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Review and new insights into wood anatomy that help understand and control oak wilt

Revue documentaire et nouvelles perspectives de l'anatomie du bois aidant à mieux comprendre et lutter contre le flétrissement du chêne

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Article abstract

Oak wilt, which develops in the outer sapwood, is the most destructive disease of oaks in the United States. Species of red oaks are more susceptible to this disease than white oak species and are more likely to facilitate the spread of the pathogen *Bretziella fagacearum*. To prevent its establishment in new areas, phytosanitary certificates are mandatory for commercial trade, as is the inspection of logs to confirm identification. A literature survey and the results of our assays with seven oak species confirm that it is easy to identify wood of the red and white oak groups using anatomical features. Specifically, earlywood vessels are generally open in red oaks, while they appear occluded with tyloses in white oaks. Such plugs are the consequence of air embolism (cavitation) and not the cause of the wilting process. Although these wide vessels are efficient for water transport, they are vulnerable to cavitation that appears to favour the growth of the oak wilt pathogen. Compartmentalization of infected wood succeeds in restraining cavitation and pathogen spread, allowing some trees to recover from oak wilt.

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Keywords: microscopy, defence reactions, sap transport, embolism, cavitation, tyloses.

Revue documentaire et nouvelles perspectives de l'anatomie du bois aidant à mieux comprendre et lutter contre le flétrissement du chêne

Le flétrissement du chêne est la maladie qui affecte le plus les chênes aux États-Unis. Les espèces de chênes rouges sont plus sensibles que le groupe des chênes blancs à cette maladie, et ainsi plus susceptibles de répandre l'agent pathogène *Bretziella fagacearum*. Pour empêcher l'établissement de la maladie dans de nouveaux territoires, l'exigence de certificats phytosanitaires est nécessaire lors d'échanges commerciaux de même que durant l'inspection des billes pour confirmer l'identification du bois. Une revue de la documentation scientifique et nos essais avec sept espèces de chêne montrent qu'il est aisé de différencier le bois du groupe des chênes rouges de celui des blancs avec quelques critères anatomiques. Un de ces critères est l'apparente absence d'occlusion dans les vaisseaux du bois initial chez les chênes rouges alors qu'ils sont obstrués par des thylles chez les chênes blancs. Ces occlusions sont une conséquence de l'embolie par l'air (cavitation), et non la cause du flétrissement. Ces gros vaisseaux sont efficaces pour transporter la sève, mais vulnérables à la cavitation, cette dernière semblant stimuler la croissance de l'agent pathogène. Le compartimentage du bois infecté réussit à freiner la cavitation et la colonisation par l'agent pathogène, permettant à certains arbres de survivre à cette maladie.

Mots-clés : microscopie, réactions de défense, transport de la sève, embolie, cavitation, thylles.

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INTRODUCTION

The genus *Quercus* (oak) comprises around 500 species worldwide (Manos *et al.* 1999), mainly distributed in the Northern hemisphere (Nixon 1993). Oaks represent the most important broadleaf group, commercially and ecologically, in North America (Sander and Rosen 1985). Ninety-one species are reported in the continental United States (US) and Canada and 172 species in Mexico (Cavender-Bares 2019). Caused by the fungus *Bretziella fagacearum* (Bretz) Z.W. de Beer, Marinc., T.A. Duong, & M.J. Wingf., oak wilt develops in the outer sapwood (de Beer *et al.* 2017). Considered by some as the most serious disease of oaks in the US, oak wilt has not yet been reported elsewhere. The Canadian Food Inspection Agency (CFIA) is the Government of Canada's science-based organization whose mandate focuses on food safety, animal health and plant health. It also regulates international market access. The CFIA is the equivalent of the Animal and Plant Health Inspection Service (APHIS) in the US. These organizations are involved in implementing measures to mitigate the spread of oak wilt, with the CFIA taking additional measures to prevent its introduction into Canada. We gave a lecture on how to differentiate red oak from white oak species using wood anatomy to CFIA officers at an unpublished oak wilt workshop in Saint-Hyacinthe (Quebec) on January 28, 2020. Following the numerous questions by attendees, we have realized that a review paper on the characteristics that allow such differentiation and the anatomical features that help understand the pathogenesis, as well as how trees react to limit damage once infected, was needed. The emphasis of this paper is mainly on red and white oaks, the two principal groups affected by this disease in the US. When data are available, the live oak group, also affected by oak wilt, is discussed even though this wood is much less likely to be traded or imported into Canada. This paper is mainly intended for regulators in North America who want to protect oak trees and/or mitigate the spread of oak wilt. It is also for academia, industries and the public interested in learning about this disease, particularly how to differentiate wood from the red and white oak groups, how sap is transported, how wilting occurs, and how oaks react to limit the colonization of the pathogen. The last two sections of this paper discuss the ascent of sap and the anatomical factors that restrict oak wilt development. Some readers might find this information too specialized. However, knowing about sap transport in the tallest trees, why white oak wood is preferred to that of red oak for cooperage, and the link between bubble structures and vessel cavitation that seems key for oak wilt development will surely arouse the curiosity of most readers.

OAK WILT: A BRIEF OVERVIEW

Oak wilt is caused by the fungus *Bretziella fagacearum*, previously known as *Ceratocystis fagacearum*. This invasive disease was reported for the first time in Wisconsin in the 1940s (Henry *et al.* 1944), but some surveys suggest it was present there and in Minnesota as early as 1912 (French and Stienstra 1980). Even though oak wilt is only found in the US, its origin remains unclear, with some researchers suggesting this introduced pathogen may have come from Central or South America, or from Mexico (Juzwik *et al.* 2008). The disease is present in 27 states (Poiré and Appleton 2018), mainly in the Northeast (Fig. 1A), but also in the South as far as Texas where it is well established (O'Brien *et al.* 2011). A list of regulated states, including the District of Columbia, is in Appendix 2 of the CFIA's Directive D-99-03 (2020). Although oak wilt has not yet been found in Canada, the pathogen's DNA was detected in 2020 on some unidentified insects collected using traps placed near the US-Canada border in southern Ontario (DiGasparro 2020).

This pathogen attacks all oaks but is particularly devastating among species of the red oak group. Once infected, trees die within a year. Mortality can occur as rapidly as 3 (O'Brien *et al.* 2011) to 6 weeks (Sinclair and Lyon 2005). Species of the white oak group are considered more resistant and may die after one year, but are usually killed over a period of several years. Species such as bur oak (*Q. macrocarpa*) that seem intermediate in susceptibility may be killed as rapidly as the red oaks or as slowly as the white (French and Stienstra 1980). American live oaks also have intermediate susceptibility to oak wilt. They are considered significant hosts in the southern US because they tend to form root-connected clones through which the pathogen can quickly spread (Appel 1995; O'Brien *et al.* 2011; Sinclair and Lyon 2005). Infected live oaks occur mainly in Texas where the most common species is *Q. virginiana* Mill., also called Texas or southern live oak and at times Live Oak (Goldman 2017).

In the *Quercus* clade, species in the red and the white oak groups belong to the sections *Lobatae* and *Quercus*, respectively, while the seven species of live oak, also called evergreens, are assigned to the section *Virentes* (Cavender-Bares 2019). The classification of live oaks has always been a difficult task. At first, they were associated with the red oak group based on the thick-walled and rounded vessels in latewood (Williams 1939). Later, they were linked to the white oak group, mainly according to leaf and acorn characters (Stein *et al.* 2003), before receiving their own name *Virentes*. Cavender-Bares (2019) mentions a fourth group, the golden cup oaks (section *Protobalanus*), which has a limited geographic range, essentially restricted to the Mediterranean climate of the western US, mainly California.

The oak wilt pathogen is disseminated from tree to tree a short distance apart via two routes: (1) underground through root grafts, and (2) above ground through spores vectored by insects. In live oaks, the pathogen can invade multiple stems that come from their clonal root systems (Sinclair and Lyon 2005). It is frequent to see circular groups of trees dying of oak wilt. Those are called infection centres and occur when trees first become infected (primary infection). Contaminated insects carry the pathogen to a single tree in a new area or it is transported over long distances via infected logs or firewood. It then spreads to other trees mainly through root grafts.

Wilting symptoms in red oak species usually appear rapidly after infection. Leaves at the top of the tree begin to turn dull green, tan or bronze in colour, along the edges of the upper part of the blade (French and Stienstra 1980; Sinclair and Lyon 2005). Infected white oak species display scattered symptoms in the crown resembling fall discolouration, in which leaves turn brown from the apex to the base of the leaf (Tainter and Baker 1996). In live oaks, leaves of infected trees usually show a yellow chlorotic pattern in or along the veins that eventually may turn brown (O'Brien *et al.* 2011; Tainter and Baker 1996). Brown to black streaks appear in the outer sapwood concomitantly with the development of foliar symptoms in many oak species, but not in live oaks where synchronicity between external and internal symptoms does not always happen (Sinclair and Lyon 2005).

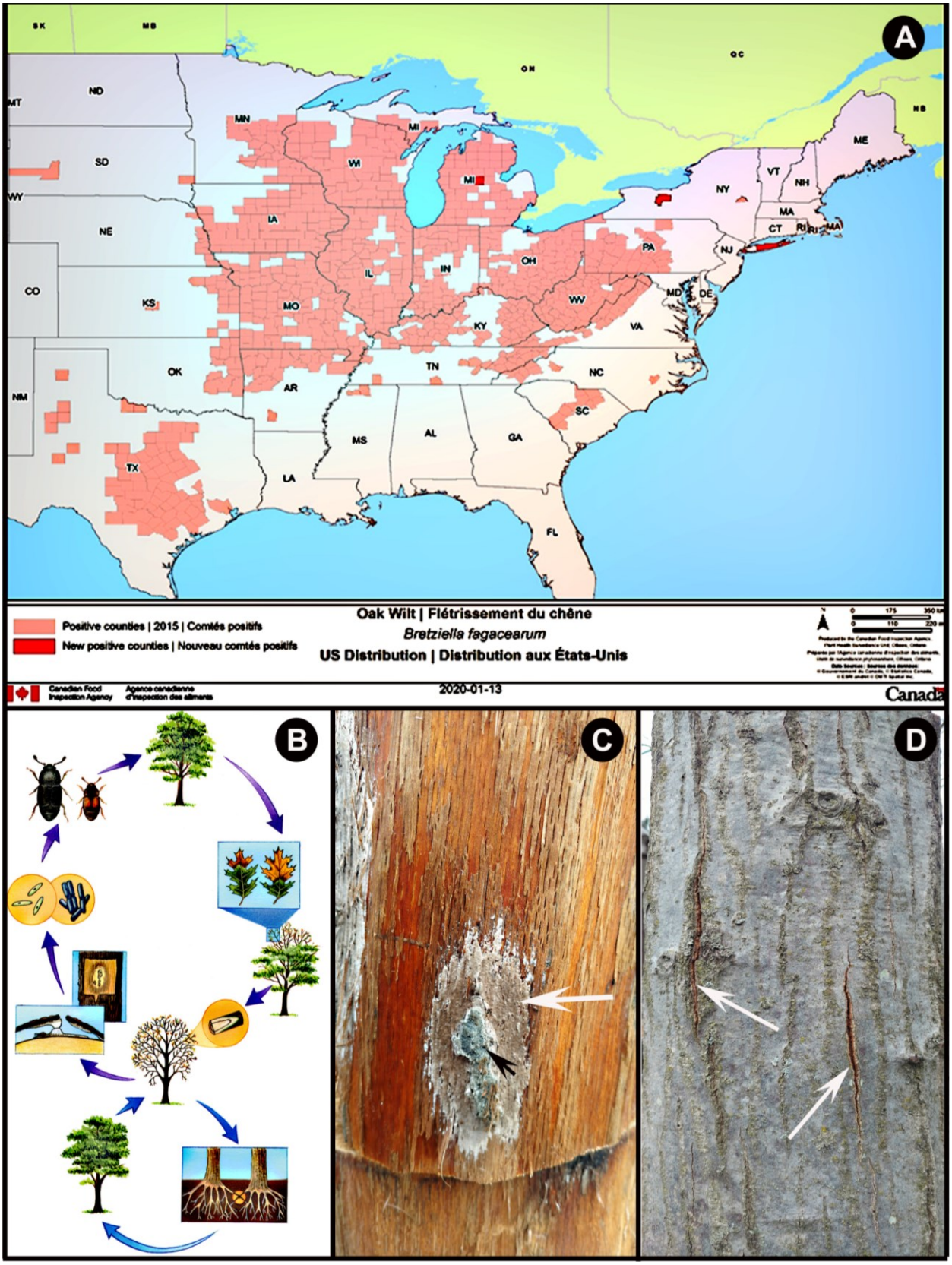

Figure 1. (A): Geographic range of oak wilt in the United States. **(B):** Cycle of oak wilt (O'Brien *et al.* 2011). The disease can also propagate over long distances by moving infected wood. **(C):** A fungal mat (white arrow) within which a pressure pad has developed (black arrow) (Phillip Kurzeja, Michigan Department of Natural Resources). **(D):** The pressure pad (shown in **C**) forces the bark to split open (arrows) (Phillip Kurzeja, Michigan Department of Natural Resources).

Figure 1B shows the oak wilt disease cycle where spores carried by insects form on a fungal cushion (Fig. 1C), also called a mat, beneath the bark at the cambial level. Around the centre of these mats, a specialized non-producing spore structure, called a pressure pad (Fig. 1C), forms and generates sufficient force to crack the bark open (Fig. 1D). Fungal mats emit a fruity odour through bark fissures that attracts many insects (French and Stienstra 1980; O'Brien *et al.* 2011). Nitidulid beetles, also called sap beetles, are the main species attracted to these mats. These primary vectors of oak wilt carry the pathogen's spores from diseased trees to wounds on healthy trees (O'Brien *et al.* 2011). Bark beetles can also disseminate the pathogen when they emerge, becoming contaminated before feeding on the crown of healthy trees (O'Brien *et al.* 2011). While nitidulid beetles are present everywhere in the geographic range of oak wilt, bark beetles are considered minor vectors, particularly in the Upper Midwest and in Texas (Juzwik *et al.* 2011). Conidia form in abundance on the fungal mats while ascospores form occasionally when two compatible strains are present in the same tree. Although fungal mats are reported for some white oak species, such as *Q. macrocarpa* Michx. (Nair and Kuntz 1963) and white oak – *Q. alba* L. (Cones 1967), they almost exclusively form on red oaks, which is why these species are more strictly regulated than white oak species. Fungal mats have never been found on live oaks (O'Brien *et al.* 2011). Although isolates of *B. fagacearum* may vary in virulence, the frequency of mat formation does not seem related to this pathogen attribute (Sinclair and Lyon 2005).

THE RESOURCE AT STAKE

In 2017, the growing stock of oaks in the US was estimated at 138,184 million ft^3 (= 3,913 million m^3), 96.2% of this volume being located in the East (Oswalt *et al.* 2019). Data about red and white oaks in the West is not provided, only that oaks represent 5,175 million ft^3 (= 147 million m^3) there. In the East, red oak species account for 55.7% of the stock. According to Stein *et al.* (2003), there is about the same number of red and white oak species across the US, for instance 25 of each in the East. These 50 species dominate 68% of the eastern hardwood forests and overall, oaks comprise 38% of the total hardwood volume in the US (Sander and Rosen 1985).

In Canada, there are 11 native oak species, distributed mostly in the eastern part of the country (Farrar 1995). Only two white oak species are present in the West: (1) Garry oak (*Q. garryana* Douglas ex Hook.) in British Columbia and (2) bur oak (*Q. macrocarpa*) ranging from a small part of southern Saskatchewan to New Brunswick (Farrar 1995). Oak wilt is a special concern in eastern Canada where all red oak species occur, with red oak (*Q. rubra* L.) being the most common species. Apart from *Q. rubra*, the other red oak species (e.g., black oak [*Q. velutina* Lam.] and pin oak [*Q. palustri*s Münchh.]) are only present in southern Ontario. In this province, *Q. rubra* is the most important species, with an estimated growing stock of 47 million m^3 (Ontario Government 2016). In Quebec, only *Q. rubra* occurs as a red oak species, representing around 24 million m^3, which was about 99% of the volume of all oak species in 2020 (Carl Bergeron [MFFP-DIF], unpublished results). *Quercus rubra* stock in this province is especially at risk as all the companies that import red oak logs from the US are located there. In 2019, this represented a total import value of Can $34,683,969 (= ~ US $27,053,000) (Government of Canada 2019). In the Maritimes, oak resources are less abundant, being present only in New Brunswick, with a total volume of 434,000 m^3 (New Brunswick Energy Resources Development, personal communication, July 28, 2020). Apart from a few isolated *Q. macrocarpa* trees, stock is almost exclusively *Q. rubra*.

In urban areas where oak species are abundant, the economic impact of this wilt is also important. In Austin, Texas, it would cost around US $150 per 1 inch (2.5 cm) diameter to replace trees infected or killed by oak wilt (Martin *et al.* 1989). In Minnesota, just the removal of trees affected by oak wilt in the mostly urban Anoka County is estimated at US $18 to $60 million (Haight *et al.* 2011). In Canada, a recent study indicates that if oak wilt became established, the disease could cost cities Can $266 to $420 million (= ~ US $207 to $328 million), not including the replacement of trees (Pedlar *et al.* 2020).

The abundance of oak trees across its geographic range nurtures a large proportion of animal biodiversity. In the eastern US, oaks are dominant in 68% of the forests (Stein *et al.* 2003) and are considered the best food, mainly their acorns, for many mammals and birds, particularly during winter when usual food sources become scarce (McShea *et al.* 2007). Thus, the impact of oak wilt appears economically and environmentally significant everywhere the disease occurs.

CONTROL OF THE DISEASE

As with most tree diseases, it is extremely difficult and costly to eradicate oak wilt once established, especially in forests and other natural areas. There is no effective cure once a tree is infected. The fungicide propiconazole achieves some success as a preventive treatment for red oak species, but its injection must be repeated every two years to provide sufficient protection (Blaedow *et al.* 2010; Osterbauer and French 1992). When the disease is circumscribed in forests and rural settings, or appears in urban areas, severing roots to prevent the transmission of the pathogen through either root grafts or common clonal root systems appears to be a successful approach (Juzwik *et al.* 2010). However, often when the disease is detected, it has already affected many trees so limiting its spread is almost impossible.

The best ways to combat a disease is to prevent its introduction and educate about its danger. As *B. fagacearum* is disseminated over long distances via wood that can produce fungal mats (e.g., firewood or logs), it is important to inspect any red oak species to detect the pathogen whenever possible. The best approach to prevent the entry of oak wilt into a region is to avoid importing oak wood products from infected areas. Permits or certificates to export logs free of oak wilt should always be produced during trading. Additionally, it is crucial for regulators to be able to differentiate wood from red and white oaks. White oak wood usually commands higher prices than red, thus the selling of red oak logs as white may be tempting. As for several diseases and undesirable insects, moving firewood from infected areas poses a high risk to spread oak wilt, especially if this wood comes from infected red oak trees.

Differentiating the oak groups – Anatomy and morphology

The main morphological differences and macroscopic wood anatomical features of the three oak groups are presented in Table 1. Most studies differentiate species in the red group from those in the white group, so the focus of this section is mainly on these two clades. Live oaks are included because

they are susceptible to oak wilt. A few of these features are morphological (e.g., leaves) and others may concern both the morphology and the anatomy (e.g., the inner bark). With wood, some of the best anatomical criteria can be seen with the naked eye or with a hand lens (macroscopic view), while others require examination at higher magnification in microscopy.

On March 28, 2019, we received from the CFIA many trunk slices (= cookies), ~ 5 cm in thickness, of seven oak species (Table 2) to help verify the features that enable the distinction between red and white oak species. The CFIA obtained these cookies from the Vexco sawmill (Saint-Ferdinand, Quebec) from a single trunk of each species. The suppliers to the sawmill are located in northeastern US, mainly in the State of New York. The trees were felled in the US during the dormant season (late fall to winter). Four days after receiving the slices, they were cut into quarters and two of each species were oven-dried for a period of 37 days: 40 °C for 10 days, 42 °C for 12 days and 52 °C for 15 days. We have also examined a piece of oak wood seized and sent to us by Environment and Climate Change Canada that we have identified as most likely live oak (*Q. virginiana*) using anatomical features and wood density. Most of our observations come from samples polished with an orbital sander (fine to ultra-fine 220-1000 grits). The resulting dust was removed with a Shop-Vac® and photos were taken with a smart phone or with a flatbed scanner (HP Scanjet 4890; 2400 to 3600 dpi scan resolution). Some samples were sectioned with a sliding microtome (thickness ~ 15-30 µm) and stained with phloroglucinol-HCl. The procedure consists of immersing the sections in water saturated with phloroglucinol (~ 3 g 100 mL^{-1}) for 1 to 2 minutes, then removing them from the solution and adding a drop of HCl 20% (Jensen 1962). Phloroglucinol stains lignin red, thus quenching its autofluorescence and facilitating the detection of suberin under ultraviolet (UV) light (Biggs 1984). Suberin autofluorescence is highlighted under UV with a Polyvar light microscope (Reichert-Jung, Vienna, Austria) or with a SZX16 stereomicroscope (Olympus Corporation, Tokyo, Japan) equipped with epifluorescence. The sections are also examined with a confocal Olympus inverted IX83 microscope (Olympus Corporation, Tokyo, Japan) mainly with the laser line 405 nm to detect suberin. Additional procedure details are provided in the legends of figures in this paper or in Rioux *et al.* (2018).

Leaves

Examining leaves is one of the best ways to discriminate red oak species from white oak species. Farrar (1995) mentions this as the first criterion for differentiating these species. Red oaks usually have leaves with acute lobes, but their most exclusive feature is the presence of bristle-tipped teeth (Table 1 and Fig. 2A). White oaks lack such bristle-tipped leaves and their lobes are usually rounded (Table 1), as shown in a schematic view (Fig. 2B) of the true white oak (*Q. alba*). The leaves of some white oak species (e.g., chinquapin oak [*Q. muehlenbergii* Engelm.]) are toothed rather than lobed. Their deciduous nature and absence of bristle tips (Table 1 and Fig. 2C) indicate that the species belongs to the white oak group. This species is rare in North America and thus of little economic value (Harlow *et al.* 1979). In Canada, it is only found in southern Ontario. In addition to the description in Table 1, persistent leaves of live oak species have a leathery appearance, a shiny green colour, and are accompanied by pubescence on their lower (abaxial) surface, as reported for the most common species *Q. virginiana* (Goldman 2017). These characteristics are usually indicative of drought-resistant plants.

Table 1. Main morphological and macroscopic anatomical features allowing differentiation of the oak groups in North America[1]

Features	Red oaks	White oaks	Live oaks
Leaves	Deciduous; lobes with pointed apex (occasionally dented or sinuate). Always bristle-tipped.	Deciduous; lobes usually rounded (occasionally margin toothed). Absence of bristle tips.	Persistent[2]; oval with pointed or rounded tips; margin entire or coarsely toothed. Absence of bristle tips.
Bark	Usually furrowed and grey; inner bark often slightly orange to reddish.	Usually scaly and brown; inner bark various shades of brown.	Share characters of both the red oaks (e.g., grey) or the white oaks (e.g., scaly), depending on the species.
Acorns	Require 2 years to mature.	Mature in one growing season.	Mature in one growing season.
Wood[3]	Ring-porous with large rays; growth rings obvious; heartwood reddish-brown.	Ring-porous with large rays; growth rings obvious; heartwood different shades of brown.	Semi-ring-porous with large rays; growth rings scarcely distinct; heartwood dull brown to grey-brown.

[1] Features synthesized mainly from Farrar (1995), Harlow *et al.* (1979) and Stein *et al.* (2003).

[2] Also called "evergreen", or even at times subevergreen, where new and old leaves may be found on the same branches over a brief period of time (Cavender-Bares 2019).

[3] See Table 3 for other details.

Table 2. Samples examined in this study[1]

Oaks	English common name[2]	French common name	Slice diameter (cm)
Red oaks			
Quercus rubra L.	Red oak - Northern red oak	Chêne rouge	34
Quercus palustris Münchh.	Pin oak - Swamp oak	Chêne des marais	36
Quercus velutina Lam.	Black oak	Chêne noir	33
White oaks			
Quercus alba L.	White oak - Stave oak	Chêne blanc	33
Quercus bicolor Willd.	Swamp white oak	Chêne bicolore	20
Quercus macrocarpa Michx.	Bur oak - Blue oak - Mossycup oak	Chêne à gros fruits	44
Quercus montana Willd.[3]	Chestnut oak	Chêne châtaignier	36

[1] Name details from Farrar (1995).
[2] First name is the preferred common name.
[3] Also known as *Quercus prinus* in the literature.

Bark

The inner bark (phloem) of many red oak species is reported as being orange to various shades of red/pink (Stein *et al.* 2003). For instance, the inner bark of pin oak (*Q. palustris*) is described as pink and was obvious in our samples (Fig. 2D). For all white oak species, Stein *et al.* (2003) mention nothing particular about the inner bark, except for Texas live oak (*Q. fusiformis* Small) displaying an orange colour. *Quercus fusiformis* was later classified within the live oak group (Cavender-Bares 2019). In our samples, the inner bark of white oak species tends to display different shades of dull brown, as shown with *Q. alba* (Fig. 2E), a colour quite similar to that of its heartwood (Table 1).

Interestingly, Stein *et al.* (2003) only mention one oak species, chestnut oak (*Q. montana* Willd.), that sometimes presents an outer dark reddish-brown bark when observed externally in mature trees. In our samples, this reddish colour is not obvious when the surface of the bark is examined. However, in transverse sections the rhytidome (= the outer bark, composed of alternating pale cork (periderm) and dark dead phloem layers) takes a weak reddish colour (Fig. 2F). This observation suggests that this red colour might be at times evident on standing trees or on logs when some of the external rhytidome is removed to expose fresh tissues. In Harlow *et al.* (1979), an external view of the bark of *Q. montana* is described as grey to nearly black, but the most closely related species, swamp chestnut oak (*Q. michauxii* Nutt.) is reported to show outer bark areas tinged with red. Otherwise, as described by Farrar (1995) and Stein *et al.* (2003), the external bark appears mostly furrowed and dark grey for red oaks and scaly and brown for white oaks (Table 1).

Wood

The wood of red and white oak species has a typical ring-porous structure together with large rays conspicuous to the naked eye, as observed in transverse sections (Table 1, Figs. 3A and 3B). The obvious contrast between the large earlywood vessels and small latewood cells makes it easy to distinguish growth rings. While all earlywood vessels are seemingly open in red oak species (Fig. 3A), they appear occluded in white oaks except in the outermost growth ring (Fig. 3B), a key anatomical feature to distinguish both groups (Table 3). One exception to this rule is *Q. montana*, where most earlywood vessels seemed open (Fig. 3C) as in red oak species. Live oaks also possess large rays, but with a semi-ring to a diffuse-porous structure. Diffuse-porous woods have vessels quite similar in size and evenly distributed in growth rings, oftentimes making it difficult to discern growth rings (Table 1 and Fig. 3D). As live oaks are usually evergreen and found where conditions are xeric, they tend to lose the ring-porous structure, as reported for other genera. For instance, *Celtis* (hackberries) species found in temperate regions of the US usually have deciduous leaves and ring-porous structure. On the outer hand, the live (= evergreen) spiny hackberry (*C. pallida* Torrey), that occurs in the South and in Mexico, displays a diffuse-porous pattern (E.A. Wheeler *et al*. 1989). Likewise, only one of the elms described by E.A. Wheeler *et al.* (1989), cedar elm (*Ulmus crassifolia* Nutt.), has earlywood vessels smaller than other elms studied. Apparently, *U. crassifolia* does not always display the typical ring-porous structure of the *Ulmus* spp., a species native to south-central America, and only found in Mississippi, southern Arkansas and Texas in the US (Harlow *et al.* 1979). Generally, ring-porous species are rather rare in tropical regions and more abundant in northern temperate zones. In North America, excluding Mexico, their occurrence is 25%, whereas in the Neotropics their incidence is 1.2% (E.A. Wheeler *et al.* 2007). Furthermore, E.A. Wheeler *et al.* (2007) point out that in the warmer and drier regions of the world (e.g., South America, South Africa, New Zealand), mean diameters of the widest vessels rarely exceed 200 µm. They are commonly 300 µm for several *Quercus* spp. of the temperate zone, and sometimes even more (e.g., 400 µm for *Q. macrocarpa*; Woodcock 1989).

The heartwood of red oaks is reported as having pinkish to light reddish-brown coloration (Panshin and de Zeeuw 1980), as shown in *Q. velutina* (Fig. 3A). In white oaks, the heartwood takes various shades of brown (Table 1), from rich light to dark (Panshin and de Zeeuw 1980), as for *Q. alba* (Fig. 3B) and *Q. montana* (Fig. 3C).

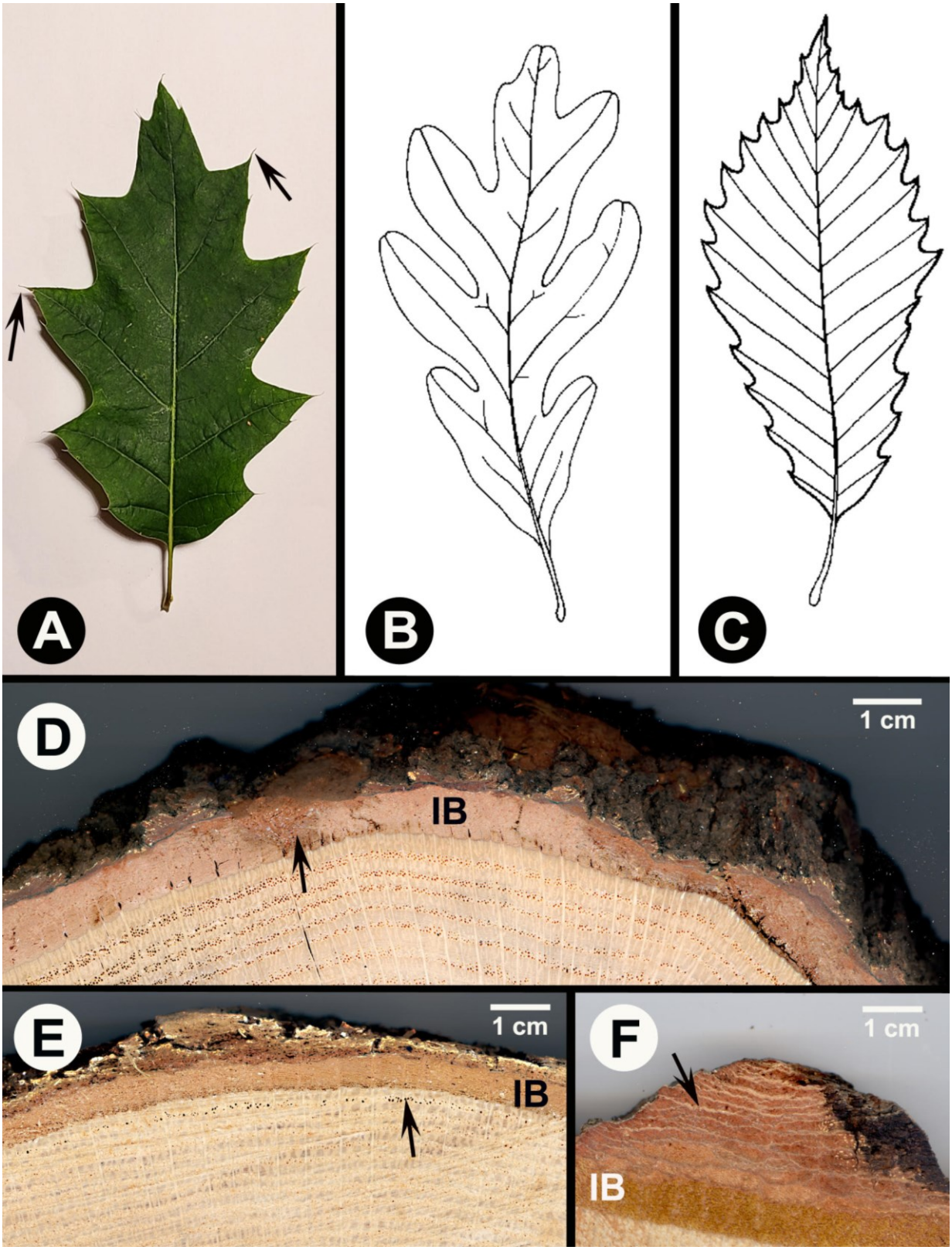

Figure 2. (A): A leaf, typical of the red oak group (here *Q. rubra*), with bristles at the tip of the lobes (arrows). **(B-C):** Leaves of white oak species lack bristle tips (Farrar 2017). **(B):** White oak species (here *Q. alba*) has usually rounded lobes. **(C):** *Q. muehlenbergii* shows a blade that is rather toothed. **(D):** The inner bark (IB) of *Q. palustris* displays a pale pink colour. Where the surface is not refreshed (arrow), the colour is rather a dull brown. **(E):** The IB of *Q. alba* takes different shades of brown. Arrow = open vessels in the last growth ring. **(F):** The IB of *Q. montana* appears yellow-brown. The outer bark presents a reddish colour intermingles with pale suberized layers (arrow).

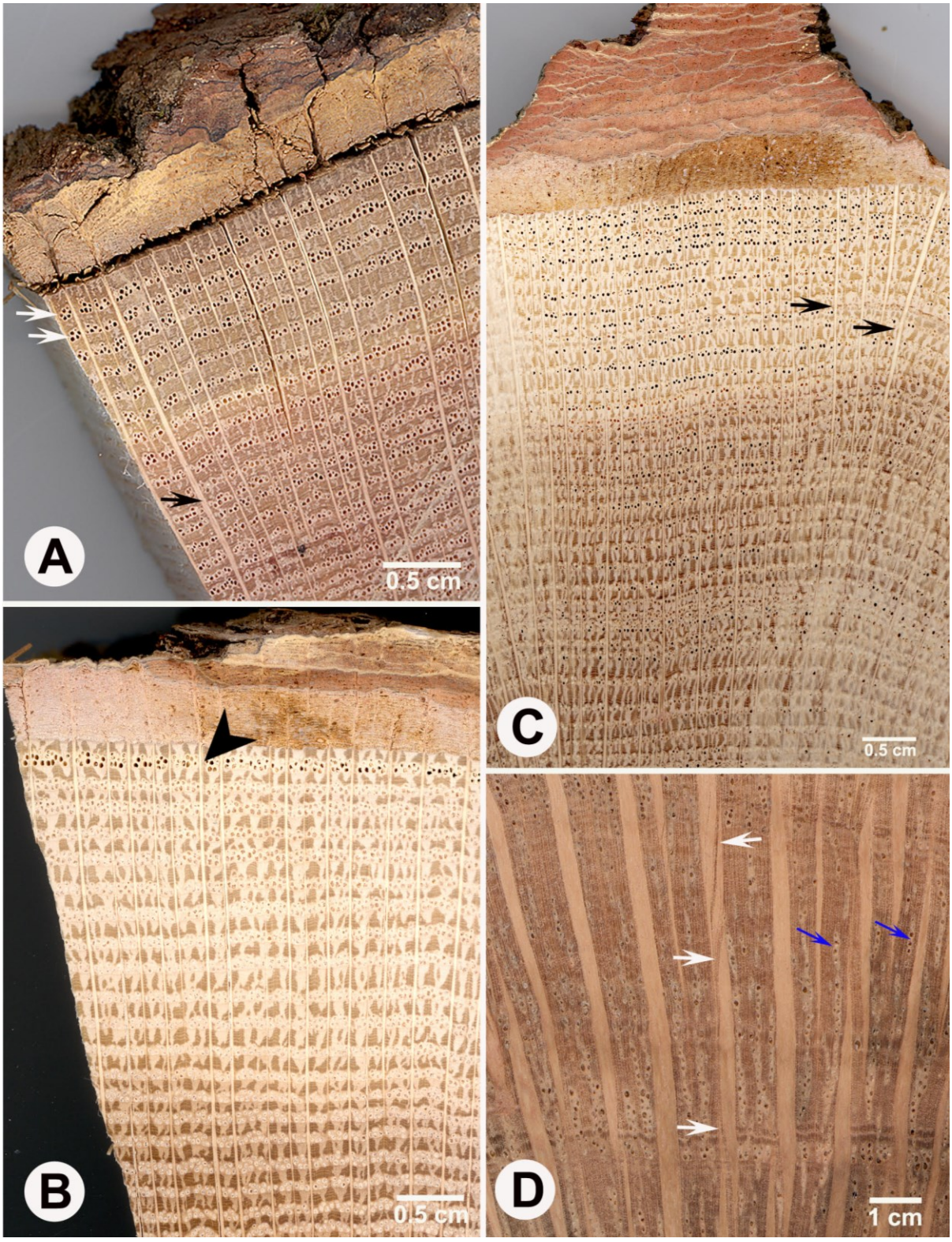

Figure 3. (A-C): Ring-porous arrangement of the wood. **(A):** In red oak species (here *Q. velutina*), most of the earlywood vessels seem open, including those of the two outermost growth rings (white arrows). Black arrow points to a wide ray and the reddish heartwood. **(B):** In white oak species (here *Q. alba*), all earlywood vessels are occluded except those of the most recent growth ring (arrow). The heartwood at the bottom of the figure displays a brown colour. **(C):** In *Q. montana*, most of the vessels appear open as in red oak species, apparently more in the sapwood than in the brown heartwood. Arrows = wide rays. **(D):** The wood of this live oak (most likely *Q. virginiana*) is semi-ring-porous. Some of the wide rays are aggregated (white arrows). Some of the rounded latewood vessels in radial files (blue arrows) appear as wide as those at the beginning of growth rings (same level as the white arrow at the bottom).

Rays

In all oak species, wood rays are of two types: (1) wide and high rays visible to the unaided eye, and (2) narrow rays (usually uniseriate) that are much more numerous, but indistinct without magnification (Panshin and de Zeeuw 1980). The broad rays, 12 to 30 + seriate and around 400 µm wide through their central portion (Panshin and de Zeeuw 1980), also reported as being usually more than 20 + seriate (Hoadley 1990), are the most distinctive anatomical feature of oaks among all temperate woods. Panshin and de Zeeuw (1980) even call these broad rays the "oak-type". When cut tangentially, it is possible to differentiate red oak species from white by measuring the height of the taller rays (Table 3). In white oaks, rays taller than 3.8 cm are usually conspicuous while they rarely reach that height in red oaks. These figures vary in the literature. For instance, Hoadley (1990) mentions that rays taller than 1 inch (2.5 cm) are rarely found in red oaks while they are generally more than 1.25 inches (3.2 cm) in white oaks. We can confirm the validity of this feature in all oaks studied (Figs. 4A and 4B), except in *Q. montana* where the taller rays rarely exceed 2.5 cm. From two cookies, we have measured 31 of the taller rays which averaged 1.14 cm in height, and only two exceeded 2.5 cm (2.8 and 3.1 cm). A sample of *Q. montana* from a collection managed by the Canadian Wood Fibre Centre reveals that most of its rays are smaller than 2.5 cm. However, a tangential view of *Q. prinus* (= *Q. montana*) can be seen in The Wood Database website (n.d.-a) with some rays appearing to go beyond 2.5 cm.

Hoadley (1990) estimates that the degree of confidence using ray height to discriminate red from white oaks is around 90-95%. He underscores the fact that red oaks with rays taller than 1 inch (2.5 cm) are more common than white oaks with rays shorter than 1.25 inches (3.2 cm). Our samples of *Q. montana* seem to be an exception to this rule. We have also seen rays in *Q. velutina* that seem taller than 2.5 cm (Fig. 4C), but a closer look at higher magnification shows a partition zone within the rays (Fig. 4D) that is composed of fibres (Fig. 4E). Such aggregate rays could cause misinterpretation of this feature at low magnification. According to Panshin and de Zeeuw (1980) and as shown in Figure 3D, aggregate rays appear common in *Quercus* spp., particularly in *Q. virginiana*. Aggregate rays in *Quercus* spp. have been known about for a long time, for example, in fossil wood described by Eames (1910). Thus, caution is required regarding this character as aggregate rays in *Quercus* spp. can appear at times as a single tall ray in tangential view. In Eames (1910), as generally discussed for *Quercus* spp., as well as specifically reported for *Q. calliprinos* Webb. (Lev-Yadun 1994), the parenchyma cells of aggregate rays are generally crossed by axial fibres like those described in Figure 4E.

Finally, a curiosity that may puzzle some observers: large rays usually appear darker in tangential views (Figs. 4A and 4B) than in transverse (Fig. 3) and radial sections. We suspect that in transverse and radial sections more walls of the procumbent (horizontal) cells are seen than in the tangential plane where the volume of dark cytoplasm would be more prevalent than that occupied by the walls. Another possibility is that different reflections of light may be obtained depending on the angle from the point of examination (E.A. Wheeler, personal communication, February 17, 2021). We can confirm that considerable variations occur in the appearance of rays. Sometimes they appear darker and other times lighter in colour, depending on the angle of the light and/or of the position of the observer, especially in tangential views.

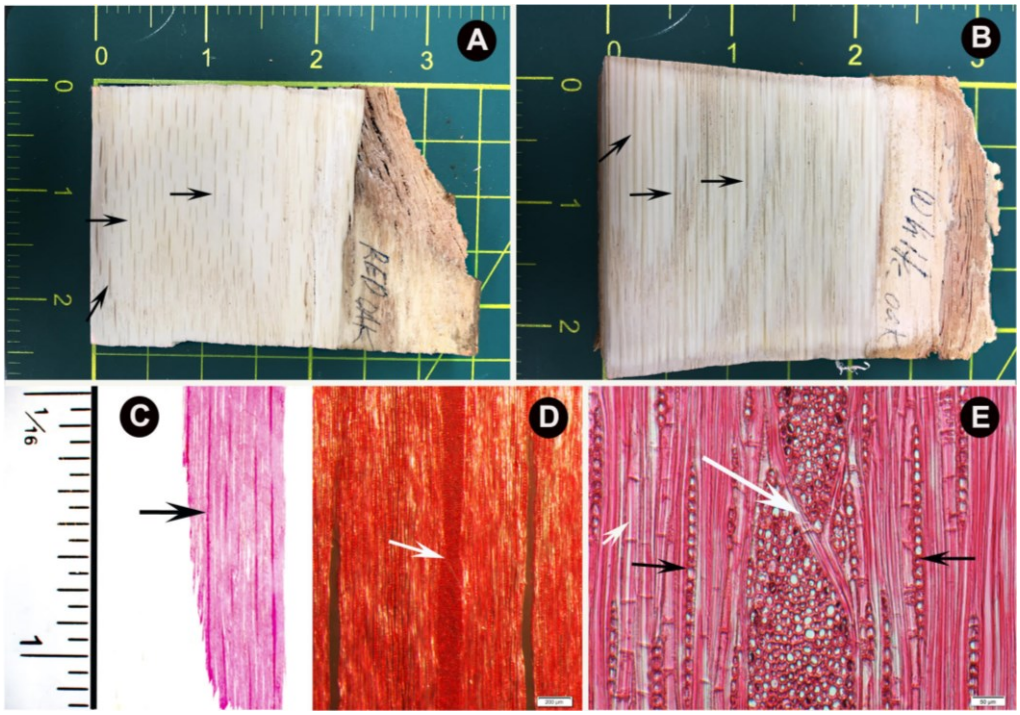

Figure 4. The height of rays (tangential sections). (A): The taller rays (arrows) do not exceed 1 inch in red oaks (here *Q. rubra*). **(B):** Rays (arrows) are generally taller than 1 inch (2.5 cm) in white oaks (here *Q. alba*). **(C-E):** The same ray in *Q. velutina* taller than 1 inch, shown C. at low magnification (arrow), **(D):** that is an aggregate of two rays (arrow), **(E):** separated by fibres (large white arrow). Black arrows = uniseriate rays. Small white arrow = an axial parenchyma cell in a strand of at least six cells.

Table 3. Main macroscopic wood features (visible to the unaided eye and/or with a 10X hand lens) allowing differentiation of the oak groups[1]

Criteria	Red oaks	White oaks	Live oaks[2]
Tangential plane			
Higher rays	$\overline{x}$[3] = 0.6-1.3 cm; rarely > 3.8 cm	$\overline{x}$ = 1.3-3.2 cm; frequently > 3.8 cm	Only described of varying length
Transverse plane			
Earlywood vessels of the heartwood	Usually open and distinct to the naked eye.	Usually occluded with tyloses and distinct to the naked eye.	Barely visible to the naked eye.
Latewood vessels[4]	Visible with a hand lens. Thick-walled and rounded.	Hardly visible with a hand lens. Thin-walled and angular (only visible at high magnification).	Likely visible with a hand lens. Thick-walled and rounded.[5]
Transition from earlywood to latewood	Gradual to more or less abrupt.	Generally abrupt.	Gradual to imperceptible.
Heartwood colour	Pinkish to pale reddish brown.	Rich light brown to dark brown.	Dull brown to grey-brown.

[1] Red and white oaks: adapted from Panshin and de Zeeuw (1980: 568).

[2] Based mainly on *Q. virginiana*, which appears the most common live oak species (Goldman 2017); denser green wood (0.81) than that of red and white oaks (~ 0.6), in fact one of the densest of North American trees (Goldman 2017).

[3] $\overline{x}$ = Average.

[4] A note added by Panshin and de Zeeuw (1980) specifies that this criterion to discriminate red from white oak species is "… by far the most reliable."

[5] Based on Williams (1939), our interpretation of the transverse section shown on page 564 of Panshin and de Zeeuw (1980) and our Figure 3D.

Vessels

It has been known for a long time that it is quite easy to discriminate between red and white oak species by examining their vessels, in particular whether or not occlusions are present in the large earlywood vessels (Table 3). In 1921, the USFS-FPL (US Forest Service-Forest Products Laboratory) published a technical note showing that the earlywood vessels of red oaks appear open while those of white oaks are usually plugged with tyloses. This difference is visible to the naked eye by an observer with minimum training. Tyloses are usually the type of occlusions that occur in the large dysfunctional vessels of ring-porous tree species. The origin and role of vessel occlusions are discussed later in this paper.

The USFS-FPL paper (1921), as mentioned in most other references, specifies that vessels must be examined in the heartwood and that occlusions may also be found in the large vessels of the inner sapwood. When examined, all wide vessels in the sapwood of our white oak cookies appeared as plugged as those of the heartwood, except in the outermost growth ring (Fig. 3B). The only exception was *Q. montana* (Fig. 3C) where most of the broad vessels appeared open, possibly a little more in the sapwood than in the heartwood. As with ray height, *Q. montana* looks irregular among the white oaks studied in respect to the plugging of its large vessels. This observation is stressed elsewhere, including on The Wood Database (n.d.-b) website where it is stated that "One exception to this rule is chestnut oak, which is still considered to be in the white oak group, even though its pores are open like red oaks." The USFS-FPL paper (1921) highlights that the presence or absence of tyloses in the earlywood vessels is not as a reliable feature to differentiate oak groups as is observing the structure of the latewood vessels. Panshin and de Zeeuw (1980) insist that the size and the shape of the latewood vessels are the most reliable anatomical feature to differentiate red oak from white oak species (Table 3). The latewood vessels are smaller in white oak species than in red oak species, actually hardly visible even with a hand lens. To discriminate between groups using this characteristic, we suggest using a small sample of *Q. rubra* and *Q. alba*, with surfaces already cleanly prepared with a sharp knife or an industrial blade, to compare with any unknown wood using a hand lens. After few identifications of this kind, together with other discriminating anatomical features (Table 3), an observer will be able to identify with great confidence which group the oak wood belongs to. At low magnification, the vessels in the latewood of red oak species, visible with a hand lens, are mostly grouped and arranged as a single band in the radial direction, although they usually appear quite small and scattered at the very end of wide growth rings (Fig. 5A). The light-coloured tissue around these latewood vessels is mostly composed of parenchyma

cells and tracheids. Between zones of latewood vessels, darker fibres with thick walls are predominant (Panshin and de Zeeuw 1980). Although not clearly visible at low magnification in white oak species, the latewood vessels and the surrounding parenchyma cells accompanied with some tracheids (Panshin and de Zeeuw 1980) often take radial to oblique shapes similar to a flame or a tornado (Fig. 5B). The earlywood vessels usually form a band 1 to 4 pores wide in red oaks, whereas this zone is no more than three vessels wide in white oaks (Panshin and de Zeeuw 1980). For more certainty during identification (Table 3), it is possible to get thin slices of latewood of the unknown specimen for microscopic examination to check whether the vessels are rounded with thick walls (red oak group) or with thin angular walls (white oak group).

Figure 5C shows the typical structure of a few isolated latewood vessels at the end of a growth ring in a red oak species. At high magnification, all of our white oak species had the expected latewood structure, including *Q. montana* (Figs. 5D and 5E). The flame (tornado) appearance is obvious (Fig. 5D), similar to that at low magnification (Fig. 5B), with thin angular vessel walls (Fig. 5E). We consider that micrographs based on the appearance of these latewood vessels combined with a chemical test (see next section), together with control samples from *Q. rubra* and *Q. alba* for visual comparisons, represent incontestable proof, even in court, for determining whether an unknown sample belongs to the red or the white oak group.

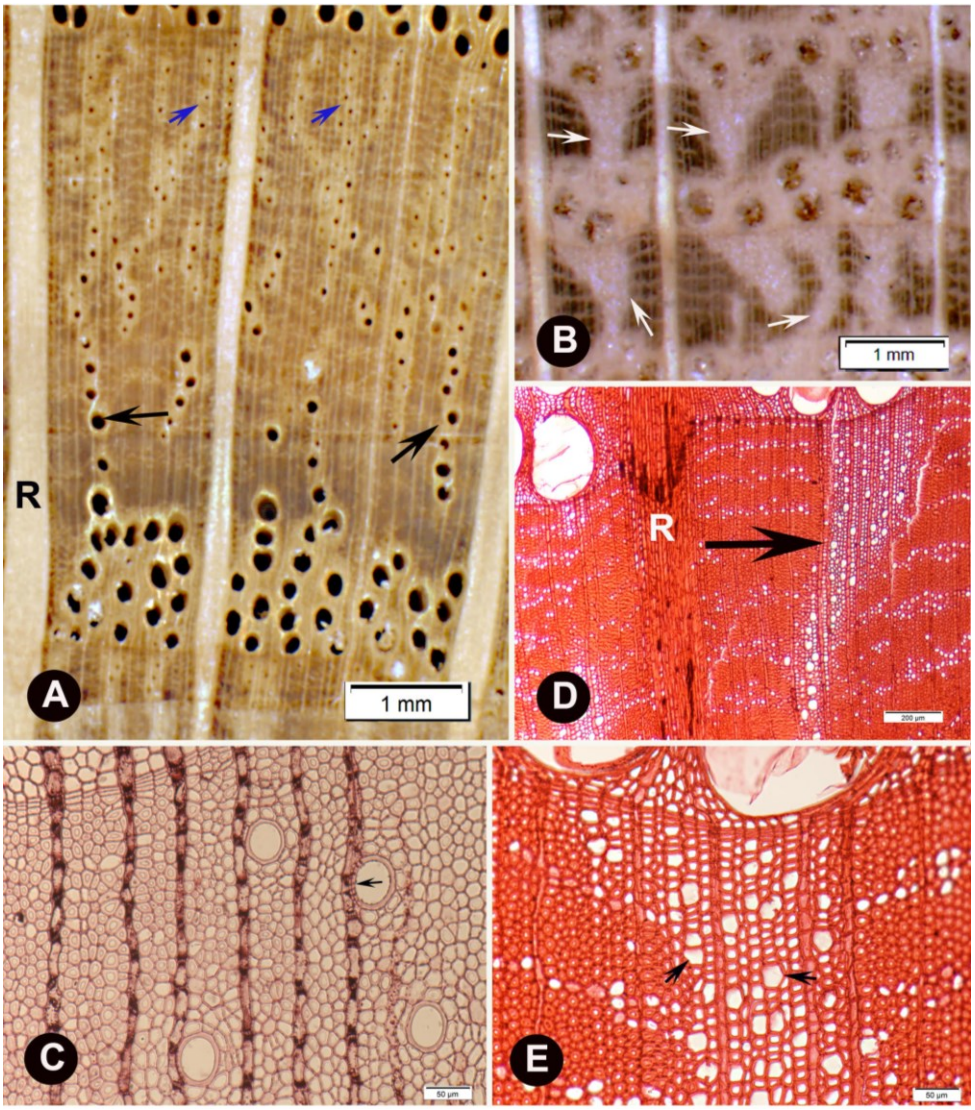

Figure 5. Vessel arrangements in latewood. (A): In red oaks (here *Q. rubra*), the round vessels in radial files (black arrows) next to radial files of at least four large earlywood vessels are visible with a hand lens. Those formed later in the growing season (blue arrows) are only observable with a stereomicroscope. The latewood vessels are surrounded by whitish parenchyma cells. **(B):** In white oaks (here *Q. alba*), the latewood vessels, barely visible with a hand lens, and the surrounding cells (mostly parenchyma) appear as flame-like (tornado-like) groups (white arrows). The earlywood vessels are no more than three vessels per radial file. **(C):** In red oaks (here *Q. velutina*), the latewood vessels are thick-walled and rounded. In one of these vessels, the pits shared with a ray cell can be seen (arrow). **(D-E):** In *Q. montana*, the arrangement of latewood vessels in a flame shape (**D**, arrow) is typical of the white oak group. At high magnification, these vessels appear angular with thin walls (**E**, arrows). At the top of these figures, some occluding material appears visible in the large earlywood vessels.

Differentiating the oak groups – The sodium nitrite test

The sodium nitrite colour test (Miller *et al.* 1985) was developed to differentiate red oak species from white for three main reasons:

(1) The macro- and microscopic methods, although accurate and efficient, need a high level of expertise.
(2) When many logs must be assessed, this is more appropriate than anatomy, as around 100 logs can be evaluated in less than an hour.
(3) Stacks of logs many meters high can be tested as the solution is sprayed, another practical advantage over sampling for anatomical examination.

As with wood anatomy, the main drawback of this test is that cleaning dirty wood or removing weathered grey ends to expose a fresh surface is required.

In Miller *et al.* (1985), a solution of 10% of sodium nitrite ($NaNO_2$) in water was tested on the heartwood of nearly 10,000 logs at sawmills and on 500 specimens from the USFS-FPL collection in Madison, Wisconsin. Every test identified the wood to the correct oak group. Once the solution is applied, the heartwood of both groups turns yellowish or brown with an orange tint. In red oaks, this colouring does not turn darker with time, but after 2 h it fades to a greenish yellow. By 16 h, it appears a light-yellowish colour. Within 5 min, the heartwood of white oaks appears dark greenish or purplish brown to black. This colour fades after 2 h. After 16 h it becomes brownish. Colour fades considerably in both groups with time. After several days, Miller *et al.* (1985) stress out that it is impossible to distinguish the species of either group. We have assessed this reaction with our samples and most of the above is confirmed, except that the heartwood of white oaks turns immediately darker after the application of the solution while the orange-brown tint is particularly obvious with red oak species (Fig. 6A). Within 10 to 15 min, the heartwood in red oaks has an amber-green colour while in white oaks it turns dark grey-black (Fig. 6B). Contrary to the results of Miller *et al.* (1985), these colours do not fade much after several hours (Fig. 6C), or even after days. We have tested *Q. montana* and within 5 min it is easy to determine that it belongs to the white oak group (Fig. 6D), as mentioned for 48 specimens of *Q. prinus* (= *Q. montana*) in Miller *et al.* (1985). From Figure 6, it is clear why this test must be conducted with heartwood, in particular Figures 6C and 6D where the difference in colour reaction is striking between heartwood and latewood. As the colour in our samples holds over time, we suggest, as with the anatomy, that testers bring samples of *Q. rubra* and *Q. alba* upon which the solution has been applied. For more certainty, use a fresh solution on them to compare unknown woods.

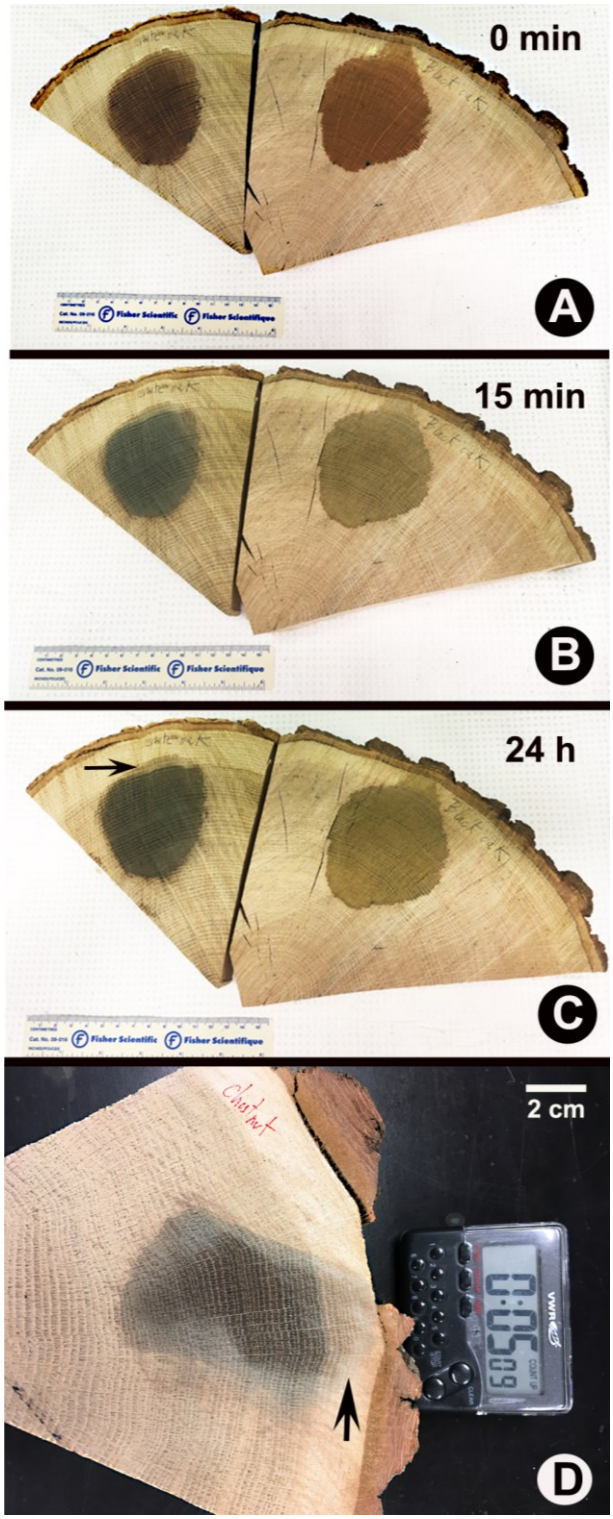

Figure 6. Chemical tests with sodium nitrite. (A-C): *Q. alba* on the left; *Q. velutina* on the right. **(A):** In white oaks, the heartwood rapidly takes a blackish colour that remains even after 24 h. In red oaks, the original brown to orange colour turns rapidly to an amber-green colour **(B)** that still holds after 24 h **(C)**. **(D):** Within 5 minutes, the heartwood of *Q. montana* presents a blackish appearance typical of the white oaks. Arrows = the sapwood is only weakly coloured following this test.

THE ASCENT OF SAP AND THE WILTING PROCESS

There is extensive literature on water relations in plants. In fact, papers are still being regularly published in many scientific journals (e.g., the review by Piao *et al.* 2019), often in relation to the challenges posed by climate change that increases water stress. For instance, a catastrophic case triggered by a record drought was described for plantations composed of Norway spruce trees (*Picea abies* (L.) H. Karst.) in Germany (Popkin 2021). As shown later in this section, the effort has also been made to explain how water moves from the roots to crowns, as this is especially intriguing in trees reaching more than 100 m in height.

Sap is mostly composed of water accompanied by some solutes such as minerals and is transported in the xylem in an upward direction. Sap moves passively through the transpiration stream in the xylem because no obvious metabolic pump, similar to a heart in animals, appears to occur in plants. Water potential helps understand how sap circulates in plants. Water potential is generally expressed by the Greek letter Ψ, where it is equal to pressure potential + osmotic potential (= solute concentration). Pressure potential is positive, whereas osmotic potential has a negative value. For instance, plants in desertic regions have leaves where solutes are highly concentrated (highly negative) and generate a high transpiration pull that is instrumental for extracting water from arid soils (Scholander *et al.* 1965). Water potential is the chemical energy of water when it moves from a region of high to a region of low water potential or, in other words, along gradients of decreasing free energy of water. If the potential of pure water is zero, which occurs in fully turgid cells, water potential at any given point in plants generally takes negative values when transpiration occurs, that is when $\Psi^{soil} > \Psi^{root} > \Psi^{stem} > \Psi^{leaf} > \Psi^{air}$. For instance, when the atmosphere is saturated with water, then Ψ^{air} is equal to, or near 0, and transpiration in plants stops.

In the past, most researchers believed that the only significant input of water came from the soil. Now, it appears that a non-negligible portion also comes from absorption through other parts of the plant, the leaves in particular. In periods of rain, fog or dew, foliar absorption is an important source of water, a supply particularly helpful for drought-stressed plants (Breshears *et al.* 2008). Although a large part of fog drips from the coast redwood (*Sequoia sempervirens* (D. Don) Endl.) canopy to help replenish soil water content (Dawson 1998), Limm *et al.* (2009) show that direct foliar absorption from wetting surfaces may account for as much as 2 to 11% of leaf water content for several plant species of this ecosystem. This foliar intake occurs through the stomata and directly across the cuticle (Limm *et al.* 2009). Coast redwoods are considered the tallest trees of the world, many reaching over 110 m in height. The tallest, called Hyperion, towers at 115.85 m (380.1 ft) (Wikimedia Foundation n.d.). In such tall trees, water must travel through long complicated channels from the roots to the leaves. However, as their canopy is frequently in contact with fog, interception of water appears as important in reducing transpiration and rehydrating leaf tissues, including during long summer droughts when advection fogs may be common (Burgess and Dawson 2004). Thick layers of humus, as well as various epiphytic plants and even salamanders (*Aneides vagrans* Wake & Jackman, 1998), may be present between large branches and the trunk in tall redwoods and adventitious roots have been reported to exploit this soil (Sillett and Van Pelt 2000). This most likely represents an important source of water and nutrients channelled to the crown of these giant trees.

Although not perfect, the cohesion theory (Dixon and Joly 1895), also called the cohesion-tension theory, appears as a generally accepted concept explaining how a column of water can be lifted up under great tension without breaking (Tyree and Zimmermann 2002). It has been shown with a centrifuge that a water column does not break until the tension reaches around 270 bars (27 MegaPascales [MPa]) at 20 °C (Briggs 1950). This was shown using glass capillaries varying from 600 to 800 µm in diameter. The diameter of xylem conduits would be sufficiently small to prevent air embolism, also called cavitation (Scholander *et al.* 1965). Knowing that the integrity of the water column is inversely proportional to the diameter of the conduits (Tyree and Zimmerman 2002), the water-conducting system of trees appears quite resistant to cavitation. This resistance is evident even in xeric plants into which the tension can reach up to 80 bars (Scholander *et al.* 1965). In these plants, larger vessels rarely exceed 150 µm in diameter (Baas *et al.* 1983). This theory proposes that evaporation from leaves pulls the water column in the xylem, generating powerful negative pressure in the conduits. This pressure is generally more negative at the top of the tree than in the roots. Capillary force helps sap move upward, but even an optimal 10 µm conduit would generate a capillary pressure of only 3 m (H.R. Brown 2013; Steudle 1995), much too insignificant to explain how sap is pulled up in tall trees.

In the mature parts of trees, water moves in the young xylem or sapwood while the older xylem, the heartwood, becomes non-functional. However, sap is usually only transported in the most external growth rings of the sapwood. Trouy (2015) emphasizes that the true definition of sapwood should be the part of the wood that contains living cells, rather than being defined in relation to the ascent of sap. Wood anatomy and sap transport vary between species, and even between different parts of the same tree. At times, it enables different wood types to be defined, usually into three main groups: (1) conifers (or softwoods), (2) hardwood diffuse-porous species, and (3) hardwood ring-porous species. In conifers, as much as 90% of the wood is composed of tracheids that ensure sap transport and structural support, with practically no axial parenchyma. For example, Panshin and de Zeeuw (1980) report ~8% ray parenchyma and, when present, ~2% resin canals (constituted of epithelial parenchyma cells). In broadleaf trees, wood is more variable in its composition. In *Q. macrocarpa* for instance, wood contains 26.6% vessels, 40.8% fibres (probably *sensu lato*, including tracheids), 12% axial parenchyma and 20.6% ray parenchyma (Panshin and de Zeeuw 1980). In diffuse-porous species (e.g., maple [*Acer* spp.], birch [*Betula* spp.] and beech [*Fagus* spp.]), there is little variation in vessel diameter within growth rings. In ring-porous species (e.g., oak [*Quercus* spp.], elm [*Ulmus* spp.] and ash [*Fraxinus* spp.]), there are wide vessels in earlywood followed quite abruptly by much smaller latewood vessels. Only some outer growth rings transport sap in hardwoods, and in ring-porous species in particular nearly all the sap is transported in the last growth ring, within which most of the sap goes through the wide earlywood vessels. According to the Hagen-Poiseuille law (see below) and with the injection of a dye solution, it has been calculated that 92% of sap would flow in the last growth ring in stems of 7 to 15-year-old American elm (*Ulmus americana* L.) saplings (Ellmore and Ewers 1986). In particular, this law states that the flow rate is proportional to the cylinder radius to the fourth power. Counting the number of vessels in their samples and measuring their diameter,

Ellmore and Ewers (1986) estimate that in the last growth ring around 96% of the sap would flow within the large earlywood vessels.

Hagen-Poiseuille equation:

$$q_v = \pi r^4 \Delta p / 8 \mu L$$

q_v = Flow rate;
r = Conduit radius;
Δp = Pressure difference between the ends of the conduit;
μ = Viscosity;
L = Length of the conduit.

Another way to comprehend the great difference in water transport using the Hagen-Poiseuille law is to directly compare the larger with the smaller diameter vessels. Early-wood vessels in oaks are often more than 300 μm in diameter while those of the latewood are frequently no more than 50 μm. Thus, all other factors being constant, vessels are around six times wider in earlywood than in latewood and conduct 1296 (= 6^4) times more water. In other words, wide earlywood vessels carry 99.9% of the water. With this calculation in mind, we roughly estimate the number of vessels in oak growth rings of the present study. Thus, we have assessed that at the very least 95% of sap moves in the wider vessels of the last growth ring, both in red or white oak species. In addition to their large diameter, earlywood vessels of ring-porous species also appear to be very long. In fact, vessel length would be positively correlated with their diameter (Tyree and Zimmermann 2002). Zimmermann and Jeje (1981) report that the earlywood vessels in white ash (*Fraxinus americana* L.) and *Q. rubra* could be around 9 to 10 m tall, often as long as the trunk. In diffuse-porous species such as red maple (*Acer rubrum* L.) and American beech (*Fagus grandifolia* Ehrh.), the longest vessels in their study rarely reach 50 cm long, actually most of them being in the 0 to 10 cm length class. The Hagen-Poiseuille equation indicates that the flow of liquid in a conduit would be inversely proportional to the length of the conduit. This is apparently caused by the turbulence greater in wide than in narrow cylinders, thus creating some resistance to the flow of liquids. Considering the general small size of vessels, it appears that such turbulence is unlikely to occur, this value being estimated as negligible in the calculation of the sap flow in vessels (Tyree and Zimmermann 2002). Whatever its importance, hydraulic resistance would appear only notable in the small (distal) parts of the tree (Tyree and Zimmermann 2002), where narrow conduits occur. Overall, vessel/tracheid pit membranes would be responsible for more than 50% of the total xylem hydraulic resistance (Rabaey *et al.* 2008). Thus, sap transport appears easier in the large vessels of ring-porous species, while most of the sap moves from short vessels to short vessels laterally through pits in diffuse-porous species. The diameter combined to the length of vessels gives a better idea of the water conduction efficiency of the vessels. Tyree and Zimmermann (2002) compare a maple tree with vessels 75 μm in average diameter and 10 cm long with an oak tree where the large vessels were 300 μm in diameter and 3 m long. Considering the size of these vessels, the maple must have 256 (= $(300/75)^4$) as many vessels as the oak to carry an equal volume of water. As the ratio of the length is 30 (= 3/0.1), Tyree and Zimmermann (2002) postulate that the maple needed 7680 (30 × 256) as many functional vessels to transport as much water as in the oak. They also stress that the contribution of latewood vessels is insignificant in ring-porous species when the earlywood vessels are wide and functional.

It is well known that cambial activity in trees resumes in the spring in a basipetal fashion, that is from the top to the base of trees. This fact is particularly accepted for conifers and diffuse-porous hardwood species (Tyree and Zimmermann 2002). Wide earlywood vessels would be only functional for one year in ring-porous species. Thus, it is assumed that new vessels are formed basipetally, more rapidly everywhere in the tree than in conifers or diffuse-porous species, at the beginning of the growing season to supply enough water for bud burst and leaf development (Takahashi *et al.* 2015; Utsumi *et al.* 1996). In diffuse-porous species, many of the old vessels remain functional for more than one season and new vessels form after the development of the current-year leaves, at times weeks after full leaf expansion (Takahashi *et al.* 2015). One of the difficulties in such studies is to determine exactly the time when the new vessels are mature/functional, and this at different heights, a huge task when trees are very tall. The maturation of such vessels is often studied in microscopy and while some researchers suggest that lignification of these vessels is the main indicator of their maturation (Takahashi *et al.* 2015), others argue that the lysis of vessel element end walls is the ultimate structural indication that the first-formed vessels are mature and functional (Kitin and Funada 2016). Using perforation occurrences between vessel members, Kitin and Funada (2016) show with a ring-porous species with one row of earlywood vessels, castor aralia (*Kalopanax septemlobus* (Thunb. ex A. Murr.) Koidz.), that these new vessels in the trunk are only mature after bud burst and just before the leaves achieve their full size.

If this were the case in other ring-porous species, the liquid still present in the large vessels of the previous growing season could represent an important reservoir of water. It would be quite helpful if the tree could recuperate some of this water at the start of the growing season to help during the expansion of the new wide vessels. As shown in Figure 3B, the wide vessels of the outermost growth ring are still at least partly open during the dormant season and likely contain a significant volume of water. We postulate that some of this water is somehow retrieved and that this may happen through the formation of plugging structures (tyloses) occurring in the spring in these earlywood vessels (see next section).

While wide and long vessels are efficient for water conduction, they appear more vulnerable to embolism than smaller conduits, particularly to cavitation (Hargrave *et al.* 1994; Tyree and Zimmermann 2002). Tyree and Zimmermann (2002) even stress that the conductivity of the vessels is inversely proportional to their propensity to embolize. In other words, as previously hypothesized, the large vessels in oak would be 7680 times more susceptible to cavitation than those in maple. It is well known that in temperate regions, the wider the conducting elements, the more likely they are to suffer freezing cavitation (Cruiziat *et al.* 2002; Davis *et al.* 1999; Tyree and Zimmermann 2002). Conifers appear quite immune to freezing, diffuse-porous species intermediate in their susceptibility, while ring-porous species are the most affected by freezing (Sperry and Sullivan 1992). In oak species of the temperate regions, the wide earlywood vessels are particularly vulnerable and are easily cavitated during freezing (Davis *et al.* 1999; Sperry and Sullivan 1992), thus remaining functional for only one year. As the gases would be around 1000 times less soluble in ice than in water (Scholander *et al.* 1953), freezing would cause air bubble formation by forcing the dissolved gas out of the solution. Once the ice melts in spring, these gas bubbles already present in wide vessels would expand by merging with each other to fill most of the conduit. This filling would be especially favoured when transpiration resumes in spring, when tension is again applied to the water column (Sperry *et al.* 1988).

Drought also causes air embolism when sap tension becomes too high and breaks the water column in conducting elements. While species that are more vulnerable to freezing also appear to be drought-intolerant, as shown for chaparral shrub species where this seems linked to the diameter of the vessels (Langan *et al.* 1997), this is not always the case. For instance, Sperry and Sullivan (1992) studied ring-porous, diffuse-porous and conifer species and observed no correlation between vulnerability to freezing and water stress treatments. While Gambel oak (*Q. gambelii* Nutt.) is clearly the most affected by freezing among the five species they investigated, its earlywood vessels being particularly vulnerable to cavitation, this species is only the second least impacted by water stress after Rocky Mountain juniper (*Juniperus scopulorum* Sarg.).

When cavitation occurs spontaneously in some vessels under high water stress or freezing, it is thereafter propagated from vessel to vessel through an air-seeding mechanism. This occurs when air is sucked into a functional vessel mainly through the pores of pit membranes. While there is no clear relation between vessel diameter and tolerance to drought between species, it appears that within an individual species the conduit diameter correlates well with vulnerability to water stress. This susceptibility of the wide conduits appears associated with pit membranes more permeable to air-seeding, as shown for instance in conifers (Sperry and Tyree 1990). Although wider vessels in several species seem to possess larger pit areas, rendering such vessels more vulnerable to cavitation (J.K. Wheeler *et al.* 2005), it has been suggested that no link exists between the pit membrane area versus the size of their pores. Rather, this porosity is positively correlated with pit membrane thickness, thinner membranes being usually more porous (Jansen *et al.* 2009). Vulnerability to air embolism in broadleaf trees may be a combination of several factors such as vessel diameter, vessel length, the number of pits per vessel, but especially the thickness/porosity of the intervessel pit membranes (Jansen *et al.* 2009).

Using scanning electron microscopy, the maximum diameter of intervessel pit membrane pores of several tree species is shown by Jansen *et al.* (2009) to vary greatly, from 10 to 225 nm. It has been calculated that, in theory, air-seeding occurs for pore diameters of 100 nm and 50 nm at pressures of -1.44 and -2.88 MPa, respectively (Tyree and Zimmermann 2002). When such pressures are applied, pores may enlarge by stretching and allow air-seeding (Jansen *et al.* 2009). Accordingly, air-seeding figures differs between species. For instance, this happens at thresholds of -2.12 MPa in box elder maple (*Acer negundo* L.) with pores around 148 nm in diameter, whereas pores as wide as 340 nm in silver birch (*Betula pendula* Roth.) are penetrated by air at pressure of only -0.95 MPa (Jansen *et al.* 2009).

A recent study indicates that ring-porous American oaks would be more tolerant to water stress than previously thought (Skelton *et al.* 2021). However, Skelton *et al.* (2021) seem to have neglected assessing the wood anatomy of the oak species they tested. Some of these species are particularly well adapted to xeric conditions, and as shown for other tree species, these oaks do not present a typical ring-porous structure. For example, the evergreen coast live oak (*Q. agrifolia* Née) and interior live oak (*Q. wislizeni* A.DC.), as well as the deciduous blue oak (*Q. douglasii* Hook. & Arn.) and Garry oak (*Q. garryana* Douglas ex Hook.), which were part of their study, are described as being diffuse-porous by Robert *et al.* (2017). It is important to note that Robert *et al.* (2017) classify their oaks as presenting either a ring-porous or a diffuse wood structure. However, it is most likely that the wood of some of their species would possess a semi-ring-porous arrangement. For example, southern live oak (*Q. virginiana*) is described as diffuse-porous in Robert *et al.* (2017) while Panshin and de Zeeuw (1980) report it as semi-ring-porous. Most of the live oak species are likely to possess a semi-ring-porous structure (as mentioned in Table 1 and shown in Fig. 3D). In the Mediterranean region, the wood of Boissier oak (*Q. boissieri* Reut.) even shifts from a ring-porous to a semi-ring-porous structure under drier conditions (Castagneri *et al.* 2020).

Xylem anatomy has often been overlooked in studies on hydraulic conductivity. For instance, common grapevine (*Vitis vinifera* L.), a species linked to oaks for a very long time (see next section), also possesses wide vessels. Embolism only occurs at what is called severe water stress, which is when water potential in the leaves drops from -0.41 to -0.77 MPa according to Schultz and Matthews (1988), as reported by Lovisolo and Schubert (1998). In the latter study, it is shown that what is considered mild water stress in grapevine (~ 0.66 MPa) also causes a reduction of vessel size and this seems to prevent embolism that occurs at more severe water stress. Studying the physiology of woody plants demonstrates that their anatomy should always be examined to get better insights into how assays are designed and mainly into the interpretation of the results.

Diseases can also cause embolism because the large earlywood vessels of the current growing season in ring-porous species are particularly vulnerable. It is perhaps not a coincidence that two of the best known and most devastating vascular tree diseases, oak wilt and Dutch elm disease (DED), affect such species. Primary infection of these trees occurs after some kind of wounding, many directly caused by insect vectors for Dutch elm disease and mechanical or storm-related wounds for oak wilt. The insect vectors appear particularly active in spring, a period of high susceptibility to these diseases. In Minnesota and Texas, peak fungal mat formation in red oaks occurs early in the growing season, a period when nitidulid beetles are particularly active (O'Brien *et al.* 2011). As a result, it is highly recommended to avoid any kind of wounding, pruning for instance, during this period (Appel *et al.* 1987; Juzwik *et al.* 2010). When wounding is unavoidable, tree wound dressing should be applied promptly to avoid attracting unwanted insects. Spring is a period of great activity for insect vectors and increased availability of fungal propagules for these two diseases. It is also a time when new earlywood vessels are formed in a superficial and attackable location, just under the bark adjacent to the cambium. These large vessels ensure most of the sap transport and studies on DED frequently state that their vulnerability partially explains why elms (*Ulmus* spp.) are so susceptible early in the growing season (Tchernoff 1965). To our knowledge, this has never been mentioned for oak wilt. Obviously, the long and wide earlywood vessels represent ideal routes for the spread of fungal propagules in elm and oak xylem. In fact, the most severe infections of DED reported coincide exactly with the maturation of earlywood vessels in the spring (Tchernoff 1965). When functional and full of water, vessels are not ideal for pathogens as a certain quantity of air is required for full growth and development to be achieved. For instance, certain woods remain quite intact for hundreds and even thousands of years when submerged in water, the wood-decaying basidiomycete fungi being particularly intolerant to high moisture and low oxygen-level conditions while soft rot fungi and bacteria tolerant of the conditions are primarily responsible for some degradation (Kim and Singh 2000). Some of the most precious sunken logs are at times even retrieved by divers (Zucchino 2014). While the ascomycete wilt fungi that causes DED and oak wilt can grow and sporulate in poor oxygen conditions (Pegg 1985), their growth is likely to be improved in the presence of air that occurs after wounding.

Cavitated vessels would also favour the spread of the pathogen downward toward the roots (Tyree and Zimmermann 2002), where secondary infection through root grafts occurs for both DED and oak wilt. Tainter and Fraedrich (1986) have collected some roots from turkey oak (*Q. laevis* Walter) artificially inoculated with *B. fagacearum* and they demonstrate that the pathogen takes about one year to reach the roots. These trees, which were part of another study (Tainter and Ham 1983), were 10 to 30 cm in diameter. It seems that some kind of cavitation is involved in stems that facilitates downward pathogen spread. Eventually, that leads to colonization of nearby healthy trees through root grafts/contacts.

When samples are taken from symptomatic trees, various occluding structures in vessels are frequently observed. Many studies have linked the presence of these plugs to the interruption of sap transport. Zimmerman and McDonough (1978) stress that many plant pathologists made this assumption while infiltration of air (cavitation) in these vessels is most likely the main cause of vessels dysfunction, in particular during wilt diseases. The occluding structures in dysfunctional vessels, such as tyloses, would then be a consequence of embolism, not the cause. Plants would not block a functional vessel with plugs to deprive themselves of this vital fluid. The formation of these plugs is triggered by cavitation, which can be equated to vessels dysfunction. While studying DED with dyes and examining hundreds of transverse sections, Newbanks *et al.* (1983) reported the presence of dysfunctional vessels before any microscopic changes in them, including the presence of pathogen cells and/or occluding structures. Air would spread from vessel to vessel through air-seeding of the unaffected pit membranes, but also when pathogens penetrate these membranes and/or through the action of their extracellular hydrolytic enzymes (Zimmermann and McDonough 1978). In particular, pectinases would degrade the pit membranes and/or widen their pores, and thus accelerate the air-seeding process. It has long been known that cellulases and pectinases are produced by wilt pathogens (Dimond 1970; Pegg 1985). Degradation and/or penetration of host walls and pit membranes have often been mentioned during the development of vascular diseases, such as oak wilt (Jacobi and MacDonald 1980; Sachs *et al.* 1970). The presence of air in vessels would clearly be detrimental to plants, and as previously discussed, would help pathogens proliferate in the xylem. For DED and oak wilt, the presence of bubbles in vessels, different in structure from tyloses, is reported in microscopy as one of the first visible anatomical changes after infection (see next section). For example, bubbles are shown to occur as early as three days after inoculation and always in close association in vessels with the oak wilt pathogen (Jacobi and MacDonald 1980). Although the role, if any, of these bubbles is unclear, we now firmly believe that they are somehow linked to cavitation. The fact that they have not been observed in water-injected controls (Jacobi and MacDonald 1980), as also reported for DED (Et-Touil *et al.* 2005), also suggests that the host-pathogen interaction is crucial for their appearance. Ouellette (1980) clearly differentiates these bubbles (= alveolar network) from tyloses in transmission electron microscopy (TEM) during DED development. They seem so closely associated with pathogen growth that during one of his first studies, Ouellette (1962) seems to have confused them at times with emptied forms of the DED pathogen in light microscopy. It is worth noting that while studying the reactions of many nonhosts to the DED pathogen, bubbles were only observed in the most susceptible nonhost, pin cherry (*Prunus pensylvanica* L. f.), as well as in the very susceptible host *U. americana* (Rioux and Ouellette 1989).

The oak wilt pathogen is a dimorphic fungus, meaning it can exist in either mycelial or yeast-like (endoconidia) forms (Brandt 1963). True yeast such as *Saccharomyces cerevisiae* can proliferate under anaerobic conditions during fermentation (Dashko *et al.* 2014), and with the filamentous fungi *Mucor* spp. yeast growth is also favoured under such conditions (Orlowski 1991). However, laboratory studies of dimorphism of the Dutch elm fungi *Ophiostoma ulmi* (Buisman) Melin & Nannf. and *O. novo-ulmi* Brasier, wilt pathogens closely related to *B. fagacearum*, have shown that maximum production of yeast-like cells occurs in defined liquid media aerated by rotary agitation (Kulkarni and Nickerson 1981; Naruzawa and Bernier 2014). Whatever the case, in addition to fungal growth occurring in cavitated vessels, we postulate that colonization of oak trees by *B. fagacearum* also occurs under poor oxygen conditions through passive transport of some propagules, whether spores such as endoconidia or fragmented hyphae, immediately after penetration of the pit membranes between a cavitated and a functional vessel. When the latter vessel also becomes cavitated, these cells would germ and proliferate, allowing the hyphae to repeat the process, that is, to colonize adjacent vessels, whether cavitated or not. This colonization process would explain how the pathogen is able to reach the crown of oak trees showing wilt symptoms (Barnett 1953), which is caused by dysfunctional (cavitated) vessels.

Plants have developed different strategies to avoid gas emboli and/or to restore hydraulic conductance following different cavitation damage. Avoidance is often related to stomatal closure that occurs before water potential reaches negative thresholds inducing cavitation. Thus, embolism tolerance appears in some species to be linked to greater safety margins between water potential causing such closure and those causing gas emboli (Martin-StPaul *et al.* 2017). Prevention or tolerance to air-seeding is linked to xylem anatomy, as shown in the present study for the greater vulnerability of the wide vessels of ring-porous species when compared with diffuse-porous species. In *V. vinifera*, the arrangement of vessels and their interconnections via pits seems to play a significant role in embolism spread (Brodersen *et al.* 2013). Embolism repair in vessels has rarely been reported or satisfactorily explained in most of the plants studied. Herbaceous and bamboo species, which do not possess axial parenchyma in their xylem, are reported to refill their cavitated vessels mainly via root pressure (Brodersen *et al.* 2013). In conifers, the wood of which usually lacks axial parenchyma, repair is only reported in the crown, as well as in small branches and leaves where absorption of water through the cuticle and stomata seems to play a significant role to restore hydraulic conductivity (Limm *et al.* 2009). In diffuse-porous species, vessels that are affected by freezing somehow restore sap transport at the beginning of the growing season. Due to the importance of the sugar maple industry, sugar maple (*Acer saccharum* Marshall) has been intensely studied for its apparent ability to generate positive stem pressure sufficient to recover from freeze-induced vessel embolism and to allow sap exudation when temperatures oscillate around the freezing point (Graf *et al.* 2015). One of the most likely mechanisms of "vessel repair" in *A. saccharum* is proposed by Milburn and O'Malley (1984) who postulate that fibres are nonconductive under growing conditions and are mostly filled with air. During freezing, some of the vessel sap is drawn into these fibres through their walls and the compressed air generates a positive pressure within them. Upon thawing, the compressed air bubbles in the fibres expand and force the sap's reversal, thus repairing vessel conductivity through a repressurizing process. To our knowledge, repair has never been reported in relation to the large vessels of ring-porous species. Using non-invasive X-ray microtomography,

Choat *et al.* (2019) have studied such potential remediation after drought in three diffuse-porous *Eucalyptus* species and the ring-porous *Q. palustris* and concluded that refilling of embolized vessels is most likely not a widespread mechanism in woody plants. In the ring-porous *F. americana*, some embolism repair is reported by Zwieniecki and Holbrook (1998), but only in relation to slight water stress occurring diurnally, and in 1-year-old branches where the ring-porous structure is not present yet and most vessels are much smaller than elsewhere in the tree. We believe that once cavitated, vessels are often invaded by pathogens and remain dysfunctional with diseases such as oak wilt. Air spreading and concomitantly pathogen development from vessel to vessel are in part restricted by tylosis formation (see next section). Additionally, trees that survive oak wilt generally compartmentalize the invaded dysfunctional xylem before the cambium resumes its normal production of functional xylem elements (see next section).

ANATOMICAL FEATURES LIMITING OAK WILT DEVELOPMENT

In trees wounded and/or invaded by different microorganisms, or when sap transport is impaired during natural aging processes, the injured tissues are often compartmentalized, that is surrounded by defence boundaries that limit the development of compromised tissues. Compartmentalization in trees has been reported numerous times when the secondary xylem (wood) is implicated (Blanchette 1992; Pearce 1996; Shigo 1984). In its broad sense in the bark, compartmentalization generally occurs when new periderm layers are formed around the damaged tissues (Biggs 1992a, 1992b; Pearce 1996).

The concept of compartmentalization has been greatly popularized by the proposition of a model called CODIT, an acronym for "compartmentalization of decay in trees" (Shigo and Marx 1977). Although it initially explained how trunk xylem reacts to decay-causing fungi, CODIT was also reported in roots (Tippett and Shigo 1981b) and as a defence response to other types of microorganisms, notably wilt pathogens (Tippett and Shigo 1981a). Compartmentalization as a defence reaction to DED is also described as an important parameter to assess when screening cultivars potentially resistant to this disease (Beier and Blanchette 2018; Beier *et al.* 2017). CODIT comprises four types of walls that help the tree defend itself. Walls 1, 2 and 3 (= reaction zones) result from responses of parenchyma cells extant in the xylem at the time of damage. They limit the longitudinal, radial and tangential spread of microorganisms, respectively. Wall 4 (= barrier zone) is formed by the vascular cambium after damage and represents the most effective boundary of the model (Shigo 1984).

Turkey oak (*Q. laevis*) is a red oak species not quickly killed after artificial inoculations with *B. fagacearum* in South Carolina (Tainter and Ham 1983). This oak reacts to oak wilt by forming densely stained parenchyma cells together with vessels blocked by tyloses, interpreted as discontinuous barrier zones (wall 4) that hinder the progression of the pathogen (Tainter and Fraedrich 1986). The latter study provided the first description of defence reactions to oak wilt associated with the CODIT model. As it occurs for other diseases (Shigo 1984), after barrier zone formation, the cambium resumes its normal differentiation of xylem, notably of new vessels that appear healthy (Tainter and Fraedrich 1986). South Carolina represents the most southeastern range of the disease (Fig. 1A), and as early as 1975, while studying six southern states, Peacher *et al.* (1975) indicated that the southern progression of oak wilt in this area appears negligible. Even though Schoeneweiss (1959) was not aware of the concept of compartmentalization, he has described an unusual layer of cells in *Q. alba* that has survived to oak wilt, quite similar to the barrier zone reported in *Q. laevis*. He mentions a band of unusual cells continuous between the infected vessels and the healthy xylem tissue subsequently formed by the cambium. Schoeneweiss (1959) also stresses that these cells may protect the tree through the isolation of the pathogen. DED has been studied more intensively than oak wilt and compartmentalization processes such as barrier zone formation has been frequently reported as an effective defence mechanism (Bonsen *et al.* 1985; Shigo and Tippett 1981). In dead or dying parts of the elm, Shigo and Tippett (1981) do not observe any barrier zone formation. We have shown (Rioux and Ouellette 1991a) that these barriers may be present in American elm (*U. americana*) in response to DED, but are often discontinuous, and that occlusions of vessels and the presence of the pathogen in elm are then noted close to the cambium. In nonhost trees, the same study shows that when barrier zones are observed, they are generally continuous. Jacobi and MacDonald (1980) report that zones of darkly stained parenchyma cells, likely corresponding to barrier zone cells, are regularly formed in response to artificial inoculations with *B. fagacearum* in two white oak species. In *Q. rubra,* these zones are described as diffuse or poorly formed, which we interpret as discontinuous barrier zones. In the absence of these barriers or when they are discontinuous, the cambium is greatly altered and the tree dies (Shigo and Tippett 1981). During oak wilt, this appears to signal the onset of fungal mat formation (Struckmeyer *et al.* 1958).

Polyphenolic substances such as lignin and suberin are shown to be abundant in barrier zones cells, for instance in response to DED (Rioux and Ouellette 1991a, 1991b). Dark substances that are likely phenols are also reported in barrier and/or reaction zone cells in response to oak wilt (Jacobi and MacDonald 1980; Tainter and Fraedrich 1986). Lignin and suberin, the latter also constituted of large amounts of fatty acids, confer antimicrobial properties to the walls, but concomitantly they render them impermeable to air propagation that precedes and/or accompanies the infection.

Most of the time cells that compose barrier zones are described in the literature as parenchymatous (e.g., see Pearce 1996). We have reported for the first time (Rioux and Ouellette 1991a, 1991b) that fibres can also be a main component of such barriers in *U. americana* and in the nonhost balsam popular (*Populus balsamifera* L.) in response to inoculations with the DED pathogen. These fibres are often suberized and it is interesting to note that while studying *U. americana* trees that survive acute stages of DED, Ouellette (1981a) describes fibres in TEM having wall layers "reminiscent of suberin layers observed in other normal or diseased plants". It is most likely that several of these suberized fibres were the main constituents of barrier zones, as described in Rioux and Ouellette (1991a, 1991b).

In the literature, the main structural change associated with wall 1 of the CODIT model is the occlusion of vessels by tyloses and/or gels (= gums). Actually, tyloses are more frequent within the large dysfunctional vessels of ring-porous trees than in the smaller vessels of other species, whatever the damage. Tyloses are also abundant in heartwood of ring-porous species (Bonsen and Bucher 1991), even though they are usually formed in the internal sapwood, since they are outgrowths from living parenchyma cells, either ray or axial. Although the discovery of tyloses is attributed to Marcello

Malpighi in 1675 (Bonsen and Bucher 1991), the first extensive study on these vessel plugs was produced in 1845 by Hermine von Reichenbach, a fascinating investigation shared by Zimmermann (1979). von Reichenbach rightly describes tyloses as outgrowths from parenchyma cells growing into the vessels through the pits. Among the taxa she has studied that produced tyloses were common oak (*Q. robur* L.) and *V. vinifera*, two species discussed below in relation to the wine industry. Tyloses are the usual occluding structures found in the wider dysfunctional vessels of plants, as reported in detail by Gerry (1914), where abundant tylosis development in many ring-porous species (*Quercus* spp. and black locust [*Robinia pseudoacacia* L.]) is particularly obvious. Gels or gums often occur in diffuse-porous species as the result of secretory activities of parenchyma cells (Bonsen and Kučera 1990; De Micco *et al.* 2016). These parenchyma cells contiguous to vessels are usually called paratracheal, but at times also contact or vessel-associated cells (Morris and Jansen 2016; Morris *et al.* 2018). There seems to be a positive correlation between wide vessels and pit aperture diameter, with vessels less than 80 µm in diameter and pit aperture less than 3 µm tend to produce gels, while those with greater sizes form tyloses (Bonsen and Kučera 1990). This holds true for most temperate tree species while there are many exceptions to this rule for tropical species (De Micco *et al.* 2016). It makes sense that pit apertures less than 3 µm would not easily allow the passage of the double tylosis walls. A few exceptions have been reported for temperate species concerning vessel sizes. For instance, *Magnolia* spp. form tyloses even though their vessel diameters are generally less than 80 µm. However, no species have been described with pit apertures below 3 µm and tylosis formation. In the case of *Magnolia* spp., pit aperture diameter is over 3 µm (Bonsen and Kučera 1990), and even as much as 10 µm according to De Micco *et al.* (2016). When examining young seedlings or saplings of species usually forming tyloses in the trunk in response to various damages, it is important to remember that their vessels are smaller than normal as are, in general, their pit apertures. This may result in the concomitant production of tyloses and gels, as reported for instance for butternut seedlings (*Juglans cinerea* L.) in Rioux *et al.* (2018). There is also the possibility that tyloses secrete material similar to gels, often of a pectinic nature, as shown using monoclonal antibodies in Rioux *et al.* (1998), which helps to completely block vessel lumina. If transverse sections examined in microscopy are through such gels without showing the tylosis walls from which they might originate, they could be misinterpreted as being directly secreted from parenchyma cells. It is also possible that some water still present in these vessels is caught by these pectic fibrils and moved through the primary wall of tyloses to help their expansion. Some of this water might be translocated elsewhere to support other cells in differentiation in trees, such as new vessels in expansion in the current growth ring (see previous section).

Vessels of smaller diameters are reported to be related to resistance to DED in some elms, such as field elm (*U. minor* Mill.) (Solla and Gil 2002). Studying offspring from different crosses between resistant and susceptible *U. minor* trees, more variable results have been reported, specifically that narrow earlywood vessels are not always related to DED resistance (Martín *et al.* 2021). However, resistant offspring with wide earlywood vessels react to inoculation with *O. novo-ulmi* by forming narrower earlywood vessels the following year. Then in the latewood, there is an increase in vessel size that likely helps water transport. It is worth noting that barrier zone formation, whether continuous or not, is only partially associated with resistance in offspring studied by Martín *et al.* (2021).

The characteristics of vessels, such as their diameter or their arrangement within growth rings, have not been assessed in relation to the susceptibility of different species to oak wilt. It is possible that future studies in that sense could shed some light on host-pathogen interactions during oak wilt development. For instance, bur oak (*Q. macrocarpa*) seems to have earlywood vessels wider than other oak species (Woodcock 1989) and this may explain why it is more susceptible to oak wilt than other white species.

In our samples, we have examined some discoloured tissues that were buried in the wood and well compartmentalized. As expected, the necrotic tissues are surrounded by reactive cells impregnated with suberin and phenols, as shown in confocal microscopy (Fig. 7A). The presence of suberin in the V-shape band of cells that covers the wound indicates that they are formed by the bark cambium, and thus is a necrophylactic periderm. This transverse section is exactly at the level of the wound that causes the damage, as discussed in Rioux *et al.* (2018) when examining *J. cinerea* reactions after inoculations with the butternut canker pathogen (*Ophiognomonia clavigignenti-juglandacearum* (Nair, Kostichka & Kuntz) Broders & Boland) that threatens the survival of this species. As in Figure 7A, Rioux *et al.* (2018) describe in detail the presence of cells that have accumulated phenols that apparently limit the tangential development of the injured xylem.

Tyloses have always been reported as a response to oak wilt in histopathological studies (Jacobi and MacDonald 1980; Tainter and Fraedrich 1986). At times, they appear adjacent internally to barrier zones, whether in *Q. alba* (Schoeneweiss 1959) or in *Q. rubra* (Tainter and Fraedrich 1986). Most of the tyloses examined under UV light in undamaged wood in our study were suberized, as in *Q. alba* (Fig. 7B). Following some unknown damage, the vessels of the wood directly exposed to air are occluded with tyloses, but they do not seem suberized (Fig. 7A). We have seen a similar phenomenon when studying DED, particularly in the nonhost *P. balsamifera* where the wounded and invaded xylem, containing several non-suberized tyloses, is however surrounded by suberized tyloses that help in the efficacy of the wall 3 reaction zone (Rioux and Ouellette 1991a). It is worth noting that the invaded xylem of *P. balsamifera* seedlings is at times completely walled off by compartmentalized barriers, even internally by a reaction zone (= wall 2) constituted of perimedullary cells that dedifferentiate and eventually become intensely suberized. This type of reaction zone was described for the first time in trees by Rioux and Baayen (1997). This reaction zone shows similarities with a suberized zone formed in carnation (*Dianthus caryophyllus* L.) in response to another wilt pathogen, *Fusarium oxysporum* f.sp. *dianthi* W.C. Snyder & H.N. Hansen (1940), the whole compartmentalization process reported for the first time in an herbaceous species by Baayen *et al.* (1996). The suberized layer is often the last formed within tylosis walls, as well as in any parenchyma cell from which they protrude, and this apparently causes their death. It is likely that the xylem parenchyma and tyloses in oak directly exposed to air (Fig. 7A) do not have sufficient time to generate such layers before dying. Understandably, when the wound surface dries out too rapidly in trees, the affected parenchyma cells cannot react quick enough to the damage, thus no plugs are formed in their vessels (Bonsen and Kucher 1991).

Tyloses are observed in the wide earlywood vessels of our samples that were dysfunctional, even in our red oak cookies. In transverse sections, 100% of the dysfunctional vessels in the white oak species studied are occluded, an indication that 100% of their length is blocked. Using similar

cross-sections for the red oaks, we are always able to see at least some tyloses in dysfunctional vessels, such as suberized ones in the sapwood of black oak (*Q. velutina*) (Figs. 7C and 7D). In *Q. rubra*, some suberized tyloses were also observed in vessels seemingly dysfunctional following sampling the branches of healthy trees in winter (Rioux *et al.* 1995). It is almost impossible with ring-porous species to follow longitudinally their long and wide sinuous vessels in large stems to visually determine the proportion that is occluded with plugs. Researchers so far have rather used various dyes to help them in such evaluations (Ellmore and Ewers 1986). We believe that with sufficient sampling and statistical analyses with transverse sections in red oaks, it would be possible to get a good assessment of the proportion of vessel lengths occluded with tyloses. For instance, if the Figure 7C were representative of sufficient sampling in *Q. velutina*, we could conclude that around 3% (1 occluded vessel out of 30) of the length of these dysfunctional vessels are obstructed with tyloses. As previously mentioned, examining numerous transverse sections has proved useful in understanding the conducting status of vessels in elms affected by DED (Newbanks *et al.* 1983). We postulate that only a small fraction of the length of dysfunctional vessels in red oaks are always occluded with tyloses. Longitudinal views would have to be checked from time to time to confirm that an obstructed vessel, such as that of Figure 7C, is only partially plugged and not the only vessel intensely clogged with tyloses on most of its length.

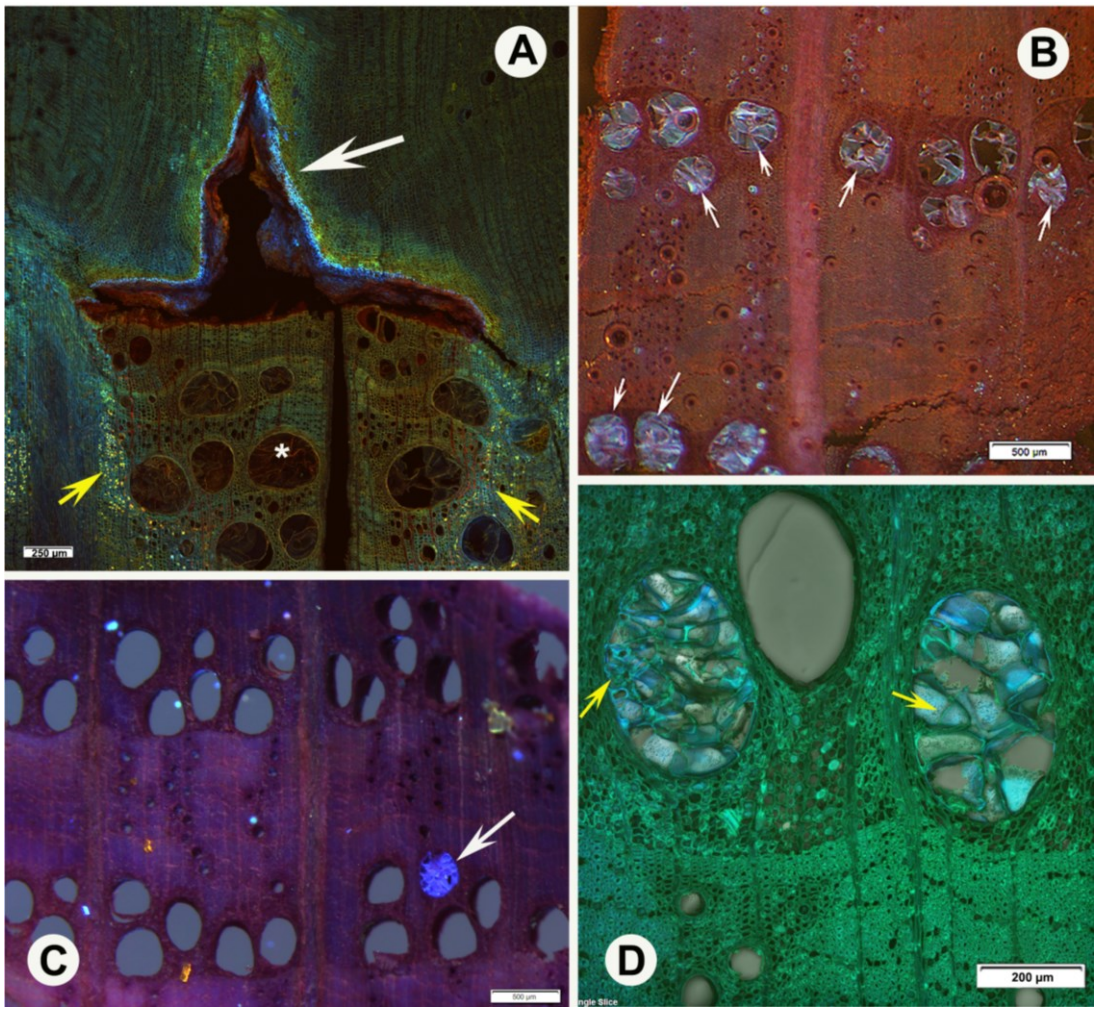

Figure 7. (A): In confocal microscopy, a merging of images taken with lasers 405, 488, 559 and 635 nm, compartmentalization of an unidentified wounding damage in *Q. macrocarpa*. The V-shape and light blue autofluorescence colour revealed under 405 nm of the layer covering the injured xylem, indicative of a necrophylactic suberized periderm (white arrow), means that this section is at the same level as the original damage. The yellow autofluorescence detected under 488 nm of the cells tangentially bordering (yellow arrows) the injured xylem suggests the presence of phenols. The vessels in the damaged xylem are plugged with non-suberized tyloses (asterisk). **(B-D):** Suberized tyloses. **(B-C):** Detected in conventional epifluorescence microscopy under UV illumination (sections stained with phloroglucinol-HCl). **(B):** All the earlywood vessels in *Q. alba* are clearly plugged with suberized tyloses (arrows). **(C):** Only one (arrow) out of 30 vessels in the sapwood of *Q. velutina* is occluded with tyloses, this autofluorescence indicating the presence of suberin. **(D):** Heartwood of *Q. velutina*. This confocal micrography, a composite of two images taken with lasers 405 and 488 nm, shows light blue autofluorescence indicative of suberin in the tylosis walls in two vessels when excited with 405 nm. This micrography also shows a thin wall in a tylosis on the right (arrow) and many small tyloses on the left (arrow) suggesting that these structures are in a division process.

Schmitt and Liese (1994) report that some tyloses can divide to produce additional daughter plugs in earlywood vessels of *R. pseudoacacia* after wounding. They suggest that a similar phenomenon might also occur in other ring-porous species. In our study, samples were not examined in TEM to disclose whether tyloses can exhibit such divisions, but the use of confocal microscopy seems to indicate that this may also occur in oak trees. In Figure 7D, the vessel on the left contains many small tyloses that may have resulted from such divisions. Within the vessel on the right, a thin wall in a tylosis can be seen, suggesting that it could have been in a current state of division.

The aptitude of white oaks to readily form tyloses might explain why they are more resistant than red oaks to oak wilt (Jacobi and MacDonald 1980). Red oaks also appear more susceptible than white oaks to other diseases, such as sudden oak death caused by *Phytophthora ramorum* Werres *et al.* 2001 (Rizzo *et al.* 2002). As this latter pathogen appears present in the wood more often than previously thought (A.V. Brown and Brasier 2007), a tissue it may exploit to colonize the tree more easily than the bark, the failure of red oaks to rapidly form tyloses might favour extensive vessel cavitation and pathogen growth during sudden oak death development. Furthermore, Smith and Stanosz (2018) report shorter stem lesions in white oaks than in red oaks following inoculations with the stem canker pathogen *Diplodia corticola* A.J.L. Phillips, A. Alves & J. Luque. They postulate that host responses such as tylosis formation might explain the apparent difference in susceptibility between these oaks.

It has been reported as early as at the beginning of the 20th century that felling a tree could induce the formation of tyloses in the outer growth rings of logs (Gerry 1914). However, this appears to occur only weeks after cutting the trees during dormancy and the logs must be kept in conducive conditions of moisture and temperature, as shown for *Q. rubra* (Murmanis 1975). In our study, the trees were cut in winter and when brought to the laboratory, they were dried within four days to avoid significant tylosis formation. We checked *Q. alba* to see if the formation of suberin in tyloses could have been induced after felling. We did not observe any noticeable suberin autofluorescence differences, or in the apparent thickness of suberized layers, related to this compound in tyloses in all sapwood growth rings, except the last one where vessels are apparently free of plugs (Fig. 3B). Another clear indication that the changes in the tree cookies we examined were unrelated to felling was that only the parenchyma cells in the transition zone between sapwood and heartwood accumulated what appears to be phenolics. This is expected under normal growing conditions, whereas similar parenchyma in the other sapwood rings were not visually different than those usually found in oaks. However, in the outermost growth ring of *Q. alba* (Fig. 3B), although most vessels seem entirely free of occlusions, we occasionally noticed some material in the large vessels that could have been tyloses in formation. In this case, we cannot entirely rule out the possibility that some may have resulted from cutting down the trees.

The abundance of tyloses in the large vessels of white oak species explains why their wood is preferred for cooperage in comparison to the leaky wood of the red oak group. The sessile oak (*Q. petraea* (Matt.) Liebl.) and pedunculate oak (*Q. robur*) are the main species used in Europe, while white oak (*Q. alba*) is the most common species used to make casks in North America. The presence of tyloses with thick walls in *Q. alba* would render its wood more watertight than that of the European species. The presence of these tyloses also explains why the wood of *Q. alba* is sawn, while that from *Q. petraea* and *Q. robur* must be split to avoid leaking problems caused by damaging their more fragile tyloses (Chatonnet and Dubourdieu 1998). Furthermore, it is difficult to obtain more than 25% of staves from split logs while 50% can be produced when logs are sawn (Chatonnet and Dubourdieu 1998). This is why European barrels are more expensive to produce than those made of *Q. alba* wood. Considering it is quite easy to detect suberin using the Biggs' method developed in 1984, as shown in our study, we are a bit surprised to realize after surveying the literature that the presence of suberin has never been taken into account during assessing the parameters associated with the tightness of wood used for cooperage.

We have never seen the role of pit membrane pores as a pathway for air propagation between cavitated and functional vessels studied in relation to plant diseases. For instance, in a recent review on wilt diseases, the porosity of pit membranes is not discussed except to stress that their reinforcement with various substances may prevent the spread of pathogens and/or block the movement of macromolecules produced by them, such as toxins and enzymes (Kashyap *et al.* 2021). Vessel coatings of a lipidic to suberin nature would occur in alfalfa (*Medicago sativa* L.) infected with *Verticillium albo-atrum* Reinke & Berthold (1879), but this is interpreted as a barrier to pathogen movement (Newcombe and Robb 1989), not in relation to hindering cavitation spreading in the xylem. In Van Alfen *et al.* (1983), macromolecules produced by vascular pathogens are linked to host susceptibility through increasing the resistance to water flow, mainly by plugging the pit membrane pores. Pit membrane thickenings have been reported in elms surviving DED (Ouellette 1981b) and in nonhost plants inoculated with its causal pathogen (Rioux and Ouellette 1989). However, it has never been envisaged that these modifications are somehow linked to prevent air propagation in xylem tissues. In Rioux *et al.* (2018), compartmentalization is discussed as a reaction to butternut canker that may have been triggered by air infiltration, preventing the drying out of the living xylem. We should have insisted that some reactions, mainly related to suberization, could be a direct impediment to cavitation propagation that subsequently helps the growth/spread of the pathogen. In Rioux *et al.* (2018), Supplementary Figure 2 shows in TEM a typical lamellar structure of a suberized coating in a vessel, this conduit being part of a barrier zone in *P. pensylvanica* in response to an inoculation with the DED pathogen. What was not mentioned is that at times this suberin also clearly covers the pit membranes, thus possibly preventing air propagation in adjacent xylem cells. Air propagation in wood saturated with water under normal physiological conditions, or the mechanisms that may help impede its spread, is likely the most promising approach to investigate host-wilt pathogen interactions in the future.

CONCLUDING REMARKS

As in other research fields (e.g., physiology, biochemistry, genetics, evolution), improving our understanding of trees by studying anatomy is still providing key findings that help better comprehend the complex and wonderful material that is wood. Unfortunately, some of the remarkable characteristics of wood, not the least its natural beauty and durability, is at the root of illegal activities worldwide. Such illegal international trade has grown to the point that the survival of many species (e.g., rosewood [*Dalbergia* spp.] and mahogany [*Swietenia* spp.]) is threatened. To combat this black market, regulators rely on different tools such as the correct identification of wood products along the supply chain. Anatomical analysis remains one of the best and cheapest ways to achieve this goal, at least the identification at the genus level (Lowe *et al.* 2016). In relation to oak wilt, using relatively simple anatomical techniques, our study shows that it is easy to identify the two main groups of oaks affected by this

disease. Such information can be used to prevent trading infected material. Additionally, microscopic examination also permits to observe many histological changes that occur during the development of this disease, notably the formation of defensive structures related to compartmentalization processes. Wood anatomy is also an important component to consider to better understand how sap moves in trees and how wilting occurs, in particular in ring-porous species such as oaks. We suggest that detecting cavitation and the propagation of pathogens in relation to vessel characteristics may well prove to be one of the best ways to provide new insights into vascular diseases.

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REFERENCES

Appel, D.N. 1995. The oak wilt enigma: perspectives from the Texas epidemic. Annu. Rev. Phytopathol. 33: 103-118. doi:10.1146/annurev.py.33.090195.000535

Appel, D.N., R. Peters, and R. Lewis, Jr. 1987. Tree susceptibility, inoculum availability, and potential vectors in a Texas oak wilt center. J. Arboric. 13: 169-173. doi:10.48044/jauf.1987.037

Baas, P., E. Werker, and A. Fahn. 1983. Some ecological trends in vessel characters. IAWA J. 4: 141-159. doi:10.1163/22941932-90000407

Baayen, R.P., G.B. Ouellette, and D. Rioux. 1996. Compartmentalization of decay in carnations resistant to *Fusarium oxysporum* f. sp. *dianthi*. Phytopathology 86: 1018-1031. doi:10.1094/Phyto-86-1018

Barnett, H.L. 1953. Isolation and identification of the oak wilt fungus. Bulletin No. 359T. West Virginia Agricultural and Forestry Experiment Station Bulletins. Available online [https://researchrepository.wvu.edu/wv_agricultural_and_forestry_experiment_station_bulletins/627].

Beier, G.L., and R.A. Blanchette. 2018. Defence responses in the xylem of *Ulmus americana* cultivars after inoculation with *Ophiostoma novo-ulmi*. For. Pathol. 48: e12453. doi:10.1111/efp.12453

Beier, G.L., B.W. Held, C.P. Giblin, J. Cavender-Bares, and R.A. Blanchette. 2017. American elm cultivars: variation in compartmentalization of infection by *Ophiostoma novo-ulmi* and its effects on hydraulic conductivity. For. Pathol. 47: e12369. doi:10.1111/efp.12369

Biggs, A.R. 1984. Intracellular suberin: occurrence and detection in tree bark. IAWA J. 5: 243-248. doi:10.1163/22941932-90000899

Biggs, A.R. 1992a. Anatomical and physiological responses of bark tissues to mechanical injury. Pages 13-40 *in* R.A. Blanchette and A.R. Biggs (eds.), Defense mechanisms of woody plants against fungi. Springer, Berlin, Germany. doi:10.1007/978-3-662-01642-8_2

Biggs, A.R. 1992b. Responses of angiosperm bark tissues to fungi causing cankers and canker rots. Pages 41-61 *in* R.A. Blanchette and A.R. Biggs (eds.), Defense mechanisms of woody plants against fungi. Springer, Berlin, Germany. doi:10.1007/978-3-662-01642-8_3

Blaedow, R.A., J. Juzwik, and B. Barber. 2010. Propiconazole distribution and effects on *Ceratocystis fagacearum* survival in roots of treated red oaks. Phytopathology 100: 979-985. doi:10.1094/PHYTO-01-10-0008

Blanchette, R.A. 1992. Anatomical responses of xylem to injury and invasion by fungi. Pages 76-95 *in* R.A. Blanchette and A.R. Biggs (eds.), Defense mechanisms of woody plants against fungi. Springer, Berlin, Germany. doi:10.1007/978-3-662-01642-8_5

Bonsen, K.J.M., and H.P. Bucher. 1991. What arborists have to know about vessel plugs. Arboric. J. 15: 13-17. doi:10.1080/03071375.1991.9746864

Bonsen, K.J.M., and L.J. Kučera. 1990. Vessel occlusions in plants: morphological, functional and evolutionary aspects. IAWA J. 11: 393-399. doi:10.1163/22941932-90000528

Bonsen, K.J.M., R.J. Scheffer, and D.M. Elgersma. 1985. Barrier zone formation as a resistance mechanism of elms to Dutch elm disease. IAWA J. 6: 71-77. doi:10.1163/22941932-90000916

Brandt, W.H. 1963. Dimorphism and interactions between the oak wilt fungus and associated fungi. Plant Dis. Rep. 47: 579-582.

Breshears, D.D., N.G. McDowell, K.L. Goddard, K.E. Dayem, S.N. Martens, C.W. Meyer, and K.M. Brown. 2008. Foliar absorption of intercepted rainfall improves woody plant water status most during drought. Ecology 89: 41-47. doi:10.1890/07-0437.1

Briggs, L.J. 1950. Limiting negative pressure of water. J. Appl. Phys. 21: 721-722. doi:10.1063/1.1699741

Brodersen, C.R., A.J. McElrone, B. Choat, E.F. Lee, K.A. Shackel, and M.A. Matthews. 2013. In vivo visualizations of drought-induced embolism spread in *Vitis vinifera*. Plant Physiol. 161: 1820-1829. doi:10.1104/pp.112.212712

Brown, A.V., and C.M. Brasier. 2007. Colonization of tree xylem by *Phytophthora ramorum*, *P. kernoviae* and other *Phytophthora* species. Plant Pathol. 56: 227-241. doi:10.1111/j.1365-3059.2006.01511.x

Brown, H.R. 2013. The theory of the rise of sap in trees: some historical and conceptual remarks. Phys. Perspect. 15: 320-358. doi:10.1007/s00016-013-0117-1

Burgess, S.S.O., and T.E. Dawson. 2004. The contribution of fog to the water relations of *Sequoia sempervirens* (D. Don): foliar uptake and prevention of dehydration. Plant Cell Environ. 27: 1023-1034. doi:10.1111/j.1365-3040.2004.01207.x

Canadian Food Inspection Agency. 2020. D-99-03: Phytosanitary import requirements to prevent the entry of oak wilt disease (*Bretziella fagacearum* (Bretz) Hunt) from the Continental United States. Government of Canada. Available online [https://www.inspection.gc.ca/plant-health/plant-pests-invasive-species/directives/date/d-99-03/eng/1323852753311/1323852907742] (Accessed in October 2020).

Castagneri, D., M. Carrer, L. Regev, and E. Boaretto. 2020. Precipitation variability differently affects radial growth, xylem traits and ring porosity of three Mediterranean oak species at xeric and mesic sites. Sci. Total Environ. 699: 134285. doi:10.1016/j.scitotenv.2019.134285

Cavender-Bares, J. 2019. Diversification, adaptation, and community assembly of the American oaks (*Quercus*), a model clade for integrating ecology and evolution. New Phytol. 221: 669-692. doi:10.1111/nph.15450

Chatonnet, P., and D. Dubourdieu. 1998. Comparative study of the characteristics of American white oak (*Quercus alba*) and European oak (*Quercus petraea* and *Q. robur*) for production of barrels used in barrel aging of wines. Am. J. Enol. Vitic. 49: 79-85. doi:10.5344/ajev.1998.49.1.79

Choat, B., M. Nolf, R. Lopez, J.M.R. Peters, M.R. Carins-Murphy, D. Creek, and T.J. Brodribb. 2019. Non-invasive imaging shows no evidence of embolism repair after drought in tree species of two genera. Tree Physiol. 39: 113-121. doi:10.1093/treephys/tpy093

Cones, W.L. 1967. Oak wilt mats on white oak in West Virginia. Plant Dis. Rep. 51: 430-431.

Cruiziat, P., H. Cochard, and T. Améglio. 2002. Hydraulic architecture of trees: main concepts and results. Ann. For. Sci. 59: 723-752. doi:10.1051/forest:2002060

Dashko, S., N. Zhou, C. Compagno, and J. Piškur. 2014. Why, when, and how did yeast evolve alcoholic fermentation? FEMS Yeast Res. 14: 826-832. doi:10.1111/1567-1364.12161

Davis, S.D., J.S. Sperry, and U.G. Hacke. 1999. The relationship between xylem conduit diameter and cavitation caused by freezing. Am. J. Bot. 86: 1367-1372. doi:10.2307/2656919

Dawson, T.E. 1998. Fog in the California redwood forest: ecosystem inputs and use by plants. Oecologia 117: 476-485. doi:10.1007/s004420050683

de Beer, Z.W., S. Marincowitz, T.A. Duong, and M.J. Wingfield. 2017. *Bretziella*, a new genus to accommodate the oak wilt fungus, *Ceratocystis fagacearum* (Microascales, Ascomycota). MycoKeys 27: 1-19. doi:10.3897/mycokeys.27.20657

De Micco, V., A. Balzano, E.A. Wheeler, and P. Baas. 2016. Tyloses and gums: a review of structure, function and occurrence of vessel occlusions. IAWA J. 37: 186-205. doi:10.1163/22941932-20160130

DiGasparro, M. 2020. Oak wilt eDNA detected in Ontario. Available online [https://www.invasivespeciescentre.ca/oak-wilt-edna-detected-in-ontario] (Accessed in August 2022).

Dimond, A.E. 1970. Biophysics and biochemistry of the vascular wilt syndrome. Annu. Rev. Phytopathol. 8: 301-322. doi:10.1146/annurev.py.08.090170.001505

Dixon, H.H., and J. Joly. 1895. XII. On the ascent of sap. Phil. Trans. R. Soc. Lond. B 186: 563-576. doi:10.1098/rstb.1895.0012

Eames, A.J. 1910. On the origin of the broad ray in *Quercus*. Bot. Gaz. 49: 161-167. doi:10.1086/33014

Ellmore, G.S., and F.W. Ewers. 1986. Fluid flow in the outermost xylem increment of a ring-porous tree, *Ulmus americana*. Am. J. Bot. 73: 1771-1774. doi:10.1002/j.1537-2197.1986.tb09709.x

Et-Touil, A., D. Rioux, F.M. Mathieu, and L. Bernier. 2005. External symptoms and histopathological changes following inoculation of elms putatively resistant to Dutch elm disease with genetically close strains of *Ophiostoma*. Can. J. Bot. 83: 656-667. doi:10.1139/B05-037

Farrar, J.L. 1995. Trees in Canada. Canadian Forest Service, and Fitzhenry and Whiteside Limited, Ottawa/Markham, ON, Canada. 502 pp.

Farrar, J.L. 2017. Trees in Canada. Canadian Forest Service, and Fitzhenry and Whiteside Limited, Ottawa/Markham, ON, Canada. 512 pp.

French, D.W., and W.C. Stienstra. 1980. Oak wilt. Report No. 310. University of Minnesota, Agricultural Extension Service, MN, USA. 6 pp.

Gerry, E. 1914. Tyloses: their occurrence and practical significance in some American woods. J. Agric. Res. 1: 445-470.

Goldman, D.H. 2017. Plant guide for live oak (*Quercus virginiana*). USDA-Natural Resources Conservation Service, National Plant Data Team. Greensboro, NC, USA. Available online [http://gulfcoastswcd.org/wp-content/uploads/2020/01/PG-Live-Oak.pdf].

Government of Canada. 2019. 440391 - Logs, of oak, not treated - Canadian Importers Database (CID). Available online [https://www.ic.gc.ca/app/scr/ic/sbms/cid/productReportHS10.html?hsCode=4403910010] (Accessed in March 2021).

Graf, I., M. Ceseri, and J.M. Stockie. 2015. Multiscale model of a freeze–thaw process for tree sap exudation. J. R. Soc. Interface 12: 20150665. doi:10.1098/rsif.2015.0665

Haight, R.G., F.R. Homans, T. Horie, S.V. Mehta, D.J. Smith, and R.C. Venette. 2011. Assessing the cost of an invasive forest pathogen: a case study with oak wilt. Environ. Manage. 47: 506-517. doi:10.1007/s00267-011-9624-5

Hargrave, K.R., K.J. Kolb, F.W. Ewers, and S.D. Davis. 1994. Conduit diameter and drought-induced embolism in *Salvia mellifera* Greene (Labiatae). New Phytol. 126: 695-705. doi:10.1111/j.1469-8137.1994.tb02964.x

Harlow, W.M., E.S. Harrar, and F.M. White. 1979. Textbook of dendrology: covering the important forest trees of the United States and Canada. McGraw-Hill, New York, NY, USA. 510 pp.

Henry, B.W., C.S. Moses, C.A. Richards, and A.J. Riker. 1944. Oak wilt, its significance, symptoms and cause. Phytopathology 34: 636-647.

Hoadley, R.B. 1990. Identifying wood: accurate results with simple tools. The Taunton Press, Newtown, CT, USA. 223 pp.

Jacobi, W.R., and W.L. MacDonald. 1980. Colonization of resistant and susceptible oaks by *Ceratocystis fagacearum*. Phytopathology 70: 618-623. doi:10.1094/Phyto-70-618

Jansen, S., B. Choat, and A. Pletsers. 2009. Morphological variation of intervessel pit membranes and implications to xylem function in angiosperms. Am. J. Bot. 96: 409-419. doi:10.3732/ajb.0800248

Jensen, W.A. 1962. Botanical histochemistry: principles and practice. W.H. Freeman, San Francisco, CA, USA. 408 pp.

Juzwik, J., D.N. Appel, W.L. MacDonald, and S. Burks. 2011. Challenges and successes in managing oak wilt in the United States. Plant Dis. 95: 888-900. doi:10.1094/PDIS-12-10-0944

Juzwik, J., T.C. Harrington, W.L. MacDonald, and D.N. Appel. 2008. The origin of *Ceratocystis fagacearum*, the oak wilt fungus. Annu. Rev. Phytopathol. 46: 13-26. doi:10.1146/annurev.phyto.45.062806.094406

Juzwik, J., J. O'Brien, C. Evenson, P. Castillo, and G. Mahal. 2010. Controlling spread of the oak wilt pathogen (*Ceratocystis fagacearum*) in a Minnesota urban forest park reserve. Arboric. Urban For. 36: 171-178. doi:10.48044/jauf.2010.023

Kashyap, A., M. Planas-Marquès, M. Capellades, M. Valls, and N.S. Coll. 2021. Blocking intruders: inducible physico-chemical barriers against plant vascular wilt pathogens. J. Exp. Bot. 72: 184-198. doi:10.1093/jxb/eraa444

Kim, Y.S., and A.P. Singh. 2000. Micromorphological characteristics of wood biodegradation in wet environments: a review. IAWA J. 21: 135-155. doi:10.1163/22941932-90000241

Kitin, P., and R. Funada. 2016. Earlywood vessels in ring-porous trees become functional for water transport after bud burst and before the maturation of the current-year leaves. IAWA J. 37: 315-331. doi:10.1163/22941932-20160136

Kulkarni, R.K., and K.W. Nickerson. 1981. Nutritional control of dimorphism in *Ceratocystis ulmi*. Exp. Mycol. 5: 148-154. doi:10.1016/0147-5975(81)90015-3

Langan, S.J., F.W. Ewers, and S.D. Davis. 1997. Xylem dysfunction caused by water stress and freezing in two species of co-occurring chaparral shrubs. Plant Cell Environ. 20: 425-437. doi:10.1046/j.1365-3040.1997.d01-94.x

Lev-Yadun, S. 1994. Radial fibres in aggregate rays of *Quercus calliprinos* Webb. – evidence for radial signal flow. New Phytol. 128: 45-48. doi:10.1111/j.1469-8137.1994.tb03984.x

Limm, E.B., K.A. Simonin, A.G. Bothman, and T.E. Dawson. 2009. Foliar water uptake: a common water acquisition strategy for plants of the redwood forest. Oecologia 161: 449-459. doi:10.1007/s00442-009-1400-3

Lovisolo, C., and A. Schubert. 1998. Effects of water stress on vessel size and xylem hydraulic conductivity in *Vitis vinifera* L. J. Exp. Bot. 49: 693-700. doi:10.1093/jxb/49.321.693

Lowe, A.J., E.E. Dormontt, M.J. Bowie, B. Degen, S. Gardner, D. Thomas, C. Clarke, A. Rimbawanto, A. Wiedenhoeft, Y. Yin, and N. Sasaki. 2016. Opportunities for improved transparency in the timber trade through scientific verification. BioScience 66: 990-998. doi:10.1093/biosci/biw129

Manos, P.S., J.J. Doyle, and K.C. Nixon. 1999. Phylogeny, biogeography, and processes of molecular differentiation in *Quercus* subgenus *Quercus* (Fagaceae). Mol. Phylogenet. Evol. 12: 333-349. doi:10.1006/mpev.1999.0614

Martin, C.W., R.C. Maggio, and D.N. Appel. 1989. The contributory value of trees to residential property in the Austin, Texas metropolitan area. J. Arboric. 15: 72-76. doi:10.48044/jauf.1989.017

Martín, J.A., A. Solla, T. Oszako, and L. Gil. 2021. Characterizing offspring of Dutch elm disease-resistant trees (*Ulmus minor* Mill.). Forestry 94: 374-385. doi:10.1093/forestry/cpaa040

Martin-StPaul, N., S. Delzon, and H. Cochard. 2017. Plant resistance to drought depends on timely stomatal closure. Ecol. Lett. 20: 1437-1447. doi:10.1111/ele.12851

McShea, W.J., W.M. Healy, P. Devers, T. Fearer, F.H. Koch, D. Stauffer, and J. Waldon. 2007. Forestry matters: decline of oaks will impact wildlife in hardwood forests. J. Wildl. Manage. 71: 1717-1728. doi:10.2193/2006-169

Milburn, J.A., and P.E.R. O'Malley. 1984. Freeze-induced sap absorption in *Acer pseudoplatanus*: a possible mechanism. Can. J. Bot. 62: 2101-2106. doi:10.1139/b84-285

Miller, R.B., J.T. Quirk, and D.J. Christensen. 1985. Identifying white oak logs with sodium nitrite. For. Prod. J. 35: 33-38.

Morris, H., and S. Jansen. 2016. Secondary xylem parenchyma – From classical terminology to functional traits. IAWA J. 37: 1-15. doi:10.1163/22941932-20160117

Morris, H., L. Plavcová, M. Gorai, M.M. Klepsch, M. Kotowska, H. Jochen Schenk, and S. Jansen. 2018. Vessel-associated cells in angiosperm xylem: highly specialized living cells at the symplast-apoplast boundary. Am. J. Bot. 105: 151-160. doi:10.1002/ajb2.1030

Murmanis, L. 1975. Formation of tyloses in felled *Quercus rubra* L. Wood Sci. Technol. 9: 3-14. doi:10.1007/BF00351911

Nair, V.M.G., and J.E. Kuntz. 1963. Mat formation in bur oaks infected with *Ceratocystis fagacearum*. Note No. 95. University of Wisconsin, College of Agricultural and Forest Research, Madison, WI, USA.

Naruzawa, E.S., and L. Bernier. 2014. Control of yeast-mycelium dimorphism in vitro in Dutch elm disease fungi by manipulation of specific external stimuli. Fungal Biol. 118: 872-884. doi:10.1016/j.funbio.2014.07.006

Newbanks, D., A. Bosch, and M.H. Zimmermann. 1983. Evidence for xylem dysfunction by embolization in Dutch elm disease. Phytopathology 73: 1060-1063. doi:10.1094/Phyto-73-1060

Newcombe, G., and J. Robb. 1989. The chronological development of a lipid-to-suberin response at *Verticillium* trapping sites in alfalfa. Physiol. Mol. Plant Pathol. 34: 55-73. doi:10.1016/0885-5765(89)90016-7

Nixon, K.C. 1993. Infrageneric classification of *Quercus* (Fagaceae) and typification of sectional names. Ann. Sci. For. 50: 25s-34s (Suppl.). doi:10.1051/forest:19930701

O'Brien, J.G., M.E. Mielke, D. Starkey, and J. Juzwik. 2011. How to identify, prevent, and control oak wilt. Publication No. NA-FR-01-11. United States Department of Agriculture, Forest Service, Northeastern Area State and Private Forestry, Newton Square, PA, USA. 38 pp. Available online [https://arborcareandconsulting.com/sites/arbor/files/oak_wilt_usda.pdf].

Ontario Government. 2016. Red oak – *Quercus rubra*. Forest Resources of Ontario. Available online [https://www.ontario.ca/document/forest-resources-ontario-2016/red-oak-quercus-rubra] (Accessed in August 2022).

Orlowski, M. 1991. *Mucor* dimorphism. Microbiol. Mol. Biol. Rev. 55: 234-258. doi:10.1128/mr.55.2.234-258.1991

Osterbauer, N.K., and D.W. French. 1992. Propiconazole as a treatment for oak wilt in *Quercus rubra* and *Q. ellipsoidalis*. J. Arboric. 18: 221-226. doi:10.48044/jauf.1992.044

Oswalt, S.N., W.B. Smith, P.D. Miles, and S.A. Pugh. 2019. Forest Resources of the United States, 2017: a technical document supporting the Forest Service 2020 RPA Assessment. General technical report No. WO-97. US Department of Agriculture, Forest Service, Washington, DC, USA. 237 pp. doi:10.2737/WO-GTR-97

Ouellette, G.B. 1962. Morphological characteristics of *Ceratocytis ulmi* (Buism.) C. Moreau in American elm trees. Can. J. Bot. 40: 1463-1466. doi:10.1139/b62-141

Ouellette, G.B. 1980. Occurrence of tyloses and their ultrastructural differentiation from similarly configured structures in American elm infected by *Ceratocystis ulmi*. Can. J. Bot. 58: 1056-1073. doi:10.1139/b80-130

Ouellette, G.B. 1981a. Ultrastructural cell wall modifications in secondary xylem of American elm surviving the acute stage of Dutch elm disease: fibres. Can. J. Bot. 59: 2425-2438. doi:10.1139/b81-293

Ouellette, G.B. 1981b. Ultrastructural cell wall modifications in secondary xylem of American elm surviving the acute stage of Dutch elm disease: vessel members. Can. J. Bot. 59: 2411-2424. doi:10.1139/b81-292

Panshin, A.J., and C. de Zeeuw. 1980. Textbook of wood technology: structure, identification, properties, and uses of the commercial woods of the United States and Canada. McGraw-Hill, New York, NY, USA. 722 pp.

Peacher, P.H., M.J. Weiss, and J.F. Wolf. 1975. Southward spread of oak wilt remains static. Plant Dis. Rep. 59: 303-304.

Pearce, R.B. 1996. Antimicrobial defences in the wood of living trees. New Phytol. 132: 203-233. doi:10.1111/j.1469-8137.1996.tb01842.x

Pedlar, J.H., D.W. McKenney, E. Hope, S. Reed, and J. Sweeney. 2020. Assessing the climate suitability and potential economic impacts of Oak wilt in Canada. Sci. Rep. 10: 19391. doi:10.1038/s41598-020-75549-w

Pegg, G.F. 1985. Life in a black hole – The micro-environment of the vascular pathogen. Trans. Br. Mycol. Soc. 85: 1-20. doi:10.1016/S0007-1536(85)80151-0

Piao, S., Q. Liu, A. Chen, I.A. Janssens, Y. Fu, J. Dai, L. Liu, X. Lian, M. Shen, and X. Zhu. 2019. Plant phenology and global climate change: current progresses and challenges. Glob. Change Biol. 25: 1922-1940. doi:10.1111/gcb.14619

Poiré, T., and E. Appleton. 2018. CFIA plant health surveillance update: eastern Ontario forest health review. Government of Canada, Canadian Food Inspection Agency. Available online [https://www.eomf.on.ca/media/k2/attachments/CFIA_-RFHN_Review_November_2018.pdf] (Accessed in September 2021).

Popkin, G. 2021. Forest fight. Science 374: 1184-1189. doi:10.1126/science.acx9733

Rabaey, D., S. Huysmans, F. Lens, E. Smets, and S. Jansen. 2008. Micromorphology and systematic distribution of pit membrane thickenings in Oleaceae: tori and pseudotori. IAWA J. 29: 409-424. doi:10.1163/22941932-90000195

Rioux, D., and R.P. Baayen. 1997. A suberized perimedullary reaction zone in *Populus balsamifera* novel for compartmentalization in trees. Trees 11: 389-403. doi:10.1007/PL00009682

Rioux, D., M. Blais, N. Nadeau-Thibodeau, M. Lagacé, P. DesRochers, K. Klimaszewska, and L. Bernier. 2018. First extensive microscopic study of butternut defense mechanisms following inoculation with the canker pathogen *Ophiognomonia clavigignenti-juglandacearum* reveals compartmentalization of tissue damage. Phytopathology 108: 1237-1252. doi:10.1094/PHYTO-03-18-0076-R

Rioux, D., H. Chamberland, M. Simard, and G.B. Ouellette. 1995. Suberized tyloses in trees: an ultrastructural and cytochemical study. Planta 196: 125-140. doi:10.1007/BF00193226

Rioux, D., M. Nicole, M. Simard, and G.B. Ouellette. 1998. Immunocytochemical evidence that secretion of pectin occurs during gel (gum) and tylosis formation in trees. Phytopathology 88: 494-505. doi:10.1094/PHYTO.1998.88.6.494

Rioux, D., and G.B. Ouellette. 1989. Light microscope observations of histological changes induced by *Ophiostoma ulmi* in various nonhost trees and shrubs. Can. J. Bot. 67: 2335-2351. doi:10.1139/b89-299

Rioux, D., and G.B. Ouellette. 1991a. Barrier zone formation in host and nonhost trees inoculated with *Ophiostoma ulmi*. I. Anatomy and histochemistry. Can. J. Bot. 69: 2055-2073. doi:10.1139/b91-258

Rioux, D., and G.B. Ouellette. 1991b. Barrier zone formation in host and nonhost trees inoculated with *Ophiostoma ulmi*. II. Ultrastructure. Can. J. Bot. 69: 2074-2083. doi:10.1139/b91-259

Rizzo, D.M., M. Garbelotto, J.M. Davidson, G.W. Slaughter, and S.T. Koike. 2002. *Phytophthora ramorum* as the cause of extensive mortality of *Quercus* spp. and *Lithocarpus densiflorus* in California. Plant Dis. 86: 205-214. doi:10.1094/PDIS.2002.86.3.205

Robert, E.M.R., M. Mencuccini, and J. Martínez-Vilalta. 2017. The anatomy and functioning of the xylem in oaks. Pages 261-302 *in* E. Gil-Pelegrín, J.J. Peguero-Pina, and D. Sancho-Knapik (eds.), Oaks physiological ecology. Exploring the functional diversity of genus *Quercus* L. Springer International Publishing, Cham, Switzerland. doi:10.1007/978-3-319-69099-5_8

Sachs, I.B., V.M.G. Nair, and J.E. Kuntz. 1970. Penetration and degradation of cells walls in oaks infected with *Ceratocystis fagacearum*. Phytopathology 60: 1399-1404. doi:10.1094/Phyto-60-1399

Sander, I.L., and H.N. Rosen. 1985. Oak, an American wood. Publication No. FS-247. US Department of Agriculture, Forest Service, Washington, DC, USA. 11 pp. Available online [https://www.fpl.fs.usda.gov/documnts/usda/amwood/247oak.pdf] (Accessed in September 2021).

Schmitt, U., and W. Liese. 1994. Wound tyloses in *Robinia pseudoacacia* L. IAWA J. 15: 157-160. doi:10.1163/22941932-90001357

Schoeneweiss, D.F. 1959. Xylem formation as a factor in oak wilt resistance. Phytopathology 49: 335-337.

Scholander, P.F., W. Flagg, R.J. Hock, and L. Irving. 1953. Studies on the physiology of frozen plants and animals in the Arctic. J. Cell. Physiol. 42: 1-56 (Suppl.).

Scholander, P.F., E.D. Bradstreet, E.A. Hemmingsen, and H.T. Hammel. 1965. Sap pressure in vascular plants: negative hydrostatic pressure can be measured in plants. Science 148: 339-346. doi:10.1126/science.148.3668.339

Schultz, H.R., and M.A. Matthews. 1988. Vegetative growth distribution during water deficits in *Vitis vinifera* L. Aust. J. Plant Physiol. 15: 641-656. doi:10.1071/PP9880641

Shigo, A.L. 1984. Compartmentalization: a conceptual framework for understanding how trees grow and defend themselves. Annu. Rev. Phytopathol. 22: 189-214. doi:10.1146/annurev.py.22.090184.001201

Shigo, A.L., and H.G. Marx. 1977. Compartmentalization of decay in trees. Bulletin No. 405. US Department of Agriculture, Forest Service, Washington, DC, USA. 73 pp. Available online [https://www.nrs.fs.usda.gov/pubs/misc/ne_aib405.pdf].

Shigo, A.L., and J.T. Tippett. 1981. Compartmentalization of American elm tissues infected by *Ceratocystis ulmi*. Plant Dis. 65: 715-718. doi:10.1094/PD-65-715

Sillett, S.C., and R. Van Pelt. 2000. A redwood tree whose crown may be the most complex on Earth. Pages 11-18 *in* M. Labrecque (ed.), L'Arbre 2000. Isabelle Quentin, Montréal, QC, Canada.

Sinclair, W.A., and H.H. Lyon. 2005. Oak wilt. Pages 238-239 *in* W.A. Sinclair, and H.H. Lyon (eds.), Diseases of trees and shrubs. Cornell University Press, Ithaca, NY, USA.

Skelton, R.P., L.D.L. Anderegg, J. Diaz, M.M. Kling, P. Papper, L.J. Lamarque, S. Delzon, T.E. Dawson, and D.D. Ackerly. 2021. Evolutionary relationships between drought-related traits and climate shape large hydraulic safety margins in western North American oaks. Proc. Natl. Acad. Sci. 118: e2008987118. doi:10.1073/pnas.200898711

Smith, D.R., and G.R. Stanosz. 2018. Occurrence of *Diplodia corticola*, including new oak host records, in Wisconsin, USA. For. Pathol. 48: e12427. doi:10.1111/efp.12427

Solla, A., and L. Gil. 2002. Xylem vessel diameter as a factor in resistance of *Ulmus minor* to *Ophiostoma novo-ulmi*. For. Pathol. 32: 123-134. doi:10.1046/j.1439-0329.2002.00274.x

Sperry, J.S., J.R. Donnelly, and M.T. Tyree. 1988. Seasonal occurrence of xylem embolism in sugar maple (*Acer saccharum*). Am. J. Bot. 75: 1212-1218. doi:10.1002/j.1537-2197.1988.tb08834.x

Sperry, J.S., and J.E.M. Sullivan. 1992. Xylem embolism in response to freeze-thaw cycles and water stress in ring-porous, diffuse-porous, and conifer species. Plant Physiol. 100: 605-613. doi:10.1104/pp.100.2.605

Sperry, J.S. and M.T. Tyree. 1990. Water-stress-induced xylem embolism in three species of conifers. Plant Cell Environ. 13: 427-436. doi:10.1111/j.1365-3040.1990.tb01319.x

Stein, J.D., D. Binion, and R.E. Acciavatti. 2003. Field guide to native oak species of eastern North America. US Department of Agriculture, Forest Service, Forest Health Technology Enterprise Team, Morgantown, WV, USA. 161 pp.

Steudle, E. 1995. Trees under tension. Nature 378: 663-664. doi:10.1038/378663a0

Struckmeyer, B.E., J.E. Kuntz, and A.J. Riker. 1958. Histology of certain oaks infected with the oak fungus. Phytopathology 48: 556-561.

Tainter, F.H., and F.A. Baker. 1996. Oak wilt. Pages 673-682 *in* F.H. Tainter, and F.A. Baker (eds.), Principles of forest pathology. John Wiley & Sons, New York, NY, USA. 832 pp.

Tainter, F.H., and S.W. Fraedrich. 1986. Compartmentalization of *Ceratocystis fagacearum* in Turkey oak in South Carolina. Phytopathology 76: 698-701. doi:10.1094/Phyto-76-698

Tainter, F.H., and D.L. Ham. 1983. The survival of *Ceratocystis fagacearum* in South Carolina. Eur. J. For. Pathol. 13: 102-109. doi:10.1111/j.1439-0329.1983.tb01313.x

Takahashi, S., N. Okada, and T. Nobuchi. 2015. Relationship between vessel porosity and leaf emergence pattern in ring- and diffuse-porous deciduous trees in a temperate hardwood forest. Botany 93: 31-39. doi:10.1139/cjb-2014-0129

Tchernoff, V. 1965. Methods for screening and for the rapid selection of elms for resistance to Dutch elm disease. Acta Bot. Neerl. 14: 409-452. doi:10.1111/j.1438-8677.1965.tb00204.x

The Wood Database. n.d.-a. Chestnut Oak sealed. Available online [https://www.wood-database.com/wp-content/uploads/chestnut-oak-sealed.jpg] (Accessed in August 2022).

The Wood Database. n.d.-b. Distinguishing red and white oak. Available online [https://www.wood-database.com/wood-articles/distinguishing-red-oak-from-white-oak/] (Accessed in August 2022).

Tippett, J.T., and A.L. Shigo. 1981a. Barrier zone formation: a mechanism of tree defense against vascular pathogens. IAWA J. 2: 163-168. doi:10.1163/22941932-90000724

Tippett, J.T., and A.L. Shigo. 1981b. Barriers to decay in conifer roots. Eur. J. For. Pathol. 11: 51-59. doi:10.1111/j.1439-032
9.1981.tb00070.x

Trouy, M.-C. 2015. Anatomie du bois : formation, fonctions et identification. Éditions Quae, Versailles, France. 151 pp.

Tyree, M.T., and M.H. Zimmermann. 2002. Xylem structure and the ascent of sap. Springer, Berlin, Germany. 283 pp.

US Forest Service-Forest Products Laboratory. 1921. Identification of oak woods. Technical note No. 125. Madison, WI, USA. 2 pp. Available online [https://www.fpl.fs.usda.gov/documnts/fpltn/fpltn-125.pdf].

Utsumi, Y., Y. Sano, J. Ohtani, and S. Fujikawa. 1996. Seasonal changes in the distribution of water in the outer growth rings in *Fraxinus mandshurica* var. *japonica*: a study by cryo-scanning electron microscopy. IAWA J. 17: 113-124. doi:10.1163/22941932-90001439

Van Alfen, N.K., B.D. McMillan, V. Turner, and W.M. Hess. 1983. Role of pit membranes in macromolecule-induced wilt of plants. Plant Physiol. 73: 1020-1023. doi:10.1104/pp.73.4.1020

Wheeler, E.A., P. Baas, and S. Rodgers. 2007. Variations in dicot wood anatomy: a global analysis based on the InsideWood database. IAWA J. 28: 229-258. doi:10.1163/22941932-90001638

Wheeler, E.A., C.A. LaPasha, and R.B. Miller. 1989. Wood anatomy of elm (*Ulmus*) and hackberry (*Celtis*) species native to the United States. IAWA J. 10: 5-26. doi:10.1163/22941932-90001106

Wheeler, J.K., J.S. Sperry, U.G. Hacke, and N. Hoang. 2005. Inter-vessel pitting and cavitation in woody Rosaceae and other vesselled plants: a basis for a safety versus efficiency trade-off in xylem transport. Plant Cell Environ. 28: 800-812. doi:10.1111/j.1365-3040.2005.01330.x

Wikimedia Foundation. n.d. Hyperion (tree). Wikipedia. Available online [https://en.wikipedia.org/wiki/Hyperion_(tree)] (Accessed in August 2022).

Williams, S. 1939. Secondary vascular tissues of the oaks indigenous to the United States - I. The importance of secondary xylem in delimiting *Erythrobalanus* and *Leucobalanus*. Bull. Torrey Bot. Club 66: 353-365. doi:10.2307/2481037

Woodcock, D.W. 1989. Distribution of vessel diameter in ring-porous trees. Aliso 12: 287-293. doi:10.5642/aliso.19891202.05

Zimmermann, M.H. 1979. The discovery of tylose formation by a Viennese lady in 1845. IAWA Bull. 2-3: 51-56.

Zimmermann, M.H., and A.A. Jeje. 1981. Vessel-length distribution in stems of some American woody plants. Can. J. Bot. 59: 1882-1892. doi:10.1139/b81-248

Zimmermann, M.H., and J. McDonough. 1978. Dysfunction in the flow of food. Pages 117-140 *in* J.G. Horsfall and E.B. Cowling (eds.), Plant disease: an advanced treatise. Vol III: How plants suffer from disease. Academic Press, New York, NY, USA.

Zucchino, D. 2014. 1800s-era sunken logs are now treasure; here are the men who find them. Los Angeles Times. Available online [https://www.latimes.com/nation/la-na-sinker-wood-20140713-story.html] (Accessed in July 2021).

Zwieniecki, M.A., and Holbrook, N.M. 1998. Diurnal variation in xylem hydraulic conductivity in white ash (*Fraxinus americana* L.), red maple (*Acer rubrum* L,) and red spruce (*Picea rubens* Sarg.). Plant Cell Environ. 21: 1173-1180. doi:10.1046/j.1365-3040.1998.00342.x