) and a scalar notation of needle
234 discoloration caused by the inoculation (from 0: no discoloration to 3: all needles around the
235 necrosis showed discoloration). *A. ostoyae* is a root pathogen of conifers that causes growth
236 cessation and eventually leads to tree mortality (Heinzelmann et al., 2018). In this study, the
237 susceptibility to *A. ostoyae* was assessed through the length of the lesion in sapwood. Finally,
238 the pine processionary moth is an insect that rapidly defoliates pines conducing to forest
239 decay (Jacquet *et al.* 2013). Biotic-stress responses were only evaluated in the Bordeaux
240 common garden trial.

241 *DNA extraction and SNP genotyping*

242 Needles were collected from one ramet per clone in the Asturias trial (*N*=523). Genomic
243 DNA was extracted using the Invisorb® DNA Plant HTS 96 Kit/C kit (Invitek GmbH, Berlin,
244 Germany). A 9K *Illumina Infinium* SNP array developed by by Plomion et al. (2016b) was
245 used for genotyping. This array comprised SNPs previously identified as well as newly
246 developed *in silico* SNPs, either randomly screened from EST sequences or specifically
247 detected in candidate gene involved in adaptation to biotic and abitoc factors (see Plomion et
248 al. 2016 for further details). Genotyped SNPs covered the 12 chromosomes of *P. pinaster*
249 genome as revealed by linkage mapping (Plomion *et al.* 2016b). For this study, 6100 SNPs
250 were finally retained after standard filtering followed by removal of SNPs with uncertain
251 clustering patterns (visual inspection using *GenomeStudio v. 2.0*). This resulted in 5,167
252 polymorphic SNPs that were included in the polygenic association study.

253 *Quantitative genetics analysis*

254 Genetic components of the phenotypic variance were estimated using Generalized Linear
255 Mixed Effects Models (GLMM) fitted in a Bayesian framework using Markov Chain Monte
256 Carlo (MCMC) methods. Model (2) was implemented for those phenotypic traits evaluated in
257 multiple sites of the CLONAPIN common garden network. In addition, to estimate the genetic
258 control of the genotype per environment interaction (G×E term), model (3) was fitted for
259 those traits measured in all sites of the CLONAPIN common garden network (i.e. height and
260 survival).

261 $$y_{ijk} = \mu + S_i + S(B)_{ij} + P_k + P(C)_{kl} + S_i * C_l + \varepsilon_{ijkl} \quad (2)$$

262 $$y_{ijk} = \mu + S_i + S(B)_{ij} + P + P(C)_{kl} + S_i * P_l + S_i * C_l + \varepsilon_{ijkl} \quad (3)$$

263 where for any trait y, μ denotes de overall phenotypic mean, S_i represents the fixed effect of
264 the CLONAPIN common garden site i, B represents the random effect of experimental block j

265 nested within site i, P is the random effect of population k, C denotes the random effect of
266 clone l nested within population k and ε is the residual effect.

267 Models (4) or (5) were implemented for those phenotypic traits measured in just one site of
268 the CLONAPIN common garden network.

269 $$y_{ijk} = \mu + B_i + P_j + P(C)_{jk} + \varepsilon_{ijk} \quad (4)$$

270 $$y_{ijk} = \mu + B_i + cov + P_j + P(C)_{jk} + \varepsilon_{ijk} \quad (5)$$

271 where for any trait y, μ denotes de overall phenotypic mean, B_i represents the fixed effect of
272 experimental block i, P_j is the random effect of population j, C denotes the random effect of
273 clone k nested within population j and ε is the residual effect. In model (5), *cov* represents a
274 covariate implemented when modeling the presence of pine processionnary moth nets (i.e
275 height in 2015) and necrosis caused by *A. ostoyae* (i.e. a categorical evaluation of the level of
276 humidity in the experimental jar).

277 All models were fitted with the R package *MCMCglmm* (Hadfield 2010). Phenotypic traits
278 that showed a Gaussian distribution where modeled using the identity link function while
279 phenotypic traits that followed a binomial distribution (i.e. survival, polycyclism, *D. sapinea*
280 needle discoloration and pine processionary moth) were modeled either with *logit* or *probit*
281 link functions (see Supplemental Table S2 for an exhaustive list of model parameter
282 specifications). Multivariate-normal prior distribution with mean centered around zero and
283 large variance matrix (10^8) were used for fixed effects. For ordinal traits a Gelman prior for
284 the variance of fixed effects was set, as suggested by Gelman et al. (2008). Inverse Wishart
285 non-informative priors were used for the variances and covariances of random effects with a
286 matrix parameter V set to 1 and a parameter n set to 0.002 (Hadfield 2010). Parameter
287 expanded priors were used to improve the convergence and mixing properties of the chain as

288 suggested by Gelman (2006) for models with near-zero variance components. Parameter
289 estimates were not sensitive to change in the priors. The models were run for at least 550,000
290 iterations, including a burn-in of 50,000 iterations and a thinning interval of 100 iterations.
291 Four chains per model were run to test for parameter estimate convergence. Gelman-Rubin
292 criterion Potential Scale Reduction Factor (psrf) was consistently below 1.02 (Gelman and
293 Rubin 1992).

294 Variance components were then used to compute broad-sense heritability (H^2) as (6):

295 $$H^2 = \frac{\sigma^2_{clone}}{(\sigma^2_{clone}+\sigma^2_e)} \qquad (6)$$

296 where σ^2_{clone} is the variance among clones within populations and σ^2_e the residual variance.
297 For the estimation of broad-sense heritability in traits with binomial distribution (i.e. survival,
298 polycyclism, *D. sapinea* needle discoloration and pine processionary moth), we included an
299 extra term in the denominator ($+\pi^2/3$) to account for implicit *logit* link function variance and
300 $+1$ for probit link function (Nakagawa and Schielzeth 2010).

301 The GLMM described above were used to estimate the genetic value by means of Best Linear
302 Unbiased Predictors (BLUPs) (Henderson 1973; Robinson 1991) The genetic value of each
303 clone was defined as the population BLUP plus the clone BLUP. BLUPs of the G×E
304 component, obtained from model (3), were calculated as the difference between the mean
305 population BLUPs for Atlantic sites minus the mean population BLUPs for Mediterranean
306 sites, plus the difference between the mean clone BLUPs for Atlantic sites minus the mean
307 clone BLUPs for Mediterranean sites, following equation (7):

308 $$G\times E\ BLUP = \left(\frac{\sum BLUP_{pop}\ atl}{N\ atl} - \frac{\sum BLUP_{pop}\ med}{N\ med}\right) + \left(\frac{\sum BLUP_{clone}\ atl}{N\ atl} - \frac{\sum BLUP_{clone}\ med}{N\ med}\right) \qquad (7)$$

309 Parameter estimates from quantitative genetics analysis were presented as the mode of the
310 posterior distribution, and 95% credible intervals were computed as the highest density region
311 of the posterior parameter distribution.

312 *Q_{st}-F_{st} comparison*

313 Population neutral differentiation (F_{ST}) was estimated according to Weir and Cockerham
314 (1984) using the diveRsity package in R (Keenan *et al.* 2013) to calculate a global F_{ST} using
315 the 5,167 SNPs. The 95% confidence intervals of the global F_{ST} were computed by
316 bootstrapping. Quantitative genetic differentiation among populations was calculated as in
317 Spitze (1993), using the variance components estimated from the models previously described
318 (equations 2-5):

319 $$Q_{\mathrm{ST}} = \frac{\sigma^2_{pop}}{\sigma^2_{pop} + 2\sigma^2_{clone}} \qquad (8)$$

320 where σ^2_{pop} is the variance among populations and σ^2_{clone} is the variance among clones within
321 populations. If the 95% credible intervals obtained from the posterior distribution of Q_{ST} were
322 not overlapping with the 95% confidence intervals of F_{ST}, both estimates were considered
323 significantly different.

324 *Polygenicity across traits, years and environments*

325 Polygenicity was evaluated by estimating the number of SNPs with non-zero effects on
326 phenotypic traits and using two alternative methods, posterior inference via model averaging
327 and subset selection (VSR), as implemented in piMASS software (Guan and Stephens 2011),
328 and a Bayesian Mixed Linear Model (MLM) developed by Zeng et al. (2018) and
329 implemented in CGTB software. This latter model simultaneously estimates SNP-based
330 heritability (H^2_{SNP}, the proportion of trait variance explained by SNPs with non-zero effects),

331 polygenicity, and the relationship between SNP effect size and minor allele frequency (*S*), a
332 common indicator of negative selection. When negative selection is operating, *S* is expected
333 to be negative, as most new mutations are deleterious and thus SNPs with high-effect on
334 phenotypes are kept at low frequencies. Previous to analyses, neutral population genetic
335 structure was removed by running a simple linear model of the trait genetic values (i.e. with
336 site and block effects removed) on the Q factors obtained in a STRUCTURE run for K=6
337 based on neutral markers (see Jaramillo-Correa *et al.* 2015 for further details), and using the
338 normalized residuals, as recommended in piMASS manual.

339 Analyses were done separately for different traits, years and environments (see Supplemental
340 Table S1). VSR models were run for 2,000,000 iterations with a burn-in of 100,000 iterations
341 and a thinning interval of 100. After several preliminary runs, the maximum number of SNPs
342 included in VSR models was fixed to 2,000 (i.e. maximum polygenicity of ~40%). MLM
343 models were run for 500,100 iterations, including a burn-in of 100 iterations and a thinning
344 interval of 10 iterations. Parameter estimates from both VSRs and MLMs were presented as
345 the median of the posterior distribution, instead of the mode in order to better handle bimodal
346 distributions (Supplemental Figure S2). The 95% credible intervals were computed as the
347 highest density region of the posterior parameter distribution, as above.

348 **Results**

349 *Genetic differentiation among populations and broad-sense heritability*

350 Global F_{ST} was 0.112 [0.090, 0.141] (estimate and 95% confidence intervals). All five groups
351 of phenotypes evaluated in this study showed at least one trait with Q_{ST} higher than F_{ST}, except
352 for survival (Supplemental Table S1). The highest Q_{ST} was obtained for susceptibility to *D.*
353 *sapinea* infection measured as necrosis length in excised branches, followed by $\delta^{13}C$ and

354 height, which also showed very stable estimates of Q_{ST} measured in different environments or
355 at different tree ages (Figure 1).

356 All traits had moderate to low broad-sense heritability (Supplemental Table S1). The highest
357 H^2 was observed for phenology-related traits and height. Heritability estimated from the
358 polygenic models (H^2_{SNP}) was consistent between the two methods, although VSR method
359 tended to render higher heritability values (Supplemental Table S3). On average H^2_{SNP} was
360 0.38 across traits, with a minimum of 0.018 for survival in the French Atlantic environment in
361 2018 and a maximum of 0.99 for *D. sapinea* necrosis.

362 *Genetic architecture (polygenicity)*

363 Polygenicity estimates, i.e. the number of SNPs with non-zero effects on phenotypes, were
364 consistent between VSR and MLM methods (Supplemental Table S3). Both methods showed
365 substantial polygenic control for most of the study traits with an average of 6% and up to 27%
366 for the upper boundary of $\delta^{15}N$ credible interval (Figure 2). Polygenicity for height was stable
367 across environments and years, when measured multiple times in the same environment (i.e.
368 French Atlantic common garden) (Figure 3). In the same line, polygenicity for phenology-
369 related traits and survival also remained stable across environments, although 95% credible
370 intervals overlapped zero for some of them. The low values of polygenicity observed for
371 survival in the French Atlantic common garden are probably a consequence of the low levels
372 of genetic variability in this site (almost no mortality, with 97.12% of trees remaining alive,
373 Supplemental Table S1). Polygenicity estimated for biotic-stress response and functional traits
374 varied depending on the studied trait (Figure 2). *D. sapinea* susceptibility presented higher
375 levels of polygenicity than *A. ostoyae* susceptibility and pine processionary moth presence.
376 For functional traits, SLA and $\delta^{15}N$ showed the highest levels of polygenicity while $\delta^{13}C$
377 showed a considerably lower number of SNPs with non-zero effects.

378 Polygenicity and H^2_{SNP} were positively and consistently correlated for both VSR and MLM
379 models (Figure 4; R^2=0.59 for VSR and R^2=0.62 for MLM, p<0.001). A positive correlation
380 between polygenicity and H^2_{SNP} suggests that heritability is mainly determined by small effect
381 genetic variants in a high number of genes, according to the polygenic genetic architecture of
382 the analysed phenotypes. H^2_{SNP} estimated by the VSR method on the G×E component of
383 height (considering Atlantic versus Mediterranean environments) was low but significant
384 (median = 0.238, 95% credible interval = 0.043-0.409), indicating some heritable component
385 of plasticity. However, this result could not be confirmed by the MLM method. Moreover,
386 H^2_{SNP} was not significant for the G×E component of survival using any of the models.

387 *Evidence of negative selection*

388 The kind of natural selection acting on the studied traits was evaluated through the correlation
389 between SNP effect size and minor allele frequency (MAF). Out of the 29 analyzed traits, we
390 were able to estimate *S* through the MLM method for 19 of them. The estimates of *S* over
391 traits ranged from -1.68 (bud burst in 2017) to 0.55 (survival in French Atlantic environment).
392 Seven traits involving four out of the five trait groups (i.e. height, survival, phenology-related
393 and functional traits) had significant *S* estimates (Figure 5). No significant effect of selection
394 was observed for any trait belonging to the biotic stress category. Remarkably, all the
395 significant estimates of *S* were negative indicating negative selection.

396 Estimates of *S* for height were consistent across years and environments. However, the impact
397 of negative selection on survival could only be observed in the Iberian Atlantic environment
398 and for phenology-related traits, only in bud burst measured in 2017 (Figure 5). These results
399 contrast with the remarkably stable degree of polygenicity for all these traits across years and
400 environments. Interestingly, our results suggest a stable polygenic architecture but an
401 environment and year-dependent impact of negative selection for some traits.

402 **Discussion**

403 Unraveling the genetic architecture of complex polygenic traits has been a challenge for
404 association studies because of the difficulty to detect variants of low effect. Here, we
405 addressed this question for an extensive number of traits belonging two five different
406 categories related with fitness and adaptation to abiotic and biotic stresses (i.e. height,
407 survival, phenology-related, biotic-stress response and functional traits). These traits were
408 measured in 36 range-wide populations of maritime pine clonally replicated (i.e. in total more
409 than 12,500 trees were evaluated) which allowed precise phenotypic estimations. In addition,
410 traits were evaluated in three different environments (i.e. French Atlantic, Iberian Atlantic and
411 Mediterranean), representative of different environmental conditions of the distribution range
412 of maritime pine, and for consecutive years for some of them (Supplemental Figure S1).
413 First, we assessed the effect of natural selection on the measured traits and we identified local
414 adaptation for most of them. Then, we estimated the degree of polygenicity that resulted
415 remarkably stable across traits, years and environments. Finally, we assessed the effect of
416 natural selection in the variants associated with these traits. This test revealed the impact of
417 negative selection in the analyzed traits.

418 *Evidence of local adaptation in maritime pine*

419 Understanding the causes and consequences of population differentiation is of broad interest
420 in evolutionary biology, ecology and genetics and provide essential knowledge to inform
421 conservation biology, breeding programs and medical applications (Leinonen *et al.* 2013).
422 Q_{ST}-F_{ST} comparison has been a useful approach to discern if population differentiation is
423 caused by selective versus neutral processes (Savolainen *et al.* 2007; Leinonen *et al.* 2008;
424 Whitlock 2008). Through this approach the amount of genetic differentiation among
425 populations relative to the total genetic variance at neutral marker loci (F_{ST}) is compared with

426 the analogous parameter Q_{ST} that estimates the amount of genetic variance between
427 populations for quantitative traits. Therefore, if $Q_{ST} \approx F_{ST}$ trait divergence among populations
428 could have been achieved by genetic drift alone, if $Q_{ST} > F_{ST}$ divergent selection would be
429 driving local adaptation and the action of uniform selection is suggested from $Q_{ST} < F_{ST}$
430 (Whitlock 1999; Whitlock and Guillaume 2009). Our results show evidences of adaptive
431 differentiation ($Q_{ST} > F_{ST}$) for most of the traits analysed in this study, involving four out of
432 the five trait categories (except survival). These results are in accordance with the pervasive
433 observation of locally adapted traits in forest trees (Savolainen *et al.* 2007, 2013; Alberto *et*
434 *al.* 2013; Lind *et al.* 2018). It was remarkable the high amount of adaptive genetic
435 differentiation found for $\delta^{13}C$ and *D. sapinea* necrosis (Figure 1). These traits deserve special
436 relevance because of the implication of water use efficiency in drought resistance (reviewed
437 by Plomion *et al.* 2016a) and the concern of disorders caused by *D. sapinea* as important
438 emerging diseases for maritime pine (Fabre *et al.* 2011; Brodde *et al.* 2019). Lamy et al.
439 (2011) did not find adaptive genetic differentiation between populations of maritime pine for
440 $\delta^{13}C$ and similar results have been reported in broad-leaved trees (Torres-Ruiz *et al.* 2019).
441 Although, this disagreement may be influenced by the higher number of populations analyzed
442 in this study compared to Lamy et al. (2011), we cannot discard the overestimation of genetic
443 differences because of the fact that we estimated total genetic variance instead of additive
444 genetic variance (analyzed trees from each population were clonal replicates). The study of
445 adaptive genetic differentiation in biotic-stress response traits has not been fully addressed in
446 trees yet (but see Ahrens *et al.* 2019). Anyway, the high adaptive genetic differentiation for
447 $\delta^{13}C$ and *D. sapinea* susceptibility suggest that maritime populations will likely require to
448 match their new environments in the context of climate change.

449 Survival is a trait directly related with a component of fitness (i.e. viability) and contrarily to
450 our results previous evidence of local adaptation for this trait in maritime has been obtained

451 (e.g. González-Martínez *et al.* 2002). Local adaptation for fitness has been also observed in
452 model plant species (e.g. Fournier-Level *et al.* 2011). The low levels of phenotypic variability
453 observed for survival in this study may explain these contrasting results. In the most favorable
454 environment, that is the French Atlantic common garden (Bordeaux, France), almost all trees
455 remained alive. In the most unfavorable Mediterranean environments, a high percentage of
456 trees were dead after the first year of plantation. The strong selective pressure in the
457 Mediterranean region exacerbated the genetic differences among clones and consequently a
458 higher heritability was estimated but quantitative genetic differentiation between populations
459 was not adaptive. Another plausible explanation is that, in this work, survival was measured
460 as a binary trait. Future studies should focus on quantitative evaluations of survival in order to
461 better gather the complexity of this trait and be able to discern genetic differences between
462 populations.

463 Height also showed high levels of adaptive genetic differentiation that were stable across
464 different environments and years (Figure 1). So, for this trait we did not find evidences of
465 increased population differentiation under more stressful environments. Although adaptive
466 genetic differentiation for height has been broadly described in maritime pine and other tree
467 species (e.g. Yang *et al.* 1996; González-Martínez *et al.* 2002; Lamy *et al.* 2011; Rodríguez-
468 Quilón *et al.* 2016; Bogdan *et al.* 2017; Ahrens *et al.* 2019), the question of the environment
469 and temporal variability of this parameter has not been addressed in depth (but see Rodríguez-
470 Quilón *et al.* 2016). Nevertheless, phenology-related traits showed contrasting estimates of
471 Q_{ST} depending on the environment and year of measurement. Phenology-related traits in trees
472 usually display local adaptation (Vitasse *et al.* 2009; Csilléry *et al.* 2014; Firmat *et al.* 2017;
473 Torres-Ruiz *et al.* 2019) prompting its susceptibility to climate change. However, high
474 phenotypic plasticity has been also detected for phenology, highlighting its potential for short-
475 term survival under climate change (Firmat *et al.* 2017). Our results highlight that the

476 evolutionary forces driving population genetic differences in phenology-related traits are
477 environmentally and temporally-dependent which can slow-down the achievement of new
478 phenotypic optima under rapidly changing environmental conditions. Our experimental
479 design, with multiple clonal replicates and contrasting trial environments should have help to
480 overcome some of the drawbacks assigned to Q_{ST}-F_{ST} comparisons (Whitlock 2008) .

481 *Insights on forest tree missing heritability*

482 SNP-based heritability is usually defined as the proportion of phenotypic variance attributable
483 to the additive effects of a given set of SNPs and it is becoming a fundamental parameter in
484 quantitative genetics (Hou *et al.* 2019). We were able to estimate H^2_{SNP} though independent
485 methodologies (VSR and MLM, see below). Despite some slight differences, notably for
486 biotic-stress response traits that were limited by low sample size, both methods presented
487 robust and consistent estimations. In this study, SNP-based heritability was calculated for the
488 genetic component of the expressed phenotype (i.e. the clonal BLUPs extracted from the
489 quantitative genetic models). Thus, H^2_{SNP}, as estimated here, should be interpreted as the
490 proportion of genetic variance explained by the additive effects of the SNPs with non-zero
491 effects. The high proportions of the genetic variance explained in this way, usually higher
492 than 50%, suggest that by adopting a polygenic analytical framework we were able to account
493 for a big part of the missing heritability commonly observed in GWAS in forest trees
494 (reviewed by Lind *et al.* 2018).

495 However, these results should be interpreted with caution as *P. pinaster* is a species with a
496 huge genome size of 28 Gbp (Grotkopp *et al.* 2004; Zonneveld 2012) and a rapid decay in
497 linkage disequilibrium (Neale and Savolainen 2004), which would need a large number of
498 genotyped SNPs to obtain a good coverage of its genome. Even though further investigations
499 are needed to draw definitive conclusions, robust and consistent estimates of polygenicity

500 across methods (see Discussion below), and with previous estimates in humans (Zeng *et al.*
501 2018), may have been fostered by precise phenotypic evaluation using a clonal common-
502 garden network covering the species environmental range. Denser genome-wide genotyping
503 in conifers, that have extremely large genomes, should be feasible rapidly as new technologies
504 such as genotyping-by-sequencing, sequence capture and direct sequencing are currently
505 being developed for these species (Neves *et al.* 2013; Pan *et al.* 2015; Vidalis *et al.* 2018).
506 The availability of new genomic resources for conifers will allow advancing our
507 understanding of polygenicity.

508 *Genetic architecture of adaptive traits*

509 To increase our knowledge on the genetic basis of phenotypic variation is crucial for the
510 understanding of species adaptive evolution. Disentangling the genetic architecture of fitness-
511 related traits allows to make inferences on the action of natural selection on traits that drive
512 evolutionary change (Fisher 1930). The development of GWAS in the last decade has
513 provided a powerful tool to unravel the genetic architecture of complex traits in humans
514 (Visscher 2008; Shi *et al.* 2016; Visscher *et al.* 2017), plants (Brachi *et al.* 2011; Li *et al.*
515 2016; De La Torre *et al.* 2019a) and animals (Karlsson *et al.* 2007; Charlier *et al.* 2008; Yang
516 *et al.* 2016), but it is not yet widespread in long-lived forest trees (but see, for example, De La
517 Torre *et al.* 2019a). The conclusions inferred from these studies motivated the development of
518 a new evolutionary framework conceptualized in the polygenic adaptation model (Pritchard *et*
519 *al.* 2010; Pritchard and Rienzo 2010). This model states that adaptation to new environments
520 results from simultaneous selection at many loci and standing genetic variation. Although the
521 expected signatures of polygenic adaptation are well-known, still little is known about how
522 genetic architecture varies across age (i.e. ontogenetic effects) and environments (see below).

523 In our study, we have used two complementary Bayesian methods that allowed us to evaluate
524 the robustness of our estimates. The VSR method (Guan and Stephens 2011) is a multi-SNP
525 analysis whose potential lies in the estimation of the overall percentage of explained
526 phenotypic variance (PEV) that might be accounted for by relevant SNPs, even when
527 confidently identifying this relevant SNPs is not possible. The MLM method (Zeng *et al.*
528 2018) estimates a SNP-based heritability as an analogous parameter of the overall PEV
529 estimated through VSR. Both methods showed a considerable degree of polygenicity for most
530 traits, 4-15% for those traits with polygenicity estimates no overlapping zero, in the same
531 order of magnitude as for humans when using the same methods (MLM, Zeng *et al.* 2018),
532 although we obtained higher credible intervals because of lower sample size. Considering that
533 *P. pinaster* genome size is more than seven times higher than that of the human genome (De
534 La Torre *et al.* 2014), our results suggest that genome size is not conditioning the genomic
535 architecture that underlie fitness-related traits. This observation contrast with the hypothesis
536 proposed by Mei *et al*. (2018), that predicts different genetic architectures as a function of
537 genome size.

538 Recent studies have suggested that polygenicity estimates for human height in Europe, one of
539 the clearest example for polygenic adaptation, are affected by subtle biases in GWAS caused
540 by population stratification (Berg *et al.* 2019a; Sohail *et al.* 2019). In our study, we devoted
541 special care to correct for the effect of neutral population genetic structure on phenotypic
542 variation. In addition, the clonal common garden network allowed us to thoroughly identify
543 the traits that have diverged due to adaptation and to separate the genetic and the
544 environmental effect on phenotypes. Upcoming polygenic association methods that consider
545 corrections for population stratification (e.g. Josephs *et al.* 2019) should be implemented in
546 different plant species with varied life-history traits in order to elucidate the mechanisms that
547 allow the maintenance of variation in adaptive traits for species with complex demographic

548 history. In this sense, our work highlights the potential of combining precise estimation of the
549 genetic effect on phenotypes with multi-locus genotype-phenotype association models.

550 The implementation of polygenic adaptation studies outside humans is gently coming out in
551 the last years (Csilléry *et al.* 2014; He *et al.* 2016; Lind *et al.* 2017; Barghi *et al.* 2019; Wisser
552 *et al.* 2019), providing increased evidence that polygenic adaptation through complex traits
553 may be pervasive and a key mode of adaptation (Sella and Barton 2019). As a result, new
554 evolutionary questions relevant for different organisms arise. In forest trees, for which local
555 adaptation is frequently observed (Savolainen *et al.* 2007, 2013; Alberto *et al.* 2013; this
556 study), how alleles of small effect and selection coefficients (and therefore more prompted to
557 be swamped by gene flow) can contribute to shape local adaptation is a question that remains
558 to be solved (Yeaman 2015). A fundamental question that arise from the application of
559 polyenic adaptation studies to conifers is the role of genetic redundancy in shaping this mode
560 of adaptation. It has been suggested that genetic redundancy favor polygenic adaptation in
561 model species (Höllinger *et al.* 2019; Barghi *et al.* 2019) and to unravel this relationship in
562 conifers, whose genomes are characterized by a high number of paralogs (Diaz-Sala *et al.*
563 2013), may shed-new light in this fundamental question.

564 *Stability of polygenicity across environments and years*

565 The temporal and spatial heterogeneity of selection can impact the evolution of genetic
566 architectures underlying adaptation (Sella and Barton 2019). Using a range wide maritime
567 pine collection replicated clonally in multiple common gardens, we were able to test whether
568 polygenicity varies across environments (Mediterranean vs. Atlantic) for traits related to
569 growth, survival and phenology. In this sense, our study is not only a validation of the
570 polygenic adaptation framework in a new organism but it also contributes to improve our
571 understanding of this kind of adaptation. Surprisingly, the estimated degree of polygenicity

572 remained stable across different environments for all trait categories analyzed, especially for
573 height (Figure 3). The genetic architecture of height, phenology and survival was also highly
574 stable across years. Patterns of genetic architecture across years are an important issue in
575 long-lived forest trees that may experience changing selection pressure along their lifetime. In
576 conifers, previous genotype-phenotype association analyses (using single-locus models) have
577 revealed different genes underlying the expression of phenology-related traits across years
578 (Pelgas *et al.* 2011; Hurel *et al.* 2020). Our study suggests that even if the causal variants
579 underlying adaptive traits may vary across years, the estimated degree of polygenicity remains
580 stable. In the context of climate change, the stability of polygenicity is an important
581 consideration because species experience rapid and abrupt deterioration of environmental
582 conditions (Seidl *et al.* 2017) and polygenic adaptation is expected to produce rapid changes
583 in phenotypic variation (Hermisson and Pennings 2017a; Jain and Stephan 2017; Dayan *et al.*
584 2019; Wisser *et al.* 2019).

585 Strikingly, the impact of selection did not mimicked the trend observed for polygenicity. That
586 is, the impact of negative selection remained stable for height but changed across
587 environments and years for survival and phenology-related traits. Our results highlight that
588 even when the impact of natural selection on SNPs was not stable, the degree of polygenicity
589 remained unchanged. This pattern has to be validated with more genotyped SNPs, specially
590 rare SNPs that can considerably affect the estimates of S (Zeng *et al.* 2018). Moreover, some
591 traits presenting evidences of negative selection did not show adaptive genetic differentiation
592 between populations (inferred from Q_{ST} - F_{ST} comparisons; for example: survival in Iberian
593 Atlantic environment, bud burst in 2017 and SLA) and viceversa (for example duration of bud
594 burst in 2017, growth phenology index and *D. sapinea* necrosis). Thus, our results suggest an
595 uncoupling of the impact of natural selection on some phenotypes and on the identified causal
596 variants, a question that deserve further investigation. It has been already suggested that

597 polygenic adaptation can accelerate differentiation between populations but generally does
598 not produce predictable directional changes in allele frequencies and the genetic response can
599 be highly stochastic (Zhang *et al.* 2013).

600 *The role of negative selection*

601 All the significant correlations between SNP effect size and frequency were negative (for
602 height, bud burst and SLA). These results are in accordance with a genetic architecture
603 designed by the action of negative selection: SNPs with large effects are rare because they
604 mostly have deleterious effects and thus are selected against (O'Connor *et al.* 2019). The
605 MLM method does not allow to elucidate whether the negative estimates of *S* are the
606 consequence of an enrichment of trait-increasing or -decreasing alleles (Zeng *et al.* 2018) but
607 it certainly suggests that these traits (height, bud burst and SLA) have been under negative
608 selection. The effect of purifying selection is widespread in model plant genomes (Wright and
609 Andolfatto 2008) and it has been also largely evidenced in trees (Krutovsky and Neale 2005;
610 Palmé *et al.* 2009; Eckert *et al.* 2013; De La Torre *et al.* 2017). Indeed, negative selection has
611 been pointed out as the main cause for the maintenance of polygenicity (Zeng *et al.* 2018;
612 O'Connor *et al.* 2019).

613 Negative selection can explain the degree of polygenicity observed in maritime pine adaptive
614 traits, as well as the absence of large-effect SNPs in previous GWAS for the species . In
615 addition, the omnigenic model (Boyle *et al.* 2017) provides a complementary reasoning to
616 explain highly polygenic architectures in maritime pine. A large number of causal SNPs in
617 this species could be explained by the very high biological complexity and interconnectivity
618 of gene expression networks, resulting in the association of virtually all expressed genes in
619 relevant tissues with the observed phenotypes (Wray *et al.* 2018).

620 **Conclusions**

621 The study of genetic adaptation is currently facing new challenges. The advancement of
622 GWAS relies on the development of methods able to detect causal variants of small effect size
623 or at low allele frequency. In our study, adopting a polygenic adaptation framework on well-
624 characterized clones planted in contrasted environments, contributed to a better understanding
625 of the heritability of complex adaptive traits and its underlying genetic architecture.
626 Moreover, we addressed the question of how genetic architecture varies over spatial and
627 temporal heterogeneity. Our results showed that most complex adaptive traits in a long-lived
628 plant species, such as maritime pine, are polygenic, with several of them showing also
629 signatures of negative selection. The degree of polygenicity was similar for traits spanning
630 different functional categories, and this genetic architecture was considerably stable over time
631 and environments. Current models to predict population trajectories of forest trees under
632 climate change are based on identification of SNP outliers with relatively large effects on
633 phenotypes and/or strong correlation with climate variables (e.g. Jaramillo-Correa *et al.* 2015;
634 Rellstab *et al.* 2016). Because polygenic adaptation can take place rapidly (see, for example,
635 Jain and Stephan 2017), current prediction models are probably underestimating the capacity
636 of natural populations of forest trees to adapt to new environments. Thus, adopting a
637 polygenic adaptation perspective could significantly improve the accuracy of species range-
638 shift predictions under climate change and provide new scenarios to inform forest
639 conservation and reforestation programs.

640 **Supplemental material**

641 **Table S1. Data summary on measured phenotypes**. Vg stands for genetic variance
642 (posterior mean of the variance explained by clone effect), H2 stands for broad-sense
643 heritability and *Q*st for genetic differentiation among populations (posterior mode and 95%
644 credible interval are presented).

645 **Table S2 . MCMCglmm Bayesian model parametrization**. Psrf stands for Gelman-Rubin
646 criterion Potential Scale Reduction Factor, a mesure of model convergence. Good
647 convergence of models is expected for psrf <1.02.

648 **Table S3. Parameter results from two multi-locus association studies: Bayesian variable**
649 **selection regression (implemented in piMASS software) and Bayesian linear mixed**
650 **model (implemented in GCTB software).** The parameters are presented as the posterior
651 median and 95% credible interval for SNP-based heritability (H2SNP), number of non-zero
652 effect SNPs (polygenicity) and coefficient of correlation between SNP effect size and minor
653 allele frequency (*S*). Non-overlapping zero estimates are marked in bold. NA values for MLM
654 models correspond to models that did not converge.

655 **Figure S1. Sampled *Pinus pinaster* populations (circles) and common garden trial sites**
656 **(other symbols).** Neutral gene pools (identified in Jaramillo-Correa *et al.* 2015) outline the
657 species natural distribution range in different colors.

658 **Figure S2. Posterior distributions of the number of non-zero SNPs for 26 traits**
659 **belonging to five categories: height, survival, phenology-related traits, biotic-stress**
660 **response and functional traits.** The number of non-zero SNPs was estimated through two
661 Bayesian methods: posterior inference via model averaging and subset selection (VSR)
662 implemented in the software piMASS (Guan and Stephens 2011) and Mixed Linear Model
663 (MLM) implemented in the software CGTB (Zeng *et al.* 2018). The posterior median is
664 indicated with a dashed line.

665

666 **Literature cited**

667 Ahrens C. W., R. A. Mazanec, T. Paap, K. X. Ruthrof, A. Challis, *et al.*, 2019 Adaptive
668 variation for growth and resistance to a novel pathogen along climatic gradients in a
669 foundation tree. Evolutionary Applications 12: 1178–1190.
670 https://doi.org/10.1111/eva.12796

671 Aitken S. N., S. Yeaman, J. A. Holliday, T. Wang, and S. Curtis-McLane, 2008 Adaptation,
672 migration or extirpation: climate change outcomes for tree populations. Evolutionary
673 Applications 1: 95–111. https://doi.org/10.1111/j.1752-4571.2007.00013.x

674 Alberto F. J., S. N. Aitken, R. Alía, S. C. González-Martínez, H. Hänninen, *et al.*, 2013
675 Potential for evolutionary responses to climate change – evidence from tree
676 populations. Global Change Biology 19: 1645–1661.
677 https://doi.org/10.1111/gcb.12181

678 Alonso-Blanco C., J. Andrade, C. Becker, F. Bemm, J. Bergelson, *et al.*, 2016 1,135 Genomes
679 Reveal the Global Pattern of Polymorphism in Arabidopsis thaliana. Cell 166: 481–
680 491. https://doi.org/10.1016/j.cell.2016.05.063

681 Aranda I., R. Alía, U. Ortega, Â. K. Dantas, and J. Majada, 2010 Intra-specific variability in
682 biomass partitioning and carbon isotopic discrimination under moderate drought stress
683 in seedlings from four Pinus pinaster populations. Tree Genetics & Genomes 6: 169–
684 178. https://doi.org/10.1007/s11295-009-0238-5

685 Barghi N., R. Tobler, V. Nolte, A. M. Jakšić, F. Mallard, *et al.*, 2019 Genetic redundancy
686 fuels polygenic adaptation in Drosophila. PLOS Biology 17: e3000128.
687 https://doi.org/10.1371/journal.pbio.3000128

688 Berg J. J., and G. Coop, 2014 A Population Genetic Signal of Polygenic Adaptation, (M. W.
689 Feldman, Ed.). PLoS Genetics 10: e1004412.
690 https://doi.org/10.1371/journal.pgen.1004412

691 Berg J. J., A. Harpak, N. Sinnott-Armstrong, A. M. Joergensen, H. Mostafavi, *et al.*, 2019a
692 Reduced signal for polygenic adaptation of height in UK Biobank, (M. Nordborg, M.
693 I. McCarthy, M. Nordborg, N. H. Barton, and J. Hermisson, Eds.). eLife 8: e39725.
694 https://doi.org/10.7554/eLife.39725

695 Berg J. J., X. Zhang, and G. Coop, 2019b Polygenic Adaptation has Impacted Multiple
696 Anthropometric Traits. bioRxiv 167551. https://doi.org/10.1101/167551

697 Björkegren J. L. M., J. C. Kovacic, J. T. Dudley, and E. E. Schadt, 2015 Genome-Wide
698 Significant Loci: How Important Are They? J Am Coll Cardiol 65: 830–845.
699 https://doi.org/10.1016/j.jacc.2014.12.033

700 Bloom J. S., J. Boocock, S. Treusch, M. J. Sadhu, L. Day, *et al.*, 2019 Rare variants
701 contribute disproportionately to quantitative trait variation in yeast, (C. R. Landry, and
702 N. Barkai, Eds.). eLife 8: e49212. https://doi.org/10.7554/eLife.49212

703 Bogdan S., M. Ivanković, M. Temunović, M. Morić, J. Franjić, *et al.*, 2017 Adaptive genetic
704 variability and differentiation of Croatian and Austrian Quercus robur L. populations
705 at a drought prone field trial. Annals of Forest Research 60: 33-46–46.
706 https://doi.org/10.15287/afr.2016.733

707 Boyle E. A., Y. I. Li, and J. K. Pritchard, 2017 An Expanded View of Complex Traits: From
708 Polygenic to Omnigenic. Cell 169: 1177–1186.
709 https://doi.org/10.1016/j.cell.2017.05.038

710 Brachi B., G. P. Morris, and J. O. Borevitz, 2011 Genome-wide association studies in plants:
711 the missing heritability is in the field. Genome Biology 12: 232.
712 https://doi.org/10.1186/gb-2011-12-10-232

713 Brodde L., K. Adamson, J. Julio Camarero, C. Castaño, R. Drenkhan, *et al.*, 2019 Diplodia
714 Tip Blight on Its Way to the North: Drivers of Disease Emergence in Northern
715 Europe. Frontiers in Plant Science 9. https://doi.org/10.3389/fpls.2018.01818

716 Bucci G., S. C. González-Martínez, G. L. Provost, C. Plomion, M. M. Ribeiro, *et al.*, 2007
717 Range-wide phylogeography and gene zones in Pinus pinaster Ait. revealed by
718 chloroplast microsatellite markers. Molecular Ecology 16: 2137–2153.
719 https://doi.org/10.1111/j.1365-294X.2007.03275.x

720 Charlier C., W. Coppieters, F. Rollin, D. Desmecht, J. S. Agerholm, *et al.*, 2008 Highly
721 effective SNP-based association mapping and management of recessive defects in
722 livestock. Nat Genet 40: 449–454. https://doi.org/10.1038/ng.96

723 Chateigner A., M.-C. Lesage-Descauses, O. Rogier, V. Jorge, J.-C. Leplé, *et al.*, 2019 To
724 what extent gene connectivity within co-expression network matters for phenotype
725 prediction? bioRxiv 523365. https://doi.org/10.1101/523365

726 Cortijo S., R. Wardenaar, M. Colomé-Tatché, A. Gilly, M. Etcheverry, *et al.*, 2014 Mapping
727 the Epigenetic Basis of Complex Traits. Science 343: 1145–1148.
728 https://doi.org/10.1126/science.1248127

729 Craine J. M., E. N. J. Brookshire, M. D. Cramer, N. J. Hasselquist, K. Koba, *et al.*, 2015
730 Ecological interpretations of nitrogen isotope ratios of terrestrial plants and soils. Plant
731 Soil 396: 1–26. https://doi.org/10.1007/s11104-015-2542-1

732 Csilléry K., H. Lalagüe, G. G. Vendramin, S. C. González-Martínez, B. Fady, *et al.*, 2014
733 Detecting short spatial scale local adaptation and epistatic selection in climate-related
734 candidate genes in European beech (Fagus sylvatica) populations. Mol Ecol 23: 4696–
735 4708. https://doi.org/10.1111/mec.12902

736 Daub J. T., T. Hofer, E. Cutivet, I. Dupanloup, L. Quintana-Murci, *et al.*, 2013 Evidence for
737 Polygenic Adaptation to Pathogens in the Human Genome. Molecular Biology and
738 Evolution 30: 1544–1558. https://doi.org/10.1093/molbev/mst080

739 Dayan D. I., X. Du, T. Z. Baris, D. N. Wagner, D. L. Crawford, *et al.*, 2019 Population
740 genomics of rapid evolution in natural populations: polygenic selection in response to
741 power station thermal effluents. BMC Evol. Biol. 19: 61.
742 https://doi.org/10.1186/s12862-019-1392-5

743 De La Torre A. R., I. Birol, J. Bousquet, P. K. Ingvarsson, S. Jansson, *et al.*, 2014 Insights
744 into conifer giga-genomes. Plant physiology 166: 1724–32.
745 https://doi.org/10.1104/pp.114.248708

746 De La Torre A. R., Z. Li, Y. Van de Peer, and P. K. Ingvarsson, 2017 Contrasting Rates of
747 Molecular Evolution and Patterns of Selection among Gymnosperms and Flowering
748 Plants. Mol. Biol. Evol. 34: 1363–1377. https://doi.org/10.1093/molbev/msx069

749 De La Torre A. R., D. Puiu, M. W. Crepeau, K. Stevens, S. L. Salzberg, *et al.*, 2019a
750 Genomic architecture of complex traits in loblolly pine. New Phytologist 221: 1789–
751 1801. https://doi.org/10.1111/nph.15535

752 De La Torre A. R., B. Wilhite, and D. B. Neale, 2019b Environmental Genome-Wide
753 Association Reveals Climate Adaptation Is Shaped by Subtle to Moderate Allele

754 Frequency Shifts in Loblolly Pine, (T. Slotte, Ed.). Genome Biology and Evolution
755 11: 2976–2989. https://doi.org/10.1093/gbe/evz220

756 Desprez-Loustau M.-L., B. Marçais, L.-M. Nageleisen, D. Piou, and A. Vannini, 2006
757 Interactive effects of drought and pathogens in forest trees. Ann. For. Sci. 63: 597–
758 612. https://doi.org/10.1051/forest:2006040

759 Diaz-Sala C., J. A. Cabezas, B. Fernández de Simón, D. Abarca, M. Á. Guevara, *et al.*, 2013
760 The uniqueness of conifers, pp. 67–96 in *From Plant Genomics to Plant*
761 *Biotechnology*, edited by Poltronieri P., Burbulis N., Fogher C. Woodhead
762 Publishing, Cambridge.

763 Eckert A. J., A. D. Bower, K. D. Jermstad, J. L. Wegrzyn, B. J. Knaus, *et al.*, 2013 Multilocus
764 analyses reveal little evidence for lineage-wide adaptive evolution within major clades
765 of soft pines (*Pinus* subgenus *Strobus*). Mol Ecol 22: 5635–5650.
766 https://doi.org/10.1111/mec.12514

767 Edge M. D., and G. Coop, 2019 Reconstructing the History of Polygenic Scores Using
768 Coalescent Trees. Genetics 211: 235–262.
769 https://doi.org/10.1534/genetics.118.301687

770 Fabre B., D. Piou, M.-L. Desprez-Loustau, and B. Marçais, 2011 Can the emergence of pine
771 Diplodia shoot blight in France be explained by changes in pathogen pressure linked
772 to climate change? Global Change Biology 17: 3218–3227.
773 https://doi.org/10.1111/j.1365-2486.2011.02428.x

774 Fady B., J. Cottrell, L. Ackzell, R. Alía, B. Muys, *et al.*, 2016 Forests and global change:
775 what can genetics contribute to the major forest management and policy challenges of

the twenty-first century? Reg Environ Change 16: 927–939. https://doi.org/10.1007/s10113-015-0843-9

Farquhar G. D., and R. A. Richards, 1984 Isotopic Composition of Plant Carbon Correlates With Water-Use Efficiency of Wheat Genotypes. Functional Plant Biol. 11: 539–552. https://doi.org/10.1071/pp9840539

Field Y., E. Boyle, N. Telis, Z. Gao, K. J. Gaulton, *et al.*, 2016 Detection of human adaptation during the past 2000 years. Science 354: 760–764. https://doi.org/10.1126/science.aah5114

Firmat C., S. Delzon, J.-M. Louvet, J. Parmentier, and A. Kremer, 2017 Evolutionary dynamics of the leaf phenological cycle in an oak metapopulation along an elevation gradient. Journal of Evolutionary Biology 30: 2116–2131. https://doi.org/10.1111/jeb.13185

Fisher R. A., 1918 The correlation between relatives on the supposition of Mendelian inheritance. Transaction Royal Society Edinburgh 52: 399–433.

Fisher R. A., 1930 *The Genetical Theory of Natural Selection.* Clarendon Press, Oxford.

Fournier T., O. Abou Saada, J. Hou, J. Peter, E. Caudal, *et al.*, 2019 Extensive impact of low-frequency variants on the phenotypic landscape at population-scale, (C. R. Landry, and N. Barkai, Eds.). eLife 8: e49258. https://doi.org/10.7554/eLife.49258

Fournier-Level A., A. Korte, M. D. Cooper, M. Nordborg, J. Schmitt, *et al.*, 2011 A Map of Local Adaptation in Arabidopsis thaliana. Science 334: 86–89. https://doi.org/10.1126/science.1209271

Fréjaville T., and M. Benito Garzón, 2018 The EuMedClim Database: Yearly Climate Data (1901–2014) of 1 km Resolution Grids for Europe and the Mediterranean Basin. Front. Ecol. Evol. 6: 31. https://doi.org/10.3389/fevo.2018.00031

Gelman A., and D. B. Rubin, 1992 Inference from iterative simulation using multiple sequences. Statistical Science 7: 457–511.

Gelman A., 2006 Prior distributions for variance parameters in hierarchical models (comment on article by Browne and Draper). Bayesian Analysis 1: 515–534. https://doi.org/10.1214/06-BA117A

Gelman A., D. A. van Dyk, Z. Huang, and Boscardin, 2008 Using Redundant Parameterizations to Fit Hierarchical Models. Journal of Computational and Graphical Statistics 17.

Gnecchi-Ruscone G. A., P. Abondio, S. De Fanti, S. Sarno, M. G. Sherpa, *et al.*, 2018 Evidence of Polygenic Adaptation to High Altitude from Tibetan and Sherpa Genomes. Genome Biol Evol 10: 2919–2930. https://doi.org/10.1093/gbe/evy233

González-Martínez S. C., R. Alía, and L. Gil, 2002 Population genetic structure in a Mediterranean pine (Pinus pinaster Ait.): a comparison of allozyme markers and quantitative traits. Heredity 89: 199–206. https://doi.org/10.1038/sj.hdy.6800114

González-Martínez S. C., K. V. Krutovsky, and D. B. Neale, 2006 Forest-tree population genomics and adaptive evolution. New Phytologist 170: 227–238.

Grattapaglia D., O. B. Silva-Junior, R. T. Resende, E. P. Cappa, B. S. F. Müller, *et al.*, 2018 Quantitative Genetics and Genomics Converge to Accelerate Forest Tree Breeding. Front. Plant Sci. 9: 1693. https://doi.org/10.3389/fpls.2018.01693

Greenwood S., P. Ruiz-Benito, J. Martínez-Vilalta, F. Lloret, T. Kitzberger, *et al.*, 2017 Tree mortality across biomes is promoted by drought intensity, lower wood density and higher specific leaf area. Ecology Letters 20: 539–553. https://doi.org/10.1111/ele.12748

Grotkopp E., M. Rejmánek, M. J. Sanderson, and T. L. Rost, 2004 Evolution of Genome Size in Pines (Pinus) and Its Life-History Correlates: Supertree Analyses. Evolution 58: 1705. https://doi.org/10.1554/03-545

Guan Y., and M. Stephens, 2011 Bayesian variable selection regression for genome-wide association studies and other large-scale problems. The Annals of Applied Statistics 5: 1780–1815. https://doi.org/10.1214/11-AOAS455

Hadfield J. D., 2010 MCMC Methods for Multi-Response Generalized Linear Mixed Models: The **MCMCglmm** *R* Package. Journal of Statistical Software 33. https://doi.org/10.18637/jss.v033.i02

Hamblin M. T., E. S. Buckler, and J.-L. Jannink, 2011 Population genetics of genomics-based crop improvement methods. Trends in Genetics 27: 98–106. https://doi.org/10.1016/j.tig.2010.12.003

Hancock A. M., D. B. Witonsky, E. Ehler, G. Alkorta-Aranburu, C. Beall, *et al.*, 2010a Human adaptations to diet, subsistence, and ecoregion are due to subtle shifts in allele frequency. Proceedings of the National Academy of Sciences 107: 8924–8930. https://doi.org/10.1073/pnas.0914625107

Hancock A. M., G. Alkorta-Aranburu, D. B. Witonsky, and A. Di Rienzo, 2010b Adaptations to new environments in humans: the role of subtle allele frequency shifts. Phil. Trans. R. Soc. B 365: 2459–2468. https://doi.org/10.1098/rstb.2010.0032

842 Hayward L. K., and G. Sella, 2019 Polygenic adaptation after a sudden change in
843 environment. bioRxiv. https://doi.org/10.1101/792952

844 He F., A. L. Arce, G. Schmitz, M. Koornneef, P. Novikova, *et al.*, 2016 The Footprint of
845 Polygenic Adaptation on Stress-Responsive Cis-Regulatory Divergence in the
846 Arabidopsis Genus. Mol Biol Evol 33: 2088–2101.
847 https://doi.org/10.1093/molbev/msw096

848 Henderson C. R., 1973 Sire evaluation and genetic trends. Journal of Animal Science 1973:
849 10–41. https://doi.org/10.1093/ansci/1973.Symposium.10

850 Hermisson J., and P. Pennings, 2017a Soft sweeps and beyond: Understanding the patterns
851 and probabilities of selection footprints under rapid adaptation. bioRxiv 49.

852 Hermisson J., and P. S. Pennings, 2017b Soft sweeps and beyond: understanding the patterns
853 and probabilities of selection footprints under rapid adaptation. Methods in Ecology
854 and Evolution 8: 700–716. https://doi.org/10.1111/2041-210X.12808

855 Höllinger I., P. S. Pennings, and J. Hermisson, 2019 Polygenic adaptation: From sweeps to
856 subtle frequency shifts. PLOS Genetics 15: e1008035.

857 Hou K., K. S. Burch, A. Majumdar, H. Shi, N. Mancuso, *et al.*, 2019 Accurate estimation of
858 SNP-heritability from biobank-scale data irrespective of genetic architecture. Nat
859 Genet 51: 1244–1251. https://doi.org/10.1038/s41588-019-0465-0

860 Huang X., and B. Han, 2014 Natural Variations and Genome-Wide Association Studies in
861 Crop Plants. Annual Review of Plant Biology 65: 531–551.
862 https://doi.org/10.1146/annurev-arplant-050213-035715

863 Hurel A., M. de Miguel, C. Dutech, M.-L. Desprez-Loustau, C. Plomion, *et al.*, 2020 Genetic
864 basis of susceptibility to Diplodia sapinea and Armillaria ostoyae in maritime pine.
865 bioRxiv 699389. https://doi.org/10.1101/699389

866 Jacquet J.-S., A. Bosc, A. P. O'Grady, and H. Jactel, 2013 Pine growth response to
867 processionary moth defoliation across a 40-year chronosequence. Forest Ecology and
868 Management 293: 29–38. https://doi.org/10.1016/j.foreco.2012.12.003

869 Jain K., and W. Stephan, 2017 Rapid Adaptation of a Polygenic Trait After a Sudden
870 Environmental Shift. Genetics 206: 389–406.
871 https://doi.org/10.1534/genetics.116.196972

872 Jaramillo-Correa J.-P., I. Rodríguez-Quilón, D. Grivet, C. Lepoittevin, F. Sebastiani, *et al.*,
873 2015 Molecular Proxies for Climate Maladaptation in a Long-Lived Tree (Pinus
874 pinaster Aiton, Pinaceae). Genetics 199: 793–807.
875 https://doi.org/10.1534/genetics.114.173252

876 Josephs E. B., J. J. Berg, J. Ross-Ibarra, and G. Coop, 2019 Detecting adaptive differentiation
877 in structured populations with genomic data and common gardens. Genetics 211: 989–
878 1004. https://doi.org/10.1101/368506

879 Karlsson E. K., I. Baranowska, C. M. Wade, N. H. C. S. Hillbertz, M. C. Zody, *et al.*, 2007
880 Efficient mapping of mendelian traits in dogs through genome-wide association. Nat
881 Genet 39: 1321–1328. https://doi.org/10.1038/ng.2007.10

882 Keenan K., P. McGinnity, T. F. Cross, W. W. Crozier, and P. A. Prodöhl, 2013 diveRsity: An
883 R package for the estimation and exploration of population genetics parameters and
884 their associated errors. Methods in Ecology and Evolution 4: 782–788.
885 https://doi.org/10.1111/2041-210X.12067

886 Koch L., 2014 An epigenetic twist on the missing heritability of complex traits. Nat Rev
887 Genet 15: 218–218. https://doi.org/10.1038/nrg3698

888 Krutovsky K. V., and D. B. Neale, 2005 Nucleotide Diversity and Linkage Disequilibrium in
889 Cold-Hardiness- and Wood Quality-Related Candidate Genes in Douglas Fir. Genetics
890 171: 2029–2041. https://doi.org/10.1534/genetics.105.044420

891 Lamy J.-B., L. Bouffier, R. Burlett, C. Plomion, H. Cochard, *et al.*, 2011 Uniform Selection
892 as a Primary Force Reducing Population Genetic Differentiation of Cavitation
893 Resistance across a Species Range, (P. K. Ingvarsson, Ed.). PLoS ONE 6: e23476.
894 https://doi.org/10.1371/journal.pone.0023476

895 Leinonen T., R. B. O'hara, J. M. Cano, and J. Merilä, 2008 Comparative studies of
896 quantitative trait and neutral marker divergence: a meta-analysis. Journal of
897 Evolutionary Biology 21: 1–17. https://doi.org/10.1111/j.1420-9101.2007.01445.x

898 Leinonen T., R. J. S. McCairns, R. B. O'Hara, and J. Merilä, 2013 QST–FST comparisons:
899 evolutionary and ecological insights from genomic heterogeneity. Nature Reviews
900 Genetics 14: 179–190. https://doi.org/10.1038/nrg3395

901 Li C., B. Sun, Y. Li, C. Liu, X. Wu, *et al.*, 2016 Numerous genetic loci identified for drought
902 tolerance in the maize nested association mapping populations. BMC Genomics 17:
903 894. https://doi.org/10.1186/s12864-016-3170-8

904 Lind B. M., C. J. Friedline, J. L. Wegrzyn, P. E. Maloney, D. R. Vogler, *et al.*, 2017 Water
905 availability drives signatures of local adaptation in whitebark pine (*Pinus albicaulis*
906 Engelm.) across fine spatial scales of the Lake Tahoe Basin, USA. Molecular Ecology
907 26: 3168–3185. https://doi.org/10.1111/mec.14106

908 Lind B. M., M. Menon, C. E. Bolte, T. M. Faske, and A. J. Eckert, 2018 The genomics of
909 local adaptation in trees: are we out of the woods yet? Tree Genetics & Genomes 14.
910 https://doi.org/10.1007/s11295-017-1224-y

911 Liu X., P.-R. Loh, L. J. O'Connor, S. Gazal, A. Schoech, *et al.*, 2018 Quantification of
912 genetic components of population differentiation in UK Biobank traits reveals signals
913 of polygenic selection. bioRxiv 357483. https://doi.org/10.1101/357483

914 Maher B., 2008 Personal genomes: The case of the missing heritability. Nature 456: 18–21.
915 https://doi.org/doi:10.1038/456018a

916 Mähler N., J. Wang, B. K. Terebieniec, P. K. Ingvarsson, N. R. Street, *et al.*, 2017 Gene co-
917 expression network connectivity is an important determinant of selective constraint.
918 PLOS Genetics 13: e1006402. https://doi.org/10.1371/journal.pgen.1006402

919 Majada J., C. Martínez-Alonso, I. Feito, A. Kidelman, I. Aranda, *et al.*, 2011 Mini-cuttings:
920 an effective technique for the propagation of Pinus pinaster Ait. New Forests 41: 399–
921 412. https://doi.org/10.1007/s11056-010-9232-x

922 Mäki-Tanila A., and W. G. Hill, 2014 Influence of Gene Interaction on Complex Trait
923 Variation with Multilocus Models. Genetics 198: 355–367.
924 https://doi.org/10.1534/genetics.114.165282

925 Manolio T. A., F. S. Collins, N. J. Cox, D. B. Goldstein, L. A. Hindorff, *et al.*, 2009 Finding
926 the missing heritability of complex diseases. Nature 461: 747–753.
927 https://doi.org/10.1038/nature08494

928 Mayol M., M. Riba, S. Cavers, D. Grivet, L. Vincenot, *et al.*, 2020 A multiscale approach to
929 detect selection in nonmodel tree species: Widespread adaptation despite population

930 decline in Taxus baccata L. Evolutionary Applications 0.
931 https://doi.org/10.1111/eva.12838

932 Mei W., M. G. Stetter, D. J. Gates, M. C. Stitzer, and J. Ross-Ibarra, 2018 Adaptation in plant
933 genomes: Bigger is different. American Journal of Botany 105: 16–19.
934 https://doi.org/10.1002/ajb2.1002

935 Nakagawa S., and H. Schielzeth, 2010 Repeatability for Gaussian and non-Gaussian data: a
936 practical guide for biologists. Biological Reviews 85: 935–956.
937 https://doi.org/10.1111/j.1469-185X.2010.00141.x

938 Neale D. B., and O. Savolainen, 2004 Association genetics of complex traits in conifers.
939 Trends in Plant Science 9: 325–330. https://doi.org/10.1016/j.tplants.2004.05.006

940 Neves L. G., J. M. Davis, W. B. Barbazuk, and M. Kirst, 2013 Whole-exome targeted
941 sequencing of the uncharacterized pine genome. The Plant Journal 75: 146–156.
942 https://doi.org/10.1111/tpj.12193

943 O'Connor L. J., A. P. Schoech, F. Hormozdiari, S. Gazal, N. Patterson, *et al.*, 2019 Extreme
944 Polygenicity of Complex Traits Is Explained by Negative Selection. The American
945 Journal of Human Genetics 105: 456–476. https://doi.org/10.1016/j.ajhg.2019.07.003

946 Pallares L. F., 2019 Searching for solutions to the missing heritability problem. eLife 8:
947 e53018. https://doi.org/10.7554/eLife.53018

948 Palmé A. E., T. Pyhäjärvi, W. Wachowiak, and O. Savolainen, 2009 Selection on Nuclear
949 Genes in a Pinus Phylogeny. Mol Biol Evol 26: 893–905.
950 https://doi.org/10.1093/molbev/msp010

951 Pan J., B. Wang, Z.-Y. Pei, W. Zhao, J. Gao, *et al.*, 2015 Optimization of the genotyping-by-
952 sequencing strategy for population genomic analysis in conifers. Molecular Ecology
953 Resources 15: 711–722. https://doi.org/10.1111/1755-0998.12342

954 Pelgas B., J. Bousquet, P. G. Meirmans, K. Ritland, and N. Isabel, 2011 QTL mapping in
955 white spruce: gene maps and genomic regions underlying adaptive traits across
956 pedigrees, years and environments. BMC Genomics 12: 145.
957 https://doi.org/10.1186/1471-2164-12-145

958 Pitchers W., J. Nye, E. J. Márquez, A. Kowalski, I. Dworkin, *et al.*, 2019 A Multivariate
959 Genome-Wide Association Study of Wing Shape in Drosophila melanogaster.
960 Genetics 211: 1429–1447. https://doi.org/10.1534/genetics.118.301342

961 Plomion C., J. Bartholomé, L. Bouffier, O. Brendel, H. Cochard, *et al.*, 2016a Understanding
962 the genetic bases of adaptation to soil water deficit in trees through the examination of
963 water use efficiency and cavitation resistance: maritime pine as a case study. Journal
964 of Plant Hydraulics 3: e008. https://doi.org/10.20870/jph.2016.e008

965 Plomion C., J. Bartholomé, I. Lesur, C. Boury, I. Rodríguez-Quilón, *et al.*, 2016b High-
966 density SNP assay development for genetic analysis in maritime pine (Pinus pinaster
967). Molecular Ecology Resources 16: 574–587. https://doi.org/10.1111/1755-
968 0998.12464

969 Pritchard J. K., J. K. Pickrell, and G. Coop, 2010 The Genetics of Human Adaptation: Hard
970 Sweeps, Soft Sweeps, and Polygenic Adaptation. Current Biology 20: R208–R215.
971 https://doi.org/10.1016/j.cub.2009.11.055

972 Pritchard J. K., and A. D. Rienzo, 2010 Adaptation – not by sweeps alone. Nat Rev Genet 11:
973 665–667. https://doi.org/10.1038/nrg2880

Rellstab C., S. Zoller, L. Walthert, I. Lesur, A. R. Pluess, *et al.*, 2016 Signatures of local adaptation in candidate genes of oaks (Quercus spp.) with respect to present and future climatic conditions. Molecular Ecology 25: 5907–5924. https://doi.org/10.1111/mec.13889

Resende M. D. V., M. F. R. Resende Jr, C. P. Sansaloni, C. D. Petroli, A. A. Missiaggia, *et al.*, 2012 Genomic selection for growth and wood quality in Eucalyptus: capturing the missing heritability and accelerating breeding for complex traits in forest trees. New Phytologist 116–128. https://doi.org/10.1111/j.1469-8137.2011.04038.x@10.1002/(ISSN)1469-8137(CAT)FeatureIssues(VI)Bioenergytrees

Robinson G. K., 1991 That BLUP is a Good Thing: The Estimation of Random Effects. Statist. Sci. 6: 15–32. https://doi.org/10.1214/ss/1177011926

Rodríguez-Quilón I., L. Santos-del-Blanco, M. J. Serra-Varela, J. Koskela, S. C. González-Martínez, *et al.*, 2016 Capturing neutral and adaptive genetic diversity for conservation in a highly structured tree species. Ecol App 26: 2254–2266.

Rosenberg N. A., M. D. Edge, J. K. Pritchard, and M. W. Feldman, 2019 Interpreting polygenic scores, polygenic adaptation, and human phenotypic differences. Evol. Med. Public Health. 26–34. https://doi.org/10.1093/emph/eoy036

Savolainen O., T. Pyhäjärvi, and T. Knürr, 2007 Gene Flow and Local Adaptation in Trees. Annual Review of Ecology, Evolution, and Systematics 38: 595–619. https://doi.org/10.1146/annurev.ecolsys.38.091206.095646

Savolainen O., M. Lascoux, and J. Merilä, 2013 Ecological genomics of local adaptation. Nat Rev Genet 14: 807–820. https://doi.org/10.1038/nrg3522

997 Seidl R., D. Thom, M. Kautz, D. Martin-Benito, M. Peltoniemi, *et al.*, 2017 Forest
998 disturbances under climate change. Nature Clim Change 7: 395–402.
999 https://doi.org/10.1038/nclimate3303

1000 Sella G., and N. H. Barton, 2019 Thinking About the Evolution of Complex Traits in the Era
1001 of Genome-Wide Association Studies. Annu. Rev. Genom. Hum. Genet. 20: 461–493.
1002 https://doi.org/10.1146/annurev-genom-083115-022316

1003 Sharma A., J. S. Lee, C. G. Dang, P. Sudrajad, H. C. Kim, *et al.*, 2015 Stories and Challenges
1004 of Genome Wide Association Studies in Livestock — A Review. Asian-Australas J
1005 Anim Sci 28: 1371–1379. https://doi.org/10.5713/ajas.14.0715

1006 Shi H., G. Kichaev, and B. Pasaniuc, 2016 Contrasting the Genetic Architecture of 30
1007 Complex Traits from Summary Association Data. The American Journal of Human
1008 Genetics 99: 139–153. https://doi.org/10.1016/j.ajhg.2016.05.013

1009 Smith J. M., and J. Haigh, 1974 The hitch-hiking effect of a favourable gene. Genetics
1010 Research 23: 23–35. https://doi.org/10.1017/S0016672300014634

1011 Sohail M., R. M. Maier, A. Ganna, A. Bloemendal, A. R. Martin, *et al.*, 2019 Polygenic
1012 adaptation on height is overestimated due to uncorrected stratification in genome-wide
1013 association studies, (M. Nordborg, M. I. McCarthy, M. Nordborg, N. H. Barton, and J.
1014 Hermisson, Eds.). eLife 8: e39702. https://doi.org/10.7554/eLife.39702

1015 Speidel L., M. Forest, S. Shi, and S. R. Myers, 2019 A method for genome-wide genealogy
1016 estimation for thousands of samples. Nat Genet 51: 1321–1329.
1017 https://doi.org/10.1038/s41588-019-0484-x

1018 Spitze K., 1993 Population Structurein Daphnia obtusa:Quantitative Genetic and Allozymic
1019 Variation. 135: 367–374.

1020 Stephan W., 2016 Signatures of positive selection: from selective sweeps at individual loci to
1021 subtle allele frequency changes in polygenic adaptation. Molecular Ecology 25: 79–
1022 88. https://doi.org/10.1111/mec.13288

1023 Torres-Ruiz J. M., A. Kremer, M. R. Carins-Murphy, T. J. Brodribb, L. J. Lamarque, *et al.*,
1024 2019 Genetic differentiation in functional traits among European sessile oak
1025 populations. Tree Physiol 39: 1736–1749. https://doi.org/10.1093/treephys/tpz090

1026 Turchin M. C., C. W. Chiang, C. D. Palmer, S. Sankararaman, D. Reich, *et al.*, 2012 Evidence
1027 of widespread selection on standing variation in Europe at height-associated SNPs.
1028 Nature Genetics 44: 1015–1019. https://doi.org/10.1038/ng.2368

1029 Valladares F., S. Matesanz, F. Guilhaumon, M. B. Araújo, L. Balaguer, *et al.*, 2014 The
1030 effects of phenotypic plasticity and local adaptation on forecasts of species range
1031 shifts under climate change. Ecology Letters 17: 1351–1364.
1032 https://doi.org/10.1111/ele.12348

1033 Vidalis A., D. G. Scofield, L. G. Neves, C. Bernhardsson, M. R. García-Gil, *et al.*, 2018
1034 Design and evaluation of a large sequence-capture probe set and associated SNPs for
1035 diploid and haploid samples of Norway spruce (Picea abies). bioRxiv 291716.
1036 https://doi.org/10.1101/291716

1037 Visscher P. M., 2008 Sizing up human height variation. Nat Genet 40: 489–490.
1038 https://doi.org/10.1038/ng0508-489

1039 Visscher P. M., N. R. Wray, Q. Zhang, P. Sklar, M. I. McCarthy, *et al.*, 2017 10 Years of
1040 GWAS Discovery: Biology, Function, and Translation. The American Journal of
1041 Human Genetics 101: 5–22. https://doi.org/10.1016/j.ajhg.2017.06.005

1042 Vitasse Y., S. Delzon, C. Bresson, R. Michalet, and A. Kremer, 2009 Altitudinal
1043 differentiation in growth and phenology among populations of temperate-zone tree
1044 species growing in a common garden - Canadian Journal of Forest Research

1045 Walker X. J., M. C. Mack, and J. F. Johnstone, 2015 Stable carbon isotope analysis reveals
1046 widespread drought stress in boreal black spruce forests. Glob Chang Biol 21: 3102–
1047 3113. https://doi.org/10.1111/gcb.12893

1048 Weir B. S., and C. C. Cockerham, 1984 ESTIMATING *F* -STATISTICS FOR THE
1049 ANALYSIS OF POPULATION STRUCTURE. Evolution 38: 1358–1370.
1050 https://doi.org/10.1111/j.1558-5646.1984.tb05657.x

1051 Whitlock M. C., 1999 Neutral additive genetic variance in a metapopulation. Genet. Res. 74:
1052 215–221. https://doi.org/10.1017/S0016672399004127

1053 Whitlock M. C., 2008 Evolutionary inference from QST. Molecular Ecology 17: 1885–1896.
1054 https://doi.org/10.1111/j.1365-294X.2008.03712.x

1055 Whitlock M. C., and F. Guillaume, 2009 Testing for Spatially Divergent Selection:
1056 Comparing QST to FST. Genetics 183: 1055–1063.
1057 https://doi.org/10.1534/genetics.108.099812

1058 Wisser R. J., Z. Fang, J. B. Holland, J. E. C. Teixeira, J. Dougherty, *et al.*, 2019 The Genomic
1059 Basis for Short-Term Evolution of Environmental Adaptation in Maize. Genetics 213:
1060 1479–1494. https://doi.org/10.1534/genetics.119.302780

1061 Wray N. R., C. Wijmenga, P. F. Sullivan, J. Yang, and P. M. Visscher, 2018 Common
1062 Disease Is More Complex Than Implied by the Core Gene Omnigenic Model. Cell
1063 173: 1573–1580. https://doi.org/10.1016/j.cell.2018.05.051

1064 Wright S. I., and P. Andolfatto, 2008 The Impact of Natural Selection on the Genome:
1065 Emerging Patterns in Drosophila and Arabidopsis. Annual Review of Ecology,
1066 Evolution, and Systematics 39: 193–213.
1067 https://doi.org/10.1146/annurev.ecolsys.39.110707.173342

1068 Yang R.-C., F. C. Yeh, and A. D. Yanchuk, 1996 A Comparison of Isozyme and Quantitative
1069 Genetic Variation in Pinus contorta ssp. latifolia by FST. Genetics 142: 1045–1052.

1070 Yang J., B. Benyamin, B. P. McEvoy, S. Gordon, A. K. Henders, *et al.*, 2010 Common SNPs
1071 explain a large proportion of the heritability for human height. Nat Genet 42: 565–569.
1072 https://doi.org/10.1038/ng.608

1073 Yang J., W.-R. Li, F.-H. Lv, S.-G. He, S.-L. Tian, *et al.*, 2016 Whole-Genome Sequencing of
1074 Native Sheep Provides Insights into Rapid Adaptations to Extreme Environments. Mol
1075 Biol Evol 33: 2576–2592. https://doi.org/10.1093/molbev/msw129

1076 Yeaman S., 2015 Local Adaptation by Alleles of Small Effect. The American Naturalist 186:
1077 S74–S89. https://doi.org/10.1086/682405

1078 Zeng J., R. de Vlaming, Y. Wu, M. R. Robinson, L. R. Lloyd-Jones, *et al.*, 2018 Signatures of
1079 negative selection in the genetic architecture of human complex traits. Nature Genetics
1080 50: 746–753. https://doi.org/10.1038/s41588-018-0101-4

1081 Zhang G., L. J. Muglia, R. Chakraborty, J. M. Akey, and S. M. Williams, 2013 Signatures of
1082 natural selection on genetic variants affecting complex human traits. Appl Transl
1083 Genom 2: 78–94. https://doi.org/10.1016/j.atg.2013.10.002

1084 Zonneveld B. J. M., 2012 Conifer genome sizes of 172 species, covering 64 of 67 genera,
1085 range from 8 to 72 picogram. Nordic Journal of Botany 30: 490–502.
1086 https://doi.org/10.1111/j.1756-1051.2012.01516.x

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Trial	Plantation year	N trees	N clones	Annual precipitation (mm)	Precipitation of the warmest quarter Jun+Jul+Aug (mm)	Annual mean temperature	Annual temperature range
Asturias	2010	4,272	535	890	126	12.9	24.0
Portugal	2010	4,272	535	1122	58	14.0	26.9
Bordeaux	2011	3,434	443	933	199	13.8	26.7
Madrid	2010	4,272	535	378	35	14.8	32.8
Cáceres	2010	4,272	535	374	21	16.7	32.6

Table 1. Description of the five sites of CLONAPIN common garden network. Climatic data correspond to the mean of each parameter for the period 2005-2014 obtained from the EuMedClim database (Fréjaville and Benito Garzón 2018).

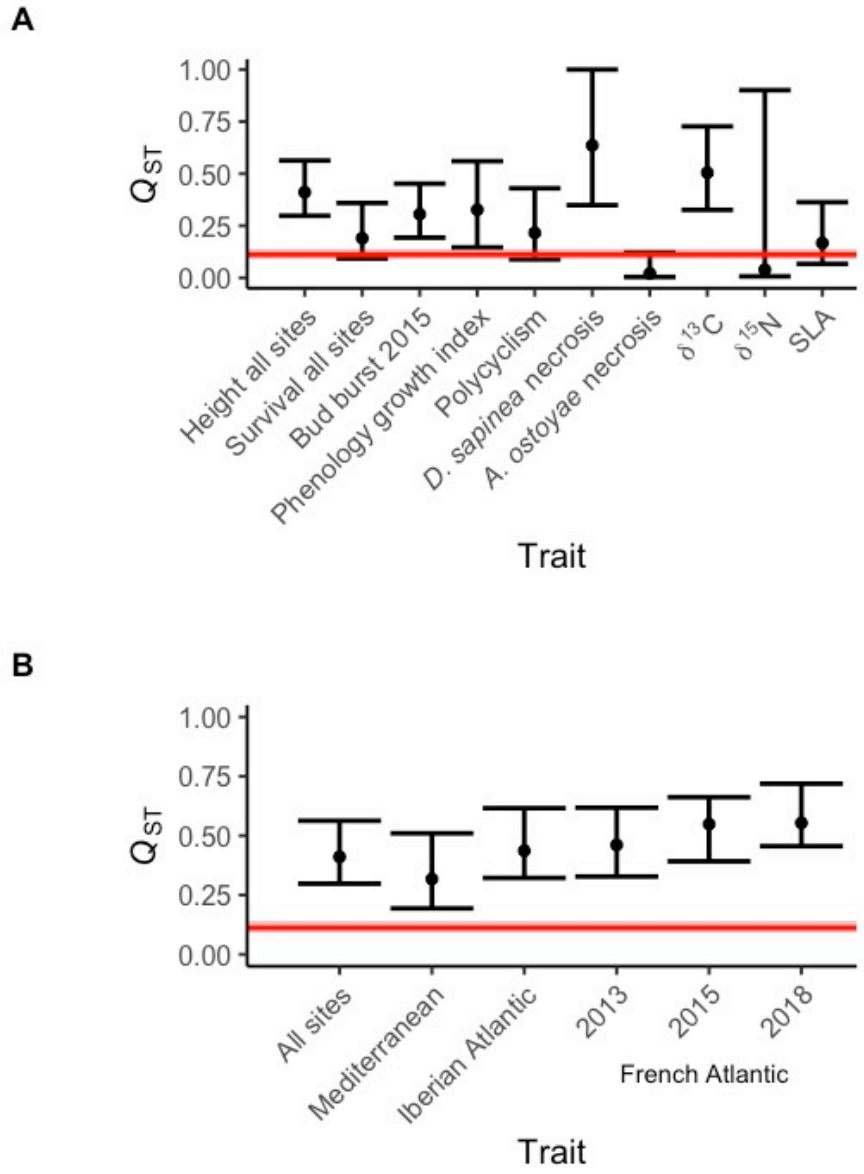

Figure 1. Q_{ST} and F_{ST} estimates. A) Q_{ST} for different traits belonging to five categories: survival, height, phenology-related traits, functional traits and biotic-stress response. B) Q_{ST} for height estimated in three different environments: Mediterranean, Iberian Atlantic, and French Atlantic, and a global Q_{ST} for the three environments together. In the French Atlantic common garden, height was measured in three different years: 2013, 2015 and 2018.

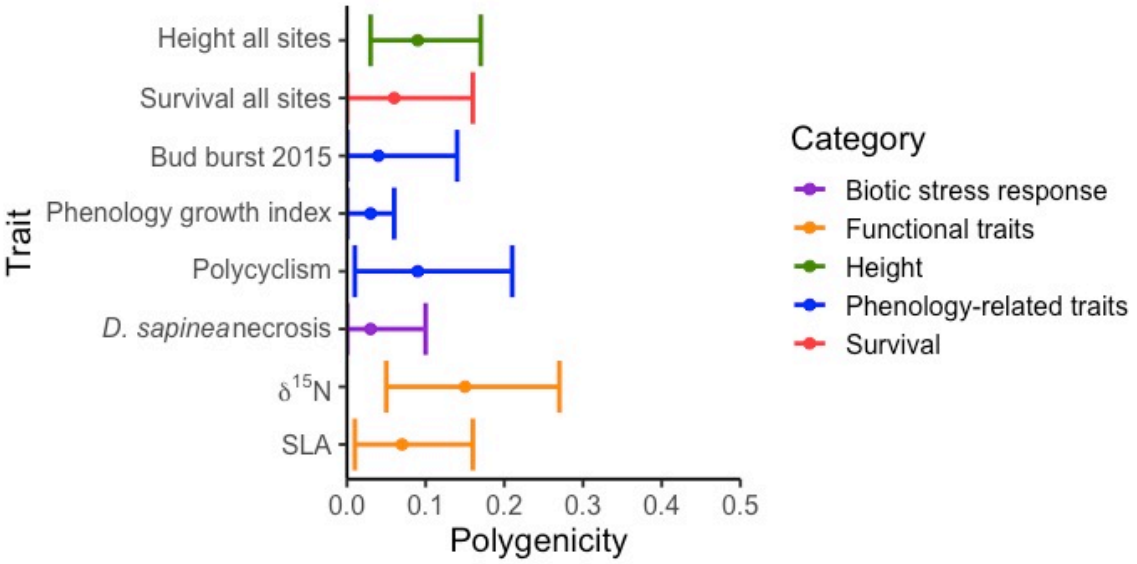

Figure 2. Polygenicity estimated from Bayesian mixed linear models (MLMs) for different traits. Polygenicity was estimated as the proportion of non-zero effect SNPs. Posterior median and 95% credible intervals are presented.

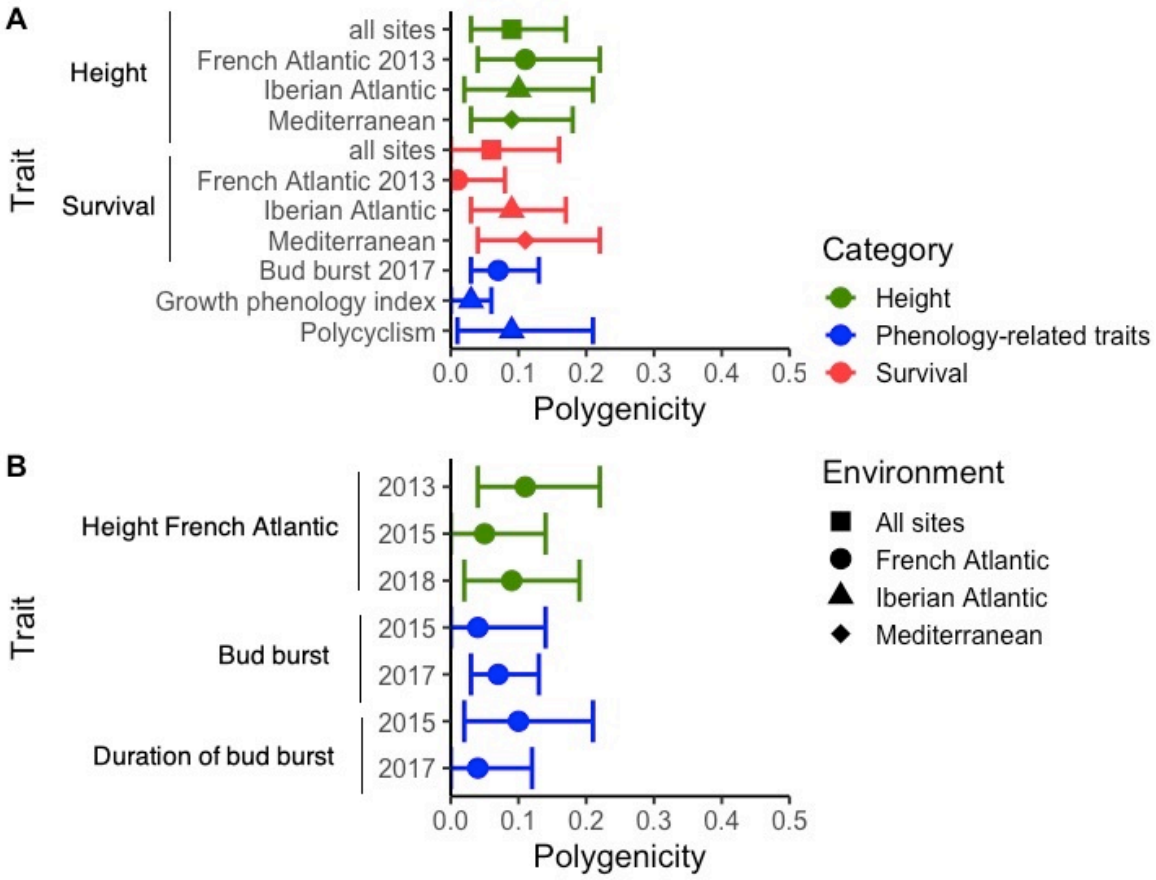

Figure 3. Polygenicity estimated from Bayesian mixed linear models (MLMs) across years and environments. A) Variation of polygenicity across environments. B) Temporal variation of polygenicity. Polygenicity was estimated as the proportion of non-zero effect SNPs. Posterior median and 95% credible intervals are presented.

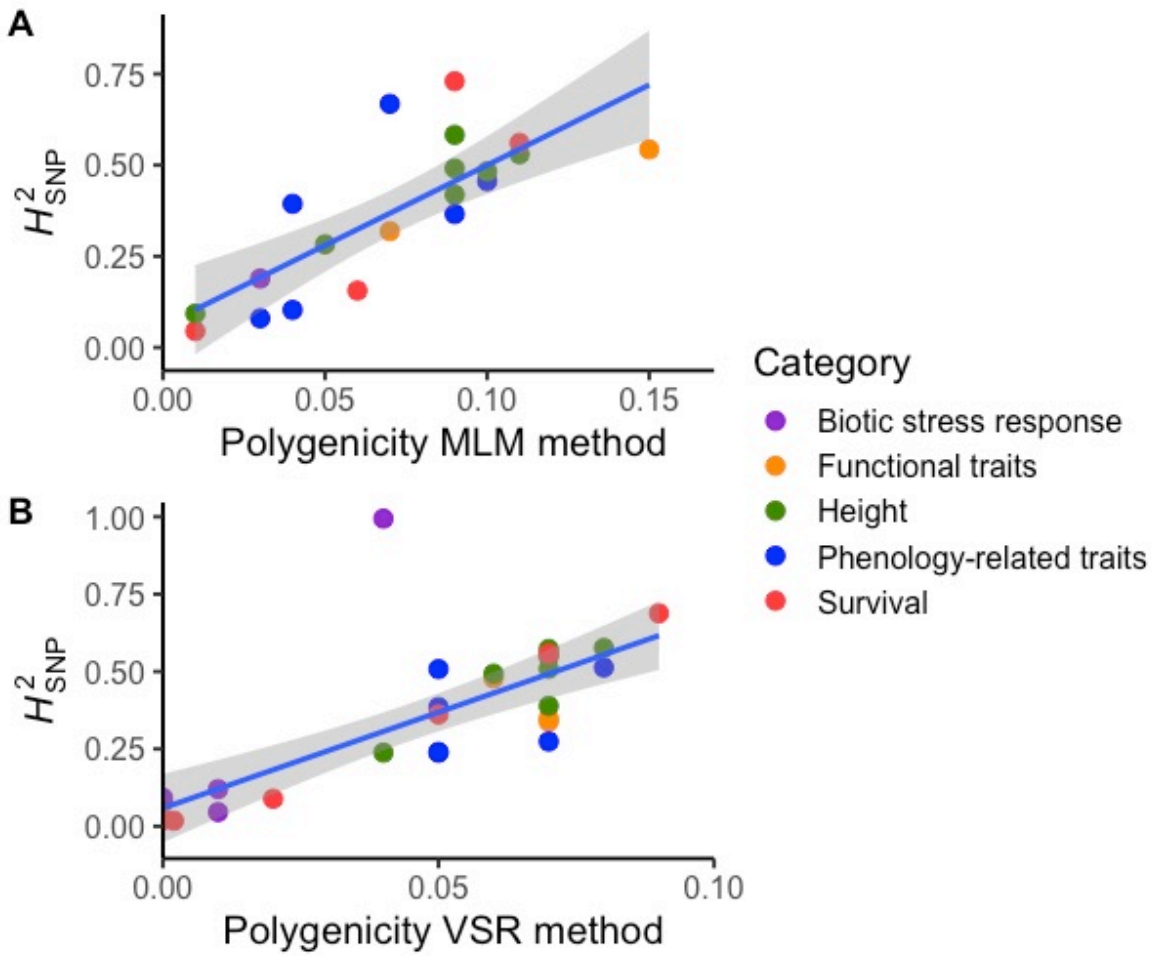

Figure 4. Correlation between polygenicity and H^2_{SNP} (SNP-based heritability). A) MLM method implemented in CGTB software. B) VSR method implemented in PiMASS software. Polygenicity was estimated as the proportion of non-zero effect SNPs. H^2_{SNP} is the proportion of the genetic variance explained by all the SNPs with non-zero effects. Each point represents the posterior median.

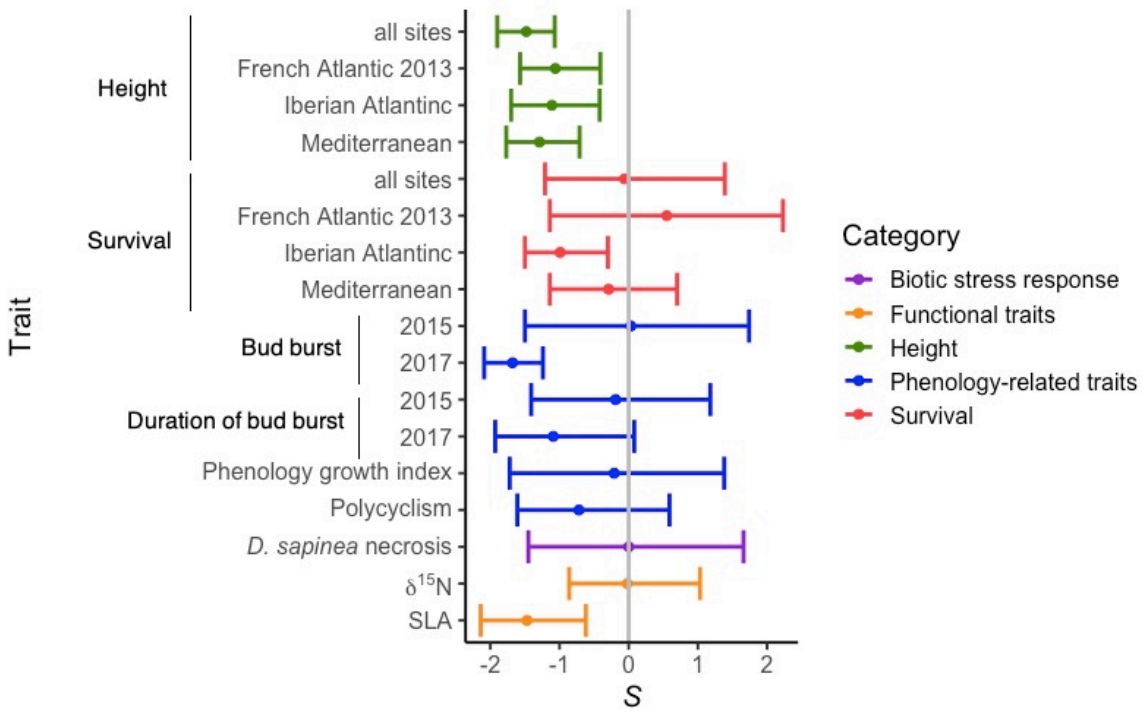

Figure 5. Correlation between SNP effect size and MAF. The coefficient of correlation between SNP effect size and MAF (*S*) was estimated through the MLM method. The posterior distribution of *S* (median and 95% credible intervals) are presented.