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Letter to the Editor

Not all lipids in xylem conduits are artefacts. A reply to Yamagishi et al.

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The paper by Yamagishi et al. (2021) represents an interesting study on the distribution and formation of incrustations on intervessel pit membranes in angiosperm wood, which have been reported in a wide range of species. The authors present clear and convincing evidence that the incrustations or coatings on pit membranes represent neutral lipids such as triacylglycerols, and are formed during dehydration of wood samples either as an artifact of drying or possibly as a process that could also occur in non-functional conduits of living plants. We are excited to see that lipids in xylem conduits were examined in detail by the authors and that novel methods such as time-of-flight secondary ion mass spectrometry were applied. We also hope that these authors or other research groups will follow up on the findings in future work.

As mentioned in the paper, lipids of pit membrane incrustations (PMIs) in dried samples cannot be compared to amphiphilic lipids found in hydrated xylem samples. However, the authors made a few omissions and unfortunate statements that could easily be misinterpreted by readers, which could lead to confusion and misinformation. Our major comment is that a clear distinction must be made between the neutral lipids studied, and polar, amphiphilic lipids such as galactolipids and phospholipids, which are more abundant in xylem of most species than neutral lipids (with the exception of fat-storing species) (Schenk et al. 2021). Unfortunately, the title and abstract of the paper do not specify that only neutral lipids were studied, which could create a false impression for readers who do not read beyond the abstract that the findings apply to polar lipids in xylem conduits as well.

There is strong and solid evidence based on careful mass spectrometry analyses and visual evidence based on confocal microscopy that most of the lipids in xylem conduits of hydrated samples are polar lipids (Schenk et al. 2017, 2018, 2021). We are well aware that earlier studies on xylem sap chemistry may have suffered from xylem sap contamination by livings cells (parenchyma or living fibres) (Schurr 1998). For this reason, it is essential in xylem sap extractions from stems to investigate contamination control samples, such as 1 ml of pure water exposed to a high-pressure-cleaned cut open stem surface for 1 minute as well as testing for potential contamination from cut cells at the other end of a stem (Schenk et al. 2017, 2021).

We disagree with the authors' statement that "... *changes in the distribution of amphiphilic lipids would still require attention because they are usually fluid at room temperature*." First, careful analyses of control samples showed that amphiphilic, polar lipids do not represent contamination artefacts of xylem sap (Schenk et al. 2017, 2021). Secondly, it is misleading to refer to amphiphilic lipids as fluid. In an aqueous environment, such lipids with hydrophilic heads and hydrophobic tails form bilayers and micelles (Marsh & King 1986). Lipid molecules can move within these structures, but a micelle is a particle, not a fluid. Because pit membranes are known to have smaller pore sizes than previously thought (Zhang et al. 2020; Kaack et al. 2020, 2021), amphiphilic lipid micelles will aggregate on and in pit membranes and typically not move across them. Unlike the neutral lipids observed in this paper, polar lipids likely originate from cytoplasmic remnants of vessel elements before these were hydrolysed (Esau et al. 1966; Schenk et al. 2021).

The paper by Yamagishi et al. (2021) provides an important advance by clarifying the nature of pit membrane coatings seen in some species and creates new questions about the origin of these neutral lipids and possibly their function if the same processes were to be found in living xylem. The findings also confirm once again that research on pit membrane structure and function must be conducted with hydrated, never-dried xylem samples, as done in recent research on polar lipids in xylem conduits and the ultrastructure of pit membranes (Schenk et al. 2017, 2018, 2021; Kotowska et al. 2020; Sorek et al. 2020; Thonglim et al. 2020; Zhang et al. 2020).

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REFERENCES

Esau K, Cheadle VI, Gill RH. 1966. Cytology of differentiating tracheary elements. II. Structures associated with cell surfaces. Am. J. Bot. 53: 765–771. DOI: 10.2307/2440178.

Kaack L, Altaner CM, Carmesin C, Diaz A, Holler M, Kranz C, Neusser G, Odstrcil M, Schenk HJ, Schmidt V, Weber M, Zhang Y, Jansen S. 2019. Function and three-dimensional structure of intervessel pit membranes in angiosperm xylem: a review. IAWA J. 40: 673–702. DOI: 10.1163/22941932-40190259.

Kaack L, Weber M, Isasa E, Karimi Z, Li S, Pereira L, Trabi CL, Zhang Y, Schenk HJ, Schuldt B, Schmidt V, Jansen S. 2021. Pore constrictions in intervessel pit membranes provide a mechanistic explanation for xylem embolism resistance in angiosperms. New Phytol. 230: 1829–1843. DOI: 10.1111/nph.17282.

Kotowska MM, Thom R, Zhang Y, Schenk HJ, Jansen S. 2020. Within-tree variability and sample storage effects of bordered pit membranes in xylem of *Acer pseudoplatanus*. Trees 34: 61–71. DOI: 10.1007/s00468-019-01897-4.

Marsh D, King MD. 1986. Prediction of the critical micelle concentrations of mono- and di-acyl phospholipids. Chem. Phys. Lipids 42: 271–277. DOI: 10.1016/0009-3084(86)90086-1.

Schenk HJ, Espino S, Romo DM, Nima N, Do AYT, Michaud JM, Papahadjopoulos-Sternberg B, Yang J, Zuo YY, Steppe K, Jansen S. 2017. Xylem surfactants introduce a new element to the cohesion-tension theory. Plant Physiol. 173: 1177–1196. DOI: 10.1104/pp.16.01039.

Schenk HJ, Espino S, Rich-Cavazos SM, Jansen S. 2018. From the sap's perspective: the nature of vessel surfaces in angiosperm xylem. Am. J. Bot. 105: 174–187. DOI: 10.1002/ajb2.1034.

Schenk HJ, Michaud JM, Espino S, Melendres T, Roth MR, Welti R, Kaack L, Jansen S. 2021. Lipids in xylem sap of woody plants across the angiosperm phylogeny. Plant J. 105: 1477–1494. DOI: 10.1111/tpj.15125.

Schurr U. 1998. Xylem sap sampling – new approaches to an old topic. Trends Plant Sci. 3: 293–298. DOI: 10.1016/S1360-1385(98)01275-8.

Sorek Y, Grinshtein S, Netzer Y, Shtein I, Jansen S, Hochberg U. 2020. An increase in xylem embolism resistance of grapevine leaves during the growing season is coordinated with stomatal regulation, turgor loss point, and intervessel pit membranes. New Phytol. 229: 1955–1969. DOI: 10.1111/nph.17025.

Thonglim A, Delzon S, Larter M, Karami O, Rahimi A, Offringa R, Keurentjes JJB, Balazadeh S, Smets E, Lens F. 2020. Intervessel pit membrane thickness best explains variation in embolism resistance amongst stems of *Arabidopsis thaliana* accessions. Ann. Bot. 128: 171–182. DOI: 10.1093/aob/mcaa196.

Yamagishi S, Shigetomi K, Fujiyasu S, Aoki D, Uno T, Fukushima K, Sano Y. 2021. Artifactual lipid coatings on intervessel pit membranes in dried xylem tissues of some angiosperms. IAWA J. 42: https://doi.org/10.1163/22941932-bja10060.

Zhang Y, Matei T, Kotowska M, Weber M, Klepsch MM, Schenk HJ, Walter P, Schmidt V, Jansen S. 2020. High porosity with tiny pore constrictions and unbending pathways characterise the 3D structure of intervessel pit membranes in angiosperm xylem. Plant Cell Environ. 43: 116–130. DOI: 10.1111/pce.13654.