A safe breeding ground: genetically improved maritime pine for growth and stem form has more efficient but not more vulnerable xylem

Yanjun Song, Xavier P. Bouteiller, Maximilian Larter, Christophe Plomion, Fabrice Sin, Sylvain Delzon

HAL Id: hal-04030010
https://hal.inrae.fr/hal-04030010

Research paper

A safe breeding ground: genetically improved maritime pine for growth and stem form has more efficient but not more vulnerable xylem

Yanjun Song [1,3], Xavier Paul Bouteiller[1], Maximilian Larter[1], Christophe Plomion [1], Fabrice Sin[2] and Sylvain Delzon[1]

[1]UMR BIOGECO, INRAE, Univ. Bordeaux, 33615 Pessac, France; [2]National Forest Office, Bordeaux, 33524 Bruges, France; [3]Corresponding author (yanjun.song2021@gmail.com)

Through repeated cycles of selection and recombination, tree breeding programs deliver genetically improved varieties for a range of target characteristics such as biomass production, stem form, resistance to biotic stresses, wood properties, etc. However, in the context of increased drought and heat waves, it is not yet known whether growth performance will impede drought resistance. To address this question, we compared the hydraulic properties, such as hydraulic efficiency and hydraulic safety, in four varieties over successive varieties of genetically improved maritime pines (i.e., *Pinus pinaster* Aït.) for growth and stem form. We measured 22 functional traits related to hydraulic efficiency, hydraulic safety, xylem anatomy and wood density. We found that improved varieties presented higher hydraulic conductivity with larger tracheid lumen size and tracheid lumen fraction, and smaller wall thickness reinforcement and tracheid density, but not at the cost of reduced embolism resistance. The reported absence of trade-off between hydraulic conductivity and embolism resistance is a strong asset to improve biomass productivity, through increased hydraulic efficiency, without impacting drought resistance, and should enable new maritime pine varieties to cope with a drier climate. Our study is one of the first to reveal the hydraulic mechanisms over successive varieties of genetic improvement for tree growth. It provides guidelines for sustainable forest management through breeding for other forest tree species.

Introduction

Tree breeding programs

Due to the high tree mortality induced by drought (Allen et al. 2010, Brodribb et al. 2020), and the world's rising population, natural forests will not be able to meet the foreseen demand for forest-related products (Nabuurs et al. 2018). To fill global wood demand and decrease the supply from natural forests, highly productive plantation forests, grown primarily for wood biomass, have been developed worldwide particularly since the 1950s.

In this context, two main factors of forest productivity have been considered and developed, i.e., the optimization of silvicultural practices and the use of genetically improved plant material (Corona 2019). Regarding the latter, recurrent selection schemes, a strategy whereby multiple breeding and production populations are produced to deliver genetic gains over successive generations, have been developed worldwide to improve stem volume growth and stem straightness (Pâques 2013, Zhao et al. 2021). The effect of high productivity on drought resistance is however not well documented.

Tree growth and xylem efficiency

The breeding system aims to improve growth, which could be explained by the underlying traits related to hydraulic efficiency, such as xylem-specific hydraulic conductivity (hereafter hydraulic conductivity, K_s). K_s indicates the capacity of water transport; high K_s advances high transpiration rate and stomatal conductance, and hence high growth rate (Domec and Gartner,

2003). Nevertheless, the direct impact of artificial selection in a breeding system on hydraulic conductivity remains untested. Comparing the hydraulic conductivity among successive generations of breeding is therefore important to understand what mechanisms improve the growth of improved genetic materials.

Wood anatomy plays an important role in explaining hydraulic efficiency. The pits and tracheids are key xylem structures for water transport, as water moves through one tracheid to another tracheid in the transverse section and through the inter-conduit pit aperture from one tracheid to adjacent tracheid in tangential section (Sterck et al. 2008, Hacke and Jansen 2009). Trees with large pits and tracheids enable relatively high lumen fraction to xylem, thus having high hydraulic conductivity (Rungwattana and Hietz 2018). However, a recent study found that there were no consistent relationships between pit/tracheid size and hydraulic conductivity (Song et al. 2022a). Instead, the pore in margo, overlapping tracheid tips or tracheid length probably determines hydraulic conductivity (Sperry et al. 2006). It remains necessary to reveal the underlying mechanisms of hydraulic conductivity.

Trade-off between growth and drought resistance

Although a high growth rate is important for timber productivity in a drier future, selecting drought-tolerant species and populations is important for designing a resilient forest to cope with the increased frequency of drought events (Lamy et al. 2014, Gleason 2015, Song et al. 2022b). Recent forest dieback events have highlighted drought-induced embolism as a driving factor in tree mortality (Choat et al. 2012, Anderegg et al. 2016, Choat et al. 2018). Hydraulic safety, the ability of trees to maintain water supply to the aerial organs during prolonged drought, is quantified as the xylem pressure inducing 50% loss of hydraulic conductivity (P_{50}): high $|P_{50}|$ means high resistance to drought-induced embolism. It is well known that breeding programs typically select genotypes with increased growth (Serrano-León et al. 2021), but it is not clear whether breeding for growth also affects drought resistance. Generally, trees with high K_s adapt a fast-growing strategy with high growth but at the cost of reduced $|P_{50}|$ (Yao et al. 2021, Wang et al. 2022), because high K_s needs high relative lumen area to wall thickness. Consequently, there is a reduced carbon investment in conduit walls and the capacity of withstanding extreme xylem tensions will be weak (Hacke and Sperry 2001). Therefore, there is a hypothesized trade-off between hydraulic conductivity (or growth) and embolism resistance (Liu et al. 2019).

Within Scots pine, faster-growing trees had lower xylem embolism resistance (Sterck et al. 2012), however other studies (e.g., in *Populus* species) found no intraspecific trade-off (Hajek et al. 2014, Guet et al. 2015), or even a positive relationship between both traits (Schuldt et al. 2016). Therefore, despite a conduit-level relationship between construction costs and embolism resistance, there is no broad consensus on the organ- or whole plant-level relationship between growth and embolism resistance, especially within a breeding context.

Embolism resistance is thought to be driven by the fine-scale structure of the inter-conduit pit anatomy. During drought, the spread of air through the xylem occurs by air-seeding from one tracheid to another through the inter-conduit pits (Tyree and Sperry 1989, Tyree and Zimmermann 2013). In broadleaf trees, microscopic air-bubbles seep through tiny pores in the pit membrane, whereas in gymnosperms the torus-margo structure acts as a valve (Hacke et al. 2004), where the impermeable torus can become deflected to seal the pit aperture when embolism occurs (Jansen and McAdam 2019). A flexible margo and a large torus diameter (relative to the pit aperture) lead to an effective sealing of the pit aperture. This likely contributes to the higher embolism resistance of conifers (Delzon et al. 2010). A reduction in pit aperture diameter, thereby increasing the torus overlap, explains the variation of P_{50} across conifer species (Hacke and Jansen 2009, Bouche et al. 2014). Although the pit size and torus-pit sealing underlie the differences in embolism resistance across species (Song et al. 2022a), it remains uncertain if they also contribute to the intraspecific differences in embolism resistance (Li et al. 2022).

Studying tree hydraulics will advance our understanding of tree growth and survival, facilitating a better prediction of biomass production under a drier future climate. Therefore, it is important to compare to what extent the improved varieties of breeding for growth affect tree hydraulic safety and efficiency. Most plantations have been and are still done with genetically improved material. In particular, maritime pine (*Pinus pinaster*) stands are widely planted over 4 million hectares in the western part of the Mediterranean region (Lamy et al. 2012, Marguerit et al. 2014). They play important roles in Mediterranean ecosystems due to high timber productivity with fast-growing characteristics. However, it has never been studied whether the artificial selection through recurrent breeding impacts the hydraulic properties of trees.

To compare the xylem resistance and efficiency of different maritime pine varieties (natural versus improved varieties), we (i) compare the hydraulic traits (i.e., embolism resistance and hydraulic conductivity) of four varieties of *P. pinaster*, and (ii) relate their hydraulic traits to the underlying anatomical tracheid and pit traits. Here, we aim to answer the following two questions and hypotheses:

(i) How has artificial selection affected the embolism resistance and hydraulic conductivity of maritime pine? We hypothesize that genetically improved maritime pine varieties have increased hydraulic conductivity allowing for higher gas exchange, but at the cost of reduced embolism resistance.

(ii) How do wood anatomical traits underlie hydraulic conductivity and embolism resistance in maritime pine under

Table 1. Overview of 23 hydraulic traits, abbreviations and units.

Type	Trait name	Abbreviation	Units
Embolism resistance	Xylem pressure inducing 12% loss in hydraulic conductance	P_{12}	MPa
	Xylem pressure inducing 50% loss in hydraulic conductance	P_{50}	MPa
	Xylem pressure inducing 88% loss in hydraulic conductance	P_{88}	MPa
	Slope	S	–
Hydraulic conductivity	Xylem specific hydraulic conductivity	K_s	kg m^{-1} s^{-1} MPa^{-1}
Wood	Wood density (earlywood)	WD_E	g cm^{-3}
	Wood density (latewood)	WD_L	g cm^{-3}
Tracheid	Hydraulic diameter (earlywood)	Dh_E	μm
	Hydraulic diameter (latewood)	Dh_L	μm
	Tracheid density (earlywood)	TD_E	# mm^{-2}
	Tracheid density (latewood)	TD_L	# mm^{-2}
	Wall thickness (earlywood)	Tw_E	μm
	Wall thickness (latewood)	Tw_L	μm
	Thickness to span ratio (earlywood)	TSR_E	μm μm^{-1}
	Thickness to span ratio (latewood)	TSR_L	μm μm^{-1}
	Tracheid lumen fraction (earlywood)	TLF_E	μm^2 μm^{-2}
	Tracheid lumen fraction (latewood)	TLF_L	μm^2 μm^{-2}
Pit	Pit membrane diameter	DPM	μm
	Torus diameter	DT	μm
	Pit aperture diameter	DPA	μm
	Margo flexibility	MF	–
	Torus overlap	TO	–
	Valve effect	VE	–

artificial selection? We hypothesize that high hydraulic conductivity of improved genetic material is associated with wider tracheids and pits, whereas high embolism resistance is triggered by strong valve effect via margo flexibility and torus overlap to limit air-seeding.

Materials and methods

Maritime pine stands

Starting in the 1960s, the maritime pine breeding program has produced three series of seed orchards based on the local Landes provenance, with an increasing improvement in growth and stem straightness from VF1 or VSEL (Vigor and Form, first series) to VF3 (third series; Mullin et al. 2011). Although VF1 = VSEL corresponds to the first generation of seed orchards (plus trees or progenies of plus-trees) with an increased volume of 15%, VF2 corresponds to a 1.5 generation (i.e., progeny tested plus-trees) with an increased volume of 30%, and VF3 corresponds to a 2.5 generation (i.e., progeny tested first-generation trees) with an increased volume of 40% (Bouffier et al. 2013). The second series VF2 (established during the 1980s–90s) is gradually being replaced by the third series VF3, which entered into production in 2011–15 (Bouffier et al. 2013, GIS GPMF 2014).

To compare the hydraulic properties (Table 1) of the aforementioned selected series (VSEL, VF2 and VF3), we sampled and measured natural populations (VNAT) from southwestern France, and the three improved maritime pine varieties (VSEL, VF2 and VF3) in spring 2021. For samples collection, we selected even-aged, monospecific stands of maritime pine with an understory consisting mainly of *Molinia caerulea* (L) Moench located in the Médoc natural park 35-km west of Bordeaux in the Landes de Gascogne Forest (Figure S1 available as Supplementary data at *Tree Physiology* Online). All stands were growing on the same soil (sandy podzol with a cemented Bh horizon limiting the root depth to −0.8 m) and climatic conditions (Figure S1b–d available as Supplementary data at *Tree Physiology* Online) in which 57–59 individuals per variety were sampled within one or four stands per variety (Table S1 available as Supplementary data at *Tree Physiology* Online). To avoid intra-individual variation (considered very low in maritime pine for hydraulic traits, see Bouche et al. 2016), we selected one fully sun-exposed branch (*c.* 50 cm) per individual. To reduce water loss and cavitation, we wrapped the branches in wet papers and sealed in black plastic bags, and then transported the branches to the lab at University of Bordeaux. The stands had extremely low soil phosphorus and nitrogen levels, and an average pH–H$_2$O of 4.0. The regional climate is maritime (mean annual temperature of 13 °C, mean annual precipitation of 977 mm) with marked drought in summer (June–August, Figure S1 available as Supplementary data at *Tree Physiology* Online). Tree age ranged between 25 and 30 years old.

Embolism resistance and hydraulic conductivity

To compare embolism resistance and hydraulic conductivity among the four levels of genetically improved maritime pines

(VNAT: unimproved material; VSEL, VF2 and VF3: gradually improved materials), we used a standard 'Cavitron' method (Cochard et al. 2005), improved by Burlett et al. (2022) through generating negative pressure (P in MPa) in branch segments. All measurements were carried out at the PHENOBOIS phenotyping platform (CavitPlace, University of Bordeaux, Talence, France). Samples were cut with a specific length of 27 cm and barks were removed to prevent resin fillings from barks. We infiltrated samples with a reference ionic solution of 10-mm 25 KCl and 1-mM $CaCl_2$ in deionized ultrapure water. To induce embolism in the xylem, we generated negative pressures by using a centrifugal force, which increased from -0.8 to -7 MPa (González-Muñoz et al. 2018). Meanwhile, hydraulic conductivity (K in m^2 MPa^{-1} s^{-1}) was recorded for 2 min when samples were exposed to the corresponding pressure. The maximum xylem hydraulic conductivity (K_{max}) was initially obtained when the pressure was near zero, and then the xylem specific hydraulic conductivity (hereafter referred to as hydraulic conductivity, K_s in kg m^{-1} MPa^{-1} s^{-1}) was calculated by dividing K_{max} by sample length and sapwood area (Larter et al. 2017). To fit the relationship of K and P, we firstly calculated the percentage loss of hydraulic conductance (PLC) and then used a sigmoid function (Delzon et al. 2010) as follows:

$$PLC = 100 \left(1 - \frac{K}{K_{max}} \right) \tag{1}$$

$$PLC = \frac{100}{\left(1 + \exp\left(\frac{S}{25} \times (P - P_{50}) \right) \right)} \tag{2}$$

where P_{50} (MPa) is the xylem pressure when 50% of hydraulic conductivity is lost, S (% MPa^{-1}) refers to the slope at the inflection point. Afterwards, xylem pressures (P in MPa) when 12 and 88% of conductance were derived and referred to as P_{12} and P_{88}. Individual values were used for further analysis.

Wood density

Wood density (WD in g cm^{-3}) indicates the capacity of mechanical support and toughness construction (Poorter et al. 2018). To assess the difference of WD among these four varieties, we measured the wood density of same samples ($\sim$40–58 samples per variety, Table S1 available as Supplementary data at *Tree Physiology* Online). We used a double-bladed saw to cut a branch segment with a thickness of 2 mm for each individual and then air-dried. Afterwards, we used an indirect-reading X-ray densitometer and measured the longest orthogonal radial density profiles by analyzing the scanned images with WinDEN-DRO software (Guay et al. 1992), inducing a series of wood density data from the bark to the pith. To get a clean dataset of wood density for all rings, we deleted the data corresponding to the bark and pith. To classify the earlywood and latewood, we used the two following criteria: (i) WindDENDRO automatically identified earlywood and latewood, and (ii) the distribution

curves were carefully checked with the images of the stem sections. For detailed methods, see Lamy et al. (2012). For each individual we calculated the mean wood density for earlywood (WD_E, g cm^{-3}) and latewood (WD_L, g cm^{-3}).

Wood anatomy

To explore how wood anatomical traits differ among these four varieties, we kept two 2–3 cm segments for three to five individuals per variety for measuring pit and tracheid traits (Table S1 available as Supplementary data at *Tree Physiology* Online). Small pit size and strong pit sealing contribute to high embolism resistance of conifer species (Song et al. 2022a), we therefore measured pit traits related to pit size (i.e., pit aperture diameter, pit membrane diameter and torus diameter) and pit sealing (margo flexibility, torus overlap and valve effect) based on earlywood for three individuals per variety. We used a scanning electron microscopy at the PHENOBOIS platform (SEM, PhenomG2 pro; Talence, France) to take pictures. Afterwards, we used ImageJ 1.53 k to measure pit aperture diameter (DPA, μm), pit membrane diameter (DPM, μm) and torus diameter (DT, μm) for 30 pits per variety. Margo flexibility (MF) was calculated [MF = (DPM − DT)/DPM], and a high value indicates higher ability of a torus to be deflected against the pit aperture. To assess the sealing capacity of pit aperture during an embolism event, torus overlap was calculated by comparing the torus against the aperture [TO = (DT − DPA)/DT]. High margo flexibility and large torus overlap both contribute to a strong valve effect [VE = MF × TO], which is an estimation of the ability of a torus to seal the pit. These composite measurements have been previously described in Delzon et al. (2010) (see also Bouche et al. 2014, Song et al. 2022a). All traits were averaged per individual.

To estimate the contribution of tracheids to water transport efficiency, we measured tracheid-level anatomical traits for five individuals per variety using light microscopy, i.e., tracheid diameter (D, μm), tracheid density (i.e., the tracheid number per area; TD, # mm^{-2}), wall thickness (Tw, μm), thickness to span ratio (TSR, μm) and tracheid lumen fraction (i.e., the proportion of tracheid lumen area over the corresponding xylem area; TLF, μm μm^{-1}). We used a sliding microtome to section samples to a thickness around 20 μm, and stained with safranine for 15 min, then washed with distilled water, ethanol 50, 75, 96 and 100%. Samples were then fixed and pictures were taken with the LAS V 3.8 software connected to a light microscope (DM2500M; Leica Microsystems, Wetzlar, Germany). For all images, tracheid traits were measured on a complete radial section using ImageJ version 1.53 k (Schneider et al. 2012), and at least 200 tracheids per individual were measured. Although earlywood in conifers contributes 90% of total hydraulic conductivity (Domec and Gartner 2002), we performed our measurements on both earlywood and latewood separately. Thickness to span ratio (TSR) was calculated as the
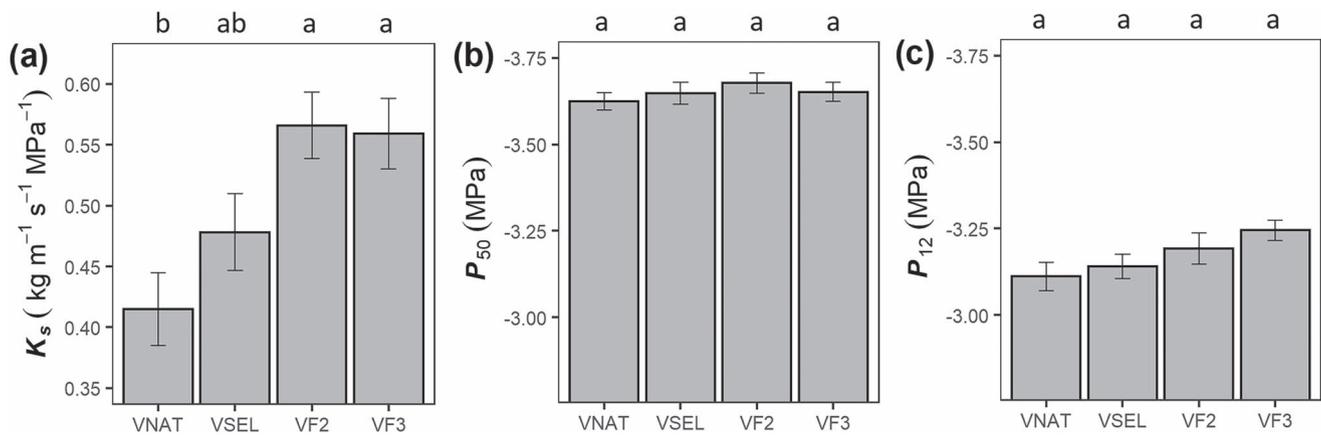
Figure 1. Mean values of (a) K_s (xylem specific hydraulic conductivity), (b) P_{50} (xylem pressure inducing 50% loss in hydraulic conductance) and (c) P_{12} (xylem pressure inducing 12% loss in hydraulic conductance) for four varieties of *Pinus pinaster* ($n = 57–59$ individuals per variety with increasing breeding intensity, i.e., VNAT, VSEL, VF2 and VF3). The bars indicate standard error. Different letters indicate significant differences ($p < 0.05$) using the post-hoc HSD test.

square of double wall thickness over tracheid diameter (Song et al. 2022a), and hydraulic diameter was calculated based on the weighted tracheid diameter (Dh, μm) for both earlywood and latewood to correct the irregular tracheid shape (Poorter et al. 2010):

$$TSR = \left(\frac{2 \times Tw}{D}\right)^2 \tag{3}$$

$$Dh = \sqrt[4]{\frac{1}{n}\sum_{i=1}^{n} Di^4} \tag{4}$$

where T_w is wall thickness, D is the adjacent tracheid diameter, and D_i is the ith measured tracheid diameter.

Statistical analyses

To assess whether different varieties of breeding intensity have different hydraulic traits (i.e., embolism resistance and hydraulic conductivity) and wood anatomical traits, two-way analysis of variance (ANOVA; variety and stand) was performed to take into account all remaining residual effects related to stand. We also used post-hoc Tukey (HSD) tests to compare varieties one by one. To further evaluate how wood anatomical traits impact hydraulic strategy, we performed Pearson correlation and regression analyses between hydraulic and anatomical traits. To meet the normality and homogeneity of variance assumptions of residuals, data were transformed when necessary using a log or square root transformation. All data were analyzed using R version 4.1.2 (R Core Team 2021).

Results

Differences in hydraulic traits between varieties of genetically improved maritime pine

Hydraulic conductivity (K_s) varied significantly among the four varieties of maritime pines (ANOVA, Figure 1a, Table 2).

The comparison of the estimated coefficients from the linear model showed that the VF2 and VF3 (coefficient of 0.23 and 0.21 kg m^{-1} s^{-1} MPa^{-1}, respectively) are significantly larger than that of VNAT and VSEL (coefficient of 0 and -0.08 kg m^{-1} s^{-1} MPa^{-1}, respectively) with an intercept of 0.34 kg m^{-1} s^{-1} MPa^{-1}. However, no difference in K_s was detected between the two 'improved' varieties. The two varieties of genetically improved *P. pinaster* (i.e., VF2 and VF3) had significantly higher hydraulic conductivity compared with the natural variety (VNAT). Embolism resistance (P_{12}, Figure 1c; P_{50}, Figure 1b; and P_{88}, Figure S2a) did not vary significantly among these four varieties. In contrast, VF3 had a higher slope of vulnerability curve than VNAT and VSEL (Figure S2b available as Supplementary data at *Tree Physiology* Online). In addition, there was no significant relationship between hydraulic conductivity and embolism resistance (Figure S3 available as Supplementary data at *Tree Physiology* Online).

Differences in wood traits between varieties of genetically improved maritime pine

Both wood density of earlywood (ANOVA, Figure 2a) and latewood (ANOVA, Figure 2b) varied significantly across these four varieties (Table 2). The wood density of earlywood was significantly lower for the natural variety compared with the other three genetically improved varieties, whereas the wood density of latewood for VNAT, VF2 and VF3 was significantly lower than VSEL. In summary, there was a trend that genetically improved varieties even had a higher earlywood density than unimproved material, but no clear trend was apparent for latewood.

We then compared the underlying wood anatomical traits such as the size of tracheids and pits that play a role in hydraulic function. Anatomical traits of earlywood varied significantly among the different varieties (Figure 3, Table 2). The most improved variety (VF3) had larger hydraulic diameter compared

Table 2. The differences in hydraulic traits related to four varieties of *Pinus pinaster* with increasing breeding intensity or study stand using two-way ANOVA. Bold and underlined values indicate significant differences. For trait abbreviations see Table 1.

| Trait	Variety		Stand	
	F-value	*p*-value	*F*-value	*p*-value
P_{12}	$F_{3,224} = 2.39$	0.07	$F_{4,224} = 1.30$	0.27
P_{50}	$F_{3,224} = 0.53$	0.66	$F_{4,224} = 2.79$	**0.03**
P_{88}	$F_{3,224} = 1.48$	0.22	$F_{4,224} = 1.72$	0.15
Slope	$F_{3,224} = 3.93$	**<0.01**	$F_{4,224} = 0.51$	0.73
K_s	$F_{3,224} = 8.42$	**<0.001**	$F_{4,224} = 7.64$	**<0.001**
WD_E	$F_{3,177} = 11.01$	**<0.001**	$F_{6,177} = 0.97$	0.45
WD_L	$F_{3,176} = 11.01$	**<0.001**	$F_{6,176} = 1.90$	0.08
Dh_E	$F_{3,13} = 6.17$	**<0.01**	$F_{3,13} = 0.53$	0.67
TD_E	$F_{3,13} = 3.71$	**0.04**	$F_{3,13} = 0.17$	0.91
Tw_E	$F_{3,13} = 0.57$	0.64	$F_{3,13} = 0.94$	0.45
TSR_E	$F_{3,13} = 6.66$	**<0.01**	$F_{3,13} = 0.94$	0.45
TLF_E	$F_{3,13} = 10.62$	**<0.001**	$F_{3,13} = 1.10$	0.38
Dh_L	$F_{3,13} = 1.17$	0.36	$F_{3,13} = 0.84$	0.50
TD_L	$F_{3,13} = 1.78$	0.20	$F_{3,13} = 0.36$	0.78
Tw_L	$F_{3,13} = 0.11$	0.95	$F_{3,13} = 0.20$	0.90
TSR_L	$F_{3,13} = 0.10$	0.96	$F_{3,13} = 0.58$	0.64
TLF_L	$F_{3,13} = 0.49$	0.69	$F_{3,13} = 0.34$	0.80
DPM	$F_{3,6} = 0.91$	0.49	$F_{2,6} = 0.26$	0.78
DT	$F_{3,6} = 1.35$	0.35	$F_{2,6} = 0.47$	0.64
DPA	$F_{3,6} = 0.67$	0.60	$F_{2,6} = 0.04$	0.96
MF	$F_{3,6} = 5.41$	**0.04**	$F_{2,6} = 1.37$	0.32
TO	$F_{3,6} = 1.55$	0.30	$F_{2,6} = 2.43$	0.17
VE	$F_{3,6} = 4.51$	0.06	$F_{2,6} = 2.70$	0.15
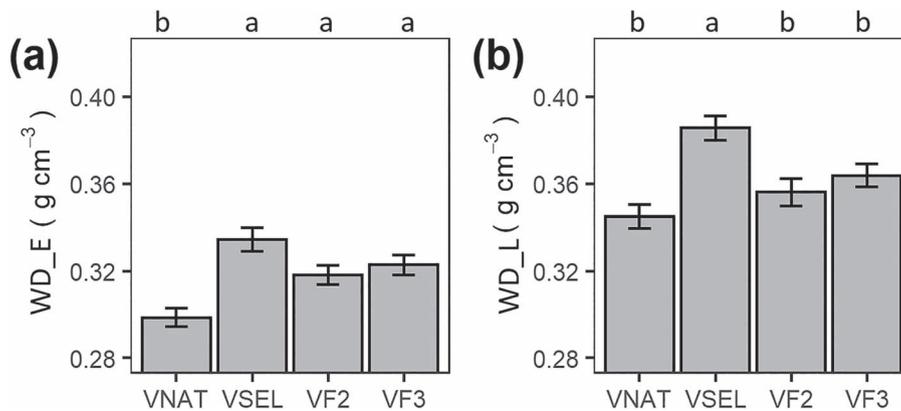
Figure 2. Mean values of wood density for (a) earlywood and (b) latewood among four varieties of *Pinus pinaster* ($n = 40$–57 individuals per variety with increasing breeding intensity, i.e., VNAT, VSEL, VF2 and VF3). The bars indicate standard error. Different letters indicate significant differences ($p < 0.05$) using the post-hoc HSD test.

with VNATA and VSEL (Dh_E, Figure 3a), a higher lumen area to whole area ratio (TLF_E, Figure 3e) than VSEL, a smaller tracheid density (TD_E, Figure 3b) than VSEL, and a smaller thickness to span ratio (TSR_E, Figure 3d) than VNAT and VSEL. For the latewood tracheid traits, there was no significant variation across the four varieties (Figure 3f–j). Overall, we observed a trend for artificial selection to increase hydraulic diameter and lumen ratio, and to decrease tracheid density and thickness to span ratio in the earlywood, rather than in the latewood.

Among the four varieties there were no significant differences of pit membrane structure and function such as pit membrane diameter (DPM, Figure 4a), torus diameter (DT, Figure 4b), aperture diameter (DPA, Figure 4c), torus overlap (TO, Figure 4e) and valve effect (VE, Figure 4f, Table 2). Margo flexibility only varied significantly across two varieties, i.e., VF2 had significantly lower margo flexibility than VSEL (Figure 4d).

Relationships between wood and hydraulic traits

The significant differences in hydraulic conductivity across these four varieties of improved material were further explained by the tracheid traits of earlywood, but not by those of latewood (Figure 5, Table 3). In particular, hydraulic conductivity (K_s)
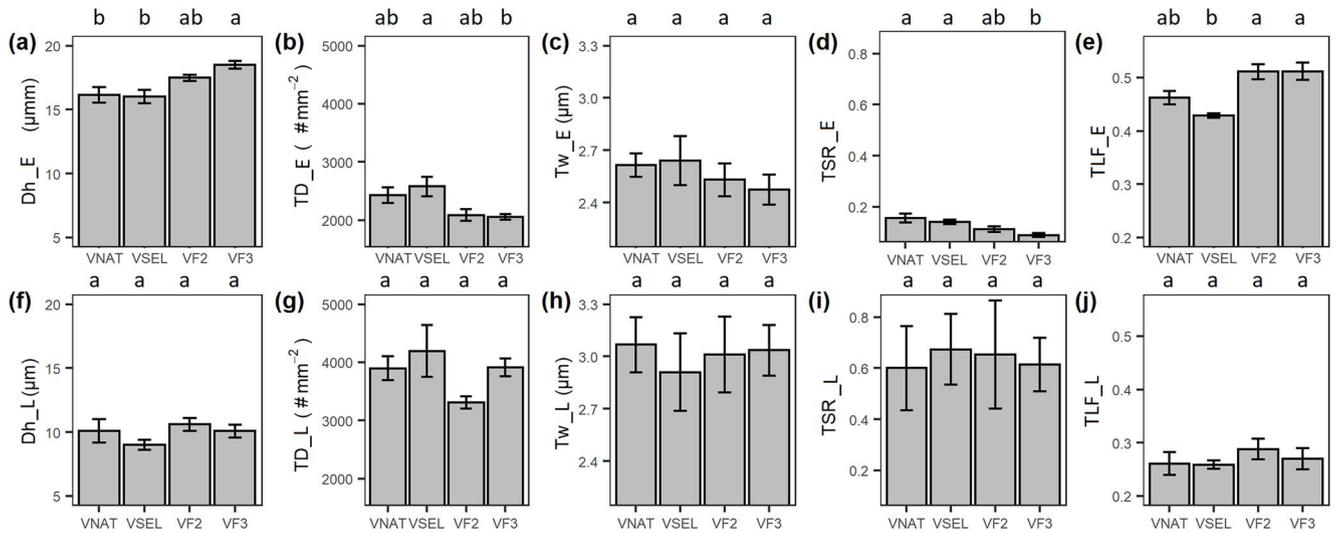
Figure 3. Mean values of tracheid traits based on earlywood (top panel, a–e) and latewood (bottom panel, f–j) for four varieties of *Pinus pinaster* ($n = 5$ individuals per variety with increasing breeding intensity, i.e., VNAT, VSEL, VF2 and VF3): (a) the hydraulic diameter of earlywood, (b) tracheid density of earlywood, (c) wall thickness of earlywood, (d) thickness to span ratio of earlywood, (e) lumen to xylem ratio of earlywood, (f) hydraulic diameter of latewood, (g) tracheid density of latewood, (h) wall thickness of latewood, (i) thickness to span ratio of latewood and (j) lumen to xylem ratio of latewood. The bars indicate standard error. Different letters indicate significant differences ($p < 0.05$) using the post-hoc HSD test. For trait abbreviations see Table 1.
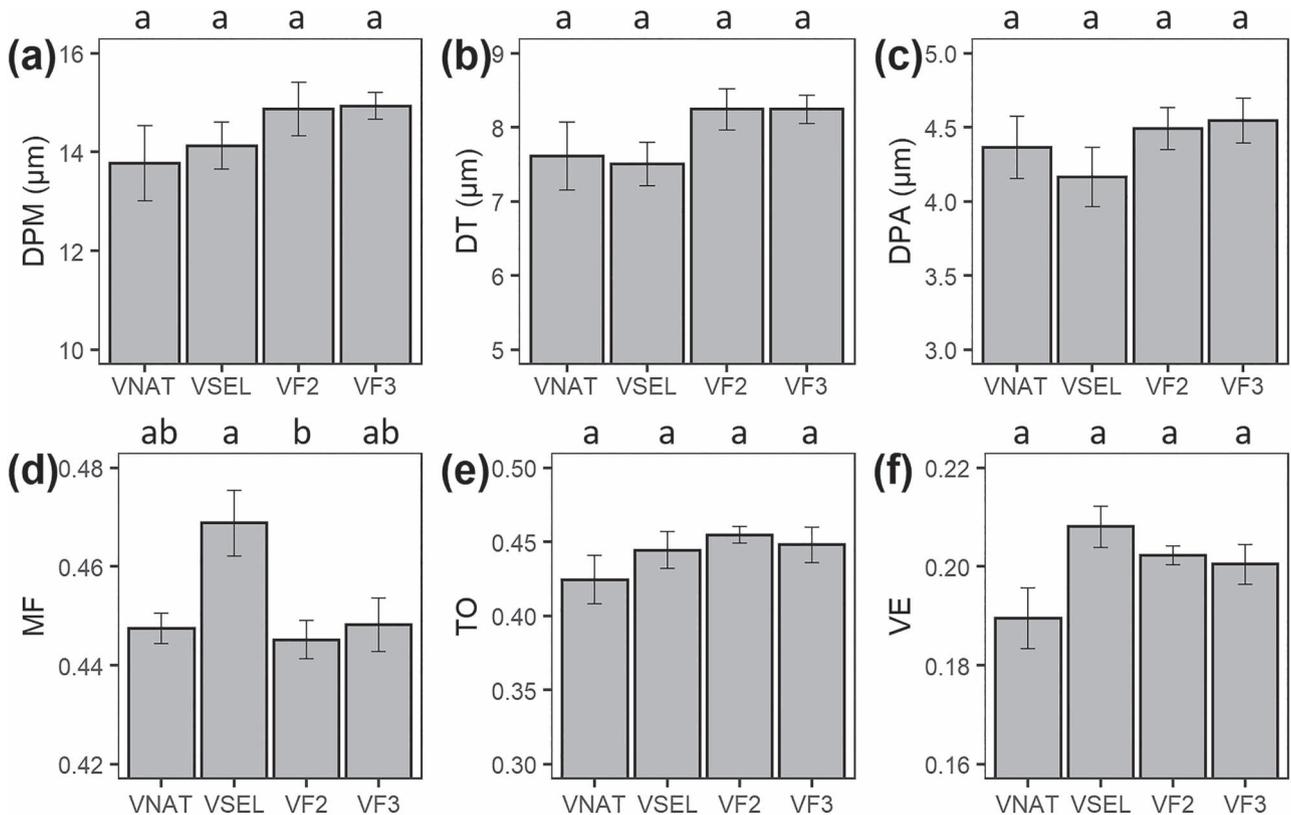

Figure 4. Mean values of pit size (a–c) and pit sealing (d–f) based on earlywood for four varieties of *Pinus pinaster* ($n = 3$ individuals per variety with increasing breeding intensity, i.e., VNAT, VSEL, VF2 and VF3): (a) DPM, pit membrane diameter; (b) DT, torus diameter; (c) DPA, pit aperture diameter; (d) MF, margo flexibility; (e) TO, torus overlap and (f) VE, valve effect. The bars indicate standard error. Different letters indicate significant differences ($p < 0.05$) using the post-hoc HSD test. For trait abbreviations see Table 1.
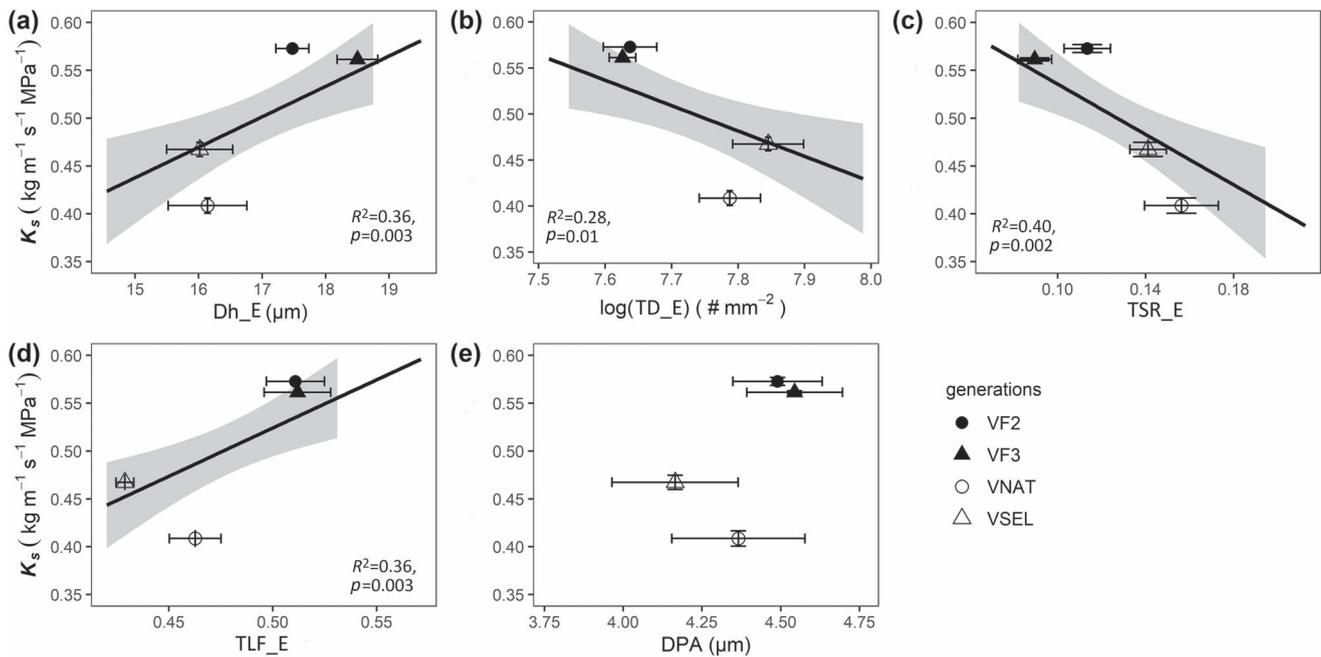
Figure 5. Bi-variate relationships between hydraulic conductivity, earlywood tracheid traits (a–d) and pit aperture diameter (e): (a) hydraulic diameter of earlywood (Dh_E), (b) tracheid density of earlywood (TD_E), (c) thickness to span ratio of earlywood (TSR_E), (d) tracheid lumen fraction of earlywood (TLF_E) and (e) pit aperture diameter (DPA). Each symbol represents mean trait values over three or five individuals per population with increasing breeding intensity, i.e., VNAT, VSEL, VF2 and VF3. The bars indicate standard error. None significant regression is not shown. Significant regression lines ($p < 0.05$) and 95 confidence intervals (gray), R^2 and p-value are shown.

increased with the hydraulic diameter ($N = 20$, $R^2 = 0.36$, $p = 0.001$) and lumen ratio ($N = 20$, $R^2 = 0.36$, $p = 0.001$), decreased with tracheid density and thickness to span ratio of the earlywood (Figure 5a–d, Table 3), but had no relationships with pit aperture size (Figure 5e) and latewood tracheid values (Figure S4 available as Supplementary data at *Tree Physiology* Online). Embolism resistance increased with torus diameter and decreased with margo flexibility, but not related to other pit traits (Table S2 as Supplementary data at *Tree Physiology* Online).

Discussion

For the first time to our knowledge, we tested whether the genetic selection of trees for better growth and straightness has had an impact on water transport efficiency and drought resistance in their vascular system. To this end, we compared both efficiency and safety of hydraulic traits (i.e., hydraulic conductivity and embolism resistance) and wood anatomical traits among four varieties of maritime pines with increasing selection intensity. Our results evidence that the most improved varieties (VF2 and VF3) had higher hydraulic efficiency than the natural (VNAT) and the first generation selected variety (VSEL), which was mostly explained by larger tracheid size and higher tracheid lumen fraction. On the other hand, improved varieties had no striking differences in embolism resistance, demonstrating that genetic improvement has not been at the expense of the drought resilience of the species. In other

words, it enables high timber productivity, but no reduced embolism resistance to cope with drought events. Here, we discuss how different varieties differ in hydraulic functions, how wood anatomical traits determine the high hydraulic conductivity of improved variety and the implication for sustainable forest management.

The improved maritime pine increased xylem hydraulic conductivity

We hypothesized that the improved varieties would have higher hydraulic conductivity, since high xylem hydraulic conductivity improves water transport, and hence high stomatal conductance and growth rate (Brodribb et al. 2009). As expected, we found improved varieties (VF2 and VF3) had higher hydraulic conductivity (i.e., K_s) than the natural variety (VNAT, Figure 1a). Therefore, the improved varieties adopt an acquisitive strategy to grow faster thanks to high K_s and this strategy is not at the cost of reduced embolism resistance (i.e., $|P_{50}|$), demonstrating the lack of a trade-off between K_s and $|P_{50}|$ in maritime pine. Previous studies failed to find this trade-off across species (Gleason et al. 2016, Larter et al. 2017), as well as the lack of intraspecific trade-off (Sáenz-Romero et al. 2013, Lamy et al. 2014). The control by a different combination of traits may explain the lack of this trade-off, since K_s is positively determined by the fourth power of tracheid size (Tyree and Ewers 1991), tracheid length, margo pores and total area of inter-conduit pits (Pittermann et al. 2006,

Table 3. Pearson correlations among embolism resistance, hydraulic conductivity and tracheid traits ($N = 20$, five individuals per variety). Bold and underlined values indicate significant correlations ($p < 0.05$). For trait abbreviations see Table 1.

Traits	K_s	$|P_{12}|$	$|P_{50}|$	$|P_{88}|$	Slope	Dh_E	TD_E	Tw_E	TSR_E	TLF_E	Dh_L	TD_L	Tw_L	TSR_L	TLF_L
K_s	1.00														
$|P_{12}|$	0.30	1.00													
$|P_{50}|$	0.15	**0.76**	1.00												
$|P_{88}|$	−0.17	−0.11	**0.56**	1.00											
Slope	0.33	**0.74**	0.17	**−0.71**	1.00										
Dh_E	**0.62**	0.13	−0.03	−0.22	0.22	1.00									
TD_E	**−0.54**	−0.04	0.23	0.40	−0.26	**−0.89**	1.00								
Tw_E	−0.23	−0.34	−0.30	−0.01	−0.29	0.13	−0.12	1.00							
TSR_E	**−0.64**	−0.25	−0.12	0.15	−0.31	**−0.58**	**0.51**	**0.60**	1.00						
TLF_E	**0.61**	0.18	0.07	−0.14	0.22	**0.73**	**−0.74**	−0.24	**−0.61**	1.00					
Dh_L	0.22	0.07	−0.23	**−0.45**	0.33	0.43	−0.41	0.04	−0.19	**0.47**	1.00				
TD_L	−0.31	−0.08	0.25	**0.49**	−0.33	**−0.54**	**0.65**	−0.42	0.06	−0.32	**−0.56**	1.00			
Tw_L	0.02	0.09	0.37	**0.46**	−0.26	0.20	−0.11	**0.47**	0.23	0.10	−0.35	−0.19	1.00		
TSR_L	0.01	0.12	**0.45**	**0.54**	−0.29	−0.09	0.12	0.20	0.17	−0.16	**−0.74**	0.15	**0.83**	1.00	
TLF_L	0.22	−0.01	−0.40	**−0.61**	0.38	0.39	−0.41	0.13	−0.13	0.38	**0.76**	**−0.47**	**−0.46**	**−0.74**	1.00

Schulte et al. 2015). These characteristics enable conifer trees to grow fast with high hydraulic efficiency. In comparison, $|P_{50}|$ is positively characterized by the strong valve effects through a flexible margo and large torus overlap to seal the pit aperture when embolism occurs (Delzon et al. 2010, Song et al. 2022a), which is strikingly different from the above-mentioned factors that affect K_s. Thus, the decoupled relationship between hydraulic conductivity and embolism resistance allows plants to grow faster without losing their capacity to tolerate drought events.

Mechanisms underlying hydraulic conductivity

We expected that improved varieties for growth would have lower wood density (WD), since improved varieties grow faster and they should have wider tracheid lumens to support increased water transport, carbon gain and growth (Poorter et al. 2010, 2019). Nevertheless, our findings challenge this assumption: instead, the improved varieties have higher wood density for earlywood (Figure 2a), suggesting that breeding higher hydraulic conductivity does not reduce wood construction cost in maritime pine. Considering that mechanically stronger wood can support against implosion by negative pressure (Hacke et al. 2001), this result indicates that xylem collapse or conduit implosion is not affected by artificial selection. The lack of relationship between hydraulic conductivity and wood density has been also reported for angiosperms (Schumann et al. 2019) and other gymnosperms (Song et al. 2022a). We therefore conclude that hydraulic conductivity is likely to be determined by anatomical conduit and pit traits for both gymnosperms and angiosperms, rather than wood density.

We further investigated whether anatomical trait values could explain the high hydraulic conductivity of improved varieties.

It was expected that hydraulic conductivity would increase with tracheid size, pit size and lumen fraction, and decrease with tracheid density, since large tracheids and pits allow for more efficient water transport. We indeed found that the most improved variety (VF3) had significantly larger tracheids than wild material (VNAT) and even the first generation of breeding (VSEL). Moreover, the linear regression reveals that hydraulic conductivity increased with earlywood hydraulic diameter and tracheid lumen fraction, decreased with thickness to span ratio and tracheid density (Figure 5a–d), supporting the theory that large conduits and lumen areas favor water transport (Russo et al. 2010). However, the different varieties did not differ in latewood tracheid trait values. Besides, latewood tracheid traits values did not correlate with hydraulic conductivity, due to the fact that earlywood contributes to 90% of total flow for hydraulic conductivity of conifer species (Domec and Gartner 2002).

We also failed to find a positive effect of pit aperture size on hydraulic conductivity (Figure 5e). Large tracheid size and high tracheid lumen fraction enable large amounts of water transport for plants, thus leading to high hydraulic conductivity (Tyree and Ewers 1991, Rungwattana and Hietz 2018). Although some studies did not find a positive relationship between tracheid size and hydraulic conductivity (Larter et al. 2017, Song et al. 2022a), the other tracheid anatomical traits still need to be further investigated, such as tracheid length and margo porosity. The lack of relationship between pit aperture and hydraulic conductivity may indicate that hydraulic conductivity is mainly determined by tracheid-level traits, whereas pit aperture has a stronger link with embolism resistance. This is also in line with the lack of trade-off between hydraulic conductivity and embolism resistance. Our study stresses the importance of tracheid traits for predicting hydraulic conductivity of improved breeding materials.

Maritime pine breeding programme had no impact on embolism resistance

Contrary to our hypothesis, the genetically improved maritime pine varieties do not display a lower resistance to embolism than non-improved varieties (similar $|P_{50}|$). We indeed found high stability of embolism resistance among the four studied varieties of maritime pines with increasing breeding selection for height growth and stem straightness, indicating that breeding programme has no effects on the drought resistance of the vascular system of *P. pinaster*. Contrary to *P. sylvestris* for which fast-growing trees adopt an acquisitive strategy at the cost of reduced embolism resistance (Sterck et al. 2012), we failed to find a reduced embolism resistance in fast-growing maritime pines. Our result is consistent with other studies showing a lack of trade-off between growth and embolism resistance among conifer species (Song et al. 2022*a*). This result implies the lack of trade-off between growth and embolism resistance is not only true across species but also within species. We found similar values of P_{50} among the four varieties of improved material, probably owing to the fact that P_{50} is a genetically constrained trait as reported by Lamy et al. (2011, 2014). The authors demonstrated that P_{50} is a canalized trait in maritime pine by showing that between-population variability of P_{50} was significantly lower than would be expected under a hypothesis of genetic drift alone. Their quantitative genetics analysis also showed that P_{50} presented a significant heritability (above 0.4), higher than that estimated for other traits such as growth. However, we did not study different genotypes of P_{50} and K_s in arid environments (e.g., in Southern Spain), where phenotypic plasticity might lead to differences. We assume that P_{50} would be still phylogenetically controlled across species and K_s would be smaller in more arid environments. It still needs to be tested in more arid areas.

We further tested whether the breeding process had an impact on pit traits, and whether pit traits related to size and sealing could explain embolism resistance. We found that artificial selection had no impact on pit size and pit sealing, but had limited impacts on margo flexibility. We found no effect of pit dimension traits on embolism resistance, likely because of the low variability in P_{50} and pit traits themselves (e.g., pit aperture size ranged from 4.16 to 4.54 μm). Hence, pit trait values explained no variation of P_{50}. The interspecific values of P_{50} could be different (Li et al. 2009, Ahmad et al. 2018, Bartlett and Sinclair 2021) or the same across crop cultivars (Stiller et al. 2003), but few studies assessed tree species. Our study suggests that improved breeding could have no impacts on embolism resistance in both crop cultivars and tree species. It confirms the approach used to obtain better performing and more resilient genotypes to better adapt to increasing drought during the vegetative period.

Implication for future study and forest management

We found that the highest yielding varieties did not have reduced resistance to drought, a benefit for designing a climate-smart forest with high timber production that can mitigate the climate warming impacts via carbon sequestration (Nabuurs et al. 2018). Given that the increased intensity and frequency of drought events have triggered tree mortality worldwide (Choat et al. 2018, Brodribb et al. 2020, McDowell et al. 2022), it is urgent to select tree species with high drought resilience to reduce mortality risk (DeSoto et al. 2020, Song et al. 2022*c*), which is critical for maintaining a resilient forest ecosystem. Based on our results we are confident about the possibility of selecting genotypes with higher drought resistance without impacting growth performance.

A breeding programme is an effective method to maintain a highly productive forest, providing guidelines for sustainable forest management. Different genotypes within and across species have been studied for comparing their embolism resistance (Sáenz-Romero et al. 2013, Larter et al. 2017, Song et al. 2022*a*), but the relationship between embolism resistance and growth remains equivocal (Guet et al. 2015). The negative relationships found in *Cedrus libani* (Ducrey et al. 2008) turns out to be positive across poplar hybrids (Fichot et al. 2010), or even disappear in both angiosperms (Guet et al. 2015, Kumar et al. 2022) and gymnosperms (Sterck et al. 2012, Song et al. 2022*a*). Providing guidelines for species selection with high drought resistance and high growth is thus urgently needed to better design the silviculture system adapted to each pedo-climatic situation. Our study shows that breeding programmes can provide genetic material with high productivity in a drier climate.

Conclusions

We compared the hydraulic strategies of different varieties of *P. pinaster* corresponding to increasing intensity of breeding for height growth and stem straightness. Compared with natural variety (unimproved material), the improved varieties had higher hydraulic conductivity, with wider tracheid lumens and narrower tracheid walls, higher lumen fraction and lower tracheid density, but no reduction of embolism resistance. The decoupled relationship between hydraulic conductivity and embolism resistance enables improved material with high hydraulic efficiency to grow fast and cope with more frequent and intense drought events without reduced drought resistance. Our study is the first study to reveal the hydraulic mechanisms of different varieties within a tree breeding scheme. It provides guidelines for forest management and breeding strategies for future studies and other forest tree species.

Supplementary data

Authors' contributions

X.P.B. and S.D. initiated the research project. X.P.B. and Y.S. contributed to data collection, and Y.S. analyzed the data. S.D., M.L. and C.P. contributed to the improvements of data analyses, and Y.S. wrote the first draft that was edited by all authors.

Acknowledgments

Conflict of interest

The authors declare no conflict of interest.

References

Ahmad HB, Lens F, Capdeville G, Burlett R, Lamarque LJ, Delzon S (2018) Intraspecific variation in embolism resistance and stem anatomy across four sunflower (*Helianthus annuus* L.) accessions. Physiol Plant 163:59–72.

Allen CD, Macalady AK, Chenchouni H et al. (2010) A global overview of drought and heat-induced tree mortality reveals emerging climate change risks for forests. For Ecol Manage 259:660–684.

Anderegg WR, Klein T, Bartlett M, Sack L, Pellegrini AF, Choat B, Jansen S (2016) Meta-analysis reveals that hydraulic traits explain cross-species patterns of drought-induced tree mortality across the globe. Proc Natl Acad Sci USA 113:5024–5029.

Bartlett MK, Sinclair G (2021) Temperature and evaporative demand drive variation in stomatal and hydraulic traits across grape cultivars. J Exp Bot 72:1995–2009.

Bouche PS, Larter M, Domec J-C, Burlett R, Gasson P, Jansen S, Delzon S (2014) A broad survey of hydraulic and mechanical safety in the xylem of conifers. J Exp Bot 65:4419–4431.

Bouche PS, Jansen S, Sabalera JC, Cochard H, Burlett R, Delzon S. (2016) Low intra-tree variability in resistance to embolism in four Pinaceae species. Annals of Forest Science 73:681–689.

Bouffier L, Raffin A, Alía R (2013) Maritime pine—*Pinus pinaster* Ait. Best practice for tree breeding in Europe. Skogforsk, Uppsala, pp 65–75.

Brodribb TJ, McAdam SA, Jordan GJ, Feild TS (2009) Evolution of stomatal responsiveness to CO2 and optimization of water-use efficiency among land plants. New Phytol 183:839–847.

Brodribb TJ, Powers J, Cochard H, Choat B (2020) Hanging by a thread? Forests and drought. Science 368:261–266.

Burlett R, Parise C, Capdeville G, Cochard H, Lamarque LJ, King A, Delzon S (2022) Measuring xylem hydraulic vulnerability for long-vessel species: an improved methodology with the flow centrifugation technique. Ann For Sci 79:1–16.

Choat B, Jansen S, Brodribb TJ et al. (2012) Global convergence in the vulnerability of forests to drought. Nature 491:752–755.

Choat B, Brodribb TJ, Brodersen CR, Duursma RA, Lopez R, Medlyn BE (2018) Triggers of tree mortality under drought. Nature 558:531–539.

Cochard H, Damour G, Bodet C, Tharwat I, Poirier M, Améglio T (2005) Evaluation of a new centrifuge technique for rapid generation of xylem vulnerability curves. Physiol Plant 124:410–418.

Corona P (2019) Global change and silvicultural research. Ann Silvicult Res 43:1–3.

Delzon S, Douthe C, Sala A, Cochard H (2010) Mechanism of water-stress induced cavitation in conifers: bordered pit structure and function support the hypothesis of seal capillary-seeding. Plant Cell Environ 33:2101–2111.

DeSoto L, Cailleret M, Sterck F et al. (2020) Low growth resilience to drought is related to future mortality risk in trees. Nat Commun 11:545–549.

Domec JC, Gartner B (2003) Relationship between growth rates and xylem hydraulic characteristics in young, mature and old-growth ponderosa pine trees. Plant Cell Environ 26:471–483.

Domec JC, Gartner BL (2002) How do water transport and water storage differ in coniferous earlywood and latewood? J Exp Bot 53:2369–2379.

Ducrey M, Huc R, Ladjal M, Guehl J-M (2008) Variability in growth, carbon isotope composition, leaf gas exchange and hydraulic traits in the eastern Mediterranean cedars *Cedrus libani* and *C. brevifolia*. Tree Physiol 28:689–701.

Fichot R, Barigah TS, Chamaillard S, Le Thiec D, Laurans F, Cochard H, Brignolas F (2010) Common trade-offs between xylem resistance to cavitation and other physiological traits do not hold among unrelated *Populus deltoides* × *Populus nigra* hybrids. Plant Cell Environ 33:1553–1568.

GIS GPMF. (2014) Matériel végétal de reboisement. Les cahiers de la reconstitution du Groupe Pin500 Maritime du Futur, n.4.

Gleason SM (2015) Evolutionary outcomes should inform strategies to increase drought tolerance. Nat Plants 1:1–1.

Gleason SM, Westoby M, Jansen S et al. (2016) Weak trade-off between xylem safety and xylem-specific hydraulic efficiency across the world's woody plant species. New Phytol 209:123–136.

González-Muñoz N, Sterck F, Torres-Ruiz JM et al. (2018) Quantifying in situ phenotypic variability in the hydraulic properties of four tree species across their distribution range in Europe. PLoS One 13:e0196075.

Guay R, Gagnon R, Morin H (1992) A new automatic and interactive tree ring measurement system based on a line scan camera. For Chron 68:138–141.

Guet J, Fichot R, Lédée C, Laurans F, Cochard H, Delzon S, Bastien C, Brignolas F (2015) Stem xylem resistance to cavitation is related to xylem structure but not to growth and water-use efficiency at the within-population level in *Populus nigra* L. J Exp Bot 66:4643–4652.

Hacke U, SperryJ 2001. Functional and ecological xylem anatomy: perspectives in plant ecology, evolution and systematics. London, UK: Springer, 4, 97–115. https://doi.org/10.1078/1433-8319-00017.

Hacke UG, Jansen S (2009) Embolism resistance of three boreal conifer species varies with pit structure. New Phytol 182:675–686.

Hacke UG, Sperry JS, Pockman WT, Davis SD, McCulloh KA (2001) Trends in wood density and structure are linked to prevention of xylem implosion by negative pressure. Oecologia 126:457–461.

Hacke UG, Sperry JS, Pittermann J (2004) Analysis of circular bordered pit function II. Gymnosperm tracheids with torus-margo pit membranes. Am J Bot 91:386–400.

Hajek P, Leuschner C, Hertel D, Delzon S, Schuldt B (2014) Trade-offs between xylem hydraulic properties, wood anatomy and yield in Populus. Tree Physiol 34:744–756.

Jansen S, McAdam S (2019) Pits with aspiration explain life expectancy of a conifer species. Proc Natl Acad Sci USA 116:14794–14796.

Kumar M, Waite P-A, Paligi SS, Schuldt B (2022) Influence of juvenile growth on xylem safety and efficiency in three temperate tree species. Forests 13:909. https://doi.org/10.3390/f13060909.

Lamy J-B, Bouffier L, Burlett R, Plomion C, Cochard H, Delzon S (2011) Uniform selection as a primary force reducing population genetic differentiation of cavitation resistance across a species range. PloS one 6:e23476. https://doi.org/10.1371/journal.pone.0023476.

Lamy J-B, Lagane F, Plomion C, Cochard H, Delzon S (2012) Micro-evolutionary patterns of juvenile wood density in a pine species. Plant Ecol 213:1781–1792.

Lamy JB, Delzon S, Bouche PS, Alia R, Vendramin GG, Cochard H, Plomion C (2014) Limited genetic variability and phenotypic plasticity detected for cavitation resistance in a Mediterranean pine. New Phytol 201:874–886.

Larter M, Pfautsch S, Domec JC, Trueba S, Nagalingum N, Delzon S (2017) Aridity drove the evolution of extreme embolism resistance and the radiation of conifer genus *Callitris*. New Phytol 215:97–112.

Li S, Li X, Wang J et al. (2022) Hydraulic traits are coupled with plant anatomical traits under drought–rewatering cycles in *Ginkgo biloba* L. Tree Physiol 42:1216–1227.

Li Y, Sperry JS, Shao M (2009) Hydraulic conductance and vulnerability to cavitation in corn (*Zea mays* L.) hybrids of differing drought resistance. Environ Exp Bot 66:341–346.

Liu H, Gleason SM, Hao G, Hua L, He P, Goldstein G, Ye Q (2019) Hydraulic traits are coordinated with maximum plant height at the global scale. Sci Adv 5:eaav1332. https://doi.org/10.1126/sciadv.aav1332.

Marguerit E, Bouffier L, Chancerel E, Costa P, Lagane F, Guehl J-M, Plomion C, Brendel O (2014) The genetics of water-use efficiency and its relation to growth in maritime pine. J Exp Bot 65:4757–4768.

McDowell NG, Sapes G, Pivovaroff A et al. (2022) Mechanisms of woody-plant mortality under rising drought, CO2 and vapour pressure deficit. Nat Rev Earth Environ 3:294–308.

Mullin TJ, Andersson B, Bastien J-C et al. (2011) Economic importance, breeding objectives and achievements. Genetics, genomics and breeding of conifers. New York, USA: Science Publishers and CRC Press, pp 40–127.

Nabuurs G-J, Verkerk PJ, Schelhaas M, González-Olabarria J, Trasobares A, Cienciala E (2018) Climate-Smart Forestry: mitigation implact in three European regions (Vol. 6). From Science to Policy 6. European Forest Institute. EFI: Helsinki, Finland, 6, 31.

Pâques LE (2013) Forest tree breeding in Europe. Netherlands: Springer, 25, pp 229–265. https://doi.org/10.1007/978-94-007-6146-9.

Pittermann J, Sperry JS, Hacke UG, Wheeler JK, Sikkema EH (2006) Inter-tracheid pitting and the hydraulic efficiency of conifer wood: the role of tracheid allometry and cavitation protection. Am J Bot 93:1265–1273.

Poorter L, McDonald I, Alarcón A, Fichtler E, Licona JC, Peña-Claros M, Sterck F, Villegas Z, Sass-Klaassen U (2010) The importance of wood traits and hydraulic conductance for the performance and life history strategies of 42 rainforest tree species. New Phytol 185:481–492.

Poorter L, Castilho CV, Schietti J, Oliveira RS, Costa FR (2018) Can traits predict individual growth performance? A test in a hyperdiverse tropical forest. New Phytol 219:109–121.

Poorter L, Rozendaal D, Bongers F et al. (2019) Wet and dry tropical forests show opposite successional pathways in wood density but converge over time. Nat Ecol Evol 3:928–934.

R Core Team (2021) R: a language and environment for statistical computing. Version 4.1. 2. Springer Nature, Vienna, Austria.

Rungwattana K, Hietz P (2018) Radial variation of wood functional traits reflect size-related adaptations of tree mechanics and hydraulics. Funct Ecol 32:260–272.

Russo SE, Jenkins KL, Wiser SK, Uriarte M, Duncan RP, Coomes DA (2010) Interspecific relationships among growth, mortality and xylem traits of woody species from New Zealand. Funct Ecol 24:253–262.

Sáenz-Romero C, Lamy J-B, Loya-Rebollar E, Plaza-Aguilar A, Burlett R, Lobit P, Delzon S (2013) Genetic variation of drought-induced cavitation resistance among *Pinus hartwegii* populations from an altitudinal gradient. Acta Phys Plant 35:2905–2913.

Schneider CA, Rasband WS, Eliceiri KW (2012) NIH Image to ImageJ: 25 years of image analysis. Nat Methods 9:671–675.

Schuldt B, Knutzen F, Delzon S, Jansen S, Müller-Haubold H, Burlett R, Clough Y, Leuschner C (2016) How adaptable is the hydraulic system of European beech in the face of climate change-related precipitation reduction? New Phytol 210:443–458.

Schulte PJ, Hacke UG, Schoonmaker AL (2015) Pit membrane structure is highly variable and accounts for a major resistance to water flow through tracheid pits in stems and roots of two boreal conifer species. New Phytol 208:102–113.

Schumann K, Leuschner C, Schuldt B (2019) Xylem hydraulic safety and efficiency in relation to leaf and wood traits in three temperate Acer species differing in habitat preferences. Trees 33: 1475–1490.

Serrano-León H, Ahtikoski A, Sonesson J et al. (2021) From genetic gain to economic gain: simulated growth and financial performance of genetically improved *Pinus sylvestris* and *Pinus pinaster* planted stands in France, Finland and Sweden. Forestry 94:512–525.

Song Y, Poorter L, Horsting A, Delzon S, Sterck F (2022a) Pit and tracheid anatomy explain hydraulic safety but not hydraulic efficiency of 28 conifer species. J Exp Bot 73:1033–1048.

Song Y, Sterck F, Sass-Klaassen U, Li C, Poorter L (2022b) Growth resilience of conifer species decreases with early, long-lasting and intense droughts but cannot be explained by hydraulic traits. J Ecol 110:2088–2104.

Song Y, Sterck F, Zhou X, Liu Q, Kruijt B, Poorter L (2022c) Drought resilience of conifer species is driven by leaf lifespan but not by hydraulic traits. New Phytol 235:978–992.

Sperry JS, Hacke UG, Pittermann J (2006) Size and function in conifer tracheids and angiosperm vessels. Am J Bot 93:1490–1500.

Sterck FJ, Zweifel R, Sass-Klaassen U, Chowdhury Q (2008) Persisting soil drought reduces leaf specific conductivity in Scots pine (*Pinus sylvestris*) and pubescent oak (*Quercus pubescens*). Tree Physiol 28:529–536.

Sterck FJ, Martínez-Vilalta J, Mencuccini M et al. (2012) Understanding trait interactions and their impacts on growth in Scots pine branches across Europe. Funct Ecol 26:541–549.

Stiller V, Lafitte HR, Sperry JS (2003) Hydraulic properties of rice and the response of gas exchange to water stress. Plant Physiol 132:1698–1706.

Tyree MT, Ewers FW (1991) The hydraulic architecture of trees and other woody plants. New Phytol 119:345–360.

Tyree MT, Sperry JS (1989) Vulnerability of xylem to cavitation and embolism. Annu Rev Plant Biol 40:19–36.

Tyree MT, Zimmermann MH (2013) Xylem structure and the ascent of sap. Springer Science & Business Media, Palo Alto, CA.

Wang Z, Ding X, Li Y, Xie J (2022) The compensation effect between safety and efficiency in xylem and role in photosynthesis of gymnosperms. Physiol Plant 174:e13617.

Yao GQ, Nie ZF, Turner NC, Li FM, Gao TP, Fang XW, Scoffoni C (2021) Combined high leaf hydraulic safety and efficiency provides drought tolerance in *Caragana* species adapted to low mean annual precipitation. New Phytol 229:230–244.

Zhao H, Jiang Z, Zhang Y, Jiang B, Cai J (2021) Hydraulic efficiency at the whole tree level stably correlated with productivity over years in 9 poplar hybrids clones. For Ecol Manage 496:119382. https://doi.org/10.1016/j.foreco.2021.119382.