Sustaining grapevines infected with eutypa dieback

FINAL REPORT to

GRAPE AND WINE RESEARCH & DEVELOPMENT CORPORATION

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1 ABSTRACT

This project developed practical and efficient methods for eutypa dieback control, determined the effect of environmental and production stresses, evaluated remedial surgery treatment for sustaining grapevines and established the presence and extent of eutypa dieback in emerging grapegrowing regions. A number of paints, pastes, fungicides and natural products were identified as effective pruning wound treatments to control eutypa dieback. Conventional spray machinery was found to be an effective means of applying fungicide to protect pruning wounds. The susceptibility of vines to eutypa dieback was influenced by water and temperature stress and deficit irrigation was shown to increase susceptibility of vines in warm, dry climates. Removal of infected wood by remedial surgery was shown to be an effective strategy to manage eutypa dieback, but success of this technique relies on removal of all infected wood. This strategy avoids long-term economic losses in affected vineyards and on-going maintenance will ensure good economic returns. Vineyard surveys revealed that eutypa dieback is wide-spread in emerging regions of South Australia, and in Tasmania, but was not detected in Western Australia. The surveys serve as a warning to grape growers to adopt control strategies to avoid production loss and increase the sustainability of grapevines.

2   EXECUTIVE SUMMARY

Eutypa dieback is a major disease of grapevines in Australia and worldwide caused by the fungus *Eutypa lata* that infects vines through pruning wounds, colonises wood tissue and causes dieback of cordons, stunting of green shoots and leaf distortion. Eutypa dieback threatens the sustainability of premium vineyards and is becoming a problem in most cool climate growing regions of southern Australia. This disease reduces growth and yield and if unmanaged eventually kills vines.

A number of pruning wound treatments have been evaluated and some controlled the disease. Physical barriers such as Gelseal, Greenseal, Garrison, Agseal and an acrylic paint were effective and are recommended for use especially on large wounds, such as those made during remedial surgery. Of the fungicides tested, Bavistin (carbendazim) and Folicur (tebuconazole) were the most effective but Scala (pyrimethanil), Switch (cyprodinil + fludioxonil) and Shirlan (fluazinam) were also effective when applied at rates higher than currently registered for use on other grape diseases. Bavistin, Folicur and Shirlan also controlled Botryosphaeria canker. Further evaluation is required to determine optimal rates for eutypa dieback control. Garlic and lactoferrin also showed promise as alternatives to fungicide application.

Spray application of fungicide to protect pruning wounds has potential to improve the efficiency of eutypa dieback control on large large-scale vineyard plantings. Bavistin applied with commercial sprayers reduced infection by *E. lata*, however, further research is required to determine the optimal fungicide rates and water volume required for adequate coverage and control with a range of sprayers and fungicides.

Growth room and field studies indicated that the susceptibility of vines to eutypa dieback was influenced by stress. Foliar symptoms of eutypa dieback increased and colonisation of the wood by *E. lata* tended to decrease when vines were subjected to water and temperature stress. Further investigation is required to test the hypothesis that although stressed vines displayed foliar symptoms, the fungus may have been inhibited. In warm, dry regions such as the Riverland, deficit irrigation may lead to increased susceptibility of pruning wounds to eutypa dieback. Further research is required to substantiate these findings and this information is important for the sustainable management of grapevines in drought conditions and with the predicted change in climate.

Removing infected cordons and trunks by remedial surgery is an effective strategy for managing eutypa dieback in grapevines. The production of new watershoots after removal of infected wood varied between grapevine cultivars. Remedial surgery reduced the incidence of vines with eutypa dieback symptoms in a vineyard but success depended on cultivar, pre-existing infection and the origin of the watershoot. It is recommended that all stained wood and a further 10 cm of healthy tissue be removed to reduce the likelihood of symptoms recurring. Cutting vines low (around 30 cm from the ground) is the most efficient means of remedial surgery and reworking a small section of a vineyard each year will minimise production losses. Production may be delayed for several years until the new canopy is established, but this is less than that required for an equivalent production from new plantings. Anecdotal evidence suggests that fruit quality is also restored within several years after trunk removal.

A preliminary decision support model was developed to extrapolate and predict economic returns of different decision scenarios in a Shiraz vineyard affected by eutypa dieback. Maintenance of vineyards in a disease-free condition ensured good economic returns for the vineyard. The cost of retrellising was insignificant for sustainability of vines. Remedial surgery undertaken when the incidence of eutypa dieback exceeds 20% in the vineyard avoids long-term economic losses. Further data need to be generated to expand and develop the model for use on other cultivars and regions.

Vineyard surveys revealed that eutypa dieback is wide-spread in the emerging regions of Adelaide Hills and Southern Fleurieu in South Australia. Although eutypa dieback was rare in the Mt Benson region, the detection of three vines during the survey serves as an early alert to growers. The youngest vines with foliar symptoms in this study were 7 years old and a greater incidence of foliar symptoms was observed in older vineyards than in younger ones. Incidence of foliar symptoms was greatest in Cabernet Sauvignon and Shiraz and very low in Merlot, Pinot Noir and Sauvignon Blanc.

A survey of vineyards in Tasmania provided the first official record of *E. lata* in grapevines in that state. It is likely that infection spread from apricot and other stone fruit orchards to new grapevine plantings. A survey of vineyards in Western Australia failed to detect foliar symptoms of eutypa dieback or identify *E. lata* in cankers on grapevine wood. The surveys serve as a warning to grape growers to adopt preventative control strategies, to avoid yield and quality loss and increase the sustainability of vines. Extension programs were initiated to inform growers of recommended strategies developed in this project for managing eutypa dieback.

3 BACKGROUND

Eutypa dieback, caused by the fungus *Eutypa lata*, is a major disease of grapevines in Australia and worldwide. It contributes to vineyard decline by reducing growth and yield (Munkvold *et al.* 1994, Creaser & Wicks 2001). In Australia, yield losses of at least 860 and 740 kg/ha have been reported for Shiraz and Cabernet Sauvignon, respectively (Wicks & Davies 1999) and in California economic losses of at least US$260 million per annum have been attributed to trunk disease (Siebert, 2001). Eutypa dieback threatens the sustainability of premium vineyards, especially those 8 years or older, and is becoming an increasing problem in most cool climate growing regions of southern Australia. The fungus infects vines through pruning wounds, colonises wood tissue and causes a characteristic wedge of dead tissue (Figure 1). The fungus is thought to produce toxic metabolites which are translocated to the foliage, causing stunting of the shoots, distortion and necrosis of leaves, reduced bunch size and uneven ripening (Figure 2; Moller & Kasimatis 1981, Tey-Rulh et al. 1991, Molyneux et al. 2002). If left unmanaged, the fungus eventually kills infected vines.

Between 1999 and 2006 the CRC for Viticulture and GWRDC funded the National Grapevine Trunk Disease project to examine ways of reducing the impact of trunk diseases in Australian vineyards. The main outcomes included:

- Foliar symptoms of eutypa dieback were directly related to yield losses although symptom expression varied from year to year. Evidence suggested that environmental factors may contribute to this phenomenon.

- The development of a bioassay where foliar symptoms are induced within 8 months and isolates of *E. lata* varied in their ability to induce foliar symptoms.

- Remedial surgery was the most effective method for treating infected vines in the short-term (up to 5 years).

- Chemical and biological alternatives were evaluated to protect pruning wounds from infection, and a number of materials are now recommended to growers, with two products currently registered and several more in the process.

- A DNA assay was developed for the identification of *E. lata* in pure culture and in wood tissue, giving more accurate and potentially cost-effective pathogen detection than morphological methods.

- Growth rate of *E. lata* in wood of various grapevine cultivars ranged from 10-18 mm per year in the field and the fungus was isolated up to 75 mm beyond staining in stems of potted vines.

Much of the information on the epidemiology and control of eutypa dieback is based on research on apricots (Carter 1991) and, more recently, on grapevine from research we have conducted through the CRCV and GWRDC. International research has also contributed to our current knowledge on management of grapevines for eutypa dieback.

Pruning wounds are susceptible to infection by *E. lata* for up to 7 weeks during winter but susceptibility decreases when pruning is delayed to late-winter or spring (Petzoldt *et al.* 1981; Munkvold & Marois 1995; Chapius *et al.* 1998). However, delaying pruning is usually impractical for growers with large-scale vineyard operations in Australia.

Chemical and biological control of eutypa dieback have been studied in countries where the disease is recognised as a significant problem in grapevines. Grapevine and apricot pruning wounds have been successfully treated with benzimidazole fungicides such as benomyl and carbendazim to control infection by *E. lata* (Carter 1991; Sosnowski *et al.* 2008). Other wound protectants effective against infection by *E. lata* include acrylic paint and pastes (Sosnowski *et al.* 2008), which produce a physical barrier to infection. Boron is an effective alternative for protection of pruning wounds (Rolshausen and Gubler 2005). Biocontrol agents, such as *Fusarium lateritium*, *Cladosporium herbarum* and *Trichoderma* spp., have also been used to protect wounds from invasion by *E. lata* spores with varying degrees of efficacy (Carter 1991; Munkvold and Marois 1993; John *et al.* 2005). To date, all treatments have been applied by hand using a paintbrush or hand sprayer. This practice is not economically viable in many large-scale vineyard operations in Australia and there is a need to evaluate commercial spray systems to provide effective spray coverage of pruning wounds.
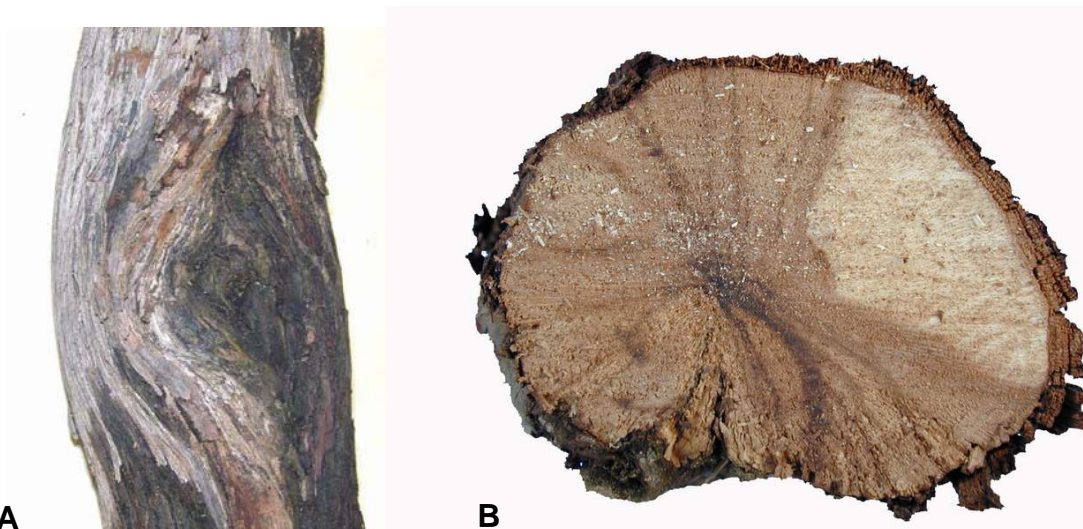
Figure 1. Wood symptoms caused by Eutypa lata, showing (A) the external canker and (B) the wedge of internal staining.

Figure 2. Characteristic foliar symptoms of eutypa dieback including (A) mature vine displaying stunted shoots, (B) single shoot with cupped, necrotic leaves and (C) uneven ripening and reduced bunch size.

In the vineyard, the incubation period between infection of wounds and appearance of foliar symptoms varies from 1 to 8 years (Carter, 1991; Tey-Rulh et al., 1991). Mycelial growth of E. lata ranges from 10 to 18 mm per year in wood of different grapevine cultivars in the field (Sosnowski et al. 2007b). Symptoms in wood develop over many years, and probably contribute to death of the infected vine, however, the severity of foliar symptom expression varies from year to year (Sosnowski et al. 2007a) possibly due to environmental factors. There is limited information on the effects of the environment on infection, growth and symptom expression. Optimal conditions for germination and growth of E. lata in vitro are 20-25ºC and at least 90% relative humidity (Carter 1957; Amborabé et al. 2005). In the field, a minimum rainfall of 1.27 mm is necessary for ascospore production from both apricot and grapevine infected with E. lata (Carter 1991). Susceptibility of grapevine wounds to ascospore infection is generally favoured by cooler winter conditions (Chapius et al. 1998; Munkvold and Marois 1995), however this may be attributed to the increased populations of epiphytic microorganisms in warmer conditions, which may reduce E. lata populations through competition.

Little is known about the effect of environmental and nutritional stress on the epidemiology of E. lata in grapevines. The development of a shadehouse bioassay in which foliar symptoms are induced within 8 months (Sosnowski et al. 2007b) provides a means of evaluating some of these stress factors to determine if it may be possible to alleviate symptoms of eutypa dieback by managing water, temperature and nutrient stress.

Research on curative control of eutypa dieback has been limited and only one strategy, remedial surgery, controls the disease. Short-term success of reworking vines is variable, gauged by production and health of watershoots and wood symptoms remaining within 5 years of surgery (Sosnowski et al. 2004). Malbec and Pinot Noir vines produced watershoots more readily than Shiraz and Cabernet Sauvignon. Vines cut at the crown produced more watershoots than vines cut 30-40 cm above the ground, however more infected wood remained below watershoots in the former. Foliar symptoms reappeared at low levels within 4 years in reworked vines, which was attributed to failure to remove all infected wood from the trunk. Long-term success of remedial surgery needs to be determined by the occurrence of foliar symptoms and, ultimately, by the yield and quality of grapes over 5 to 10 years.

Other approaches to managing vines with eutypa dieback have included foliar application of fertiliser which failed to alleviate foliar symptoms but increased yield following three years of treatment (Sosnowski et al. 2005). Injecting fungicides into trunks of vines also failed to reduce eutypa dieback symptoms (Sosnowski et al. 2004; unpublished data).

E. lata is widespread in most areas of the world where grapevines or apricots are cultivated. In Australia, eutypa dieback has been officially reported in South Australia and Victoria, but was not observed in a limited survey of 20 vineyards in the Hunter Valley and Mudgee wine regions of New South Wales (Creaser et al., 2003). The extent of eutypa dieback has been documented only for established wine regions of South Australia. In 1968, survey results indicated that up to 75% of Grenache vines and 48% of Cabernet sauvignon vines in some Barossa Valley vineyards showed symptoms of eutypa dieback (Wicks 1975). Up to 58%, 40% and 30% of Grenache vines displayed symptoms in Langhorne Creek, McLaren Vale and the Riverland, respectively. In a 1997 survey, the average incidence of vines displaying foliar symptoms of eutypa dieback was much less, ranging from 1% in Langhorne Creek to 6% in the Barossa Valley, and Grenache and Shiraz were the most likely to express symptoms with 8-9% incidence across all regions surveyed (Highet and Wicks 1998). This indicated that foliar symptoms vary from year to year, since confirmed by Sosnowski et al. (2007a), and that infected vines do not necessarily display foliar symptoms each year. Eutypa dieback has not been considered a problem in emerging wine regions such as the Adelaide Hills, Fleurieu Peninsula, Mt Benson and Robe in South Australia or in the states of Tasmania and Western Australia and, to date, little preventative action is being taken to control the disease.

This project built on the knowledge acquired over the 6 years of CRCV research and aimed to develop practical application methods for wound protection, investigate the factors underlying disease expression and effects of production stresses, evaluate long-term effectiveness of control methods and determine the extent of eutypa dieback in emerging grapegrowing regions. The objective was to improve management strategies and enhance the efficiency and sustainability of grape production for the benefit of the industry.

4 PROJECT AIMS

1. Develop practical and efficient methods for protecting wounds from infection by *E. lata*

2. Determine the effect of environmental and production stresses on the epidemiology of eutypa dieback

3. Evaluate remedial surgery treatment for the long-term sustainability of grapevines infected with *E. lata*

4. Establish the presence and extent of eutypa dieback on grapevines and alternative hosts in emerging grapegrowing regions of South Australia

Output 1

Practical and efficient application methods for wound protectants against eutypa dieback

Performance targets:

- Evaluate potential alternative fungicides and biocontrol agents for prevention of infection by *E. lata* for wound protection

- Evaluate commercial spray equipment for the application of fungicides for wound protection for efficient prevention of eutypa dieback infection in large-scale vineyards

Output 2

Improved management practices based on knowledge of effects of environmental and production stresses on eutypa dieback

Performance targets:

- Assess the effect of moisture, temperature and nutritional stresses on rooted cuttings infected with *E. lata* in controlled environments

- Inoculate field trials already established at Nuriootpa and Loxton Research Centres to assess the effect of deficit irrigation on eutypa dieback infection

- Elucidate relationships between foliar symptoms and climate

Output 3

Curative treatments to restore long-term productivity of eutypa dieback-infected vines

Performance targets:

- Monitor 12 remedial surgery trials established in the CRCV project for recurrence of eutypa dieback symptoms up to 9 years after treatment to assess long-term sustainability

- Cost-benefit analysis of remedial surgery for sustainable control of eutypa dieback

Output 4

Early alert for emerging wine regions to the incidence and threat of eutypa dieback

Performance targets:

- Survey and establish the incidence of eutypa dieback in emerging cool climate regions of South Australia

5 WOUND PROTECTION

5.1 *In vitro* evaluation

5.1.1 Introduction

Experiments were conducted in the laboratory to determine the effect of 12 fungicides and 6 alternative treatments on ascospore germination and mycelial growth of *E. lata* (Table 1). The fungicides chosen were those presently registered on grapevines for the control of other diseases and that have shown potential for eutypa dieback control in previous trials in Australia or overseas. Similarly, alternative products were chosen based on reports of their anti-microbial activity. The aim of these experiments was to identify potential products to be evaluated as pruning wound treatments in field trials.

5.1.2 Methods

Commercially formulated fungicides, listed in Table 1, were added to molten potato dextrose agar (PDA) prior to pouring into plastic Petri dishes (85 mm diam.) to provide concentrations of one and ten parts per million (ppm). Garlic (extracted from fresh garlic cloves), tea tree oil, lactoferrin (milk by-product), Sard (laundry powder) and honey were also added to molten PDA at concentrations of 1 and 10%.

E. lata ascospores were obtained from fruiting bodies, using methods based on those described in Carter (1991), by soaking dead grapevine wood in water for 1 h and leaving overnight in a plastic container, suspended by attaching to the lid. A suspension of spores was prepared and adjusted to 4000 spores/ml using a haemocytometer. Five 100 µL aliquots were placed on each of 12 replicate PDA plates per treatment including controls of unamended PDA. Plates were incubated under continuous fluorescent light at 23ºC for 48 hours. Germination percentage was determined for at least twenty-five spores randomly selected and counted from each plate.

Mycelium plugs (5 mm diam.) of *E. lata*, confirmed as such by DNA analysis (Lardner *et al.* 2005), were placed in the centre of amended PDA plates (12 replications) and incubated under fluorescent light for 12 h each day at 23ºC. Plates were assessed 7 days later by measuring and recording colony diameter.

Table 1. Treatments evaluated *in vitro* for effect on spore germination and mycelial growth of *Eutypa lata*.

Product	Active ingredient	Manufacturer
Fungicide		
Acrobat	Dimethomorph (500 g/Kg)	NuFarm Australia
Bavistin	Carbendazim (500 g/L)	BASF Australia
Domark	Tetraconazole (40 g/L)	Sipcam Pacific Australia
EcoCarb	Potassium bicarbonate (940 g/kg)	Organic Crop Protectants
Folicur	Tebuconazole (430 g/L)	Bayer Crop Science
Foli-R-Fos	Phosphonic acid (200 g/L)	U.I.M. Agrochemicals Australia
Legend	Quinoxyfen (250 g/L)	Dow Agrosciences Australia
Liquicop	Copper ammonium acetate (80 g/L)	Ekko Australia
Prosper	Spiroxamine (500 g/L)	Bayer Crop Science
Rubigan	Fenarimol (20 mL/L)	Du Pont Australia
Switch	Cyprodinil (375 g/Kg) + Fludioxonil (250 g/Kg)	Syngenta Crop Protection
Systhane	Myclobutanil (200 g/L)	Dow Agrosciences Australia
Alternatives		
Garlic	Fresh garlic juice extract	N/A
Honey	N/A	Capilano Honey
Lactoferrin	Lactoferrin (99.9%)	MG Nutritionals Australia
Proxitane sanitiser	Hydrogen peroxide (250 g/L)	Solvay Interox
Sard laundry powder	N/A	Colgate-Palmolive
Tea tree oil	*Melaleuca alternifolia* oil (15% w/w)	Herron Pharmaceuticals

N/A not applicable

5.1.3 Results and discussion

On control plates, 100% of ascospores germinated after 48 hours of incubation. The fungicides Folicur and Rubigan reduced germination to 7% and 28%, respectively, at 1 ppm (Figure 3). At the increased concentration of 10 ppm, Folicur, Rubigan, Switch, Domark and Systhane were most effective and reduced germination to between 5 and 28%. Most other fungicides had little effect on spore germination at 1 or 10 ppm. Spore germination was significantly reduced by the alternative treatments, garlic juice (germination = 2 and 1%) and lactoferrin (28 and 0%) when amended at concentrations of 1 and 10%, respectively, while tea tree oil reduced germination to 14% when included at a concentration of 10%.

Colony diameter of mycelium on control plates was 56 mm after 7 days incubation. Mycelial growth was completely inhibited by Switch and Bavistin at both 1 and 10 ppm (Figure 4). Folicur, Rubigan, Domark and Systhane completely inhibited growth at 10 ppm and at 1 ppm reduced the colony diameter to between 1 and 16 mm. Mycelial growth was inhibited by lactoferrin at 10% concentration and at 1% concentration reduced colony diameter to 17 mm. Garlic and tea tree oil inhibited mycelial growth at the higher concentration of 10% but did not significantly reduce colony diameter at 1%. Photographs of variation in colony growth between treatments are shown in Figure 5.

Folicur (tebuconazole), Rubigan (fenarimol), Switch (cyprodinil + fludioxonil), Domark (tetraconazole), garlic and lactoferrin were most inhibitory to spore germination and also reduced hyphal growth of *E. lata.* Bavisitin (carbendazim) and Systhane (myclobutanil) reduced only fungal growth significantly, however benomyl, which is effective against *E. lata* (Carter 1991, Sosnowski *et al.* 2008) also acts upon fungal growth only. These results confirm a previous report that tebuconazole, fenarimol and myclobutanil inhibited growth of *E. lata* (Halleen *et al.* 2001). Furthermore, Bester *et al.* (2007) showed that tebuconazole was also effective against fungal species associated with Botryoshphaeria canker.

Bavistin, Switch, Rubigan, Domark, Folicur, Systhane, lactoferrin and garlic were selected, amongst others, for evaluation in the field on established grapevines (Section 5.2).
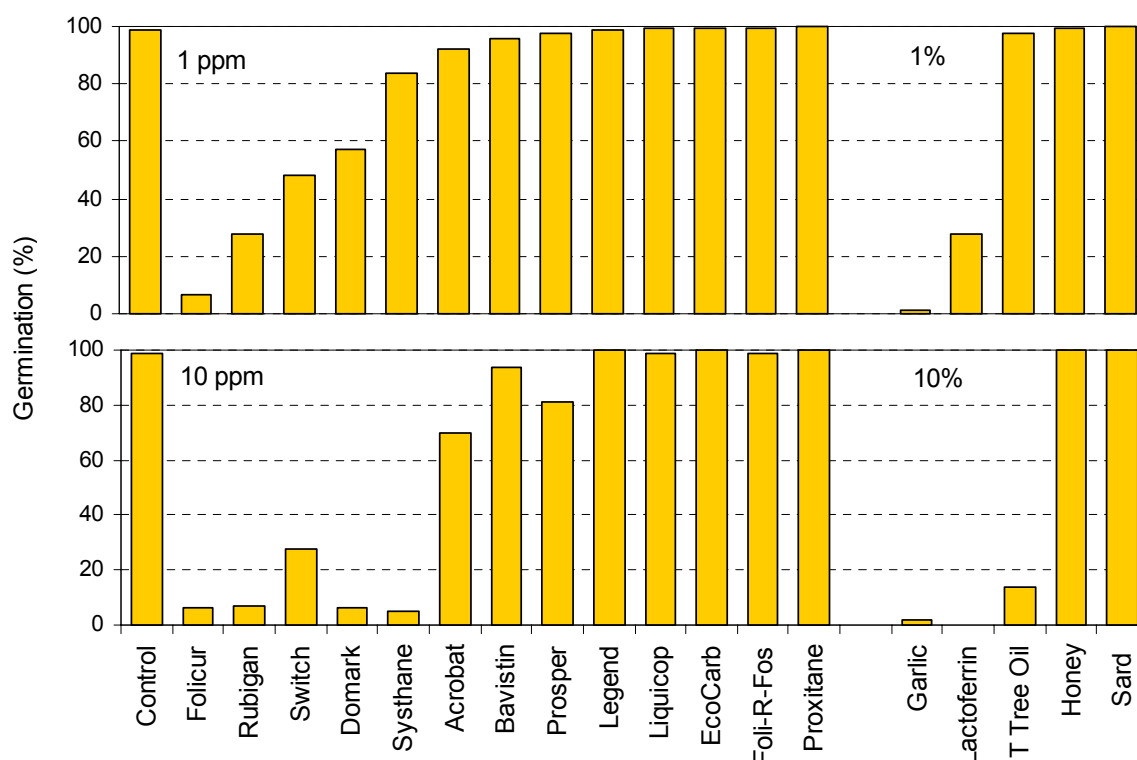

Figure 3. Effect of treatments *in vitro* on spore germination of *Eutypa lata* (1 and 10 ppm a.i. for fungicides or 1 and 10% for alternative treatments).
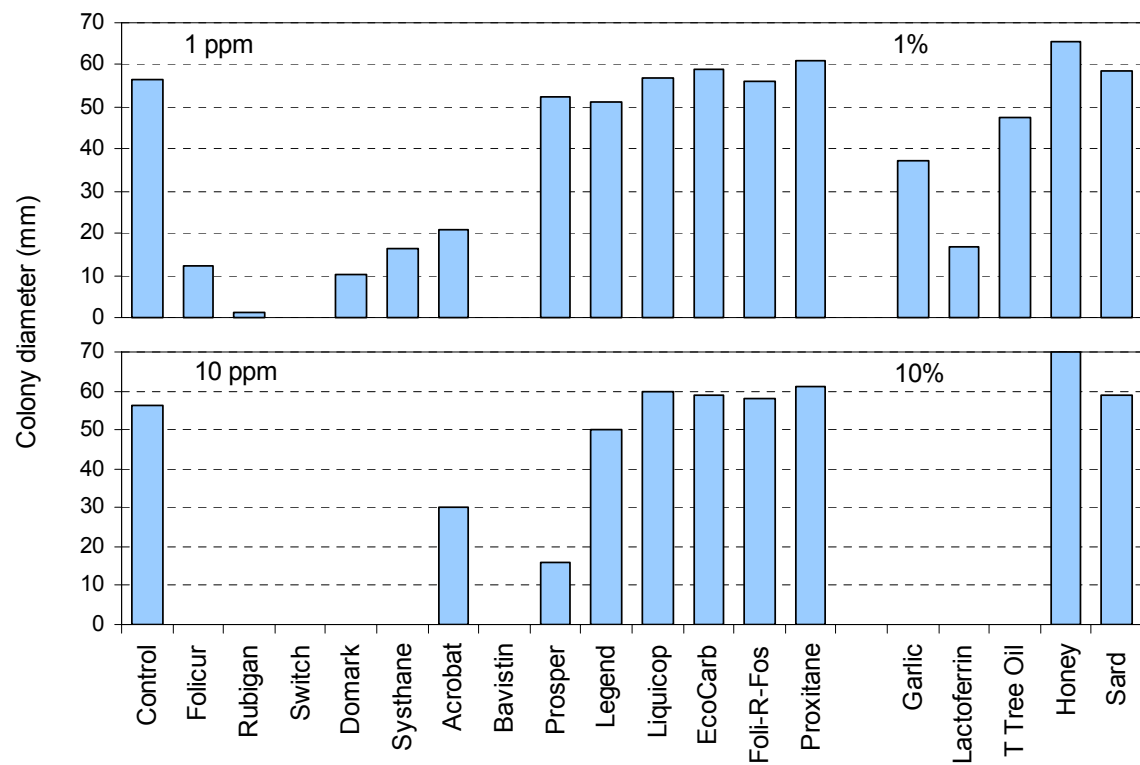
Figure 4. Effect of treatments *in vitro* on mycelial growth of *Eutypa lata* (1 and 10 ppm a.i. for fungicides or 1 and 10% for alternative treatments).
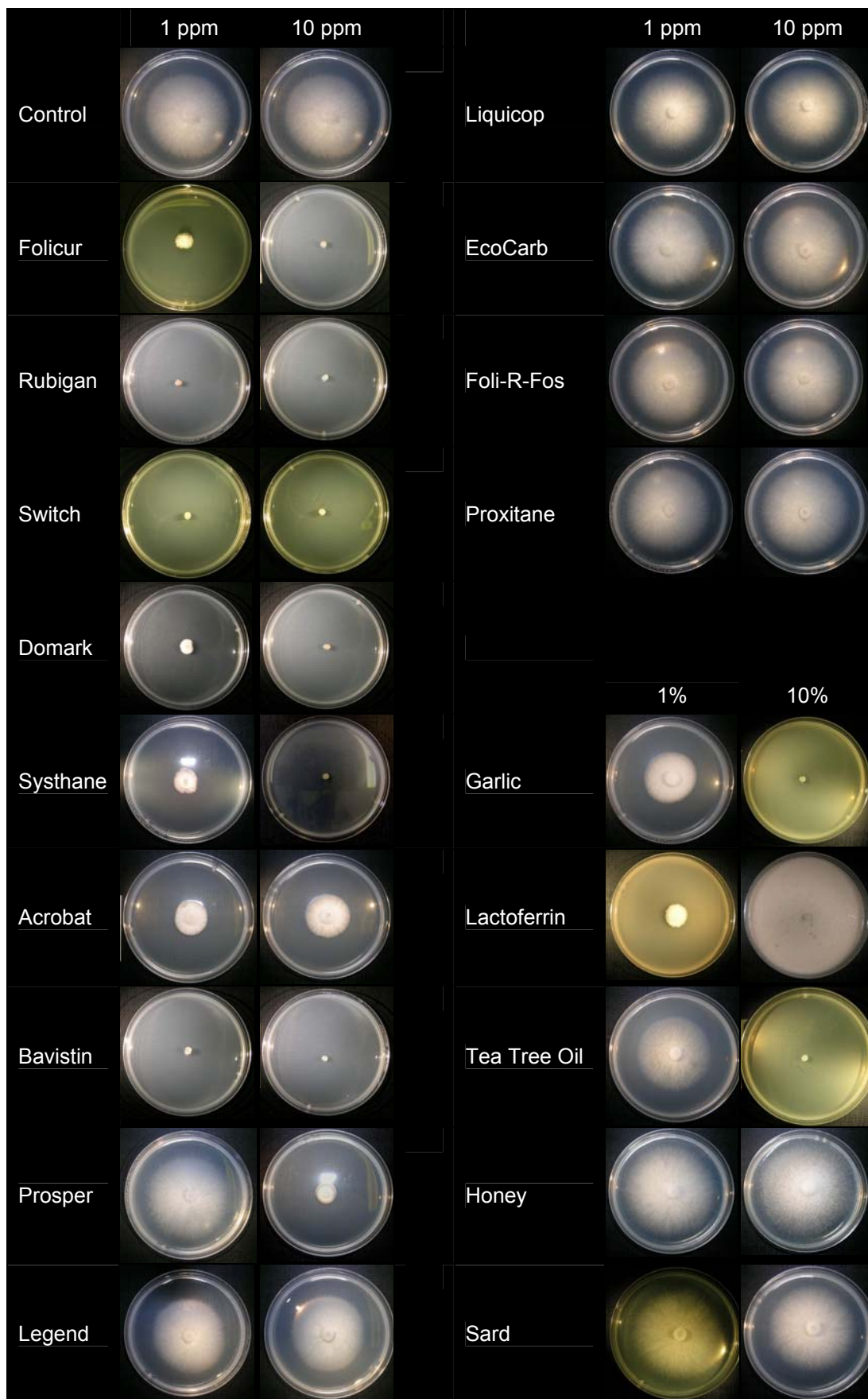
Figure 5. Photographs of colony growth of *Eutypa lata* after 7 days incubation under fluorescent light for 12 h each day at 23⁰C on potato dextrose agar plates amended with fungicides at 1 and 10 ppm and non-chemicals at 1 and 10% concentrations.

5.2 Field evaluation

5.2.1 Introduction

Three field trials were conducted to evaluate the efficacy of 11 fungicides, 3 physical barriers and 2 alternative treatments as pruning wound protectants against *E. lata* (Table 2). Treatments were selected from results of *in vitro* evaluation (Section 5.1), along with other promising treatments based on overseas results. The aim of these trials was to identify effective treatments and provide data to aid the registration or label extension of fungicides to be used for the control of eutypa dieback. A fourth trial, in collaboration with Charles Sturt University, aimed to evaluate some of these fungicides for the control of botryosphaeria canker.

5.2.2 Methods

Trials 1-3 were conducted between 2005 and 2009 on Cabernet sauvignon vines planted in 1984 at the Nuriootpa Research Centre, approximately 100 km north of Adelaide, South Australia.

In July, 1-year old canes were pruned to two buds using secateurs (Figure 6A). Using a paintbrush, treatments were applied liberally to 10 pruning wounds on each vine within 2 hours of wounding (Figure 6B). Wounds were sprayed with sterile distilled water (SDW) and then each wound inoculated with a 10-μl droplet containing *E. lata* ascospores (Figure 6C). Each trial was set up as a randomised block design.

In **Trial 1**, canes were pruned on 20 July 2005 and treated with Bavistin (2 & 10 mL/L of product), Cabrio (1.6 mL/L), Switch (8 g/L), Gelseal, Greenseal, Agseal or water as a control. The following day each wound was inoculated with approximately 1000 *E. lata* ascospores. Treated spurs were removed on 22 June 2006.

In **Trial 2**, canes were pruned on 18 July 2006 and treated with Bavistin (1 & 2 mL/L), Cabrio (0.8 & 4 mL/L), Shirlan (2 & 10 mL/L), Scala (4 & 20 g/L), Systhane (0.5 & 2.5 mL/L) or water as a control. Two hours later, each wound was inoculated with approximately 500 *E. lata* ascospores. Treated spurs were removed on 5 June 2007.

In **Trial 3**, canes were pruned on 8 July 2008 and treated with Bavistin (2 mL/L), Switch (0.8 g/L), Rubigan (0.2 mL/L), Domark (0.3 mL/L), Folicur (3 mL/L), Lactoferrin (1%), Garlic juice extract (1%), Mycloss (0.16 mL/L), Mycloss (0.16 mL/L) + Pentrabark (25 mL/L), Pentrabark (25 mL/L) or water as a control. Pentrabark (polyalkyleneoxide; Agrichem Australia) is a penetrating surfactant which is reported to assist penetration of fungicides into bark and wood. Two hours later, each wound was inoculated with approximately 500 *E. lata* ascospores and then on 12 July each wound was reinoculated with approximately 500 *E. lata* ascospores. Treated spurs were removed on 22 June 2009.

For **Trials 1-3**, during June of the following winter, canes were harvested from vines and returned to the laboratory (Figure 6D) where they were tested for the presence of viable *E. lata*. Bark was removed from each cane using a sharp knife (Figure 6E). The exposed wood was surface sterilised in 2.5% sodium hypochloride (NaOCl) containing a drop of Tween 20 surfactant for 12 minutes and then washed twice in SDW. Using sterilised secateurs, canes were cut into chips ($3 \times 2 \times 2$ mm) taken from each side of the margin between live and dead wood tissue (Figure 6F). Five wood chips were plated in each plastic Petri dish filled with PDA amended with antibiotic (streptomycin sulfate 25 μg/L), with two replicates per spur. Petri dishes were incubated at 23°C under fluorescent light for 12 h each day for 7 days and then assessed for presence or absence of *E. lata* cultures (Figure 6G). Efficacy was based on the mean percent recovery (MPR) of *E. lata* from the treated canes by isolation on PDA. Data are presented as mean percent disease control which was calculated as the reduction in MPR as a proportion of the inoculated control.

Trial 4 was conducted in collaboration with Dr Wayne Pitt (Charles Sturt University) at the Nuriootpa Research centre on Sauvignon Blanc vines (planted in 1985) which were grafted to Shiraz in 2001. On 3 July 2008, 1-year-old canes were pruned to two buds using secateurs. Using a paintbrush, treatments were applied liberally to 10 pruning wounds on each vine within 1-2 hours of wounding. Treatments included; Folicur (0.3mL/L), Nustar (0.1g/L), Shirlan (1mL/L), Bavistin (1mL/L), Switch (0.8g/L), Topas (0.125mL/L), Garrison, Bacseal, ATCS tree wound dressing, Domark (0.3mL/L) or SDW as a control. The following day each wound was inoculated with a 10-μL droplet of approximately 500 conidial spores in SDW suspension of *Diplodia mutila* (one of the causes botryosphaeria canker).

On 17 July 2009, treated canes were harvested from vines and returned to the laboratory where bark was removed from each cane using a sharp knife. The exposed wood was surfaced sterilised in 1%

w/v sodium hypochlorite (NaOCl) for 2 minutes and then washed twice with SDW. Using sterilised secateurs, canes were cut into small pieces approximately 2-3 mm^2 and five pieces from each cane transferred to plastic Petri dishes containing PDA supplemented with 25 μg./mL streptomycin sulphate. Petri dishes were incubated at 25°C in the dark for 3-5 days and then assessed for the presence or absence of *D. mutila*. Efficacy was based on the mean percent recovery (MPR) of *D. mutila* from treated canes by isolation on PDA. Data are presented as mean percent disease control which was calculated as the reduction in MPR as a proportion of the inoculated control.

Table 2. Pruning wound treatments evaluated in the field.

Product	Active ingredient	Manufacturer
Fungicide		
Bavistin	Carbendazim (500 g/L)	BASF Australia
Cabrio	Pyraclostrobin (250g/L)	BASF Australia
Domark	Tetraconazole (40 g/L)	Sipcam Pacific Australia
Folicur	Tebuconazole (430 g/L)	Bayer Crop Science
Mycloss Extra	Myclobutanil (200g/L)	Dow Agrosciences Australia
Nustar	Flusilazole (200 g/kg)	Du Pont Australia
Rubigan	Fenarimol (20 mL/L)	Du Pont Australia
Scala	Pyrimethanil (400 g/kg)	Bayer Crop Science
Shirlan	Fluazinam (500 g/L)	Syngenta Crop Protection
Switch	Cyprodinil (375 g/Kg) + Fludioxonil (250 g/Kg)	Syngenta Crop Protection
Systhane	Myclobutanil (200 g/L)	Dow Agrosciences Australia
Topas	Penconazole (100 g/L)	Syngenta Crop Protection
Physical barrier		
ATCS tree wound dressing	Acrylic paint	Hortitape Australia
Agseal	Paste	OneChem Australia
Bacseal	Paste + tebuconazole (10 g/L)	Bayer Crop Science
Gelseal	Gel + tebuconazole (10g/L)	Omnia Primaxa New Zealand
Greenseal	Paint + tebuconazole (10 g/L)	Omnia Primaxa New Zealand
Garrison	Paste + cyproconazole (2.5 g/L) + iodocarb (1 g/L)	Chemcolour Industries New Zealand
Alternatives		
Garlic	Fresh garlic juice extract	N/A
Lactoferrin	Lactoferrin (99.9%)	MG Nutritionals Australia

N/A not applicable

Figure 6. Techniques used to evaluate treatments for the prevention of wound infection: (A) Cutting canes to two-bud spurs on Cabernet Sauvignon vines at Nuriootpa; (B) Applying treatment to wound with a paint brush; (C) Inoculating treated wounds with ascospores of *E. lata*; (D) Spurs removed from vines during the following winter; (E) Spurs with bark removed; (F) Cutting chips from either side of the necrotic margin and (G) Cultures of *E. lata* growing from wood chips.

5.2.3 Results

Trial 1. E. lata was recovered from 79% of untreated control wounds (data not shown). Gelseal and Greenseal, which each contain 10 g/L tebuconazole, provided the best disease control, 100 and 94%, respectively (Figure 7). Bavistin (10 mL/L) and Agseal provided approximately 80% disease control and Bavistin (2 ml/L) and Switch controlled disease by 55-60%. Cabrio provided the least control (22%) of all treatments in Trial 1.

Trial 2. E. lata was recovered from 27% of untreated control wounds (data not shown). Bavistin provided 57 and 77% disease control when applied at 1 and 2 mL/L respectively (Figure 8). Mean percent disease control increased when the product application rate was increased 5-fold for Shirlan (from 27 to 56%), Scala (27 to 79%) and Systhane (0 to 29%).

Trial 3. E. lata was recovered from 62% of untreated control wounds (data not shown). Folicur provided 76% disease control, similar to that of Bavistin with 74% disease control (Figure 9). Mycloss + Pentrabark, Garlic, Switch and Lactoferrin provided 20-30% disease control with the other treatments providing little or no control of disease.

Trial 4. D. mutila was recovered from 59% of untreated control wounds (data not shown). A number of treatments provided control of botryosphaeria canker; Garrison (55%), Shirlan (53%), Folicur (45%), Bacseal (37%), Bavistin (32%) and ATCS acrylic paint (31%) (Figure 10).
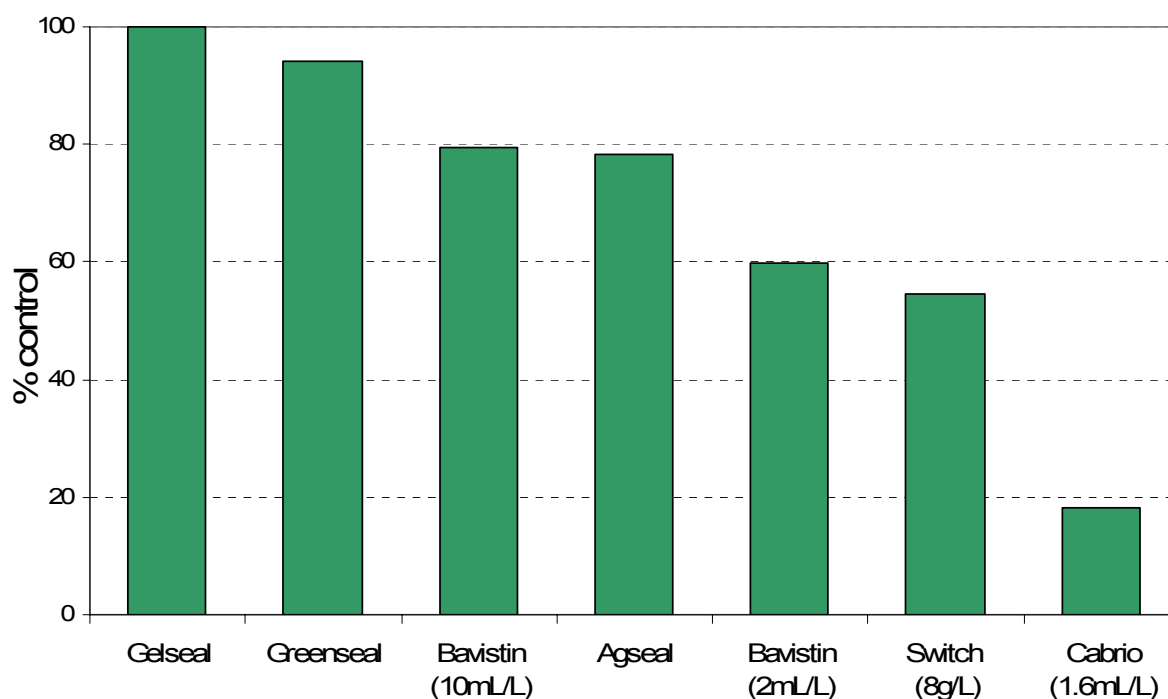

Figure 7. Trial 1 (2005/6) – mean percent control of eutypa dieback with different pruning wound treatments in the field.
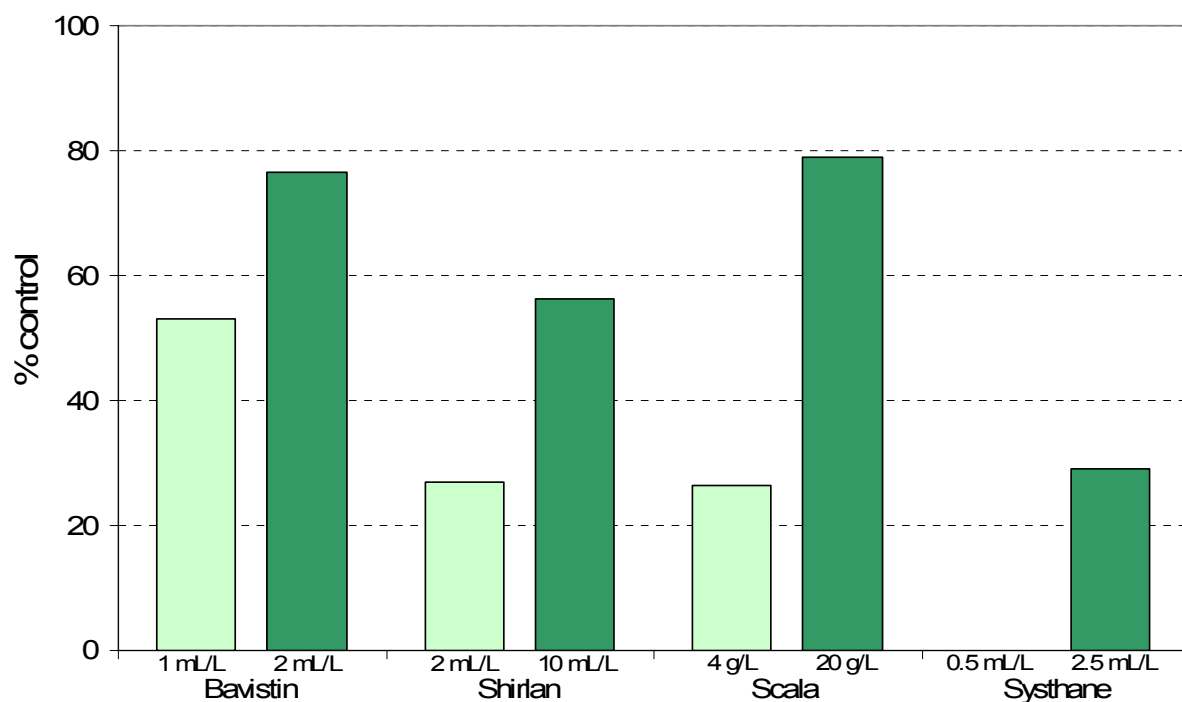
Figure 8. Trial 2 (2006/7) – mean percent control of eutypa dieback with different pruning wound treatments in the field.
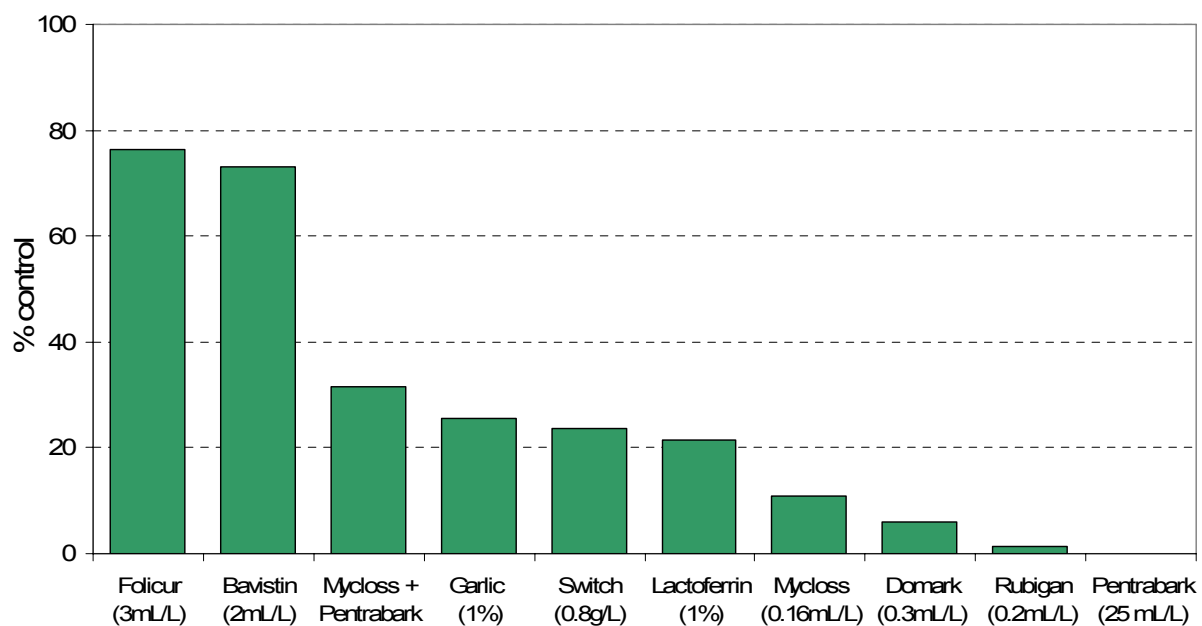

Figure 9. Trial 3 (2008/9) – mean percent control of eutypa dieback with different pruning wound treatments in the field.
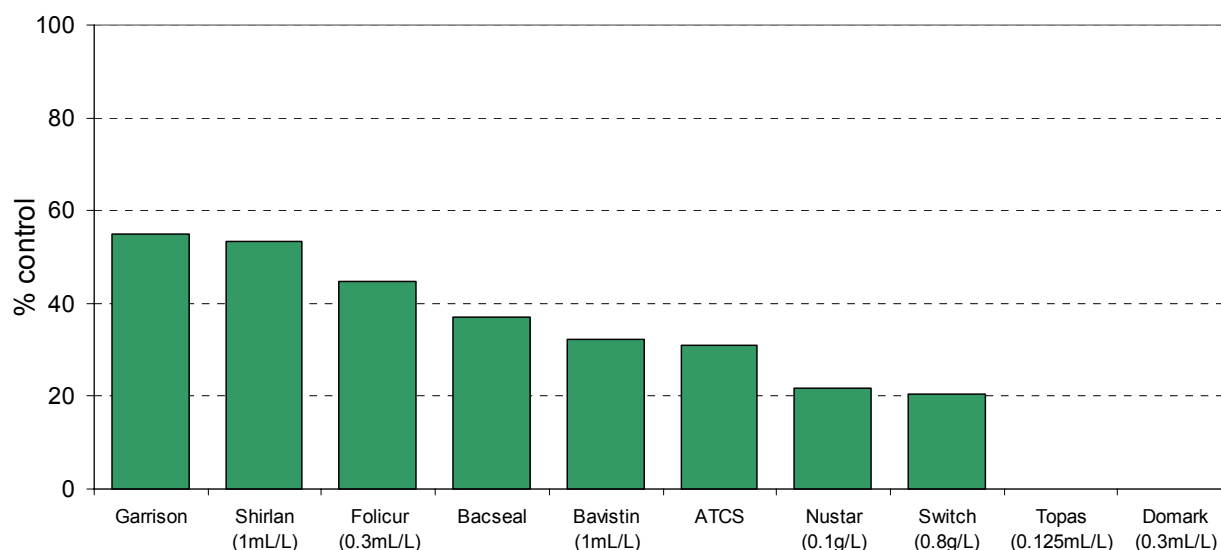
Figure 10. Trial 4 (2008/9) - Mean percent control of botryosphaeria canker with different pruning wound treatments in the field.

5.2.4 Discussion

Gelseal and Greenseal were the most effective pruning wound treatments in field trials, possibly due to the physical barrier to entry of spores into wounds provided by the gel and paint. Both products contain the fungicide tebuconazole at 10 g/L which is likely to inhibit spore germination and growth (Section 5.1) from of any spores landing on the wound prior to application or should any cracks or openings occur in the wound treatment upon drying, that would allow spores to enter wounds. The product Agseal also provided a level of control equivalent to that achieved with Bavistin. These data have supported the registration of Greenseal by the Australian Pesticides and Veterinary Medicines Authority (APVMA) for eutypa dieback control and a similar procedure is currently underway for Gelseal registration. Along with other effective products such as acrylic paint and Garrison which were identified in previous CRCV research, the use of physical barriers have been shown to be an effective strategy to protect large wounds such as those made during vine restructuring and remedial surgery (Section 7).

Of the fungicides evaluated, Bavistin (carbendazim) and Folicur (tebuconazole) were the most effective for the control of eutypa dieback. Scala (pyrimethanil), Switch (Cyprodinil + Fludioxonil) and Shirlan (fluazinam) were also effective but only when applied at 10 times the label rate. The addition of Pentrabark to Mycloss (myclobutanil) improved disease control from 10 to 30%. Pentrabark had no effect on disease control alone indicating that it enhanced the efficacy of Mycloss and will be considered as an additive to other fungicides in future evaluation trials. Although Systhane (myclobutanil) provided only 25% disease control when applied at 15 times label rate, it is possible that disease control may have been considerably more had Pentrabark been added.

Garlic and lactoferrin provided little control when applied at concentrations of 1%, however based on the *in vitro* evaluation (Section 5.1) when applied at 10% are likely to provide better control of eutypa dieback so will be included in future trials. These treatments might provide an excellent alternative to fungicides if found to effectively control *E. lata*.

The efficacy of treatments for botryosphaeria control were less than for eutypa dieback conrol. In Trial 4, the fungicides Shirlan, Folicur and Bavistin were applied at lower rates than in Trials 1-3 for eutypa dieback and so by increasing these rates it is likely that the level control will be improved. The performance of paints and pastes was also less than that for eutypa dieback and this may be due to presence of the fungus in the shoots before inoculation, as recovery of *D. mutila* from non-inoculated controls was 32% (data not shown). Research is underway to further evaluate these treatments against other fungal species associated with botryosphaeria canker and there is potential for several options of dual control of both trunk diseases which are prevalent in Australian vineyards.

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5.3 Spray application

5.3.1 Introduction

Fungicides, such as carbendazim, applied to grapevine wounds with a paint brush provide excellent control of eutypa dieback (Section 5.2). However, in large-scale viticulture enterprises this is not economically viable due to the labour costs and alternative methods of applying fungicides to wounds are required. The possibility of applying pruning wound protection using commercial sprayers was conceived from discussions with growers. Trials were established to assess the efficacy of spray application for pruning wound protection.

5.3.2 Methods

Trials 1 and 2 were set up on Cabernet Sauvignon vines at the Nuriootpa Research Centre in 2004 and 2005, respectively. One-year-old canes were pruned to two buds on 5 August 2004 and 19 July 2005. Bavistin (5 mL/L) was applied to pruned spurs using either a SARDI fan sprayer (prototype) with 596 L/ha output (Figure 11A), a Hardi air-assisted sprayer with 366 L/ha output (Figure 11B) or by hand with a paint brush and controls were treated by hand with SDW. Treated wounds were each inoculated with 1000 ascospores in 20 µL of water 1 (Trial 2) or 14 (Trial 1) days after treatment. Treated spurs were removed in June 2005 and 2006 and evaluated as described in Section 5.2.2.

Trials 3 and 4 were set up on Riesling vines in the Coonawarra in 2007 and 2008, respectively. One-year-old canes were pruned to two buds on 17 July 2007 and 23 July 2008. Bavistin (2mL/L) was applied to pruned spurs using either a Silvan Turbomiser with 200 L/ha output (Figure 11C) or a Croplands Quantum Mist with 200 L/ha output (Figure 11D). Treated wounds were each inoculated with 500 ascospores in 20 µL of water; once, 24 h after treatment for Trial 3 and twice, 12 and 24 h after treatment for Trial 4. Treated spurs were removed in June 2008 and 2009 and evaluated as described in Section 5.2.2.

5.3.3 Results

Trial 1. *E. lata* was recovered from 7% of untreated control wounds (data not shown). The fungicide spray coverage obtained with the SARDI fan sprayer was sufficient to provide 100% control of wound infection by *E. lata* when inoculation occurred 14 days after treatment, and was equal to that of application using a paint brush (Figure 12). By comparison, application with the Hardi air-assisted sprayer provided 77% control.

Trial 2. *E. lata* was recovered from 60% of untreated control wounds (data not shown). Where inoculation occurred 1 day after treatment, the SARDI fan sprayer provided 92% control, similar to that of application with a paint brush which provided 96% control. Application with the Hardi air-assisted sprayer provided 57% control. In addition, spray drift from the Hardi air-assisted sprayer was noticeably greater than that of the SARDI fan sprayer (Figure 11 A&B).

Trial 3. *E. lata* was recovered from 19% of untreated control wounds (data not shown). Application of fungicide with the Silvan and Croplands sprayers provided 49% and 30% disease control, respectively, compared to 78% control provide by application with a paintbrush (Figure 13).

Trial 4. *E. lata* spores and *E. lata* was recovered from 60% of untreated control wounds (data not shown). The Silvan and Croplands sprayers provided 43% and 53% disease control, respectively, compared to 94% control provide by application with a paintbrush.

5.3.4 Discussion

In Trials 1 and 2, control achieved with the application of Bavistin using the SARDI fan sprayer was similar to that achieved with Bavistin applied using a paint brush. The Hardi air-assisted sprayer was less effective providing 60-80% control. The output from the Hardi sprayer was less than that of the SARDI fan sprayer which most likely contributed to this difference. In Trials 3 and 4, application of Bavistin with the Silvan and Croplands sprayers had similar efficacies but were considerably less effective than application with a paintbrush. The reduction in efficacy from Trials 1 and 2 to trials 3 and 4 is probably due to the decreased fungicide application rate from 5 to 2 mL/L along with the output rates of 596 and 366 L/ha to 200 L/ha, respectively. Therefore, more research is required to determine the optimal fungicide and output rates for individual sprayers.

One of the issues that needs to be addressed is reducing spray drift whilst maximising deposition on wounds. In a recent visit to Australia, international spray application expert Dr Andrew Landers (Cornell University USA), suggested that nozzles and fans can be directed carefully and, in some cases turned off, to ensure the spray covers only the pruning wound zone. This would result in maximum deposition on the wounds with reduced output rate.

Spray application of fungicide to protect pruning wounds has shown potential to improve the efficiency of preventing eutypa dieback on large large-scale vineyard plantings. In these trials Bavistin was used but there is a need to evaluate sprayers using other fungicides and formulations, such as Folicur, as well as bio-control agents and alternative products effective against *E. lata*. It is also important to evaluate a range of sprayers typically used in Australian vineyards including recycle sprayers. To date, there is no other efficacy data on spray application of pruning wound protectants, so it is crucial that this research continues here in Australia.

Figure 11. Sprayers used for trials at Nuriootpa Research Centre in 2004 and 2005; (A) SARDI fan sprayer (prototype) and (B) Hardi air-assisted sprayer and Coonawarra in 2007 and 2008; (C) Silvan Turbomiser and (D) Croplands Quantum Mist.
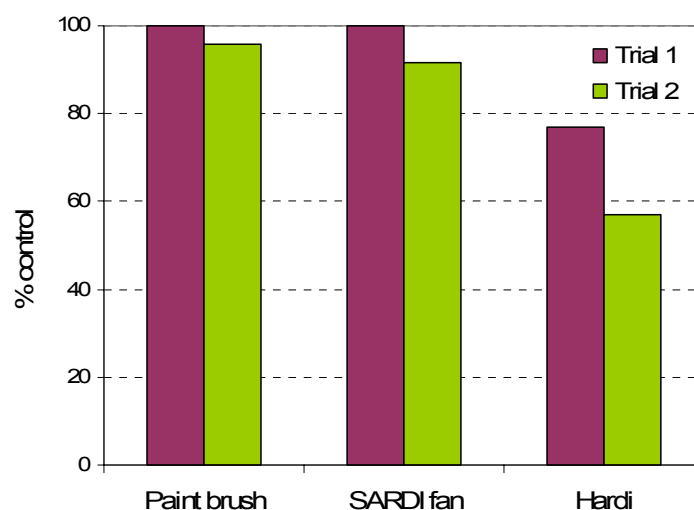
Figure 12. Trials 1 and 2 - Percentage control of *E. lata* infection of pruning wounds by application of Bavistin (5 mL/L) with either a paint brush, SARDI fan sprayer (596 L/ha output) or Hardi air-assisted sprayer (366 L/ha output).
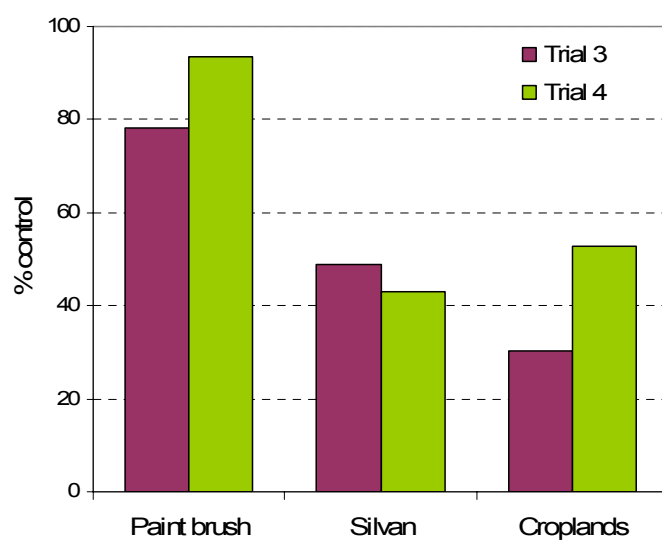

Figure 13. Trials 3 and 4 - Percentage control of *E. lata* infection of pruning wounds by application of Bavistin (2 mL/L) with either a paint brush, Silvan Turbomiser sprayer (200 L/ha output) or Croplands Quantum Mist sprayer (200 L/ha output).

6 ENVIRONMENTAL AND PRODUCTION STRESS

6.1 Effect of temperature, soil water content and nutrition on eutypa dieback

6.1.1 Introduction

Environmental conditions are thought to be an important factor in the expression of foliar symptoms of eutypa dieback and variation in severity of symptoms has been recorded from year to year in Australia (Creaser & Wicks 2001, Sosnowski *et al.*, 2007a), France (Dumot *et al.*, 2004) and the USA (Butterworth *et al.*, 2005). Sosnowski *et al.* (2007a) reported an increase in symptom expression to be associated with higher winter rainfall and suggested that increased water availability may facilitate transport of toxins to the foliage in spring. Sosnowski *et al.* (2007a) also reported that decreased disease incidence was associated with increased temperature in spring. As vines grow more vigorously in warmer conditions, it was proposed that the ability of fungal toxins to reach the foliage might be reduced and the increased amount of foliage may dilute the toxins.

Research has also demonstrated that low soil water content (Levitt, 1980, Smart & Coombe, 1983), high temperature (Kriedemann & Smart, 1971) and plant nutrient imbalance (Clarkson, 1985) can dramatically reduce grapevine health by influencing the severity of other diseases. Environment and plant health, therefore, are likely to influence the expression of foliar symptoms of eutypa dieback and the growth rate of the pathogen.

6.1.2 Methods

Experiments were undertaken to evaluate the effect of soil water content, air temperature and nutrient availability on the expression of foliar symptoms of eutypa dieback and the growth of *E. lata*. In early February of 2007 and 2008 (Table 3), rooted grapevine cuttings (cv. Grenache) obtained from a commercial nursery (Orlando Wines, Rowland Flat, South Australia) were planted into pots filled with either potting mix for the water and temperature experiments or infertile sand (Sloan's Sands, Dry Creek, South Australia) for the nutrient experiments. Potted vines were transferred to a shade house, pruned to two-bud spurs, watered by hand as required and fertilised with Thrive liquid fertiliser as per the manufacturer's label directions.

E. lata was cultured on PDA and incubated at 23ºC under fluorescent light for 12 h each day. A mycelium plug (5 mm diameter) taken from the actively growing margin of 7-day-old cultures was inserted into a 5-mm diameter hole drilled into the stem of each cutting as described by Sosnowski *et al.* (2007a). Inoculation sites were sealed with Parafilm. All vines were inoculated with *E. lata* in 2007 while in 2008, vines were inoculated with *E. lata* or blank PDA plugs as controls. Timing of inoculations are given in Table 3.

In **2007/08**, three experiments were conducted.

Temperature experiment: 14 weeks after inoculation and 4 weeks after budburst 66 potted vines were transferred to one of three controlled environment rooms set at 14°C, 22°C or 30°C with 12 hour day/night light cycles (400W metal halide globes) (Figure 14A) and temperature was monitored using Tinytag data loggers. Pots were regularly irrigated and fertilised with Thrive. One leader shoot per pot was selected and trained around a bamboo cane and lateral shoots were trimmed.

Soil water experiment: 54 potted vines received the same amount of rain and irrigation until 17 weeks after inoculation and 8 weeks after budburst. At this time, when shoots were 30-50 cm long, different soil water regimes were imposed by watering pots by hand with the same volume of water at varying time intervals to maintain the soil at high (20-40% volumetric water content), moderate (10-25%) and low (5-20%). Volumetric soil water content was measured periodically using time domain reflectometry (TDR). On 9 April 2008, a pressure chamber (pressure bomb) was used to measure grapevine water status.

Nutrient experiment; 48 potted vines were planted into sand and fertilised immediately with Yates Thrive soluble all purpose plant food (NPK analysis 27:5.5:9) as per directions on container. Thereafter, the nutrient treated vines were fertilised every 14 days and the control vines were not fertilised.

In **2008/09**, two experiments were conducted.

Soil water and temperature experiment: 162 potted vines were maintained in the shade house with regular irrigation and fertilised until 8 weeks after inoculation, when a canopy made of clear polyethylene sheets was placed over the vines to protect them from rain. Pots were then irrigated by hand to maintain high, moderate and low soil water content during dormancy, as described above.

Twenty-two weeks later (onset of bud burst) pots were transferred into one of three controlled environment rooms set at 14°C, 22°C or 30°C as described above. Irrigation continued by hand to maintain high, moderate and low soil water content. Soil water and temperature were monitored as described above. On 4 Dec 2008, a pressure chamber (pressure bomb) was used to measure grapevine water status.

Nutrient experiment: 56 potted vines were planted into sand and the experiment repeated as described above.

Vines were assessed for foliar symptoms when shoots of control vines were between 50 and 70 cm long (Table 3). Severity of eutypa dieback symptoms was expressed as the difference in length of stunted shoots on inoculated and control vines as a percentage of the length of shoots on control vines (Sosnowski *et al.* 2007b).

In both years, prior to destructive sampling for internal wood assessment (Table 3), vines were pruned and all cane pieces collected and weighed.

The extent of stained wood and spread of mycelium of *E. lata* in the stem of each plant was assessed. Bark was removed using a sharp knife and the distance of staining above and below the inoculation point was measured. Stems were surface sterilised for 12 minutes in 2.5% sodium hypochlorite (NaOH) containing a drop of Tween 20 surfactant (Sosnowski *et al.* 2007b). Cross-sections (2 mm thick) of stem were cut at 5-mm intervals above and below the inoculation point using secateurs, sterilized between samples by immersing the blades in ethanol followed by flaming. Wood sections were then placed on PDA amended with streptomycin sulphate and incubated at 23°C for 7 days under fluorescent light for 12 h each day and then assessed for presence or absence of *E. lata* cultures.

Table 3. Timing of methods from potting of vines to destructive sampling for wood assessment for experiments over two years

Experiment	Vines potted	Inoculation	Treatment imposed	Foliar assessment	Wood assessment
2007/8					
Temp	9 Feb 2007	15 Jun 2007	26 Sep 2007	n/a	23 May 2008
Soil water	9 Feb 2007	15 Jun 2007	12 Oct 2007	n/a	10 Jun 2008
Nutrient	9 Feb 2007	15 Jun 2007	9 Feb 2007	n/a	18 July 2008
2008/9					
Soil water & temp	8 Feb 2008	21 Feb 2008	15 Jun 2008* 17 Sep 2008^	7 Nov 2008	22 Apr 2009
Nutrient	12 Feb 2008	21 Feb 2008	12 Feb 2008	1 Dec 2008	22 Apr 2009

* water treatments imposed
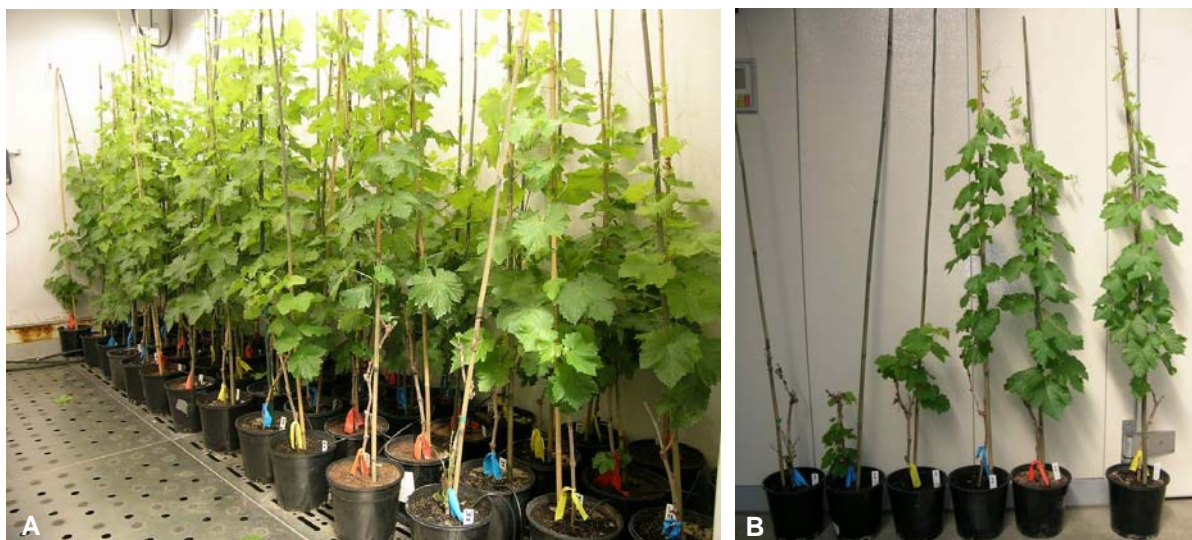^ temperature treatments imposed
n/a not applicable

Figure 14. (A) Controlled environment room with potted vines and (B) vines showing foliar symptoms of eutypa dieback (left).

6.1.3 Results

In 2007/8, foliar symptoms were not observed on vines in the temperature and soil water experiments although staining was observed in the inoculated stems. The mean extent of staining was significantly less than that of mean mycelial growth in the stems (up to 45 mm), except when temperature was 14ºC (Figure 15). Mean mycelial growth ranged from 50 to 60 mm in the temperature experiment and from 45 to 50 mm in the soil water experiment, however no differences were observed between any of the treatments. In 2008/9, when temperature and soil water treatments were combined, foliar symptoms were observed (Figure 14B) and they tended to be greater for the extreme treatments (eg. high and low temperature coupled with high and low soil water indicated by the green coloured columns in Figure 16A) with the greatest mean severity of foliar symptoms (27%) occurring on vines at 30ºC with high soil water (Figure 16A). In the same experiment, mean mycelial growth ranged between 20 and 60 mm and tended to be less, although not significantly, in the extreme treatments indicated by the blue columns (Figure 16B).

The water status of grapevines was determined once during each experiment using a pressure chamber and results are shown in Table 4.

Table 4. Water status (pressure in Bars) of grapevines under three irrigation regimes at one time for each experiment involving variable irrigation levels.

Irrigation regime	9 Apr 2008	4 Dec 2008		
		14ºC	22ºC	30ºC
Dry	-5.5	-8.2	-6.5	-8.7
Medium	-5.0	-6.0	-4.6	-4.7
Wet	-4.4	-4.5	-3.8	-4.0

In the nutrient trials, mean pruning weights were significantly different between vines treated and untreated with nutrients. In 2007/8, mean fresh weight of control vines was 3 g and for nutrient treated vines was 23 g. In 2008/9, mean fresh weight of control vines was 7 g whilst for nutrient treated vines was 32 g. Foliar symptoms were not observed in 2007/8. In 2008/9, foliar symptoms occurred in inoculated vines, with no significant difference in mean foliar symptom severity between control vines and nutrient treated vines (Figure 17). No symptoms were observed in non-inoculated vines. Following destructive sampling and assessment, there was no obvious relationships between mean extent of staining and mean extent of colonisation by *E. lata,* with staining of up to 30 mm occurring in non-inoculated vines and in inoculated vines mycelium was isolated up to 70 mm in advance of the staining. In 2007/8, mean mycelial growth was slightly greater in vines that received nutrients (62 mm) than in controls (50 mm) and in 2008/9, mean mycelial growth was significantly greater in the nutrient-treated vines (100 mm) than in controls (70 mm).
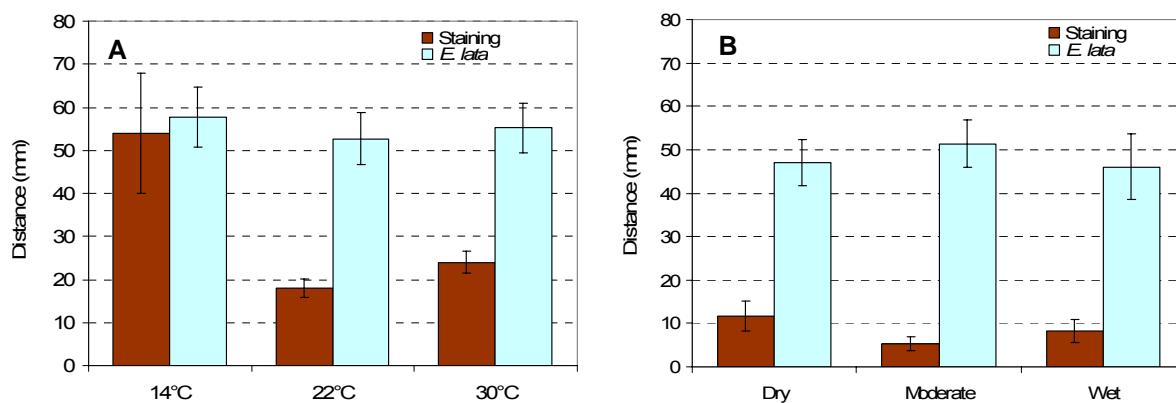
Figure 15. Effect of temperature (A) and soil water (B) on the extent of staining and the isolation of *E. lata* (blue) above and below the point of inoculation point (Bars = Standard error of the mean).
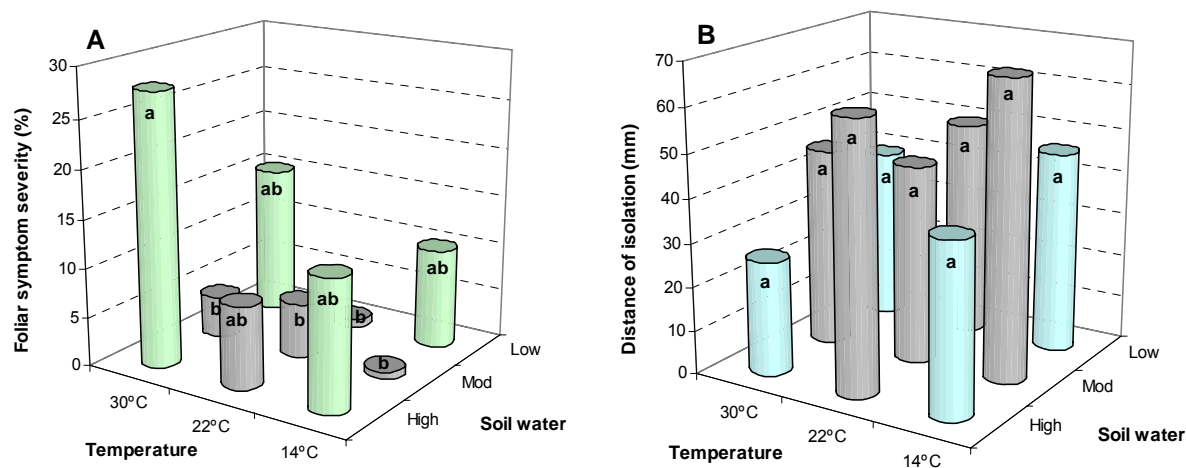

Figure 16. Effect of temperature and soil water on foliar symptoms (A) and the total spread of mycelium of *E. lata* along stems (B) in one combined experiment in 2008/9 (Treatments with the same lower case letter are not significantly different from each other)
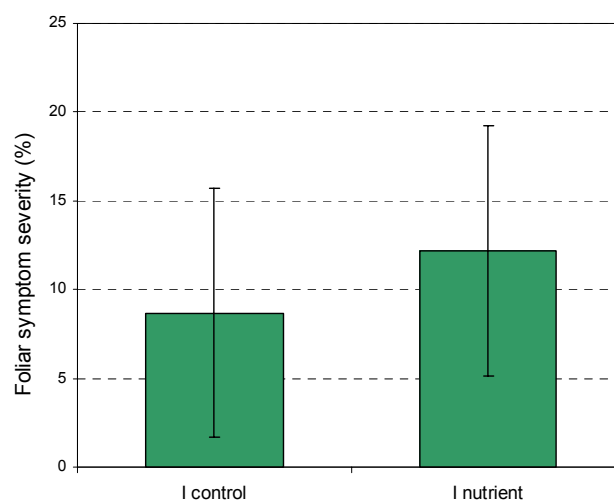

Figure 17. Effect of nutrition on foliar symptom severity in inoculated (I) vines (Bars = Standard error of the mean)
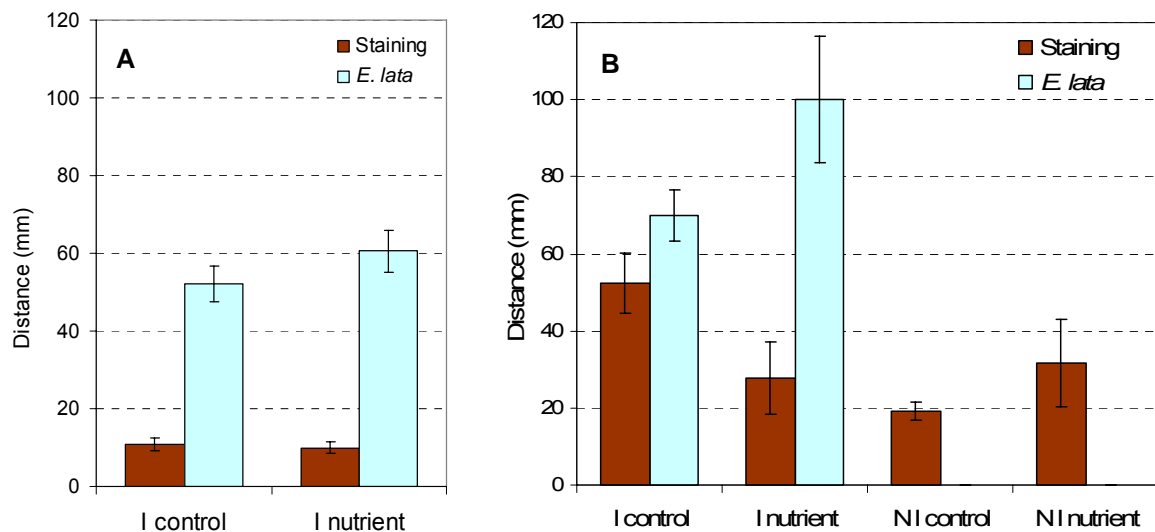
Figure 18. Effect of nutrition on the extent of staining and isolation of *E. lata* (blue) along stems, above and below the point of inoculation in inoculated (I) and non-inoculated (NI) vines in 2007/8 (A) and 2008/9 (B) (Bars = Standard error of the mean)

6.1.4 Discussion

These experiments show that there was poor correlation between staining and mycelial growth of *E. lata*, indicating that staining is not an accurate reflection of the spread of *E. lata* in woody grapevine tissue. Other factors such as wound response may also play a role in the presence of staining (Sosnowski *et al.* 2007b).

Stress imposed by high or low temperature or soil water independently did not affect the development of eutypa dieback symptoms, however when combined, high or low temperature and soil water appears to cause stress which tended to increase foliar symptoms and slow mycelial growth. The pressure chamber results reflect the higher stress (low pressure) experienced by the vines under the dry irrigation regime at 30ºC (-8.7 Bar) and 14ºC (-8.2 Bar) in the 2008/9 experiment compared to the higher pressure (-5.5 Bar) in dry vines from the 2007/8 experiment. Vines are considered to be in water deficit at a pressure of around -10 Bars (Hellman, 2010). The lack of significance in the results may be due to experimental error. In some cases, *E. lata* was isolated from the section taken furthest from the point of inoculation, meaning any further growth was not included in the analysis. In future experiments using the bioassay, attempts to isolate *E. lata* should be made at intervals up to 100 mm in advance of stained wood to ensure accurate detection of the extent of mycelial growth. Furthermore, the experiment was conducted for 1 year and extending this to 2 years may allow further development of foliar symptoms and mycelial growth and hence reveal any significant differences among treatments, as was shown by Sosnowski *et al.* (2007b).

In the nutrient trial, fresh pruning weights were significantly lower for the unfertilised control vines than for vines treated with nutrients, revealing the extent of stress on the vines. Nutrient treatment did not influence foliar symptoms in this study. However, mycelial growth was greater in vines that received nutrients than in controls. As *E. lata* requires nutrients to grow, it is possible that the lack of available nitrogen in vascular tissue of the controls limited the growth of the fungus as well as the vine.

6.2 Effect of regulated deficit irrigation on infection

6.2.1 Introduction

Regulated deficit irrigation (RDI) involves the control and management of water stress by irrigating at less than the full requirement of the vines and maintaining soil moisture at a relatively dry level to control vigour and increase the fruit quality while conserving water. Spores of *E. lata* infect vines through pruning wounds and natural wound response of the vine plays a role in reducing the ability of spores to colonise the tissue. The aim of these field trials was to evaluate the ability of *E. lata* to infect and establish in wounds of vines under different levels of irrigation.

6.2.2 Methods

Two trials were established at the Nuriootpa Research Centre in the Barossa Valley, SA and Loxton Research Centre in the Riverland, SA. In the Barossa Valley, Ruby Cabernet vines planted on own roots in 1993 were either irrigated fully (100% of the allocation in each year) or not irrigated at all (0%) beginning in 2006. In the Riverland, Cabernet Sauvignon vines grafted on Ramsey rootstock and planted in 1992 were subjected to three irrigation levels; 100, 60 and 25% of the standard irrigation program beginning in 2006 (Figure 19). In both vineyards, vines were spur-pruned with cordons.

Long term average data on the two locations were provided by the Bureau of Meteorology. Mean annual rainfall in the Riverland and Barossa Valley are 261 and 472 mm, and mean maximum temperature 23.8 and 21.5ºC, respectively.

One-year-old shoots were pruned to two spurs on 7 and 8 July 2008 in Barossa Valley and Riverland, respectively. In both trials 50 shoots were inoculated for each treatment. Within an hour of pruning, wounds were inoculated with a suspension of 500 spores/ml of *E. lata* as described in Section 5.2.2. On 12 July, wounds were re-inoculated with a suspension of 500 spores/ml of *E. lata*.

On 12 June 2009, treated canes were excised from vines and returned to the laboratory. Samples were assessed by isolating on PDA as described in section 5.2.2. Susceptibility to infection by *E. lata* was determined based on the mean percent recovery of *E. lata* from the treated canes by isolation on PDA.

Figure 19. Deficit irrigation trial at Loxton in the Riverland SA, (A) following pruning and treatment in winter and (B) during summer.

6.2.3 Results and discussion

In the Riverland, *E. lata* was recovered most frequently from vines with reduced irrigation (Figure 20A). The frequency of recovery increased from 74% in fully irrigated vines to 95% in vines that received 25% of the standard irrigation program. In the Barossa Valley, there was no effect of irrigation, with *E. lata* isolated from around 90% of inoculated shoots in both irrigated and non-irrigated vines (Figure 20B).

In the Riverland, reducing the irrigation level from 100 to 25% of the standard irrigation program significantly increased the susceptibility of pruning wounds to infection by *E. lata*. However, in the Barossa Valley, there was no difference in between irrigated and non-irrigated vines. The reason for

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this may be that the mean annual rainfall in Barossa Valley is almost twice as that in the Riverland and mean maximum temperature is 2°C lower so vines are under less stress due to environmental factors and therefore may be able to maintain natural defence against infection when under severe deficit irrigation. In a warm, dry region, deficit irrigation might increase the susceptibility of pruning wounds to infection by *E. lata*. Further research is required to substantiate these suggestions and large scale trials which exist in Langhorne Creek and Riverland regions of SA with a series of different irrigation levels provide an excellent opportunity to further evaluate infection and rate of growth of *E. lata* in stressed vines. In addition, assessment of the extent of colonisation by *E. lata* may provide further evidence of the effect of water stress on increasing the susceptibility of vines to *E. lata*.
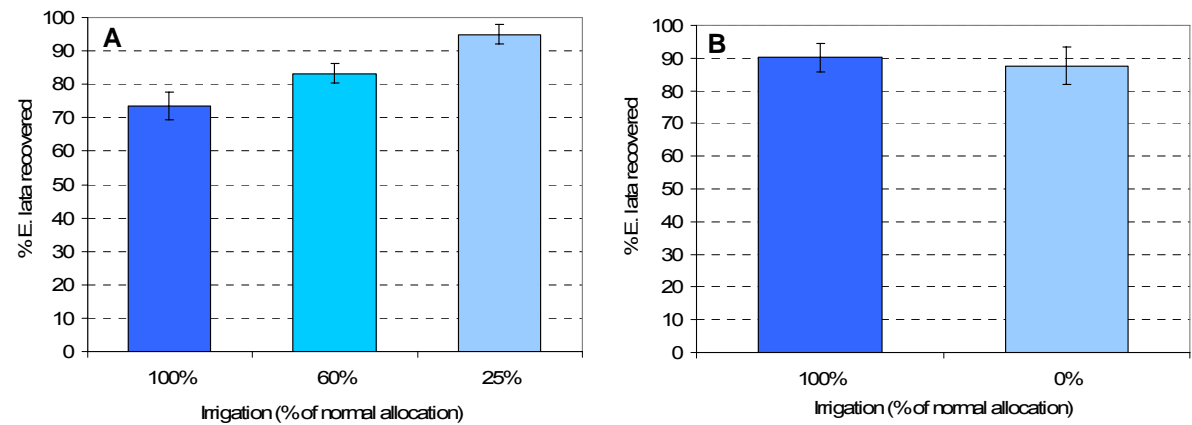

Figure 20. Effect of irrigation level on susceptibility of pruning wounds to infection by E. lata in (A) the Riverland and (B) the Barossa Valley regions of South Australia (bars represent standard error of the mean)

6.3   Relationships between foliar symptoms and climate

Prof Dani Shtienberg (Agricultural Research Organisation, Volcani Center, Israel) met with Mark Sosnowski on 19 Sept 2007 during a visit to South Australia funded by GWRDC. The purpose of the meeting was to discuss validation of a eutypa dieback disease model that Prof Shtienberg developed in collaboration with the South Australian eutypa dieback project team during a sabbatical visit to University of Adelaide and SARDI in 2004/05. The model was designed to predict the incidence of foliar symptoms using data from SARDI trial assessments of five sites over a 6-year period (Sosnowski *et al.* 2007a). Trial assessments continued for the following 2 years and included two new sites. The model was run using the new data and a relationship between predicted and observed disease incidence was evident (Figure 21). This suggests that the factors; rainfall events in early winter and temperature in spring, do influence the incidence of eutypa dieback foliar symptoms. However, the relationship was not consistent with the 1:1 line which implies inaccuracy of the timeframe over which these factors are relevant. This is most likely due to the variability of seasonal phenology in any given region. It was concluded that the current investigation on the effects of water and temperature on the development of disease is important to improve our understanding of the disease and its management.
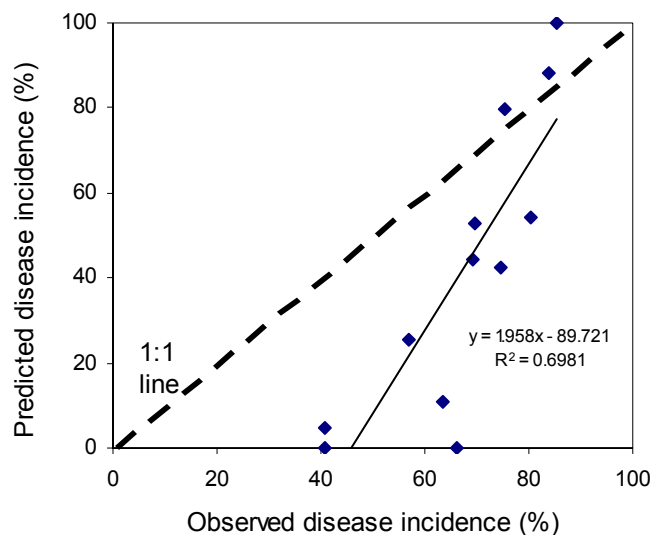

Figure 21. Validation of the model for predicting the incidence of foliar symptoms of eutypa dieback based on initial symptoms and climate. The 1:1 line is a theoretical line presenting a perfect coincidence between predicted and observed values.

7 REMEDIAL SURGERY

7.1 Medium-term effects of remedial surgery treatment of grapevines

7.1.1 Introduction

Eradicating *E. lata* from infected grapevines is very difficult. The application of foliar nutrients as well as injecting fungicides, nutrients and biocontrol agents into the trunk have not controlled eutypa dieback (Sosnowski *et al.* 2006). Many growers 'renew' infected vines by removing diseased wood and reworking, which we have termed remedial surgery. It is important to detect symptoms of eutypa dieback early, so that less wood needs to be removed. When infection is restricted to part of one or both cordons, wood is removed until the cross-sectional wedge of discoloured wood is no longer visible and then a further 10 cm cut out to ensure that all infected wood is removed. A cane is then trained to replace the section of cordon removed. Where cordons are infected and the fungus has progressed into the trunk, two methods may be used to restore vines; low cut or high cut. Thirteen trials were established to monitor the production of watershoots and recurrence of foliar symptoms of eutypa dieback following remedial surgery using low and high cut techniques. Trials were conducted in commercial vineyards where re-working of vines was in progress, which confounded statistical analysis, but did address the need to provide information to assist growers in decision making for disease management.

7.1.2 Methods

Between 1999 and 2004, remedial surgery trials were established on 13 commercial vineyards located in five wine regions of South Australia (Eden Valley, McLaren Vale, Coonawarra, Barossa Valley and Clare Valley). Shiraz, Cabernet Sauvignon, Pinot Noir and Malbec were used, with vines ranging in age from 13 to 43 years. The number of vines in each trial varied between 128 and 1135 and vines were subjected to either low cut or high cut methods of remedial surgery.

The **low cut** method of remedial surgery involved removing the trunk 30-40 cm above the ground, inducing and training a watershoot to replace the trunk and cordons (Figure 22A). The **high cut** method of remedial surgery involved removing the cordons from the top of the trunk (crown) and then selecting and training the lowest available shoots to form a new trunk and cordons (Figure 22B). Therefore shoots arose from either low (bottom third), mid (middle third) or high (top third) on the trunk (Figure 23). Once new shoots were established, any excess wood was removed several years after surgery.
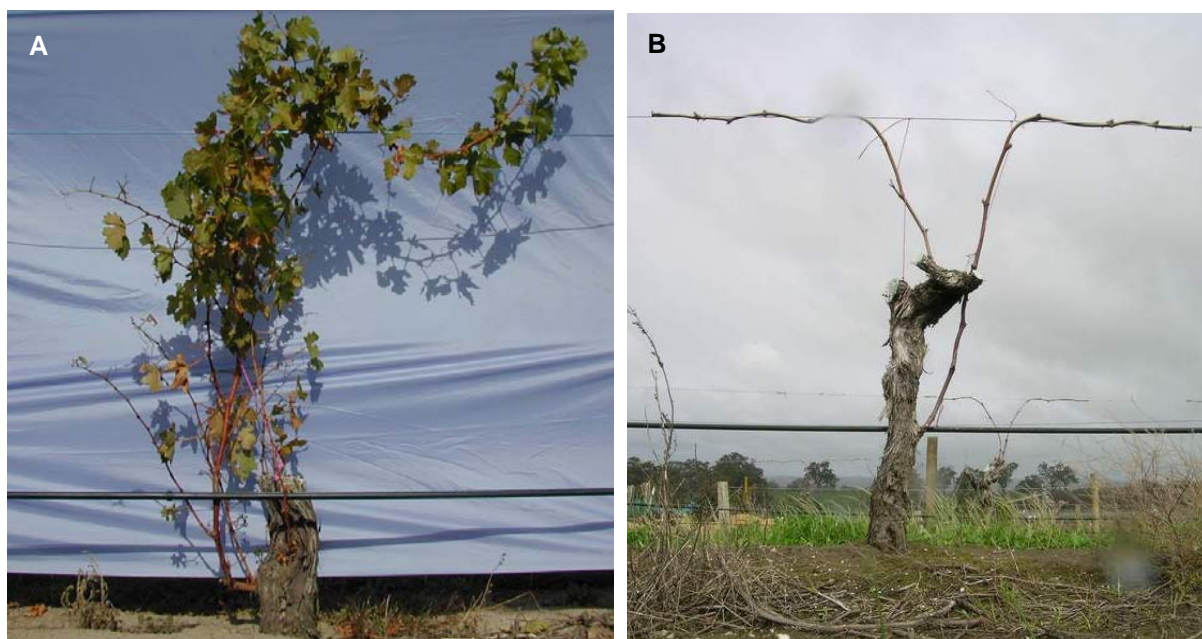

Figure 22. Two methods of conducting remedial surgery; Low cut (A) and High cut (B). In both cases the lowest watershoot is trained up as a new trunk.
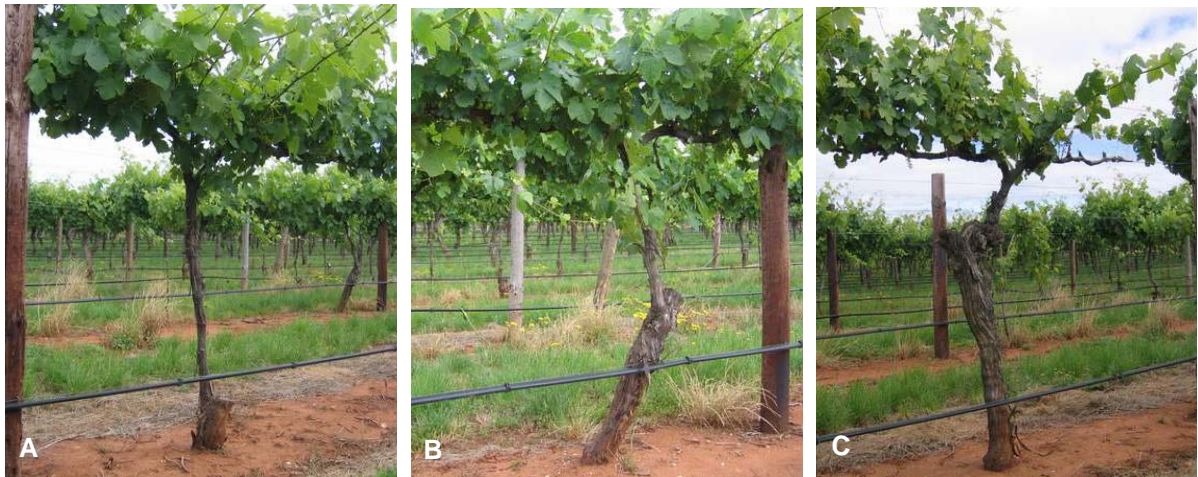
Figure 23. Position on trunk from which the lowest available watershoot arose from; low (A), mid (B) and high (C) following remedial surgery by high cut at the top of the trunk.

Trial 1 was established on Shiraz vines in Eden Valley. In spring 1999, vines were assessed for foliar symptoms of eutypa dieback. During the following winter, vines were cut low (Figure 24A) and wood symptoms were recorded in trunk cross sections. Wounds were treated with either Benlate, Nustar or Trichoseal.

Trials 2 and 3 were established on Shiraz and Malbec vines in Mclaren Vale (Figure 24B). In winter 2000, vines were cut low and then wounds were protected with Benlate mixed into acrylic paint.

Trials 4 and 5 were established on Shiraz vines in the Coonawarra (Figure 24C). Cordons were removed at the crown (high cut) in 2002 and 2003 for trials 4 and 5, respectively. Benlate was applied to wounds immediately after surgery. During the following spring, the lowest available watershoots were trained up to the wire to form new cordons.

Trial 6 was established on Cabernet Sauvignon vines in McLaren Vale (Figure 24D) and **Trials 7 and 8** were established on Shiraz vines in McLaren Vale (Figure 24E) and Eden Valley, respectively. Vines were cut low during the winters of 2002 (Trial 6) and 2003 (Trials 7 and 8). Immediately after cuts were made, wounds were assessed for presence of wood discolouration typical of eutypa dieback. Samples (2-3 cm slices of trunk) were collected for fungal isolation and molecular diagnosis of *E. lata* before wounds were treated with Benlate mixed into acrylic paint (Trial 6) or with Garrison (Trials 7 and 8).

Trial 9 was established on Pinot Noir vines in the Barossa Valley. In spring 2002, vines were assessed for foliar symptoms of eutypa dieback and in winter 2003, vines were cut low (Figure 24F) and cross sectional wood on stumps assessed for wedge shaped staining. Wounds were treated with one of four pruning wound protectants (Benlate, Garrison, Vinevax and acrylic paint) or left untreated (controls).

Trials 10 and 11 were established on Shiraz vines in Eden Valley (Figure 24G) and cordons were removed at the crown (high cut). Vines were cut in winter 2003 and wounds treated with Garrison for Trial 10 and vines were cut in winter 2004 and wounds treated with Vinevax or Spin-Flo (carbendazim) for Trial 11. During the following spring in each trial, the lowest available watershoots were trained up to the wire to form new cordons.

Trial 12 was established on Cabernet Sauvignon vines in Clare Valley (Figure 24H). In winter 2004, cordons were removed at the crown and if a wedge of stained tissue was visible, further cuts were made down the trunk until no stained tissue was visible and then a further 10 cm of trunk was removed.

Each spring following surgery, vines were assessed for presence of watershoots and foliar symptoms of eutypa dieback up until 2008. For vines which were cut high, the position from which the lowest available watershoot originated (low, mid or high) was also recorded (Figure 23).

Trial 13 was established on Shiraz vines in the Eden Valley. In spring 1998, vines were assessed for foliar symptoms of eutypa dieback. In the following winter of 1999, 18 vines (with symptom severity between 10 and 70%) were cut low and a further 18 vines (with similar symptom severity) were left untreated. During spring each year between 2004 and 2006, incidence of foliar symptoms in all 36 vines was assessed.

Figure 24. Remedial surgery trials; A. Trial 1 in Eden Valley, cutting vines in with hydraulic loppers, B. Trials 2 and 3 in McLaren Vale, 4 years after surgery in, C. Trial 4 in Coonawarra, 3 years after surgery, D. Trial 6 in McLaren Vale, 1 year after surgery, E. Trial 7 in McLaren Vale, immediately following lopping of vines, F. Trial 9 in the Barossa Valley, cutting vines with a chainsaw, G. Trial 10 in Eden Valley, 1 year after surgery and H. Trial 12 in Clare Valley, 6 months after surgery.

7.1.3 Results

Watershoot production and recurrence of foliar symptoms in the 13 remedial surgery trials up to 9 years after trunk removal are shown in Table 5.

In the first spring following surgery, watershoot production was variable (Figure 25). Of the low cut vines, incidence of watershoots on Pinot noir and Malbec was 100 and 97%, respectively. On Cabernet Sauvignon and Shiraz, the mean incidence was 55 and 58%, respectively, whereas the mean incidence of watershoots on high cut vines was 90 and 84%, respectively.
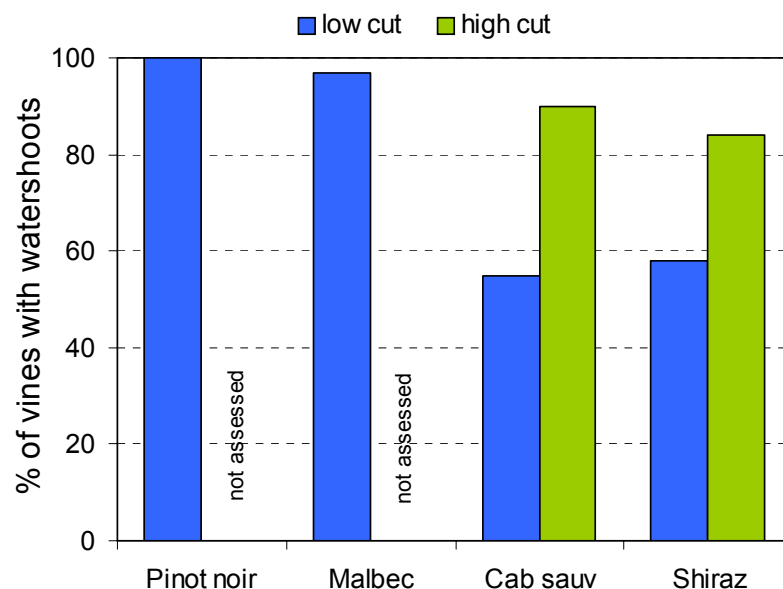

Figure 25. Mean incidence of watershoot production during the spring following remedial surgery of four grapevine cultivars and comparing low cut (blue) and high cut (green) vines. Data is consolidated for all trials involving these cultivars.

The incidence of watershoots developing on vines each spring for 6 years following surgery on Trials 8 and 10 in the Eden Valley is shown in Figure 26. Remedial surgery was conducted in winter of 2003 and the spring of 2003 incidence of water shoots on low cut and high cut vines was 42 and 68%, respectively, and in spring of 2004, 76 and 92%, respectively. For the following four seasons, incidence remained more or less the same, with 75 and 87% in 2008.
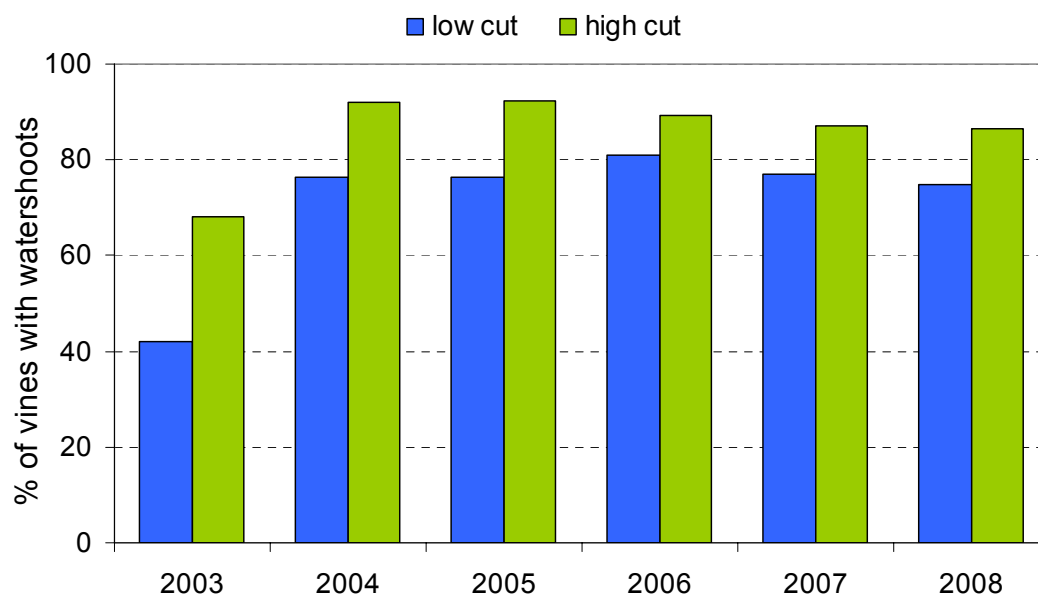

Figure 26. Incidence of watershoot production during spring of each year for 6 years following remedial surgery in winter 2003 on Shiraz vines in Eden Valley and comparing low cut (Trial 8, blue) and high cut (Trial 10, green) vines.

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Table 5. Remedial surgery trials conducted in five regions of South Australia using four cultivars between 2000 and 2004. Pre-surgery foliar and wood symptoms were recorded for some trials and production of watershoots and recurrence of foliar symptoms were assessed for all trials each spring post-surgery until 2008.

Trial	Region	Cv.	Year planted	No. of vines	Cut	Year of surgery	Pre-surgery symptoms (%)		Watershoots (%) years after surgery									Foliar symptoms (%) years after surgery								
							Foliar	Wood	1	2	3	4	5	6	7	8	9	1	2	3	4	5	6	7	8	9
1	EV	Sh	1971	141	low	2000	35	71	65	62	75	76	77	80	80	80	80	0	0	0	1.4	5.8	5.8	3.6	5.1	5.1
2	MV	Ma	1970	871	low	2000	-	-	95	95	94	94	94	94	95	94	94	0	0	0	0.1	0.7	1.0	1.6	1.1	1.4
3	MV	Sh	1970	1135	low	2000	-	-	76	76	83	80	81	80	80	80	80	0	0	0	1.1	5.1	8.4	13	8.5	7.1
4	CW	Sh	1967	200	high	2002	-	-	91	86	87	87	86	85	85			0	0	0	3.6	3.5	8.9	8.9		
5	CW	Sh	1970	144	high	2003	56	-	79	89	89	91	92	91				4.0	0.9	5.2	16	21	29			
6	MV	CS	1971	356	low	2002	-	17	55	52	53	52*						0	0	4.0	11*					
7	MV	Sh	1960	200	low	2003	-	37	51	57	56	55	54	54				0	0	0	0.6	1.5	4.2			
8	EV	Sh	1971	157	low	2003	-	36	42	76	76	81	77	75				0	0	1.3	1.1	5.0	8.8			
9	BV	PN	1990	128	low	2003	0	0	100	98	98	98	98	98				0	0	0	0	0	0			
10	EV	Sh	1971	202	high	2003	-	-	92	92	93	89	87	87				0	0	8.1	11	39	59			
11	EV	Sh	1971	355	high	2004	-	-	68	76	79	80	80					0	1.1	3.4	11	15				
12	CV	CS	1984	151	both	2004	-	0	61	78	77	78	77					0	0	0.9	0.9	3.5				
13	EV	Sh	1971	18	low	1999	89	-	100	100	100	100	100	100	100			-	-	-	5.6	17	0			

EV= Eden Valley, MV = McLaren Vale, CW = Coonawarra, BV = Barossa Valley and CV= Clare Valley
Cv. = cultivar; Sh = Shiraz, Ma = Malbec, CS = Cabernet Sauvignon, PN = Pinot Noir
*Vineyard removed
- = data not collected

In the 13 trials, symptoms developed with varying incidence on reworked vines after remedial surgery (Table 5). With the exception of Trial 5, in which foliar symptoms were evident in the first spring following surgery, it generally took 2 to 4 years before the first foliar symptoms were observed and then the incidence increased at varying rates each year onwards. The highest incidence of symptom recurrence was 59%, observed 6 years after high cut surgery in Trial 10.

The incidence of foliar symptoms before surgery on Shiraz vines in Trials 1 and 5 was 35 and 56%, respectively. Six years after surgery, 6% of low-cut vines in the Eden Valley (Trial 1) developed symptoms compared with 29% on high-cut vines in Coonawarra (Trial 5).

In Trials 2 and 3 in McLaren Vale, where Malbec and Shiraz vines were cut low, 9 years after surgery symptoms were observed on 1 and 7% of vines, respectively.

Trials 8 and 10 were established in adjacent blocks on Shiraz vines planted in 1971. Six years after surgery, 9% of vines cut low (Trial 8) developed foliar symptoms, compared with 59% of vines cut high (Trial 10).

In both Trials 4 and 10, vines were cut high and the lowest available shoots were trained to replace the trunk and cordons. Six years after surgery, watershoots occurred on 184 of 200 vines in Trial 4 and the majority (68%) of vines had shoots arising from high on the trunk (Table 6). In Trial 10, 169 of 202 vines produced watershoots, the majority (72%) of which arose from low on the trunk. Overall, foliar symptoms were recorded on 7 and 18% (Trial 4 and 10, respectively) of vines with shoots arising from low on the trunk, compared with 21 and 85% on vines with shoots arising from the mid trunk and 83 and 72% on vines with shoots arising high on the trunk (Figure 27). The effect of watershoot height on recurrence of foliar symptoms can be seen in Figure 28.

Table 6. Watershoot production on Shiraz vines cut high during remedial surgery in Trial 4 (Coonawarra) and Trial 10 (Eden Valley), showing the number of vines from which shoots arose from either low (bottom third), mid (middle third) and high (top third) on the trunk and the percentage of vines in each category.

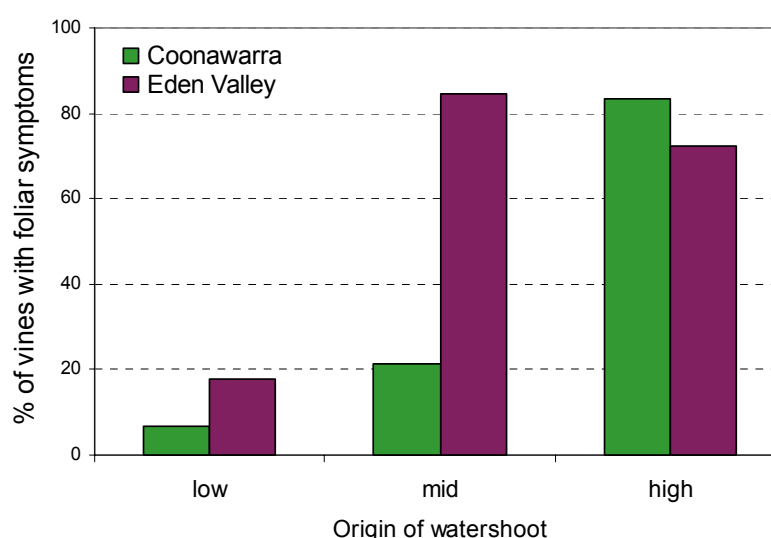| Watershoot height	Trial 4		Trial 10	
	No. of vines	% of total	No. of vines	% of total
Low	45	24	121	72
Mid	13	7	42	25
High	126	68	6	4
Total	184	-	169	-

Figure 27. Recurrence of foliar symptoms on shoots of Shiraz vines (Trials 4 and 10 at Coonawarra and Eden Valley, respectively) which originate from low (bottom third of trunk), mid (middle third) and high (top third) on the trunk, six years after remedial surgery.

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Figure 28. Cabernet Sauvignon vines in the Coonawarra which were subjected to remedial surgery in 2002; A. In spring 2004, healthy, symptomless vines in foreground with high shoot in background with low and mid shoots (arrows), B. In spring 2007, dead vine in foreground and healthy symptomless vines in background (arrows).

In Trial 13, 89 and 94% of vines assigned at random to be untreated or cut low (trunk removed), respectively, exhibited foliar symptoms of eutypa dieback in 1998, prior to remedial surgery (Figure 29). Between 5 and 7 years after surgery, foliar symptoms were recorded on 72 to 100% of untreated vines, compared with 0 to 17% on vines where the trunks and cordons were removed.
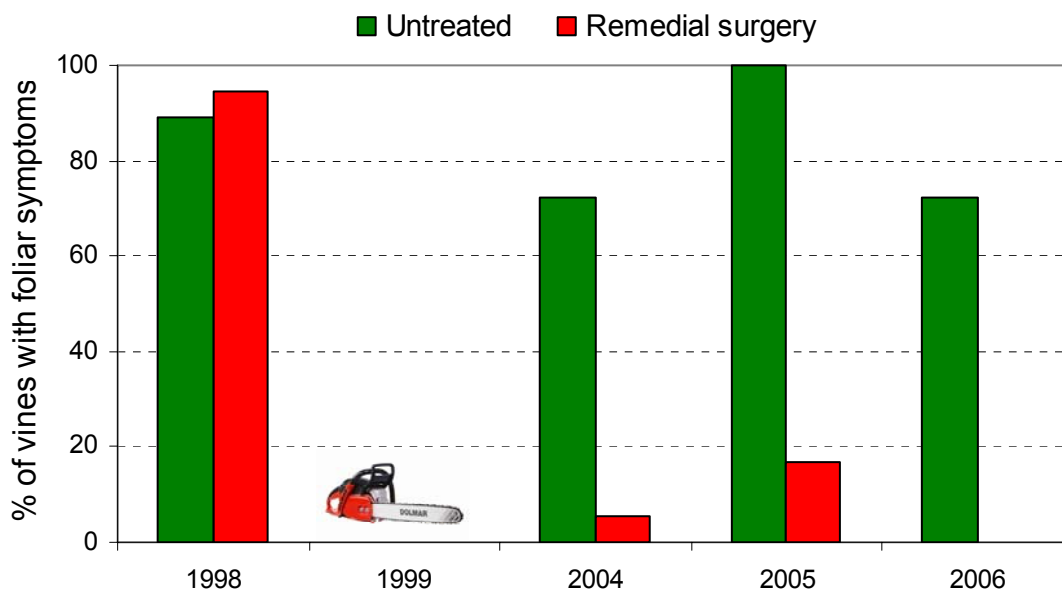

Figure 29. Foliar symptoms of eutypa dieback pre and post-remedial surgery (red) conducted with low cuts in 1999 on Shiraz vines in the Eden Valley, compared with vines left untreated (green).

7.1.4 Discussion

In this study, remedial surgery was shown to be an effective means of managing eutypa dieback affected grapevines in the medium-term. The production of new watershoots varied for different cultivars following removal of cordons and trunks. The greater amount of trunk retained after surgery, the greater the likelihood that watershoots would be produced in the first season but this difference decreased by the second season and beyond. The incidence of watershoot production generally plateaued by the second season and, in the case of Shiraz, reached a maximum of about 80% in

these trials. When watershoots do not appear after two seasons, it is generally recommended to either replant or use a layering technique to replace vines. Layering is the use of a healthy cane from a neighbouring vine which is buried in the soil to induce root growth and hence create a new vine in the position of the old dead stump (Nicholas *et al.* 2001). The new vine is severed from the mother vine once the root system is sufficient to maintain the young vine. Layering results in a fully producing vine in a shorter period of time than replanting, due to the established root system of the mother vine.

In general, remedial surgery reduced the incidence of vines with eutypa dieback symptoms. However the reduction depended on a number of factors including; cultivar, incidence and extent of pre-existing infection and, most importantly, the origin of the watershoot.

Foliar symptom expression varies between cultivars (Section 8; Carter 1991), which may explain differences seen in these trials, however, as this study focussed mainly on Shiraz, other cultivars were poorly represented. In some trials, pre-existing incidence of foliar symptoms appeared to influence the recurrence of symptoms after surgery. However, most evident from this study was the effect of position on the trunk from which watershoots arose. The lower the watershoot, the less likely symptoms were to recur in a vine. The reason for this may have been that *E. lata* infection extended varying distances down into the trunk in most vines, evident by the varying incidence of wedge-shaped staining remaining in the cross section of stumps following surgery. If shoots arose from above the infected wood, symptoms were most likely to recur in the new shoots (Figure 30). Therefore it is recommended to make cuts down the trunk until no staining can be seen and then cutting a further 10 cm or more into healthy tissue, to remove any fungus advancing ahead of stained wood (Sosnowski *et al.* 2007b). Furthermore, simply cutting all vines low (around 30 cm from the ground) will minimise time to undertake remedial surgery on a large scale, and growers might rework a small section of a vineyard each year to minimise production losses.

Remedial surgery maintains the existing rootstock, but the roots may suffer from a loss of photosynthates as a result of the canopy removal. Commercial yield is delayed for several years until the new canopy is established compared to a longer period to equivalent production from new plantings. Anecdotal evidence suggests that fruit quality is also restored within several years, which is not the case when replanting.

Remedial surgery proved to be an effective method of controlling eutypa dieback and maintaining the longevity of premium vines. However, there is no guarantee that the disease is eradicated. It is important to rework from vines which all stained wood, plus an additional 10 cm of healthy wood, is removed, to minimise the recurrence of symptoms. Finally, all wounds should be protected from further infection by *E. lata* (Section 5).

Figure 30. Shiraz vine in Eden Valley showing eutypa foliar symptoms (left) arising from high on the trunk and healthy shoot growth (right) arising from low on the trunk.

7.2   Cost-benefit analysis of remedial surgery

Remedial surgery is labour-intensive and the decision on whether to proceed needs to be made on a case by case basis. The benefits of this management strategy may outweigh the costs if the quality and value of the fruit is above average and if the disease has not progressed to ground level in trunks of vines. This study aimed to conduct a cost-benefit analysis based on data collected during GWRDC and CRCV eutypa dieback projects which would contribute to developing a decision support system to aid growers manage eutypa dieback effectively. Economic modelling was conducted by Ian Black, (SARDI Economist), Ian Trengrove (PIRSA Farm Management Economist) and Chris Dyson (SARDI Statistician) in consultation with members of the eutypa project team.

A full report can be found in Appendix 1 and summary points are provided below:

- The cost-benefit analysis has provided a preliminary decision support model which is able to extrapolate and predict economic returns of different decision scenarios in a Shiraz vineyard affected by eutypa dieback.

- Maintenance of vineyards in a disease-free condition following remedial surgery by expending approximately $50 per hectare each year to monitor and remove any diseased vines that appear will ensure continued good economic returns for the vineyard.

- The cost of retrellising made little difference to the outcome of the model, suggesting it is a small investment for sustainability of vines.

- In general, if the incidence of foliar symptoms is below 10%, remedial surgery can be delayed several years without incurring significant losses, however once incidence exceeds 20% it is imperative action is taken to avoid long-term economic losses.

- However, the model is limited to Shiraz vines as most data available concerned this cultivar and, due to variable susceptibility to eutypa dieback, further data need to be generated for a range of commonly grown cultivars.

8   INCIDENCE AND THREAT OF EUTYPA DIEBACK

8.1   Emerging regions in South Australia

8.1.1   Introduction

E. lata is widespread in most areas of the world where grapevines are cultivated. At the beginning of this project, eutypa dieback had been officially reported in South Australia and Victoria (Wicks 1975, Carter 1991, Highet and Wicks 1996) and since then Pitt *et al.* (2010) have reported *E. lata* in New South Wales. In 1968 and 1997, the extent of eutypa dieback symptoms was documented for established wine regions of South Australia such as Barossa Valley, McLaren Vale, Coonawarra, Langhorne Creek, Riverland and Clare. The incidence of eutypa dieback in emerging wine regions such as the Adelaide Hills, Fleurieu Peninsula and Mt Benson in South Australia has not been determined, so little preventative action is being taken. Determining the presence and extent of the disease in these regions would alert and encourage growers to monitor for disease symptoms and undertake preventative action, if warranted.

8.1.2   Methods

The incidence of eutypa dieback was assessed in the wine regions of Adelaide Hills (AH), Mt Benson (MB) and Southern Fleurieu (SF) in October and November of 2006, 2007 and 2008, respectively.

Vineyards aged 6 years and older were selected and 200 vines per block visually assessed by walking along the vine rows (4 rows x 50 vines). At least one block of each cultivar planted was assessed in every vineyard visited. Vines with foliar symptoms characteristic of eutypa dieback (Section 3) on at least two shoots were recorded and the incidence of symptomatic vines per block calculated. Many vineyards were visited in each region and cultivar, vine age and location co-ordinates were recorded for each block surveyed using a hand-held Garmin 60 Global Positioning System (GPS) unit.

To confirm that foliar symptoms were caused by *E. lata*, wood samples with necrotic staining in cross-sections were collected from 33 blocks, representing most vineyards surveyed. Samples were returned to the laboratory, surfaced sterilised and small pieces plated onto PDA (Sosnowski *et al.*, 2007b). Cultures were incubated at 23°C for 7 days and those which were identified as *E. lata* based on morphology were confirmed using PCR analysis of DNA as described by Lardner *et al.* (2005).

8.1.3   Results

The severity of foliar symptoms ranged from very mild, with only one spur position with typical symptomatic shoots, to severe, with most shoots on the vine showing symptoms.

In **Adelaide Hills**, foliar symptoms of eutypa dieback were recorded on 4% of all vines and in 90% of all vineyards surveyed (Table 7; Figure 32). The greatest incidence of symptoms recorded was 27% in 10-year-old Cabernet sauvignon with incidence of above 20% in a further six blocks, including Shiraz, Cabernet Sauvignon and Chardonnay vines between 16 and 25 years old.

In **Mt Benson**, foliar symptoms of eutypa dieback were recorded on 0.02% of all vines and in 15% of all vineyards surveyed (Table 7; Figure 32). One vineyard had two vines with symptoms in 12-year-old Cabernet Sauvignon block and a second vineyard had one vine with symptoms in a 14-year-old Cabernet Sauvignon block.

In **Southern Fleurieu**, foliar symptoms of eutypa dieback were recorded on 2% of all vines and in 15% of all vineyards surveyed (Table 7; Figure 32). The highest incidences recorded in individual blocks were 34 and 16%, both in 19 yo Cabernet Sauvignon, with all other blocks having less than 10% of vines with symptoms.

The youngest vines with foliar symptoms were 7-year-old Tempranillo in AH and Sauvignon Blanc in SF. In general, the incidence of foliar symptoms increased with vine age, as is shown for AH in Figure 31, although there was no such correlation in SF or MB (data not shown).

The incidence of foliar symptoms was observed to be greatest in Cabernet Sauvignon (up to 14%) and Shiraz (up to 9%) compared with all other cultivars assessed (Table 8). Of the most common cultivars represented in this study (highlighted in grey), Merlot, Pinot Noir and Sauvignon Blanc were recorded to have the lowest incidence of foliar symptoms (up to 2%).

Table 7. Number of vines, blocks, vineyards, cultivars and age ranges of vines assessed for foliar symptoms of eutypa dieback in Adelaide Hills, Mt Benson and Southern Fleurieu wine regions of South Australia

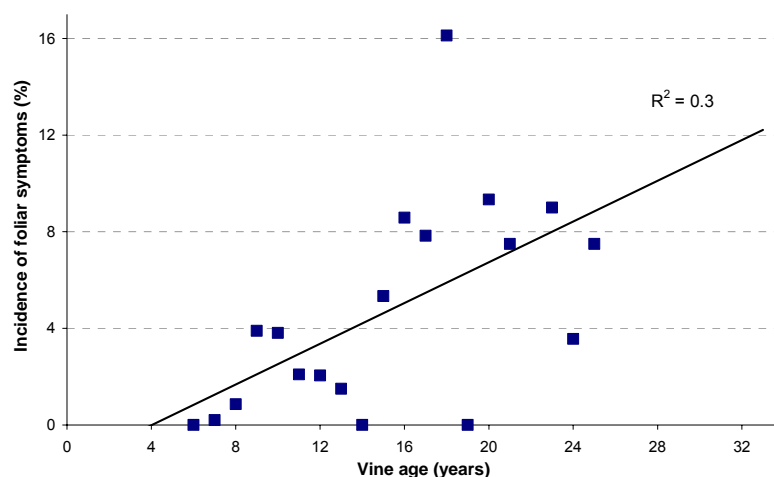|	Adelaide Hills		Mt Benson		Southern Fleurieu	
	No. Assessed	No. with Symptoms	No. Assessed	No. with Symptoms	No. Assessed	No. with Symptoms
Vines	25800	958 (4%)	18400	3 (0.02%)	13800	252 (2%)
Blocks	129	74 (57%)	92	2 (2%)	69	38 (55%)
Vineyards	29	26 (90%)	13	2 (15%)	20	16 (20%)
Cultivars	18	13 (72%)	13	1 (8%)	12	9 (75%))
Age range (y)	6-100	7-100	6-18	12-14	7-33	7-19

Figure 31. Influence of vine age on foliar symptom expression of eutypa dieback in the Adelaide Hills wine region during spring 2006.

Table 8. Grape cultivars surveyed and incidence of foliar symptoms of eutypa dieback in the Adelaide Hills (surveyed 2006), Mt Benson (2007) and Southern Fleurieu (2008) wine regions. Most common cultivars (>10 blocks assessed) are highlighted in grey.

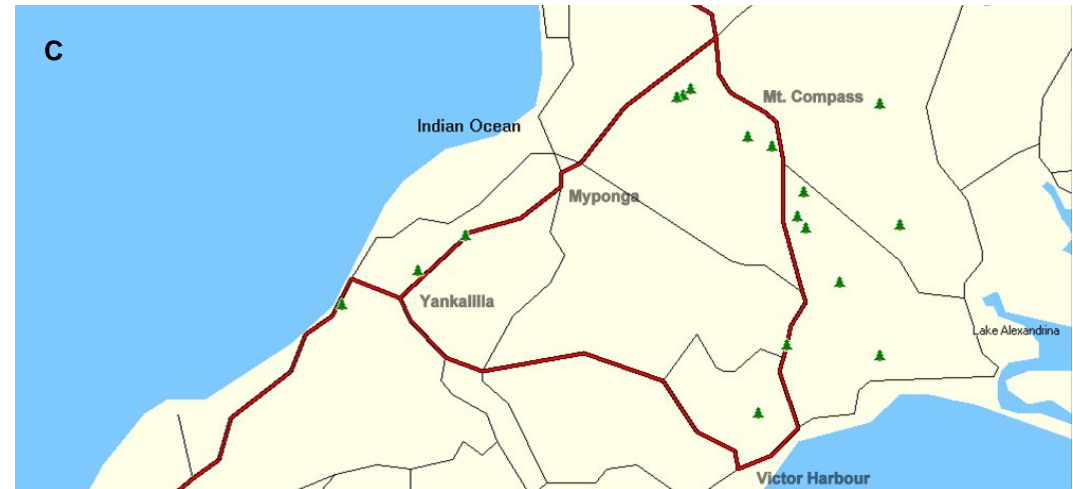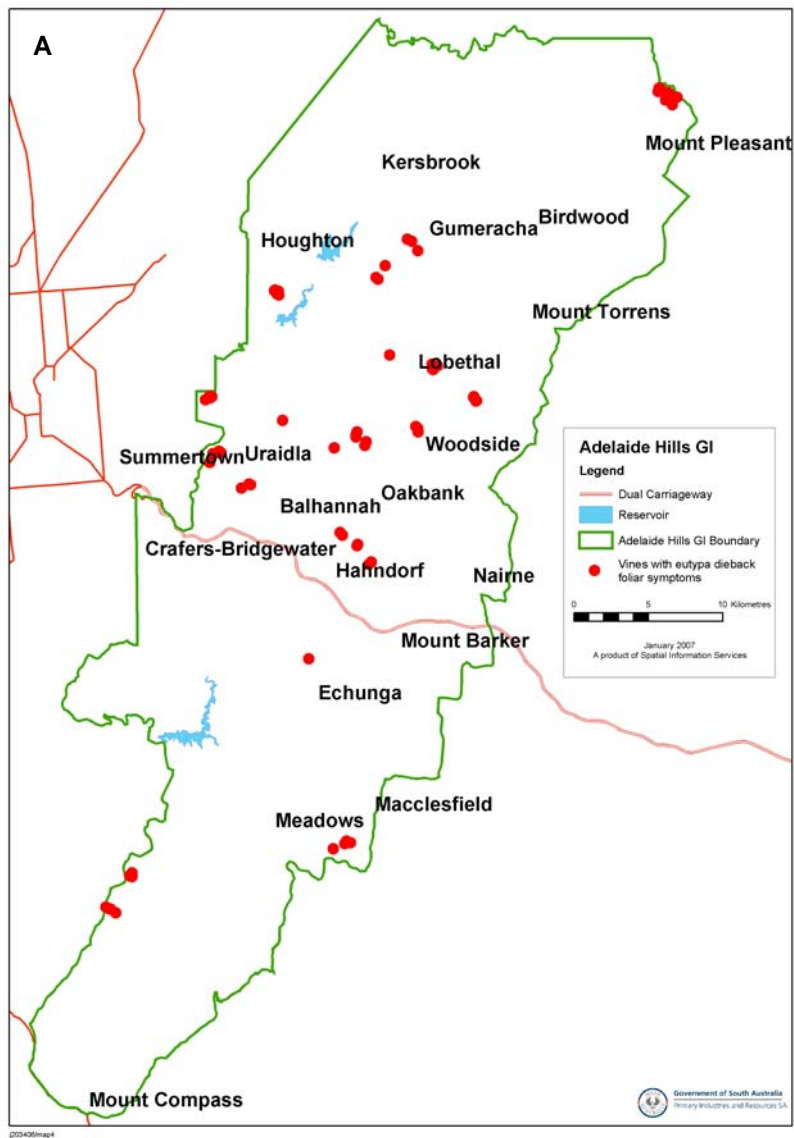| Cultivar	Adelaide Hills		Mt Benson		Southern Fleurieu	
	Blocks assessed	% symptomatic	Blocks assessed	% symptomatic	Blocks assessed	% symptomatic
Cabernet Franc	1	6	1	0	n/a	n/a
Cabernet Sauvignon	10	14	12	0.06	12	5
Chardonnay	27	4	9	0	12	1
Grenache	n/a	n/a	n/a	n/a	1	5
Merlot	9	0.06	13	0	7	0.2
Petite Verdot	1	0	1	0	n/a	n/a
Pinot Gris	5	1	2	0	1	0
Pinot Meunier	1	0	n/a	n/a	1	0
Pinot Noir	17	2	1	0	4	0.3
Riesling	4	2	1	0	2	0
Sangiovese	1	0	n/a	n/a	n/a	n/a
Sauvignon Blanc	28	2	11	0	7	1
Semillon	2	1	4	0	6	2
Shiraz	12	9	21	0	14	3
Tempranillo	2	3	n/a	n/a	n/a	n/a
Traminer	1	2	n/a	n/a	n/a	n/a
Verdelho	1	0	2	0	n/a	n/a
Viognier	5	1	1	0	2	3
Zinfandel	2	0	n/a	n/a	n/a	n/a
Mean	n/a	4	n/a	0.02	n/a	2

n/a = not applicable

Figure 32. Maps of the Adelaide Hills (A), Mt Benson (B) and Southern Fleurieu (C) wine regions, indicating the location and frequency of grapevine blocks (red dots) and vineyards (green triangles) which were recorded as having symptoms of eutypa dieback.

E. lata was isolated from 71, 93 and 100% of the wood samples collected in AH, SF and MB, respectively. All isolates were identified by morphology and confirmed as *E. lata* using PCR analysis. An isolate of *E. lata* from AH (DAR 79094) was lodged with the NSW Plant Pathology Herbarium.

8.1.4 Discussion

Foliar symptoms were found on 4% of vines in the Adelaide Hills, which was similar to the incidence of 4.9% of all vines surveyed across established South Australian wine regions by Highet and Wicks (1998). This suggests that eutypa dieback is now a major disease problem in the Adelaide Hills and growers are recommended to apply control strategies accordingly (Sosnowski *et al.* 2009). In Southern Fleurieu, the incidence of eutypa dieback symptoms was 2%, indicating that growers in this region should be alerted to the problem that is likely to increase in the future unless appropriate management strategies are adopted. Eutypa dieback was rare in the Mt Benson region, which may be attributed to a combination of young vineyards, geographical isolation from other vineyards, lack of infected alternative hosts and the prevailing wind direction (from the ocean) resulting in decreased distribution of ascospores of *E. lata*. At the time of the survey, diseased wood of infected vines was removed, however, non-symptomatic vines may still remain so wound protection is imperative in this region to maintain this low incidence of disease.

The youngest vines observed to express foliar symptoms in this study were 7 years old compared with the survey by Wicks (1975) who assessed vines from 3 yo, and the youngest symptomatic vines were 16 years old. This may reflect an increase in prevalence of eutypa dieback in South Australia along with the increase in grapevine plantings over the past two decades. The greater incidence of foliar symptoms observed on older vines compared with younger vines, which was also reported by Wicks (1975), is likely to reflect repeated exposure to infection over many years, an extended incubation period and increased number and surface area of pruning wounds, which present an increasingly large infection court as the vine ages.

The difference in foliar symptoms observed between cultivars confirmed previous reports that Grenache, Cabernet Sauvignon and Shiraz express more severe symptoms than other cultivars (Carter 1991, Wicks 1975, Highet and Wicks 1998). In the current survey, only one block of Grenache was surveyed, reflecting the reduction in planting area of this cultivar in the past decade. It is important to note also that a delay of 1 to 8 years can occur between infection and expression of foliar symptoms (Moller and Kasimatis 1978, Tey Rulh *et al.* 1991) and symptoms vary from year to year (Sosnowski *et al.* 2007b). Results of this survey based on foliar symptoms, may be conservative, and the actual incidence of infected vines may be greater.

As a result of surveys, workshops were presented in each region to inform growers of recommended strategies for managing eutypa dieback disease in grapevines. Information from the surveys serves as a warning to grape growers of the disease incidence in each region, and such awareness may help to alleviate future economic losses through implementation of disease management strategies (Sosnowski *et al.*, 2009).

8.2 Tasmania

Dowson (1931) and Wade (1960) reported symptoms of dieback disease of apricots in Tasmania and Carter (1957) isolated *Eutypa armeniacae* from apricot wood samples from Tasmania; the fungus was later renamed *E. lata* (Carter 1991). However, eutypa dieback has not been officially recorded in grapevines in Tasmania, although anecdotal evidence suggested the disease occurs in some vineyards.

From 3-7 December, 2007, 16 vineyards were visited in five wine regions; Tamar Valley, North East, East Coast, Coal River Valley and Derwent Valley as part of the GWRDC-funded project RT 0702-5. In total 44 blocks were surveyed, initially by looking for foliar symptoms. Cultivars included Cabernet Franc, Cabernet Sauvignon, Chardonnay, Merlot, Pinot Gris, Pinot Noir, Riesling, Sauvignon Blanc, Shiraz and Traminer. In each block in which foliar symptoms were observed along with four in which no symptoms were observed, cordons and trunks were cut to expose any necrotic staining and samples were collected and returned to SARDI for isolation of *E. lata* (Sosnowski *et al.* 2007b), morphological identification (Carter 1991) and confirmation with PCR analysis (Lardner *et al.* 2005). An apricot orchard in the Coal River Valley was also visited.

Foliar symptoms were recorded in all 5 of the wine regions visited, 10 of the 16 vineyards and 18 of the 44 blocks assessed in Tasmania. Symptoms ranged from very mild, with one or two shoots affected, to severe with the majority of shoots affected (Figure 33). *E. lata* was positively identified in all but two samples from vines with foliar symptoms and two samples from vines without foliar symptoms also tested positive for *E. lata*. Dieback symptoms were observed in the one apricot orchard visited and PCR analysis confirmed the presence of *E. lata*. One isolate of *E. lata* from each region surveyed in Tasmania was lodged with the NSW Plant Pathology Herbarium (DAR 79096-79100).

Potted Grenache vines were inoculated in a shadehouse, using methods described by Sosnowski *et al.* (2007b), with one isolate from each of the five Tasmanian regions visited. Foliar symptoms were observed 8 months after inoculation and *E. lata* was reisolated and confirmed by PCR from vines inoculated with each of the isolates.

Table 9. Samples collected in Tasmanian wine regions and from which *E. lata* was isolated and confirmed

Sample ID	Region	Cultivar	Foliar symptoms	*E. lata* culture	*E. lata* DNA*
TAS24	Tamar Valley	Cabernet Sauvignon	yes	yes	+
TAS25	Tamar Valley	Cabernet Sauvignon	yes	yes	+
TAS19	Tamar Valley	Chardonnay	yes	yes	+
TAS8	Tamar Valley	Sauvignon Blanc	yes	yes	+
n/a	Tamar Valley	Pinot Noir	yes	no	n/a
n/a	Tamar Valley	Pinot Noir	yes	no	n/a
TAS17	North East	Chardonnay	yes	yes	+
TAS18	North East	Pinot Noir	yes	yes	+
TAS28	North East	Sauvignon Blanc	yes	yes	+
TAS1	East Coast	Cabernet Sauvignon	yes	yes	-
TAS2	East Coast	Cabernet Sauvignon	yes	yes	+
TAS7	Coal River Valley	Apricot	yes	no	+
TAS16	Coal River Valley	Cabernet Franc	yes	yes	+
TAS5	Coal River Valley	Cabernet Sauvignon	yes	yes	+
TAS14	Coal River Valley	Cabernet Sauvignon	yes	yes	-
TAS21	Coal River Valley	Chardonnay	no	yes	+
TAS22	Coal River Valley	Pinot Gris	yes	yes	+
TAS4	Coal River Valley	Pinot Noir	yes	yes	-
TAS11	Coal River Valley	Pinot Noir	no	yes	+
TAS15	Coal River Valley	Shiraz	yes	yes	-
TAS27	Derwent Valley	Cabernet Sauvignon	yes	yes	+
TAS29	Derwent Valley	Pinot Noir	yes	yes	+

*PCR assay (Lardner *et al.* 2005)

n/a = not applicable

This is the first confirmed report of *E. lata* in grapevines in Tasmania. General observations were that many of these vines had large pruning wounds which had not been protected. Although there are previous reports of apricot dieback symptoms in Tasmania, this study has provided positive identification of *E. lata* from apricot. It is likely that the pathogen has spread from apricots and other stone fruits and alternative hosts to new grapevine plantings. It is imperative that growers manage vines to protect from infection by *E. lata* (Sosnowski *et al.* 2009)*.*

Figure 33. Tasmania survey; A. Three symptomatic Chardonnay vines in a vineyard in the Tamar Valley, B. Characteristic foliar symptoms on Pinot Noir vines in the North East, C. Symptomatic Cabernet Sauvignon on the East Coast, D. Symptomatic Cabernet Sauvignon in the Coal River Valley, E. Cross section of an infected trunk showing wedge-shaped staining, F. External trunk canker.

8.3 Western Australia

Carter (1975) observed "dying arm" symptoms in two vineyards near Wanneroo, WA and wood samples were confirmed as containing *E. armeniacae* in the laboratory. More recently, "*Eutypa*" was reported in a diagnostic sample with esca disease symptoms from the Margaret River region, although this was not confirmed as *E. lata* with DNA analysis (Edwards and Pascoe 2004). In 2003, a survey was undertaken in south-western WA for trunk pathogens associated with dieback of grapevines; *Botryosphaeria* spp. were isolated from wood, revealing that botryosphaeria canker occurred in many vineyards (Taylor *et al*. 2005). *E .lata* was not identified in any samples assessed in that survey.

In November 2009, 17 vineyards were visited in the Swan Valley, Margaret River and Great Southern wine regions of WA to look for symptoms of trunk diseases as part of the GWRDC-funded project RT 08/02-1. In each vineyard, 2 or 3 blocks were surveyed with at least 200 vines in each block assessed. No foliar symptoms of eutypa dieback were observed. Dieback and wedge shaped stained wood were present in all areas, particularly in older vines, suggesting that Botryosphaeria canker is widespread in WA, which concurs with the previous survey by Taylor *et al*. (2005).

Investigation of fruiting bodies on dead grapevine wood collected in vineyards was carried out by Dr Florent Trouillas, visiting from University of California, Davis USA on a GWRDC travel grant (GWT 09/05). Together with isolations from cankers in wood carried out at SARDI, species of *Bortryosphaeria*, *Eutypella* and *Cryptovalsa* were identified, but not *E. lata*.

The lack of a substantial stone fruit industry in WA may provide a possible reason for the scarcity of eutypa dieback. Stone fruit are highly susceptible to *E. lata* and have been implicated as a source of inoculum in other southern states of Australia (Carter 1991).

9 OUTCOMES AND RECOMMENDATIONS

Output 1

Practical and efficient application methods for wound protectants against eutypa dieback

Performance targets:

- Evaluate potential alternative fungicides and biocontrol agents for prevention of infection by *E. lata* for wound protection

- Evaluate commercial spray equipment for the application of fungicides for wound protection for efficient prevention of eutypa dieback infection in large-scale vineyards

Twenty-nine treatments were evaluated in the laboratory and field for control of infection by *E. lata*. Of these, five physical barriers, six fungicides and two natural products controlled eutypa dieback disease in grapevines.

Physical barriers such as Gelseal, Greenseal, Garrison, Agseal and an acrylic paint were effective and are recommended for use especially on large wounds, such as those made during restructuring and remedial surgery on grapevines. Greenseal, which contains the fungicide tebuconazole, has been registered for use against *E. lata* in grapevines in Australia and steps are being made towards similar registration for Garrison and Gelseal. Acrylic paint does not require registration for wound protection in grapevines. The use of paint alone provides a physical barrier to infection by *E. lata* spores, which plays a major role in the efficacy of paints and pastes.

Bavistin (carbendazim) was the most effective fungicide in these trials, supporting previous work (S2.2.4 - CRV 03/06S). Following submission of an application for label extension for *E. lata* on grapevines, it has been announced that carbendazim is under review due to concerns about operator safety and, therefore, could be removed from the market (as occurred with the related fungicide, benomyl, in 2002). In our trials, Folicur (tebuconazole) was as effective as Bavistin for the control of eutypa dieback, suggesting that this may be a potential product for future registration for eutypa dieback control. Furthermore, Scala (pyrimethanil), Switch (cyprodinil + fludioxonil) and Shirlan (fluazinam) were also effective when applied at rates higher than currently registered for other diseases on grapevines. Further evaluation of these fungicides is required to determine optimal rates and to provide data to support an application to APVMA for label extension for eutypa dieback control.

Natural products, garlic and lactoferrin, were not effective when applied at 1% concentration, however, field evaluation of higher concentrations may provide a better indication of their control of eutypa dieback, in the light of *in vitro* results which showed garlic and lactoferrin were effective at 10%. If effective in the field, these treatments might provide an excellent alternative to fungicides.

Collaborative trials with the botryosphaeria canker research team at the National Wine and Grape Industry Centre in NSW have revealed potential fungicides for dual control of eutypa dieback and botryosphaeria canker. Shirlan, Folicur and Bavistin are all candidates for dual control and further evaluation of these treatments against other Diatrypaceaous trunk pathogens as well as fungi which cause Petri disease and esca, could eventually lead to recommendations of products for the control of all trunk diseases which occur in Australian vineyards.

Spray application of fungicide to protect pruning wounds has shown potential to improve the efficiency of eutypa dieback control on large large-scale vineyard plantings. Trials have shown that similar levels of control are achieved with Bavistin applied either with a commercial spray machine or by hand with a brush. However, the control was variable when different rates of fungicide and water volumes were applied to the dormant vines. Further research is required to determine the optimal fungicide rates and water volume required for air-blast, air-shear, fan and recycle sprayers. Other promising fungicides, such as Folicur, should also be tested for efficacy when sprayed on vines, as well as bio-control agents and alternative products effective against *E. lata*. Efficacy data for spray application of pruning wound protection are not available, yet this information is critical if this method is to be developed and used by vineyard operators. It is crucial that this research continues in Australia.

Output 2

Improved management practices based on knowledge of effects of environmental and production stresses on eutypa dieback

Performance targets:

- Assess the effect of moisture, temperature and nutritional stresses on rooted cuttings infected with eutypa dieback in controlled environments

- Inoculate field trials already established at Nuriootpa and Loxton Research Centres to assess the effect of deficit irrigation on eutypa dieback infection

- Elucidate relationships between foliar symptoms and climate

Controlled environment experiments suggested that the susceptibility of vines to eutypa dieback is influenced by water and temperature stress. Independently, water or temperature stress did not alter susceptibility of the vines to foliar symptoms or colonisation of the wood by *E. lata*. However when combined (e.g. high or low water + high or low temperature), foliar symptoms of eutypa dieback were increased. In contrast, colonisation of the wood by *E. lata* tended to be less with combined stress, although differences were not statistically significant. This shows that although stressed vines display foliar symptoms, the fungus may be inhibited when the vine is under stress. To test this hypothesis, further investigation is required in which vines would be subjected to these stress factors over at least two seasons.

A water deficit trial in the Riverland suggested that water stress increased the susceptibility of vines to pruning wound infection by *E. lata*. At the Barossa Valley site, there was no difference between irrigated and unirrigated vines, whereas in the Riverland, vines with a 75% reduction of irrigation had a 21% increase in infection of pruning wounds. It is likely that the susceptibility to pruning wound infection of vines under deficit irrigation was exacerbated by the lower rainfall and higher temperatures experienced in the Riverland compared to Barossa Valley. Therefore, in warm, dry regions, deficit irrigation may lead to increased susceptibility of pruning wounds to infection by *E. lata*.

Further research is required to substantiate these findings. Large scale trials have been set up in the Langhorne Creek and Riverland regions of SA with a series of different irrigation levels and provide an opportunity to further evaluate infection and rate of growth of *E. lata* in water stressed vines. This information is especially important for the sustainable management of grapevines in drought conditions currently being experienced in southern Australia and with the predicted climate change in the future.

Nutrition deficiency did not influence expression of foliar symptoms in this study. However, mycelial growth was greater in vines receiving nutrients compared to controls. As the *E. lata* fungus requires nutrients to grow, it is possible that the lack of available nitrogen in vascular tissue of the controls may have limited the growth of the fungus as well as the vine.

Output 3

Curative treatments to restore long-term productivity of eutypa dieback-infected vines

Performance targets:

- Monitor 12 remedial surgery trials established in the CRCV project for recurrence of eutypa dieback symptoms up to 9 years after treatment to assess long-term sustainability

- Cost-benefit analysis of remedial surgery for sustainable control of eutypa dieback

Remedial surgery is an effective strategy for managing eutypa dieback in mature grapevines. The production of new watershoots after removal of infected wood varied depending on cultivar and the amount of trunk retained after surgery. When watershoots do not appear after two seasons, vines could be replaced by replanting or by layering from a neighbouring vine using a vigorous shoot. Layering results in a fully producing vine in a shorter period of time than replanting due to the established root system of the mother vine.

Remedial surgery reduced the incidence of vines with eutypa dieback symptoms in a vineyard but results indicated that success depended on a number of factors including; cultivar, incidence and extent of pre-existing infection and, most importantly, the origin of the watershoot. The lower the watershoot develops on the trunk the less likely it is that the wood below it is infected with *E. lata* (which may result in recurrence of symptoms in vines). Therefore it is recommended that cuts are made down the trunk until no staining can be seen and then a further cut is made 10 cm or more into healthy tissue. This ensures that any fungus advancing ahead of stained wood is removed. Cutting all vines low (around 30 cm from the ground) minimises the time to undertake remedial surgery on a large scale and reworking a small section of a vineyard each year will minimise production losses.

Commercial yield is delayed for several years until the new canopy is established, but this is less than that required for an equivalent production from new plantings. Anecdotal evidence suggests that fruit quality is also restored within several years after trunk removal since the established root system is retained, which is not the case when replanting.

Remedial surgery can maintain longevity of premium vines, however, there is no guarantee that the disease is eradicated. It is important to rework vines from shoots below infected wood to minimise the recurrence of symptoms, and wounds should be protected from further infection by *E. lata*.

The cost-benefit analysis has provided a preliminary decision support model which can be used to extrapolate and predict economic returns of different decision scenarios in a Shiraz vineyard affected by eutypa dieback. Maintenance of vineyards in a disease-free condition ensured good economic returns for the vineyard. The cost of retrellising was insignificant for sustainability of vines. Remedial surgery avoids long-term economic losses once incidence of eutypa dieback exceeds 20% in the vineyard. However, the model is limited to Shiraz vines so further data need to be generated to expand and develop the model for use on other cultivars.

Output 4

Early alert for emerging wine regions to the incidence and threat of eutypa dieback

Performance targets:

- Survey and establish the incidence of eutypa dieback in emerging cool climate regions of South Australia

- Early alert for emerging wine regions to the incidence and threat of eutypa dieback

Surveys showed that eutypa dieback is wide-spread in the Adelaide Hills and in the Southern Fleurieu regions. Eutypa dieback was rare in the young Mt Benson region, however, the detection of three vines during the survey serves as an early alert to growers.

The youngest vines with foliar symptoms in this study were 7 years old, which is less than in past surveys and may be a reflection of the increase in prevalence of eutypa dieback along with the increase in grapevine plantings over the past two decades. The greater incidence of foliar symptoms observed on older vines than on younger vines, provides a warning to those with younger plantings. Incidence of foliar symptoms was greatest in Cabernet Sauvignon and Shiraz and very low in Merlot, Pinot Noir and Sauvignon Blanc. However, it is important to note that results of this survey may be conservative, being based on foliar symptoms, and the actual incidence of infected vines may be greater.

A survey was undertaken in Tasmania and this provided positive identification of *E. lata* in grapevines in Tasmania. It is likely that infection has spread from apricots and other stone fruits and alternative hosts to new grapevine plantings.

In an extensive survey of Western Australia in 2003 where species of Botryosphaeriaceae were identified, *E. lata* was not reported. In the most recent survey in 2009 where over 10,000 vines were surveyed in WA, vines with foliar symptoms of eutypa dieback were not detected and no fruiting bodies of the fungus were found on dead grapevine wood.

As a result of surveys, workshops were presented in each region surveyed to inform growers of recommended strategies for managing eutypa dieback disease in grapevines. Information from the surveys serves as a warning to grape growers of the disease incidence in each region, and such awareness may help to alleviate future economic losses through implementation of disease management strategies. Growers are recommended to apply control strategies to remove any existing diseased wood and prevent new infection of pruning wounds.

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11 COMMUNICATION

11.1 Publications

11.1.1 Scientific publications

Sosnowski MR, Wicks TJ, Lardner R and Scott ES (2007) The influence of grapevine cultivar and isolate of *Eutypa lata* on wood and foliar symptoms. *Plant Disease* 91, 924-931.

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12 ACKNOWLEDGEMENTS

13 APPENDICES

Appendix 1

A decision analysis model for the rehabilitation of Shiraz vineyards affected by Eutypa dieback

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Introduction

The practical control of Eutypa dieback affected vines in Australian vineyards is currently limited to remedial surgery; the removal of the infected wood of affected vines, protective treatment of wounds, followed by retraining of a watershoot onto the trellis to rehabilitate the vine. Such labour intensive, expensive treatment clearly should not be done without the grower being convinced that a treated vineyard will be more profitable than an untreated vineyard, taking into account future cash flows.

In this report we describe a decision tool for intervention in disease affected Shiraz vineyards. The tool is a spreadsheet financial model of the benefits of treatment compared to no treatment, expressed as comparative accumulated future cash flow streams. A screen capture of the front page of the model is displayed in Fig 1.

Model variables

- Targeted grape yield (i.e. without the disease) in the present vineyard, and in the rehabilitated vineyard
- Disease level in the present vineyard and expected in 40 years time. Disease level in 20 and 40 years time in the rehabilitated vineyard.

With this information the model computes yield reductions due to the disease, according to the empirical formula: %YL = 0.7*%DR +5%, where YL is yield loss and DR is disease rating. This formula is derived from the available Australian data for Shiraz.

- Change in grape quality and hence price due to the disease in the present vineyard and in the rehabilitated vineyard.
- The grower has the opportunity to substitute their own quality/price scale

The disease score corresponding to the grade represents our estimate of the maximum possible disease score for grapes in the grade before downgrading occurs.

- The cost of rehabilitating the diseased vineyard
- The grower has the opportunity to substitute their own costs

We note that for older vineyards the grower may take the opportunity to replace the trellising in the vineyard, particularly for high disease levels. Although, strictly, retrellising cannot be ascribed to disease removal, opportunity has been given for the grower to incur these costs within the model.

- The discount rate used

The model accumulates cash flow over 30 years for each year in a diagrammatic time frame (50 years), in terms of net present value. The diagrammatic results come with guidelines for interpretation.

Three examples of the model in use

- A grower produces C2 grapes (on a 16 point scale, A1….D4) at a winery gate price of $1400/t, with a targeted yield of 11 t/ha. He believes that after rehabilitation he will continue to produce C2 grapes at 11 t/ha. After rehabilitation he intends to monitor and remove any diseased vines that appear on an annual basis, at a cost of $50/ha. The vineyard has a disease rating of 20% now and it is expected to increase to 50% in 40 years time. Full retrellising is involved in his rehabilitation plan. The model suggests that rehabilitation should take place as soon as possible – the long-term cash flow benefits of the rehabilitated vineyard exceed those of the diseased vineyard from year 0.

- A grower produces D3 grapes (on a 16 point scale) at a winery gate price of $ 400/t, with a targeted yield of 15 t/ha. He believes that after rehabilitation he will continue to produce D3 grapes at 15 t/ha. He does not intend to monitor and remove any diseased vines that appear on an annual basis, after rehabilitation. The vineyard has a disease rating of 10% now and it is expected to increase to 50% in 40 years time. After rehabilitation he expects to see a disease rating of 20% in year 20 and 50% in year 40. The model suggests that rehabilitation can be deferred 12 years – continue monitoring disease levels to ensure that the model predictions are approximately correct.
- A grower produces A3 grapes (on a 16 point scale) at a winery gate price of $ 3300/t, with a targeted yield of 7 t/ha. He believes that after rehabilitation he will continue to produce A3 grapes at 7 t/ha. After rehabilitation he intends to monitor and remove any diseased vines that appear on an annual basis, at a cost of $50/ha. The vineyard has a disease rating of 5% now and it is expected to increase to 20% in 40 years time. Full retrellising is involved in his rehabilitation plan. The model suggests that rehabilitation can be deferred 10 years – continue monitoring disease levels to ensure that the model predictions are approximately correct.

General principles emerging from a number of scenario analyses

- It is always worthwhile to maintain the vineyard in a disease-free condition after rehabilitation, by expending $50/ha/year to monitor and remove any diseased vines that appear.
- The extra cost of retrellising makes little difference to the model outcome in the majority of situations
- If there is a relatively low incidence of disease in the vineyard (10% or less), the model indicates that rehabilitation can be delayed for a number of years in the majority of cases. Conversely if there is a high incidence of disease in the vineyard (20% or more), the model indicates that rehabilitation should be carried out as soon as possible in the majority of cases.

Model limitations

- The model is limited to Shiraz grapes, because there are insufficient data on the impact of Eutypa dieback on other varieties, in terms of yield reductions, at this time. This lack of data is coupled with the observation that the disease expresses itself differently according to variety, in terms of yield and longevity of affected vines.
- Furthermore, there is a lack of useful data on the rate of spread of the disease, as expressed in increasing yield reductions over time, even for Shiraz.
- Thirdly, we have guessed the impact of the disease on grape quality reductions, as expressed in grade of grape achievable at the winery, because there are no data that we are aware of on this critical variable.

Finally, we note that yield reductions from a given level of the disease in a vineyard can vary markedly from year to year, according to environmental conditions. For this reason alone, the model is necessarily approximate.

The economics of Dieback in cool climate grapes
(when is it most profitable to control dieback by lopping vines & retrellising?)

Critical Variables

Normal Grape Yield (t/ha)	**11**
Discount Rate	**15%**
Present Vineyard	
Disease Rating Now	**20**
Expected Rating in 20 yrs	**40**
Rehabilitated Vineyard	
Expected Disease rating Y20	**10**
Expected Disease rating Y40	**20**

Change in grape quality & Price as yield reduces

% Yield reductn	Quality class	GM = Yield x price - costs	
		Price	Costs
<45%	C2	$1,287	$3,716
45 -50%	C3	$1,087	$3,716
50-55%	C4	$887	$3,716
55-60%	D1	$687	$3,722
60-70%	D2	$487	$3,722

Rehabilitation Costs

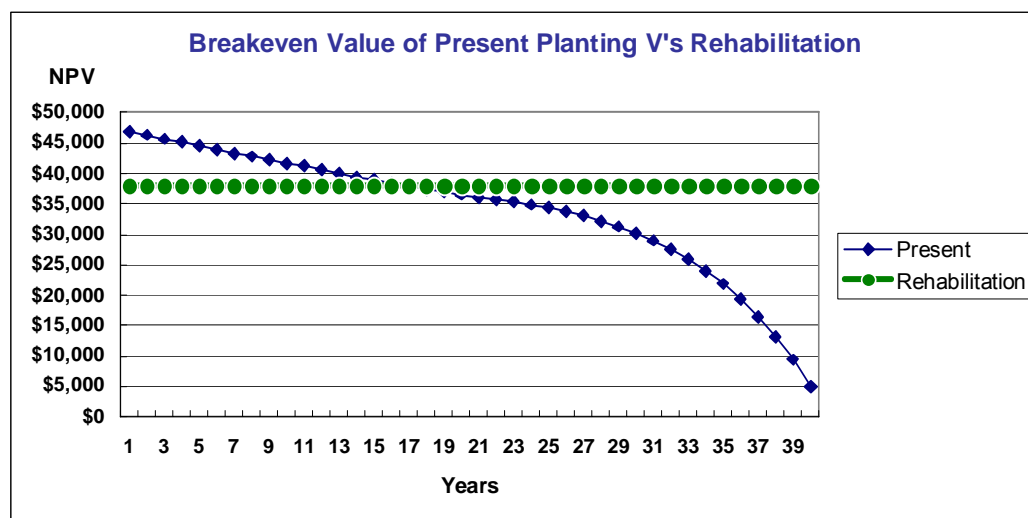|	Year 1	Year 2
Trellis materials	$2,386	
Trellis installation	$2,120	
Labour	$2,510	
Retraining cordons	$3,010	$1,500

Fig 1. Front page of eutypa dieback remedial surgery decision analysis model. Under the assumptions shown above, the model suggests that rehabilitation should take place in 16 years time.