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Spread and control of pistachio dieback in Australia

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SUMMARY – The probable causal pathogen of bacterial dieback of pistachios in Australia has been tentatively identified as *Xanthomonas translucens*. Bacterial dieback of pistachio affects orchards across the growing region of south-eastern Australia. Symptoms include decline, dieback, internal staining, trunk and limb lesions and can lead to tree death. *Xanthomonas* has been recovered from pruning tools after pruning infected orchard trees. Experiments were conducted to determine possible pathogen entry points, mechanisms of spread and to assess control strategies. *Xanthomonas* was recovered from the leaves, shoots and trunk of trees inoculated by placing drops of bacterial suspension on pruning wounds on one year-old shoots of potted trees. Copper sprays did not prevent infection when applied immediately after inoculation of pruning wounds in the glasshouse.

Key words: *Xanthomonas*, pistachio, dieback, pruning.

RESUME *– "Dissémination et contrôle du dépérissement du pistachier en Australie".* Xanthomonas translucens *a été identifié provisoirement comme étant la cause pathogène probable du dépérissement d'origine bactérienne des pistachiers en Australie. Le dépérissement des pistachiers d'origine bactérienne touche la région de culture du sud-est de l'Australie. Les symptômes en sont le déclin, le dépérissement, la pourriture intérieure, des lésions du tronc et des branches et peuvent entraîner la mort de l'arbre. Des* Xanthomonas *ont été récupérés sur des outillages utilisés pour la taille après que des arbres de vergers infectés aient été taillés. Des expériences ont été menées pour déterminer la cause pathogène possible, les mécanismes de propagation et pour mettre au point des stratégies de contrôle. Des* Xanthomonas *ont été récupérés sur des pousses, des feuilles et des troncs d'arbres inoculés par des gouttes de suspension bactérienne mises sur les plaies causées suite à la taille de jeunes boutures. La pulvérisation de cuivre en serre, effectuée immédiatement après l'inoculation sur les plaies causées par la taille, n'a pas empêché l'infection.*

Mots-clés : Xanthomonas, *pistachiers*, *dépérissement, taille.*

Introduction

Pistachio dieback is a serious disease in Australia caused by *Xanthomonas translucens* and results in yield and tree loss (Facelli *et al.,* 2001). Symptoms were first observed in the mid-1990s and coincided with the production of the first commercial crops. Symptoms include dieback of twigs and branches, excessive production of resin, black, sooty lesions on the trunk and major branches and staining in the conductive tissue. No lesions are observed on the leaves or fruit. More than 40% of Australia's pistachio plantings are affected by dieback with affected orchards having 10-20% symptomatic trees. Little is known about the mechanisms of spread, pathogen entry points and movement of the pathogen through the tree. Currently there are no control methods for pistachio dieback. We report here preliminary results of studies into the epidemiology of pistachio dieback.

Mapping of symptoms in infected orchards indicated that trees with symptoms tend to be distributed along rows suggesting that along-row cultural practices may be involved in spread (Edwards *et al.,* 1998). Transmission via pruning tools may be one of the mechanisms for spread of the pathogen in orchards. We sampled pruning tools for the presence of *Xanthomonas* after pruning infected trees.

Pruning causes wounds to trees that may then become entry sites for pathogens. Pruning wounds made on potted trees were inoculated to determine if they are possible entry points and the movement of the pathogen in the tree was then assessed.

Copper sprays have been used to control *Xanthomonas* diseases in other crops including walnut (Olsen *et al.,* 1976) tomato and pepper (Cooksey, 1990). Copper was applied to pruning wounds immediately after inoculation in an attempt to prevent infection.

Materials and methods

Pruning cuts were made on infected trees, growing in a commercial orchard, using long handled secateurs (loppers). The blades of the loppers were washed with sterile distilled water and the wash water plated onto growth media. Plates were incubated and the growth of *Xanthomonas* colonies recorded. Samples of air in the orchard were taken with a Merck MAS -100 Air Sampler.

Matched pairs of dormant potted *Pistacia vera* var. 'Sirora' trees were inoculated with *Xanthomonas* by cutting one-year-old twigs and placing inoculum onto the cut surface. Trees received either a suspension of *Xanthomonas* or sterile distilled water (SDW). The inoculated trees were maintained in a glasshouse and periodically five pairs were destructively assessed to determine the presence and location of *Xanthomonas*.

Dormant potted 'Sirora' trees were inoculated as above with either buffer or *Xanthomonas* suspended in buffer. Copper hydroxide was applied to the wounds either immediately after or 8 hours after the bacteria. Trees were maintained in the glasshouse for 6 months and then sampled to determine the presence or absence of *Xanthomonas*.

Results and discussion

Xanthomonas was recovered from 37% of pruning tools sampled. No *Xanthomonas* was detected in air samples from the orchard. There is a reasonable chance that after pruning infected trees, the bacteria are present on the pruning tools. Further studies need to be undertaken to determine if the inoculum levels present on the tools will cause infection when these tools are used to prune uninfected trees.

Two months after inoculation, *Xanthomonas* was recovered from the inoculation points, new shoots growing from buds near the inoculation points and the petioles of leaves on these shoots. *Xanthomonas* was also detected in the inoculated branches, and the trunk above and below the point where the inoculated branches joined the trunk. The rootstocks occasionally yielded *Xanthomonas*.

Trees assessed 18 months after inoculation yielded *Xanthomonas* from the inoculation points, the inoculated branches and from the trunk including, occasionally, the rootstock. *Xanthomonas* was not detected in leaves, new shoots or from the branches that grew from buds close to the inoculation points.

It appears that after inoculation *Xanthomonas* moved quickly through the tree, including into new tissue. As this new tissue (leaves and shoots) aged the rate of isolation declined and in the subsequent season (17 months later) bacteria were found only in tissue that was present at the time of inoculation. It appeared that the bacteria initially moved into the new tissue but did not survive. New tissue may not provide suitable conditions for persistence of the bacteria or host defence mechanisms may have caused the bacteria to die. The final assessments of this trial will be conducted in spring and summer 2003-04.

Xanthomonas was detected at 97% of the inoculation points and had spread into the trunks of the inoculated trees. Copper applied to trees after inoculation did not prevent infection. The levels of inoculum applied may be much higher than occurs in the field. Further research is planned to determine the level of inoculum present on tools used to prune infected trees. This information will then be used to determine if the inoculum present on tools can result in infection and also if copper will prevent infection.

Conclusions

Xanthomonas is present on tools after pruning infected trees. Pruning wounds are possible entry points for the pathogen. The pathogen moved quickly through the tree, but appeared to be confined to woody tissue in the season following infection. Copper applied to pruning wounds after inoculation did not prevent infection. The viable number of bacteria, present on tools after pruning diseased trees, needs to be determined. It will then be necessary to determine whether or not this inoculum is sufficient to cause disease.

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