THE ROLE OF HONEY BEES IN THE SPREAD OF AVOCADO SUNBLOTCH VIROID (ASBVd)

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This article has been published as:
John M.K. Roberts, Anna E.C. Jooste, Lara-Simone Pretorius, and Andrew D.W. Geering. Surveillance for Avocado Sunblotch Viroid Utilizing the European Honey Bee (*Apis mellifera*), *Phytopathology*® 2023 113:3, 559-566, https://doi.org/10.1094/PHYTO-08-22-0295-R
This SAAGA Yearbook article is a summary of the original article.

INTRODUCTION

Avocado sunblotch disease (ASBD), caused by the avocado sunblotch viroid (ASBVd), is one of the most important diseases of avocado worldwide that affects yield and quality. Typical symptoms are found on leaves, fruit, and bark of the tree. However, some trees do not display any visible symptoms and these are referred to as symptomless carrier trees. These symptomless carrier trees are currently the main concern for the avocado industry. To understand the spread of ASBVd transmission in orchards, the involvement of honeybees, as potential carriers of pollen from infected to healthy trees, was studied. Pollen and bees collected from hives were tested for the presence of ASBVd and the implications thereof will be discussed. In the published article, the use of honey bees for ASBVd surveillance was discussed in more detail.

MATERIALS AND METHODS

Bee and pollen samples were collected from pollination hives in two ASBVd orchard locations, one in Australia where only four trees in a block were known to be infected, and a second in South Africa where the estimated incidence of infection was 10%. In this article summary the focus will be on the results from the South African study.

Samples of stored pollen and adult worker bees were collected in October 2020 from managed bee hives at two avocado orchards in the province of KwaZulu-Natal of South Africa (Fig. 1). Hive sites SA1, SA2, and SA3 were at one farm and SA4 was at the second farm, 37 km away (see Fig. 3 in the original article). In addition, anthers were carefully removed from the flowers of trees using forceps and transferred to small Petri dishes, sealed with Parafilm and cold stored until use. South African leaf, bee, and pollen samples were extracted using a dsRNA method (Luttig and Manicom, 1999) from 400 mg of starting material. Pollen samples were weighed and mixed with the extraction buffer using the required weight:buffer ratio. Whole bee samples were macerated and used for dsRNA extraction. Pollination hives were also positioned underneath infected trees in 2021 (see Fig. 4 in the original article).

Figure 1: Sample collection from bee hives.

RESULTS AND DISCUSSION

ASBVd was detected consistently in pollen and bees from pollination hives at four South African orchard sites in 2020. Detection was similar in pollen and bees at each site, except for SA4 where there was no ASBVd detection in any bee sample (see Fig. 6 in the original article). The prevalence of ASBVd-infected trees at SA4 was unknown but was presumed low because the site belongs to a certified nursery that undergoes regular indexing of orchards. ASBVd levels in hive pollen at SA1 were significantly higher than other sites ($p = 0.0112 – 0.0001$), reflecting the higher prevalence of ASBVd-infected trees within 100-200 m from these hives (see Fig. 6 in the original article).

Pollination hives positioned underneath infected trees in 2021 were also tested and showed a significant difference between pollen and bee samples ($p = 0.017$). The position of the hives that were placed under infected trees are shown in Figure 4 in the original article. The spatial distribution of positive plants increased in the 2021 season. All but one pollen sample was ASBVd-positive whereas only three corresponding bee samples were ASBVd-positive. Testing of flowers from each tree confirmed high ASBVd levels in all trees, except for one sample testing negative, and all trees had adjacent ASBVd-positive trees.

CONCLUSION

Results showed that bees can carry ASBVd from pollen of symptomless carrier trees to bee hives. Although transmission of ASBVd via pollen is known to be relatively low (1.8% and 3.125% according to Desjardins *et al.*, 1979), this is a concern as it represents yet another avenue for transmission of the disease. When a symptomatic fruit is detected on a tree and occurs as single infected fruit, it is most likely that the infection derived from pollen transmission. The rest of the tree will remain negative and only the fruit will be infected. These trees should be marked and monitored over time. Symptomatic fruit that originated from infected pollen should be removed from the tree to ensure that these fruit are not used as seed source.

Trees showing fruit symptoms should be marked and the fruit should not be used as seed source. A tree with only a few symptomatic fruit was probably infected through pollen and the rest of the tree will remain healthy. These trees will test negative with molecular detection methods. When trees with fruit symptoms are detected, the probability of finding symptomless carrier trees in close proximity are high. Systematic testing of orchards is recommended. In the published paper, biomonitoring with honey bees, particularly in combination with high-throughput sequencing, was discussed as a powerful complementary strategy to existing plant biosecurity efforts. In the South African context, where ASBVd is an established disease, the monitoring of bee hives will only be effective in newly established avocado growing regions.

Acknowledgements

This project has been funded by Hort Innovation, using the avocado research and development levy and contributions from the Australian Government under Project AV18007 - Avocado sunblotch viroid survey. Co-funding was received from the South African Avocado Growers' Association (SAAGA).

REFERENCES

DESJARDINS, P.R., DRAKE, R.J., ATKINS, E.L. & BERGH, B.O. 1979. Pollen transmission of avocado sunblotch virus experimentally demonstrated. *Calif. Agric.* 33:14-15.

LUTTIG, M. & MANICOM, B. 1999. Application of a highly sensitive avocado sunblotch viroid indexing method. *SAAGA Yearb.* 22: 55-60.