



Title: **“Functional xylem in the post-veraison grape berry”**

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In this paper, the authors challenge the idea that the xylem – the bundle of vessels that conduct water and solutes from the roots to the berry – becomes disrupted after veraison. Instead, they prove that these conduits are alive and well, even after fruit ripening.

- The **xylem** are vascular bundles formed by dead cells that transport sap from the roots to the rest of the plant. In grapes – as in tomato, kiwi, or apples – there is a clear reduction in the amount of water that enters the fruit via the xylem as the fruit matures. This water cessation happens during berry growth stage III, and coincides with an increase in sugar in the berry. Because of this phenomenon, post-veraison growth and ripening of the berry is entirely dependent upon what enters the berry via the phloem. The **phloem** are a second type of vascular bundles, this time formed by living cells, that transport photosynthates between the different parts of the plant, mainly between the leaves and the berries.
- Researchers up until now believed that, from veraison on, the grape xylem started to lose its structure and functionality. They based their belief on 3 observations: 1) When they dipped pedicels attached to berries into a dye, the dye was taken up into the berry xylem if the berry was immature, but it was not taken up if the berry had gone through veraison. 2) Photomicrographs of the xylem vessels showed stretched-out helical thickenings, or gaps, which were interpreted as a disruption in xylem continuity. Finally, 3) when flow rates were measured for each pathway, they indicated a clear change of water influx from predominantly xylem to predominantly phloem just near veraison.
- The current authors believed that the fact that the xylem flow was reduced did not necessarily imply that a physical blockage existed. Instead, the explanation might be that there was not enough water pressure – tension, or hydrostatic gradient- to drive the flow. This research was an attempt to prove whether this was true.
- When the authors compared 2 methods of dye uptake, 1) *passive dye infusion*, versus 2) *pressure membrane method*, they found that, when they used the passive infusion, pre-veraison berries soaked up the dye whereas post-veraison berries did not. But when the authors gave the water in the vessels “a little push” with the pressure membrane method, both pre- and post-veraison berries took up the dye. This meant that, **even after veraison the xylem conduits were functional**, not blocked or broken as previously thought.
- The pressure membrane method consisted of using gas to push the dye through a very special membrane, something that the authors feared may have created some sort of artifact. To test this, they repeated the experience using yet another dye infusion technique: 3) the *wick method*. In this method, a paper pad is placed between the dye and the berry, which facilitates the absorption of dye without the

need for gas pressure. When the authors compared pre- and post-veraison berries, they found the same results they had found with the pressure membrane method: **post-veraison berries absorbed the dye even in the absence of a gas push**. That meant there had been no artifact due to air bubbles. It also meant that embolisms –air bubbles- in the xylem were not responsible for the reduced flow after veraison. (Embolisms would have been expected to be removed with the pressure membrane method but not with the wick method.)

- To test whether the dye was indeed entering through the xylem and not through some other unpredicted pathway, the authors next compared submerging whole cut berries into the dye versus submerging just the pedicels. The result was that the amount of dye taken up was the same in both cases. This meant that all the dye entering the berry was doing so through the xylem bundles of the pedicel.
- Finally, the authors further tested the hypothesis put forward by others that the xylem may have broken during normal growth due to excessive elongation of its vessel units (called tracheids). If this were true, and the authors could find a way of restricting berry growth, then the break down would be prevented, and post-veraison berries would be able to continue taking up dye by passive diffusion. So they placed each immature berry in a solid box of 1x1x1 cm to prevent expansion (think “foot binding”!). Then they compared the ability of these berries to take up the dye. They found that the berries that had their growth restricted showed the same pattern as the control berries. This meant that, even in the absence of xylem elongation due to growth, xylem flow still ceased after veraison.

So with this work, the authors gathered enough evidence to prove that when we see the xylem flow stop after veraison, it is not because the “hose” (*xylem*) is broken, but because the “pump” (*hydrostatic pressure*) is set on “low”. Even though the application of this study might not seem immediate, being able to document the intact nature of the post-veraison xylem is an important scientific breakthrough. Through elegant experimental design, the authors not only were able to rectify what was originally accepted, but also to prove that the post-veraison berry has a very low water potential. This will bring us closer to understanding complex processes such as “water berry”, “berry shrivel”, and the most complex of all: the process of “ripening” itself. Make sure you check out the beautiful color plates in Fig.2 of the original text.

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