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Canker and wood rot pathogens in young apple trees and propagation material

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Over the past nine years, occurrences of dieback on young apple trees have been reported shortly after orchard establishment. For instance, between 2010 and 2016 diseased trees representing 60 commercial apple orchards in the Western Cape were sent to the Disease Clinic at the Department of Plant Pathology for analyses. These trees typically had constricted cankers, well-developed root systems (Fig. 1) and dark-brown streaking in the vascular tissue. The symptoms indicated that replant is not the cause of the death of these trees but rather canker and dieback. An in-depth investigation into the reasons for the deaths of these newly established trees was launched. With a specific emphasis on assessing the phytosanitary status of nursery apple trees, as well as its propagation material. The pathogens causing latent infections in these plant materials were compared to those being expressed in young commercial trees.

What do we do?

Plant material was sampled from one-year-old commercial apple orchards showing signs of dieback, certified apple nursery trees, as well as scion and rootstock plant material used during propagation.

The sampling strategy has been summarised in Table 1. From the 13 commercial orchards a soil sample was collected to analyse physiochemical characteristics. Isolations were made from symptomatic vascular discolouration.

In the case of green scion shoots used for budding, buds were removed to investigate possible latent infections. From asymptomatic rootstock shoots isolations were made by cutting disks along the shoot. Additionally isolations were also made from trees of one-year-old nursery blocks that exhibited cankers on the rootstock shoot.

Table 1. Different apple plant material types collected in the Western Cape to assess for canker and wood rotting fungi

Plant Material	Total # plants	Total # units	Number of cultivars	Plants sampled per orchards	Type of plant material isolated per tree
One-year-old commercial orchards	130	13 orchards	N/A	ten trees per orchard	Cankerous section on tree
Certified nursery trees	480	four nurseries	three rootstocks	160 per rootstock	Wounds on scion shoot Wounds on rootstock Pruning wound on rootstock Bud union
Scion mother blocks	310	18 blocks	three scions	15-20 trees per orchard block	Canker Pruning wound Green scion shoot
Rootstock mother blocks Layer blocks one-year-old nursery shoots	405 87	nine layer blocks two blocks	three rootstocks two rootstocks	45 shoots per block 45 or 42 shoots	Asymptomatic shoots Cankerous shoot

Figure 1. One-year-old commercial apple trees with canker symptoms and well-developed root systems

Fungal identification and pathogenicity testing

Fungal species were identified based on morphological and molecular techniques. Species not yet associated with dieback on apple trees were selected to undergo pathogenicity testing. Pathogenicity trials were conducted with 39 species and an agar control onto two-year-old branches of 14-year-old Golden Delicious trees. After four months, inoculated branches were removed from the orchards. The internal lesion lengths were measured and isolations were made to confirm fungal species identity.

What did we find?

Forty-five fungal species associated with canker and wood rot on apple trees were identified from different plant material. All the species of the pathogenicity trials had significantly longer lesions compared to the agar control, indicating that the reported fungal species have the potential to cause dieback of apple trees. Similar fungal species were found causing latent infections in the propagation material and nursery trees, as what was expressed as cankers on one-year-old trees and older mother block trees. Infections were found in all plant material types. Most importantly, latent infections were found in 65% of investigated certified nursery trees, 21% of rooted rootstock cuttings from layer blocks and 5% of green scion shoots used for budding. Infected rooted rootstock cuttings and scion shoots play a direct role of infection for the nursery trees during propagation, while aerial inoculum present at the time of budding and pruning back plays an important indirect inoculum for infection.

The five fungal species causing the highest number of cankers and dieback on young commercial apple trees were also the most frequently encountered fungal species causing latent infection in nursery trees. These species are considered the most important in this study and were *Didymosphaeria rubi-ulmifolii* sensu lato, *Diplodia seriata, Schizophyllum commune, Didymella pomorum* and *Coniochaeta fasciculata. Didymosphaeria rubi-ulmifolii* sensu lato was the dominant fungus in nurseries and young commercial orchards. This species is lesser-known as a canker causing pathogen but has proved to cause significant lesion in our pathogenicity trials, as well as in a trial conducted on apple and pear trees (Cloete et al., 2011). In Ethiopia this pathogen has also caused the death of 13 928 young apple trees (57% of the orchard). Very little is known regarding the biology of this pathogen, therefore further investigation is necessary.

Scion mother blocks

Several fruiting bodies were found on cankers, as well as dead trees (Fig. 2) in these orchards, which not only provide aerial inoculum for the mother block, but also to orchards and nurseries in close proximity. Cultivar A had higher infection levels compared to cultivar B, but the average age of the orchards of cultivar A was 20 years, whereas cultivar B was eight years old. The difference of age would rather account for the difference in infection level since older trees have been pruned more, accumulating more wounds on the trees and being exposed to aerial inoculum for longer. This highlights the importance of using

Figure 2. Fruiting structures of wood rot pathogens identified in apple scion mother block orchards

younger scion mother blocks to obtain scion buds. Low infection levels were observed from the new green shoots. However, this study does prove that infection can occur on newly grafted plants through budding.

Rootstock mother blocks

There were no differences in infection levels between the rootstock cultivars. Rooted rootstock cuttings proved to be another source of direct inoculum. The latent infection found in the rooted rootstock cuttings can be expressed when experiencing stress during planting. This is identical to the canker development in one-year-old nursery trees observed (Fig. 3). Fruiting bodies removed from these cankers confirmed the pathogen that caused the dieback on the young tree. These fruiting bodies provide aerial inoculum during budding and pruning back of the rootstock.

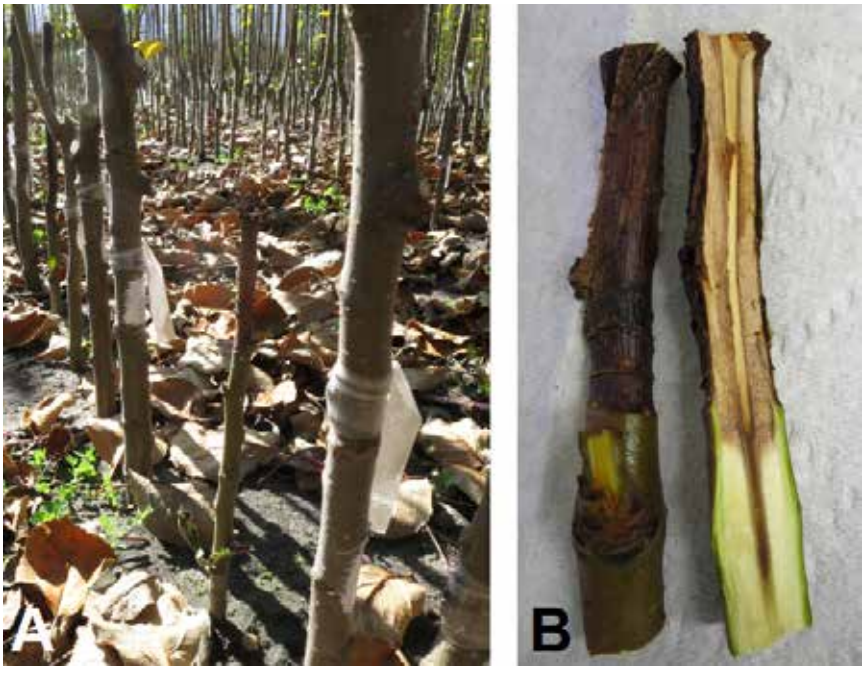

Figure 3. Left: Cankerous one-year-old nursery trees (A, B) detected between recently budded rootstock plants (A)

Certified nursery trees

Certified nursery trees did not exhibit any external symptoms, though internal vascular discolouration was frequently observed especially at the bud union or pruning wound on the rootstock (Fig. 4). Overall high levels of infection were obtained from nursery trees with the highest infection obtained from the bud union and pruning wound on the rootstock (Table 2). Therefore, these are important sites to prevent infection, since 19% of one-year-old trees from commercial orchards developed cankers from these areas. Infection in the different nurseries and rootstock cultivars were similar suggesting that the problem is not restricted to a specific rootstock or nursery.

Table 2. Infection of different plant parts isolated from nursery trees

Plant Part	Infection (%)[a]
Bud union	38a
Pruning wound	31ab
Scion	20ab
Rootstock	16c

[a] Means followed by different letters are significantly different at P <0.05

Figure 4. Dark brown vascular discolouration (A) and white soft rot symptoms originating from the pruning wound and bud union of external asymptomatic nursery apple trees

Figure 5. External (A, B) and internal canker symptoms (B, C) exhibited on the scion shoot of one-year-old commercial apple trees. Multiple pathogens were isolated from various parts of the same tree (C).

One-year-old commercial orchards

Cankers formed on four different parts of the tree specifically on the 1) scion shoot, 2) pruning wound found on the rootstock, 3) the rootstock and 4) from the bud union. The majority of the cankers formed on the scion shoot (Fig. 5A, B) and infection most likely occurred during the removal of the lateral shoots from the main stem. In many cases, more than one pathogen was isolated from the same tree (Fig. 5C). Fruiting bodies were present on the cankers which belonged to pathogens that were isolated from both the nursery trees and propagation material. The majority of the orchards were established on suboptimal soils, resulting in either a mineral deficiency or toxicity stress for the tree.

What can we do?

Our results indicated that nursery trees can harbour latent infections of canker and wood rot pathogens. Another study artificially inoculated mature apple trees with a canker pathogen and the trees only exhibited typical dieback symptoms after two years (Fujita et al., 1988). In general, canker and wood rot symptoms require time to develop, which explains why no symptoms were observed in the nursery and only developed after establishment.

Stress management

Despite high infection levels in apple nursery trees, apple producers luckily do not detect matching dieback levels or death of trees in the newly established orchards. This is because canker pathogens are opportunistic and will only display symptoms once the tree experiences stress. Therefore, it is important to ensure that stress factors are kept to the minimum, especially during lifting, storage and establishment of the apple trees. Abiotic stresses that could influence disease