## Aphid reproduction is modulated by phloem ascorbic acid – novel targets for pest control?

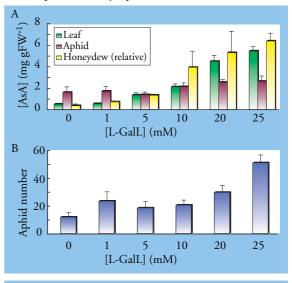
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L-Ascorbic acid (AsA; Vit C) is the most abundant soluble antioxidant found in plants and is also an essential nutrient for humans and a few other animals. Together with flavonoids, polyphenolics and lipophilic compounds such as  $\alpha$ -tocopherol (vitamin E), AsA contributes to the overall intake of free radical scavengers in the human diet. Diets rich in fruit and vegetables are associated with decreased risk of certain cancers and cardiovascular diseases and this has been attributed to the high concentrations of antioxidants in such foods. Many of the plant foods consumed by humans are sink organs (e.g. fruits, tubers), non-photosynthetic tissues that receive carbohydrates and other compounds from leaves via the phloem, the transport tissue of the plant. As part of our ongoing investigations into factors contributing towards AsA accumulation in these organs, we undertook a number of studies to investigate the contribution of AsA delivered via the phloem<sup>1,2</sup>. Work undertaken at SCRI and in other laboratories demonstrated that AsA was a major component of the phloem sap and hence transport of AsA from leaves to fruits or tubers was a mechanism by which the compound may accumulate in these organs. An additional implication of our findings was that phloem feeding insects such as aphids would be exposed to a diet rich in AsA. This led us to consider what impact phloem AsA might have on phloem feeding insects.



Figure 1 Aphid cage used in this study. The leaf petiole is placed into the reservoir shown at the foot of the image.

Aphids are major world pests on many food and commodity crops. The peach-potato aphid (*Myzus persicae*) is a pest on many crops worldwide and is of particular relevance to the UK potato industry as it represents the major vector for a number of viral diseases affecting the seed crop. We therefore chose to examine the role of AsA in modulating the reproductive success of M. persicae colonies grown on potato leaves. Single potato leaves were transferred to small aphid cages and the leaf petiole passed through the bottom of the cage into a reservoir (Fig. 1). The reservoir contained either water or solutions of L-galactono-1,4-lactone (L-GalL) the immediate precursor of AsA in plants. A single one-day-old nymph was transferred to the potato leaf and allowed to develop for 15 days. At the end of the culture period, the nymph had matured to an adult and



**Figure 2** Increasing concentrations of L-galactono-1,4-lactone in leaf reservoir buffer result in increased AsA concentrations in leaves and aphids and in the capacity of aphids to grow and reproduce.

Panel A: The x-axis shows the concentration of L-GalL in the reservoir buffer and the y-axis shows the concentration of AsA found in leaves (mg gFW<sup>-1</sup>), aphids (mg gFW<sup>-1</sup>) or aphid honeydew (relative amount). Bars

represent mean values  $\pm$  SE (n=10).

Panel B: The x-axis shows the concentration of L-GalL in the reservoir buffer and the y-axis shows the number of aphids recovered 15 days after inoculation of leaves with a single one-day-old nymph. Bars represent mean values  $\pm$ SE (n=10).

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had started reproducing. The number of aphids present was recorded and the AsA content of leaves, aphids and aphid honeydew (material secreted by the aphid after nutrient absorption) was determined. Addition of L-GalL to the reservoir resulted in a dose-dependent increase in the leaf AsA. Increased leaf AsA concentration resulted in an increased AsA concentration in both aphids and aphid honeydew (Fig. 2A). This demonstrates that there was increased availability of AsA to aphids from the phloem. Furthermore, increasing the AsA concentration in potato leaves had a dramatic effect on the number of aphids recovered at the end of the experiment rising five-fold between the lowest and highest leaf AsA concentrations (Fig. 2B). These data show for the first time that the ability of aphids to grow and reproduce on potato is enhanced when higher concentrations of AsA are present in the phloem.

In insects AsA has been implicated in a number of physiological functions however, despite many years of research, there is still no consensus regarding the capacity of insects to synthesise AsA with contradictory reports regarding insect AsA requirements. Our data suggest that when feeding directly from plants the quantity of AsA present in the phloem may be insufficient to support maximal growth and reproduction in *M. persicae*. However, there was a close correlation between leaf AsA concentration and the amount of AsA secreted in aphid honeydew suggesting that surplus AsA was available to aphids even at the lowest leaf AsA

concentrations. An alternative mechanism by which AsA could modulate aphid success is through the removal of reactive oxygen species that may be present in the phloem and act as a deterrent to aphid feeding. These compounds are highly reactive and may damage the aphid causing the insect to stop feeding to allow recovery. Due to its antioxidant properties, increased phloem AsA would reduce the prevalence of free radicals. In turn, this could allow the aphids on AsA rich diets to feed for longer periods and therefore absorb more nutrients over the same time period as the control insects. The net result would be an increase in reproductive potential due to a shorter development period, increased availability of nutrients for reproduction or a combination of the two.

Current work is aimed at further understanding the mechanism by which phloem AsA modulates aphid success. A thorough understanding of the interaction at the biochemical, molecular and physiological levels will provide novel targets for breeding aphid resistant cultivars and for the development of novel pesticides.

## References

<sup>&</sup>lt;sup>1</sup> Hancock, R.D., McRae, D., Haupt, S. & Viola, R. (2003). *BMC Plant Biology* **3**, 7.

<sup>&</sup>lt;sup>2</sup> Tedone, L., Hancock, R.D., Alberino, S., Haupt, S. & Viola, R. (2004). *BMC Plant Biology* **4**, 16.