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Title: **"Solute transport into Shiraz berries during development and late-ripening shrinkage"**

By: Suzy Rogiers, Dennis Greer, Jo Hatfield, Beverly Orchard, and Markus Keller

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Syrah (Shiraz) shrinkage has occurred in south-west Australia in six of the past seven vintages, with losses of up to 25% each harvest. In this article the authors study the sap flow within the vascular system of Syrah grapes, to try to understand what causes shriveling. *[Reviewer's note: We wish to clarify that the "shrinkage" the authors refer to in this paper is considered different from the "cluster shriveling" observed in California, which also involves the drying up of the rachis.]*

• During fruit development, the vascular tissues contribute to solute and water accumulation. We know that, after veraison, phloem flow predominates, and the current hypothesis is that it slows down during the late stages of berry ripening. Xylem flow also contributes to the growth of the berries, mainly before veraison, but it is unclear at what stage it stops. The authors wanted to find out whether changes in vascular flow could be causing late-ripening shrinkage.

• Calcium (Ca) has low mobility in the phloem, and its import into fruit is almost exclusively through the xylem. We therefore call Ca a "**xylem element**". Potassium (K) has mobility in both xylem and phloem, but it is 10 times more concentrated in the phloem than in the xylem. We can call K a "**phloem element**". The authors reasoned that the accumulation of each of these elements into the berry should give an indication of xylem and phloem functionality. So they tracked Ca and K in Syrah from early berry development to shriveling (Wagga Wagga, New South Wales, Australia).

• For two seasons (2000, 2001) the authors collected clusters from two Syrah sites, 3 times a week, from bloom to harvest, and –after thawing the frozen samples- measured the following: berry fresh weight, berry dry weight, total soluble solids, and berry K and Ca (using atomic emission spectrometry). They also monitored potted Syrah plants and measured: berry diameter, water potential, and solute potential. (Solute potential measures the ability of the solutes in the berry to drive water from the vine to the berries due the osmotic pressure, and was calculated here by formula after knowing the weight of water and the weight of solutes in the berry).

• **Berry weight and sugar accumulation**. There were 3 years of data available for this parameter (2000-2002).There were many fluctuations in berry size, berry weight loss, sugar accumulation, and harvest date (measured as days after bloom) from block to block and year to year. The authors don't show us all the data, however. In all three years, berry fresh weight reached a maximum before the commercial harvest took place (13% loss over 16 days before harvest, in 2001). In general, sugar accumulation per berry remained constant, or increased slightly, after reaching maximum weight. But because of berry water loss, total soluble solids (or Brix) increased every year as harvest approached (for example, from 22.5 to 24°Brix in 2001; from 21.9 to 26.6 in 2002).

• **Ca and K accumulation.** As mentioned earlier, the authors use these elements to indicate xylem (Ca) and phloem (K) flows. Calcium accumulation ("xylem element") was strong before veraison. Two weeks after veraison, it either remained stable (site A), or continued increasing until harvest (site B). As for potassium ("phloem element"), it moved into the berry both before and after veraison, but post-veraison accumulation was much stronger. However, when the berry reached its maximum weight (which, as we have seen, took place before commercial harvest), the rate of K accumulation declined.

• What does this mean in terms of xylem and phloem flows? The authors emphasize that, regardless of whether calcium remained constant (site A) or increased (site B), the berries continued to shrink well after veraison in both sites. This means that the occurrence of **shriveling does not depend on xylem flow**. On the other hand, the decline in K accumulation, as well as that of sugars and dry matter, suggests to the authors that the **shriveling may be due to a decrease in phloem flow**. What it is not clear is how this shriveling (loss of water) took place despite the strong negative water potentials and negative solute potentials that they measured in the post-veraison berry.

• To further study the contributions of phloem and xylem to the berry, the authors performed an interesting experiment. They either girdled or excised the berry stems (or pedicels) and looked at how this affected the above parameters (berry weight, sugar, Ca and K accumulation). When they **girdled** the pedicels, what they were doing was eliminating the phloem flow. They did that by scraping a ring of outer pedicel tissue away from the pedicel. When they completely **excised** the pedicels, obviously what they were doing was eliminating both phloem and xylem. To perform the "excision", they pulled the berries away from the clusters, and after sealing the pedicels with silicone (to prevent water loss), they resuspended them in the cluster with the aide of cotton thread! This way the authors could compare measurement on both types of berries -girdled and excised- while under the same microclimate parameters (light, temperature, evaporation, etc).

• They found that girdling the berry pedicel caused an immediate cessation in sugar accumulation. Girdling also inhibited color formation, consistent with pigment formation requiring sugars to be completed. Additionally, K accumulation was inhibited. And finally, berry growth was arrested, reinforcing the role of the phloem as the main route for water and solute uptake into the berry after veraison. However, some water appeared to still be entering the post-veraison berry through the xylem, judging by the completely excised berries (no phloem, no xylem) being smaller and having lost more water than the girdled berries (no phloem, but xylem intact).

My only comment on this paper is that the authors are making the assumption that Ca and K uptake from the soil remain constant throughout the season, and so, that their accumulation in the berry is a reflection of changes in sap flow, not of changes of concentration of these elements in the sap. But do we really know that? It would also be interesting to study next how the flow patterns in Syrah compare with those of varieties which do not shrivel. Does phloem flow also decrease during late ripening of, say, Cabernet?

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