

of Australian plants with horticultural potential either as cut flowers or for the amenity nursery industry.

We are conducting an investigation into the possibility of using somatic hybridization to double up the chromosome numbers in some of our F1 hybrids to restore fertility and to continue the wide hybridization of genotypes within the *Chamaelaucium* alliance. This involves asexually fusing two protoplasts from two plants, which would normally not sexually hybridize.

## CONCLUSION

The Floriculture Group relies on the application of modern plant propagation practices for the selection, development, and breeding activity. Good propagation is the cornerstone to achieving the objective of the group, which is the development of novel plant products for commercial application. The group uses both sexual and asexual propagation techniques as well as conventional and sophisticated propagation technologies.

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## Shoot Dieback of Geraldton Wax®

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**Geraldton wax has been observed exhibiting signs of shoot dieback. This disease causes the plant to die back from the tips, often leading to whole plant death. This disease is a problem in all stages of production, but is worst during propagation when the young plants are most susceptible to disease.**

**Isolation studies proved the fungus *Colletotrichum* sp. to be the cause of this disease. It was found that *Colletotrichum* sp. is not influenced by wounding; however a heat stress period and humidity are requirements for infection to occur. Fungicide trials indicated that a preventative treatment is needed for this disease to be managed. Control of this pathogen was found to be effective with the use of Amistar® when applied as a preventative treatment. This article describes a part of the experimental work carried out as an honours project: Shoot Dieback of Geraldton Wax (Diplock, 2004).**

## INTRODUCTION

*Chamaelaucium uncinatum* Shauer (Geraldton wax) is one of Australia's most important native floriculture products. One of the limitations growers face when producing a high quality product is the limited amount of information available on diseases of this plant. There is a recognized need for pathogen identification and control of diseases of Geraldton wax (Fuss et al., 1992). Shoot dieback was observed

during the summer months of 2002–2004 in south east Queensland. Geraldton wax was showing signs of shoot dieback exhibit shriveled brown leaves which remain attached to the plant for a long period of time before dropping naturally (Fig. 1), often followed by plant death. The aim of this project was to identify the causal agent of this disease, the environmental factors that may influence infection, and the potential for chemical control. This work was carried out over a series of experiments.

## GENERAL METHODS

Plant material showing signs of dieback was collected from a range of production locations in south east Queensland. This material was surface sterilised and placed on agar plates and incubated. Fungal colonies that grew were subcultured and preliminary identification was carried out on these. The original fungal isolations were *Alternaria*, *Rhizoctonia*, and *Colletotrichum* species. Inoculum was prepared from these cultures. These were then applied directly to plants with a hand sprayer. Plants were rated for disease over a period of 30 days.



**Figure 1.** Left: healthy cutting; Right: cutting showing symptoms of shoot dieback.

## EXPERIMENT 1

The objective of this experiment was to identify the pathogen responsible for the shoot dieback in Geraldton wax and to develop a method for inducing disease in previously healthy plants.

**Materials and Methods.** Plants were wounded by two methods (stem wound, leaf wound) or left intact (no wound). Some plants were placed in bags to increase humidity. Plants were placed in a growth cabinet with temperatures ranging from 38–45 °C during the day and 29–34 °C for the night phase. Plants were taken from the cabinet and bags were removed after 48 h and placed in the glasshouse.

**Results and Discussion.** Following Koch's rules, it was proven that the cause of the stem dieback observed in Geraldton wax is *Colletotrichum* sp. [tentatively identified as *C. gloeosporioides* (Penz.)], this supports the findings of Arnett (1987) who found *Rhizoctonia* and *Colletotrichum* sp. to be responsible for damping off in Geraldton wax in propagation. Symptoms on the plants took approximately 4 days to begin to show after inoculation.

Levels of dieback of Geraldton wax was greatest in treatments that were bagged for 48 h after inoculation, plants that were not bagged showed very little infection. Wounding of the plant had very little influence on levels of disease in all of the treatments. This is likely to be because most species of *Colletotrichum* sp. penetrate the plant directly, after the production of appressoria rather than entering through a wound (Bailey et al., 1992).

These trials proved that infection by *Colletotrichum* sp. on Geraldton wax can be induced in plants that are exposed to high temperatures, unwounded, and bagged to increase the humidity. Unfortunately these conditions are often met during propagation, making a favorable environment for disease to occur.

## EXPERIMENT 2

The objective of this experiment was to evaluate a selection of fungicides for their effectiveness in controlling shoot dieback of Geraldton wax when applied either before or after inoculation with *Colletotrichum* sp.

### Materials and Methods.

**Trial 1.** Application of fungicides after inoculation.

Unwounded Geraldton wax plants were inoculated with *Colletotrichum* sp. as described earlier. All plants were bagged and placed in the growth chamber for 48 h at 39–44 °C (day time temperature) and 27–29 °C (night time temperature). Plants were taken from the cabinet and bags removed after 48 h. Plants were sprayed with the following treatments (Table 1), then placed in the glasshouse and rated for disease over a period of 20 days.

**Table 1.** Fungicide treatments used in Trials 1 and 2.

Fungicide	Active constituents	Company	Rate used
Benlate® WP	500 g·kg <sup>-1</sup> Benomyl	Dupont® Australia Ltd.	0.1 g/200 ml
Amistar® WP	500 g·kg <sup>-1</sup> Azoxystrobin	Syngenta Crop Protection Pty. Ltd.	0.1 g/200 ml 0.1 g/200 ml
Banrot® 400 WP	250 g·kg <sup>-1</sup> Thiophanate- methyl, 150 g·kg <sup>-1</sup> Etridiazole	Scotts Australia Pty. Ltd.	0.1 g/200 ml
Pro-Teck®	(19.8% w/v) Copper sulfate Pentahydrate	Magna-Bon Corp. Florida, U.S.A.	0.2 ml/200 ml

**Trial 2,** Application of fungicides before inoculation.

Unwounded Geraldton wax plants were sprayed with the treatments as per Table 1. Forty eight hours later, plants were inoculated with *Colletotrichum* sp. as described earlier, bagged, and placed in the growth chamber for 48 h at 39–44 °C (day-time temperature) and 27–29 °C (night-time temperature). Plants were taken from the cabinet and bags removed after 48 h and placed in the glasshouse and rated for disease over a period of 20 d.

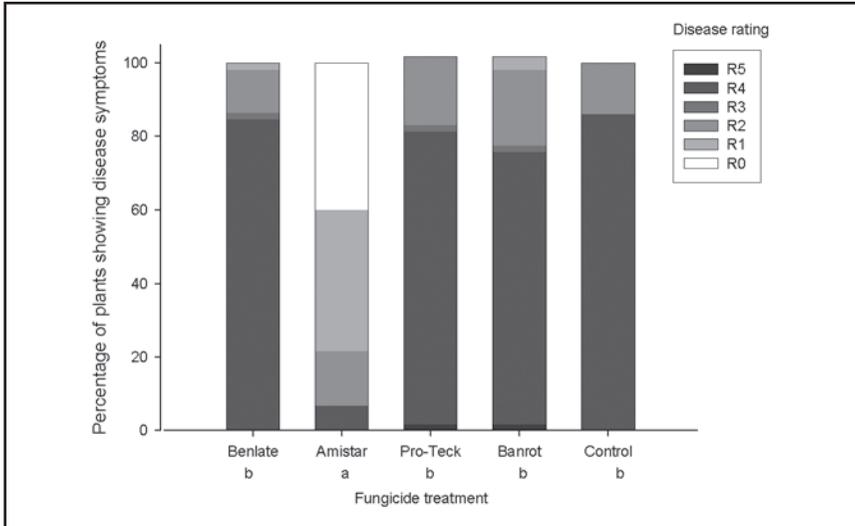
### Results.

**Trial 1.** Application of fungicides after inoculation.

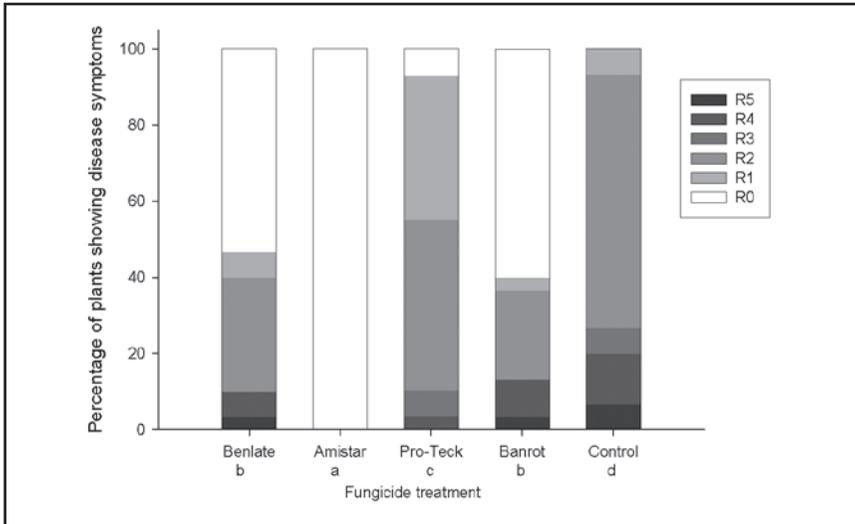
Levels of shoot dieback of Geraldton wax plants inoculated with *Colletotrichum* sp. were significantly reduced by the application of Amistar® 48 h after inoculation. All other fungicides were ineffective and not different to the untreated control.

**Trial 2.** Application of fungicides before inoculation.

Infection by *Colletotrichum* sp. on Geraldton wax was fully prevented by the application of Amistar® 48 h before inoculation (Fig. 3). Levels of shoot dieback were significantly reduced by the application of Benlate® and Banrot®. Pro-Teck® appeared to slow infection down and was significantly better than the control. The treatments when ranked from best to worst are as follows: Amistar® > Banrot® and Benlate® > Pro-Teck® > Control.



**Figure 2.** Effect of fungicide treatments (applied after inoculation) on infection by *Colletotrichum* sp. on Geraltion wax plants. (R0 = healthy, R1 = 1 mm spot on stem, R2 = lesion on stem >1 mm, R3 = lesion surrounding circumference, tip wilted, R4 = segment of shoot dead, R5 = entire shoot dead). Different letters indicate a significant difference ( $P = 0.05$ ) (Fisher's exact test SAS v8.2).



**Figure 3.** Effect of fungicide treatments (applied before inoculation) on infection by *Colletotrichum* sp. on Geraltion wax plants. (R0 = healthy, R1 = 1 mm spot on stem, R2 = lesion on stem >1 mm, R3 = lesion surrounding circumference, tip wilted, R4 = segment of shoot dead, R5 = entire shoot dead). Different letters indicate a significant difference ( $P = 0.05$ ) (Fisher's exact test SAS v8.2).

**Discussion.** Amistar® prevented disease when applied as a preventative treatment (Fig. 3) and significantly reduced disease when applied as a post-infection treatment (Fig. 2). However, it needs to be noted that Amistar is not currently registered for use on ornamental plants. Comparisons between the results from Trial 1 and Trial 2 indicate the need for the use of a preventative treatment in Geraldton wax propagation when temperatures are warm and humidity is high.

## CONCLUSION

Geraldton wax is one of Australia's most important native plants grown for the floriculture industry. Growers of this product face limitations in the area of plant protection from disease. Shoot dieback has been observed to cause significant economic losses in propagation and the field. The need for identification and understanding of pathogens of Geraldton wax has been discussed. The aim of this project was to identify the cause of the disease, control using fungicides, and factors, which may influence infection. Isolation trials and infection studies proved that shoot dieback of Geraldton wax is caused by *Colletotrichum* sp. It was demonstrated that wounding was not necessary for infection to occur, however high humidity and heat period are needed for infection to occur. Control of this disease can be achieved with the use of Amistar® when applied as preventative treatment.

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