By the narrowest of margins: nano-scale modification of pit membranes and the fate of plants during drought. A commentary on: ‘Intervessel pit membrane thickness best explains variation in embolism resistance amongst stems of Arabidopsis thaliana accessions’

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Recent incidences of forest mortality and their direct connection to climate change underscore the importance of a mechanistic understanding of how plants die as a result of drought stress (Adams et al., 2017). Consensus has crystallized around the central role that hydraulic failure plays in the long-term survival of plants as air temperatures rise, soil moisture dwindles and plants fail to replace water lost to transpiration (Brodribb et al., 2020). As the tension in the xylem sap increases, the vascular system exists in a highly vulnerable state in which gas bubbles can enter the xylem, in a process known as air-seeding of xylem conduits (Fig. 1). When left unchecked, that air can rapidly spread between xylem conduits in a process known as runaway cavitation. All plant tissues containing vascular elements are at risk of hydraulic failure, and the formation and spread of gas bubbles in the xylem has now been clearly documented in roots, stems and leaves (Brodribb et al., 2020). Significant losses in hydraulic conductivity due to cavitation ultimately lead to death of the meristematic tissues. Each species has its own mortality threshold, and the water potential resulting in a loss of 50% of the hydraulic conductivity is a now common metric for comparing the relative drought tolerance of species (Choat et al., 2012). Understanding the underlying mechanisms for both the spread of air through the vascular system and the environmental conditions that push species up to and beyond these thresholds is of critical importance. In this issue, Thonglim et al. (2021) provide novel evidence of how nano-scale differences in xylem anatomy can have a cascading effect on the overall vulnerability of plants during drought.

Establishing a set of reliable metrics for quantifying the resistance to the spread of gas bubbles in the xylem and putting those values within a practical context (Choat et al., 2012) now allows more certainty in our predictions of how climate change will affect different plant communities (Skelton et al., 2015; Anderegg et al., 2016). Yet, despite this work aimed at scaling tissue- and whole-plant physiology up to forest and ecosystem level, we still lack robust empirical evidence to support our fundamental assumptions and theory that describe how, when, where and why gas bubbles propagate through xylem networks. Much of our understanding of this process is based on the idea of air-seeding, the physical processes of gas aspirating across the air–water interface between adjacent vascular conduits, which takes place within the microscopic channels embedded in the pit membranes (Fig. 1). Whether or not air moves across these barriers is a function of the physical properties of the pit membranes and the pressure gradient. Thus, despite their diminutive size, often <200 nm in width, pit membranes are a major determinant of plants’ fitness in drought situations (Kaack et al., 2019).

Over the past few decades, investigations into the structure and function of the pit membrane, the geometry of the pit chambers and the conduits themselves have shown important anatomical differences between species (Choat et al., 2008). Pit membrane thickness likely determines both the radius of the smallest pore, and therefore the air-seeding threshold at the air–water interface, but also the tortuosity of that pathway (Kaack et al., 2019). The implications of these seemingly minor differences in thickness and structure have profound implications for how pit membranes function in planta, with strong relationships to drought and freezing tolerance, but also less obvious traits such as branch longevity (Roskilly et al., 2019). These studies, and others both theoretical and empirical, reveal how small variation in pit membrane pore dimensions and xylem network connections affect when and where gas bubbles can spread, and ultimately the vulnerability of the entire network (Mrad et al., 2018; Wason et al., 2021). Emerging from this work is a new appreciation for how xylem networks are constructed, and the relative contribution of the individual components. For the field of plant hydraulics to progress, what is needed is to obtain empirical evidence that supports our foundational principles, including the quantification of traits that are plastic, and then to establish linkages between that plasticity and the functional significance.

Herbaceous plants have recently been shown to have much higher cavitation resistance than one might expect for species without woody stems (Lens et al., 2016), and this strongly implicates pit membrane thickness as a major component maintaining xylem function during drought. In Thonglim et al. (2021), the authors provide a nuanced approach for disentangling the various anatomical traits that allow increased resistance to embolism spread, taking advantage of variability within a model system, various wild-type accessions, and a mutant. While numerous xylem traits were evaluated between the wild-type accessions and mutant, pit membrane thickness stands out as the best predictor of $P_{50}$, and therefore the vulnerability of the vascular system. At first glance, this finding is not entirely surprising given that this relationship is documented in other species, including angiosperms, gymnosperms and seedless vascular plants. The novelty of the study by Thonglim et al. (2021) is that pit membrane thickness can vary considerably within a species, but also that that variability has a direct connection to critical functional traits. The range in reported $P_{50}$ values for the Arabidopsis thaliana accessions is impressive, but so is the shape of the resulting vulnerability curves, i.e. changing pit membrane thickness affects not only $P_{50}$ but also how early gas bubbles enter the xylem and how quickly a plant might reach complete hydraulic failure. The finding that a double mutation can increase $P_{50}$ and the resistance to embolism spread by >1 MPa is notable, and opens up new opportunities for further increasing drought tolerance in A. thaliana, but also in agricultural species with genetic control over pit membrane thickness. With their results from this model species, understanding the underlying genetic control of pit membrane structure, and with it drought tolerance, becomes possible. While pit membrane thickness is not the only relevant trait for drought
tolerance, it may be a more mutable trait than others. Given the direct connection between pit membrane thickness and resistance to air-seeding, and the role that air-seeding plays in embolism spread, this marks an important point in our broader understanding of plant vascular systems and how they perform under stress. A lingering question, however, is whether there is a cost associated with increasing pit membrane thickness. If pit membrane thickness increases, the benefit of greater resistance to air-seeding may come with the penalty of increased hydraulic resistance, thereby decreasing maximum flow rates across the same interface. As shown in Thonglim et al. (2021), these trade-offs, and the advantages that thicker pit membranes confer, may have already pushed A. thaliana accessions in very different directions as they adapt to regional environmental constraints.

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