

Sustainable control of powdery and downy mildew diseases of grapevine and impacts of control on wine quality and vineyard health.



## **FINAL REPORT to**

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Research Organisation: **University of Adelaide**

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## **GRAPE AND WINE RESEARCH & DEVELOPMENT CORPORATION**

Project Number: **UA 03/03**

### **Sustainable control of powdery and downy mildew diseases of grapevine and impacts of control on wine quality and vineyard health**

#### **Authors**

**Dr Peter Crisp, Ms Carol Walker, Dr Paul Grbin and Assoc Prof Eileen Scott**

University of Adelaide, School of Agriculture, Food and Wine, Waite Campus, PMB 1,  
Glen Osmond, South Australia 5064, Australia

**Dr Katherine Evans**

University of Tasmania and Tasmanian Institute of Agricultural Research (TIAR), New  
Town Research Laboratories, 13 St. Johns Avenue, New Town, Tasmania 7008, Australia

**Dr Sandra Savocchia and Dr Roger Mandel**

National Wine and Grape Industry Centre, Charles Sturt University, Locked Bag 588,  
Wagga Wagga, NSW 2650, Australia

**Dr Trevor Wicks**

South Australian Research and Development Institute, GPO Box 397, Adelaide, South  
Australia 5001, Australia

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<b>Table of contents</b>	<b>Page</b>
<b>Authors and acknowledgements</b>	<b>1</b>
<b>1. Abstract</b>	<b>4</b>
<b>2. Executive summary</b>	<b>5</b>
<b>3. Background</b>	<b>7</b>
<b>3.1. Sustainable management of powdery and downy mildew</b>	<b>7</b>
<b>3.2. Focus of research in South Australia</b>	<b>7</b>
<b>3.3. Focus of research in Tasmania</b>	<b>8</b>
<b>3.4. Focus of research in New South Wales</b>	<b>9</b>
<b>4. Project aims and performance targets</b>	<b>10</b>
<b>5. Methods</b>	<b>11</b>
<b>5.1. Research in South Australia</b>	<b>11</b>
<i>Peter Crisp, Carol Walker, Paul Grbin, Trevor Wicks and Eileen Scott</i>	
<b>5.2. Research in Tasmania</b>	<b>20</b>
<i>Katherine Evans</i>	
<b>5.3. Research in New South Wales</b>	<b>30</b>
<i>Sandra Savocchia</i>	
<b>6. Results and Discussion</b>	<b>36</b>
<b>6.1. Research in South Australia</b>	<b>36</b>
<i>Peter Crisp, Carol Walker, Paul Grbin, Trevor Wicks and Eileen Scott</i>	
<b>6.2. Research in Tasmania</b>	<b>53</b>
<i>Katherine Evans</i>	
<b>6.3. Research in New South Wales</b>	<b>80</b>
<i>Sandra Savocchia and Roger Mandel</i>	
<b>7. Outcomes/Conclusions</b>	<b>99</b>
<b>8. Recommendations</b>	<b>101</b>
<b>Appendix 1. Communication</b>	<b>102</b>
<b>Appendix 2. Intellectual property</b>	<b>104</b>
<b>Appendix 3. References</b>	<b>105</b>
<b>Appendix 4. Project staff</b>	<b>107</b>
<b>Appendix 5. Microbial populations on leaves</b>	<b>108</b>
<b>Appendix 6. Budget reconciliation</b>	<b>109</b>

## 1. Abstract

When disease pressure was low-moderate, milk, whey and Ecocarb<sup>®</sup> plus Synertril Horti-Oil<sup>®</sup> controlled powdery mildew as effectively as sulfur on Cabernet Sauvignon in NSW and Shiraz and Verdelho in SA but were less effective on Pinot noir and Chardonnay. TA, pH and °Brix of juice and wine quality were not affected. The Gubler-Thomas index was not a reliable indicator of disease severity or spray timing. Flexible spray programs based on monitoring and cultivar susceptibility are recommended.

Ferrous formulations, tea tree oil products, Brotomax<sup>®</sup> and Ecocarb<sup>®</sup> plus Synertril Horti-Oil<sup>®</sup> prevented downy mildew as well as the standard copper treatment.

## 2. Executive summary

Powdery mildew, caused by *Erysiphe necator*, is generally controlled by the application of sulfur and synthetic fungicides in conventional vineyards, and by sulfur and vegetable oils in organic vineyards. However, sulfur is toxic to beneficial mites and insects, including natural antagonists of *E. necator*, and may contribute to environmental pollution. Furthermore, some vineyard workers may have an adverse reaction to sulfur. As the demand for alternatives to sulfur and synthetic fungicides grows in Australia and elsewhere, there has been increased effort to develop new disease management strategies and several new products are available or undergoing registration. Previous research by Crisp, Scott and Wicks identified whey, milk, potassium bicarbonate and canola-based oils as contact fungicides with potential in the management of powdery mildew. The need to assess new, sustainable strategies for powdery mildew control on a range of cultivars in various climatic regions, to assess effects of these strategies on wine quality, and to develop new materials for control of downy mildew was addressed in this project.

The efficacy of the novel treatments varied according to cultivar, canopy structure, vigour and coverage. Grape cultivars vary from very susceptible to powdery mildew, such as Chardonnay and Verdelho, to less susceptible, such as Shiraz and Grenache. To date, the novel controls have provided excellent control of powdery mildew on Shiraz, acceptable control (defined here as < 5% of the bunch affected) on Verdelho and variable or marginal control on Cabernet Sauvignon, Chardonnay and Pinot noir. Trials with Chardonnay have generally failed to provide commercially acceptable control of powdery mildew, however, in most cases factors such as canopy density and vigour were conducive to the disease.

In South Australian field trials, the severity of powdery mildew on Verdelho vines sprayed with a 1:10 dilution of milk, 45 g/L whey powder and programs comprising rotations of canola-based oil plus potassium bicarbonate (Synertrol Horti-Oil® and Ecocarb®, Organic Crop Protectants) and whey was not significantly different from that on vines sprayed with sulfur (wetable powder, 3 g/L). Likewise, disease severity on Shiraz vines sprayed with a 1:10 dilution of milk, 25 g/L whey powder and programs of potassium bicarbonate plus oil and whey was not significantly different from that on vines sprayed with sulfur or Topas®. In Tasmania, in one of two trials, the novel materials provided control of powdery mildew on Chardonnay equivalent to that of sulfur. Season-long application of milk or whey to Pinot noir resulted in disease severity on bunches of 5 and 5.4%, respectively, considered unacceptable for this high-value cultivar. Novel materials applied to Cabernet Sauvignon in New South Wales generally provided adequate control, whereas trials with Chardonnay were largely inconclusive as neither sulfur nor the novel materials provided commercially acceptable control. This was attributed to a combination of a sprawling canopy that impaired spray coverage and extended intervals between spray applications in spring caused by inclement weather.

In all three states, spray coverage was a major factor in the success of alternative control measures. Trellis systems such as Smart-Dyson and Scott-Henry, which provide an open canopy less conducive to powdery mildew, are better suited to alternative control measures than are those which promote dense canopies. Furthermore, the efficacy of spray equipment should be checked regularly and coverage optimised using water sensitive papers or fluorescent dyes.

Varying disease control programs according to susceptibility and disease pressure should allow reduced sulfur inputs. Less susceptible cultivars, such as Shiraz, need fewer fungicide applications and alternative options can be used. For example, at Temple Bruer Wines, Langhorne Creek in the 2004-2007 growing seasons the use of sulfur on a range of cultivars has been restricted to times of severe disease pressure and only the susceptible Verdelho was

sprayed regularly. In a “whole of block” experiment in Tasmania, the inclusion of one or two sulfur applications around capfall provided commercially acceptable control of powdery mildew on Pinot noir. Thus an option to reduce risk associated with “softer” powdery mildew control programs for susceptible cultivars is to apply sulfur or synthetic fungicides between capfall and pea size berries.

Monitoring for disease, rather than calendar-based spraying, can permit fewer spray applications and, therefore, reduce costs without increasing the risk of crop loss due to powdery mildew. Concentrated monitoring of known disease “hot spots” and susceptible cultivars in addition to general vineyard monitoring is an important part of any disease control program. Again using Temple Bruer Wines as an example, in 2004/05 and 2005/06 powdery mildew disease pressure was low and only three spray applications were required in all but a patch of approximately 40 vines where the disease appeared to have developed from one or two flag shoots each season. The Gubler-Thomas disease risk index predicted the onset of powdery mildew epidemics in one season of two seasons in New South Wales but not in South Australia or Tasmania.

A ‘best bet’ spray program for management of powdery mildew in Pinot noir grown organically in southern Tasmania was developed. A ‘whole-of-block’ experiment demonstrated that a spray program beginning at E-L stage 16 and based on a mixture of Synertril Horti-oil® and Ecocarb® with two applications of sulfur plus Horti-oil® during fruit set resulted in a mean maximum disease severity at veraison of 1.5%. By harvest, the grower/cooperator judged the powdery mildew control to be commercially acceptable. The spray programs evaluated did not appear to prevent colonization of buds by *E. necator* and, hence, subsequent development of flag shoots.

Applications of milk and whey generally increased the population of indigenous bacteria, yeasts and filamentous fungi on leaves and bunches, compared with vines treated with sulfur, but had no obvious effect on species diversity. While increased microbial populations may contribute to the reduction of powdery mildew on the vine surfaces, there was no evidence that they impaired grape or wine quality. Assessment of juice from grapes sprayed with alternative materials did not reveal any differences that could readily be attributed to the treatments, with no significant differences in pH, TA or °Brix. Chemical and sensory evaluation of experimental wines made from grapes sprayed with milk, whey, Synertril Horti-Oil® and Ecocarb or sulfur revealed no differences attributable to treatment. Furthermore, no off flavours or loss of quality in wines at Temple Bruer Wines have been detected since the introduction of alternative controls across the vineyard in 2001.

In summary, when disease pressure is low to moderate, commercially acceptable control of grapevine powdery mildew on cultivars that are not highly susceptible can be achieved using a range of novel materials including milk, whey and mixtures of canola oil plus potassium bicarbonate. As these compounds act as contact fungicides excellent cover of leaf and berry surfaces is required for effective control, and spray intervals should not exceed 14 days during flowering, early berry development and periods of rapid shoot growth. For growers or vineyard managers considering using such materials, it is suggested that a small trial be established first to ensure that the treatments are suited to the cultivars and canopy architecture on their vineyard. If the fruit is contracted to an external winemaker, the grower should first confirm with the winemaker that they will accept the fruit.

Ferrous sulfate, Timor, Timorex, Ecocarb plus Synertril Horti-Oil, and Brotomax, applied prior to inoculation with *Plasmopara viticola*, reduced the severity of downy mildew compared with uninoculated controls. However, leaves treated with ferrous sulfate or Brotomax and kept in a humid environment developed symptoms of phytotoxicity. With further testing and refinement, the above-mentioned materials may provide alternatives to copper for the control of downy mildew.

### 3. Background

Powdery and downy mildew can reduce both yield and quality of the fruit and wine produced from affected grapes. Powdery mildew has been estimated to cost the Australian grape and wine industry about \$30 million each year (Wicks et al. 1997). In years with wet spring weather, downy mildew can cause widespread devastation, such as occurred in 1992-93. Powdery mildew is controlled mainly by sulfur and synthetic fungicides in conventional vineyards and by sulfur in organic vineyards. Downy mildew may be controlled by a range of fungicides, including the phenylamide group and copper-based chemicals, in conventional vineyards and by copper in organic vineyards. Reducing inputs of sulfur and copper in both organic and conventional systems will reduce the possible risk of environmental contamination, such as has occurred for copper in Europe.

Organic viticulture is increasing in profile in Australia. There is large, unmet demand for organic wine in Europe (see GWRDC report UA 00/7, travel support, organic viticulture, P. Crisp), and the Australian industry is well-placed to target that market. The potential withdrawal of sulfur and copper from the acceptable inputs in organic viticulture (EC regulation 2092/91; International Federation of Organic Agriculture Movements, 1998), and the need to reduce environmental and ecological impacts in conventional viticulture, provide incentives for the development of alternative, environmentally friendly controls for powdery and downy mildew.

Previous research at the University of Adelaide, supported by the Australian Research Council (ARC), identified milk, whey and canola oil as having potential to control powdery mildew, even in seasons of high disease pressure such as 2001-2002 (Crisp et al. 2006b). Peter Crisp, under the supervision of Eileen Scott and Trevor Wicks and in close collaboration with leading organic viticulturists David Bruer (Temple Bruer Wines) and Leigh Verral (Glenara Wines), developed protocols for the use of novel controls for powdery mildew, including applications of milk, whey, vegetable oils and bicarbonates (Crisp and Bruer 2001, Crisp et al. 2000, 2002). In greenhouse trials, Synertrol Horti-Oil (a canola oil-based product), milk and whey reduced disease by 92%, 70% and 64%, respectively, compared with untreated controls (Crisp et al. 2006a). In trials with young Shiraz vines at Langhorne Creek in 2001/2002, milk and whey were as effective as the DMI fungicide Topas (Crisp et al. 2006b). Laboratory studies showed that milk and oils caused collapse of hyphae of the fungus and rupture of conidia (asexual spores), and suggested that free radical formation in light and antimicrobial proteins may be involved (Crisp et al. 2006c).

While the above materials have provided excellent control of powdery mildew on Grenache, Shiraz, Riesling and Sauvignon Blanc (D. Bruer pers. com.), they appear to be less effective on other cultivars, such as Verdelho and Viognier (Crisp et al. 2006 b). Prior to 2003, the protocols had been tested on a limited range of cultivars, mainly in the Langhorne Creek area of South Australia. The primary objective of this research, therefore, was to evaluate protocols for reducing inputs of sulfur in a range of climatic conditions, cultivars of *Vitis vinifera* and management systems.

#### 3.1. Focus of research in South Australia

Refinement of the protocols developed in the ARC project for the control of powdery mildew was attempted at several sites in South Australia, including Langhorne Creek, Lenswood and McLaren Vale. Spray programs were based on integrated pest management (IPM) principles, i.e. monitoring for disease, and disease assessment data and weather data were used to compare the efficacy of monitoring, calendar-based spray programs and the use of the Gubler-Thomas powdery mildew risk assessment model (Gubler et al. 1999). In addition, whereas milk, whey, oils and potassium bicarbonate were shown to act as contact fungicides, with curative properties (Crisp et al. 2006 a, c), their suitability as protectant fungicides for the



management of powdery mildew had not been assessed. Furthermore, their potential to control downy mildew had not been examined.

Little was known about the effects of milk, whey, oils and bicarbonate on beneficial microorganisms and arthropods in the grapevine canopy. The microorganisms present on the bunch surface enter the winemaking process. Indigenous microorganisms, predominantly yeast species, may persist and grow well into vinification (Capece *et al.* 2003; Fleet 2003), where they may produce secondary metabolites that contribute to the aroma and flavour of the wine. Preliminary studies by Palmer (2003), an honours student allied to the ARC project, suggested that treatment with milk or Synertrol Horti-Oil increased the population of the yeast-like fungus, *Aureobasidium pullulans*, on bunches at maturity compared with sulfur treatment. The lack of information on the effects of novel powdery mildew management strategies on wine quality was raised in discussions following Peter Crisp's presentation at the Organic Viticulture Congress in Canada in August 2002 (Crisp *et al.* 2002). In recognition of the oenological significance of the grape surface microbiota, research was undertaken to assess the effects of milk, whey and sulfur on grapevine microbial populations.

Research undertaken in South Australia, therefore, focussed on: (i) refinement of protocols for the control of powdery mildew using milk, whey, oils, bicarbonate and other novel materials; (ii) the effects of such protocols on the populations of microorganisms on leaves and berries and on grape and wine quality; and (iii) the identification of alternatives to copper for the control of downy mildew. The second objective was addressed by PhD student, Carol Walker.

### **3.2. Focus of research in Tasmania**

The main grape varieties grown in Tasmania are Pinot noir, Chardonnay and Riesling and the number of bearing hectares in 2006 was approaching 1,000 ha. Chardonnay and Riesling are very susceptible to powdery mildew.

The primary objective of the trial work in Tasmania, namely, the reduction of sulfur inputs in viticulture, was the same as for research conducted in South Australia and New South Wales. In addition, the research provided the first opportunity to document the epidemiology of powdery mildew in Tasmania. Tasmania has never had a powdery mildew management strategy developed specifically for local conditions. Growers have tried to modify recommendations applied elsewhere, for example, the 2, 4, 6 rule applied in the Riverland is sometimes extended to 3, 6, 9 (weeks after budburst) in Tasmania to account for slow rates of shoot development. Furthermore, high rates of sulfur, commonly 8 to 10 g/L, are applied to compensate for cool temperatures that can occur at any time. Despite these modifications, poor spray timing and coverage still lead to control failures. Therefore, an additional objective was to monitor powdery mildew in untreated control plots in relation to weather and crop stage. The third objective, in common with research in SA and NSW, was to investigate the correlation between disease progression and the cumulative Gubler-Thomas risk index for powdery mildew (Gubler *et al.* 1999). This objective was also explored and reported in GWRDC final report RITA 04/04-1 (Evans, 2005). In addition the powdery mildew control provided by season-long applications of sulfur at 6 or 12 g/L was investigated.

#### ***Evaluating organic spray programs in Tasmania using spatial information***

Results of small plot trials in Tasmania provided evidence that inputs of sulfur for powdery mildew control can be reduced based on improved knowledge of disease progression. Given the tendency for contact materials such as milk or whey to be less effective than sulfur in conditions of high disease pressure, elimination of sulfur from organic spray programs does not yet appear to be feasible in southern Tasmania. Even if alternative materials were applied in well-defined conditions of vine cultivar and vigour, the results of small plot trials would still need to be verified on a larger scale. A major limitation of small plot trials is interpreting the gap between the effective dose, as determined by hand spraying single panels of vines,

and the actual dose achieved over a large and variable area using commercial equipment. While randomisation of small plots attempts to accommodate the effects of underlying vine variation, it is assumed that the variation is also distributed randomly. However, variation in vine vigour, for example, is unlikely to be distributed randomly within rows that constitute the 'blocks' of small plot trials. Powdery mildew is often severe in dense, shaded canopies. Vine vigour is likely to be correlated positively with disease severity, although this phenomenon does not appear to have been quantified scientifically.

The Tasmanian grower co-operator was aware of the results of the small plot trials, yet he wanted to reduce sulfur applications with commercial equipment and available materials. Through consultation, a whole-of-block experiment was developed with the aim of investigating if application of only one or two sulfur sprays during the critical flowering/fruit set period would provide commercially acceptable control of powdery mildew. The experiment was designed specifically to evaluate the *magnitude* of treatment effects accounting for variation in inherent vine vigour. A second objective was to evaluate a 'whole-of-block' approach to experimentation (Bramley et al. 2005), which until now has not been applied in development of disease management strategies for grapevine.

### **3.3. Focus of research in New South Wales**

The protocols developed in South Australia were evaluated in field trials conducted in the warm, dry climate of Wagga Wagga, NSW. Field trials were located in the Charles Sturt University commercial vineyard during three growing seasons (2003-06) using adjacent blocks planted to Chardonnay and Cabernet Sauvignon. The treatments tested at the Wagga Wagga vineyard included combinations of sulfur, milk, whey, potassium bicarbonate (Ecocarb<sup>®</sup>), Synertrol Horti-oil<sup>®</sup> and Surround<sup>™</sup> WP (a.i. kaolin). Surround<sup>™</sup> WP comprises 95% kaolin and was chosen as a treatment due to its ability to form a barrier film and to act as a broad spectrum protectant, limiting damage from various insect and disease pests. It is also able to provide protection from sunburn and heat stress. As this product forms a white film on the surface of berries and leaves, the impact on vine physiology was also assessed. As in SA and Tasmania, temperature and rainfall data were collected to determine if the Gubler-Thomas powdery mildew risk index (Gubler et al. 1999) could be applied to the region.

## 4. Project aims and performance targets

The aims and performance targets were as set out in the original application, except that the completion of the PhD research was delayed due to surgery, maternity leave and contamination of cultures. The student has not yet completed her PhD thesis.

### Aims

1. Refine protocols for the use of novel treatments in the control of powdery mildew, including milk, whey and canola oil-based products, to provide commercially acceptable control on the main cultivars of grapevine.
2. Identify the characteristics of grapevines that impact on the efficacy of the novel treatments for powdery mildew and modify protocols to address any variations in efficacy associated with different cultivars.
3. Develop potential replacements for copper in the control of downy mildew.
4. Assess the impact of the treatments on juice, the fermentation process and on wine quality.
5. Assess the impact of the treatments on populations of beneficial microorganisms and arthropods within the grapevine canopy.
6. Conduct efficacy trials in various regions to ensure that treatments are appropriate Australia-wide.

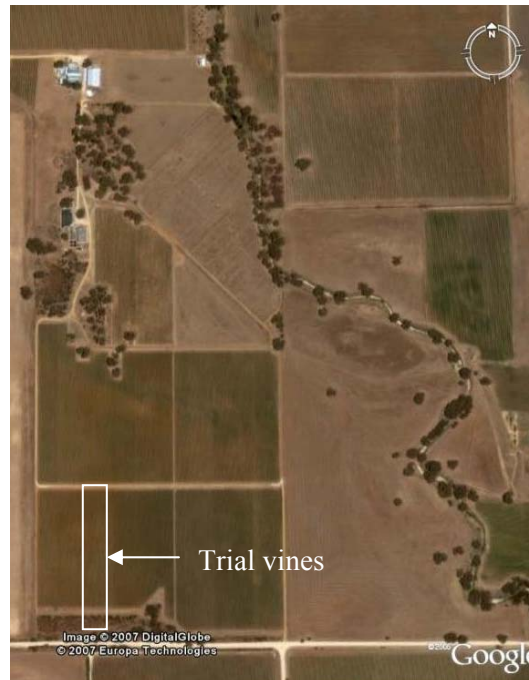
**Table 1.** Performance targets as set out in original application.

Output	Performance Targets	Date
1. Refined protocols for control of powdery mildew using milk and oils.	Evaluation of protocols for use of milk and oils to control powdery mildew.	June 2005
2. Information on response of major cultivars to treatments for powdery mildew.	Comparison of the levels of powdery mildew control achievable on selected cultivars over 3 years.	June 2006
3. Information on alternatives to copper for control of downy mildew in controlled conditions.	Data on efficacy of milk, oils and ferrous formulations for control of downy mildew in the greenhouse.	Jan 2005
4. Information on alternatives to copper for control of downy mildew in the vineyard.	Data on efficacy of milk, oils and ferrous formulations for control of downy mildew in the vineyard.	June 2006
5. Information on effects of milk and oils on the microflora of grapes.	Assessment of microflora on grapes over three seasons.	June 2006
6. Information on effects of treatment with milk and oils on wine yeast activity and the fermentation process.	Assessment of rates and characteristics of fermentation of wines made from grapes subjected to experimental and standard treatments over 3 years.	May 2004 May 2005 May 2006
7. Information on effects of milk and oils on chemical and sensory properties of juice and wine quality.	Completion of analysis of juices and wines made with treated grapes over 3 years (sensory evaluation for years 1 and 2 only).	June 2006
8. Completion of postgraduate training.	Completion of PhD thesis.	June 2006
9. Communication of results to industry.	Results presented at industry meetings and workshops. Submission of final report.	June 2006

## 5. Methods

### 5.1. Research in South Australia

Vineyards at Langhorne Creek and Lenswood were used for field trials. The vineyard at Langhorne Creek is located about 60 km to the south-east of Adelaide, South Australia ( $35^{\circ} 20' 57.46''$  S  $138^{\circ} 59' 07.68''$  E, elevation 15 m, annual rainfall 490 mm) (Figure 1). Two cultivars of *Vitis vinifera*, Shiraz and Verdelho, were selected to represent low and high susceptibility to powdery mildew, respectively. The vines were trained on a Smart-Dyson trellis system, with the canopy rarely exceeding 30 cm in width, and were drip-irrigated (Figure 2).



**Figure 1.** Aerial view of the Langhorne Creek vineyard, South Australia, with trial area indicated.



**Figure 2.** Verdelho vines on Smart-Dyson trellis, Langhorne Creek, South Australia.

The second site was a SARDI research vineyard at Lenswood, located about 30 km to the north-east of Adelaide (34° 56' 39.56" S 138° 48' 21.27" E, Elevation 515 m, annual rainfall 1032 mm). Nebbiolo, which is susceptible to both powdery and downy mildew, was used. The vines were established on a trellis system similar to the Langhorne Creek vines and were drip-irrigated.

Crop stage was identified using the modified Eichorn-Lorenz (E-L) system described by Coombe (1995).

Additional trials were undertaken by staff of several South Australian vineyards and wineries. However, most of these trials were compromised by difficulties with spray scheduling and will not be discussed further.

## **Powdery mildew**

### **Field trials: Langhorne Creek**

The field trials were set out in a completely randomized block design with six replicates of eight vines per plot. Two buffer rows were placed between the trial plots and the rest of the vineyard and there was a two-vine buffer between plots. Spray drift was monitored using water-sensitive papers and was below levels that could have confounded results. Treatments were applied using a Silvan<sup>®</sup> Selecta 50 L hydraulic spray with a hand-held wand. Spray applications were made from both sides of each row. Total water volumes ranged from 300 to 900 L/ha as the canopy developed to ensure the best possible coverage of leaf and bunch surfaces (Table 2). Each season the first application was when vines reached approximately E-L stage 11; subsequent applications were on a 10-14 day cycle adjusted to suit weather conditions for spray application and the severity and incidence of powdery mildew detected during monitoring.

Treatments applied were milk (full cream pasteurised), whey powder (Dairy Gold nonhygroscopic whey powder, Murray Goulburn Co-operative Co. Ltd), potassium bicarbonate (Ecocarb<sup>®</sup>, Organic Crop Protectants Pty Ltd) either alone in 2003/04 or mixed with canola-based horticultural oil (Synertrol Horti-Oil<sup>®</sup>, Organic Crop Protectants Pty Ltd) in 2004/05 and 2005/06, and a mixed program of whey and bicarbonate plus oil (Table 2). Sulfur was applied as an industry standard and untreated vines were used as a control. In 2004/05 and 2005/06 the area of untreated vines was halved to reduce the risk that dispersal of conidia from severely diseased vines might spread disease to the rest of the vineyard. The other half of the untreated blocks was sprayed with a mixture of diluted milk and *Bacillus subtilis* (Table 2). As powdery mildew was detected in at least three of the previous seasons and the vineyard was a commercial enterprise, inoculation with *E. necator* was not attempted. The vines were not treated with any other pesticides or foliar nutrients.

In September and October each year vines in the experimental area, including buffer rows, were checked for flag shoots, which were recorded and removed immediately, as differences in the number of flag shoot among the various treatments might have confounded the results.

Powdery mildew on leaves and bunches was assessed approximately fortnightly throughout the three growing seasons, immediately prior to application of test materials. Twenty leaves and bunches selected at random from each of the six centre vines were scored for disease using a 0 to 100 scale, based on the percentage of the surface area of the leaf or bunch with sporulating colonies of powdery mildew. Leaves and bunches were also assessed for evidence of phytotoxicity using a similar scale.

**Table 2.** Date of spray applications to Shiraz and Verdelho at Langhorne Creek, South Australia, and rate applied using a 50 L hydraulic spray tank with a hand-held wand. SH - Synertrol Horti-Oil, E – Ecocarb.

Season							
2003/2004	15/10/2003	24/10/2003	5/11/2003	25/11/2003	5/12/2003	24/12/2003	9/01/2004
Program 1	SH 3 ml/L Eco 5 g/L	SH 3 ml/L Eco 5 g/L	Whey 25 g/L	SH 3 ml/ Eco 5 g/L	SH 3 ml/L Eco 5 g/L	Whey 25 g/L	Whey 25 g/L
Milk (dilution)	1:10	1:05	1:05	1:10	1:10	1:5	1:5
Whey (g/L)	30	30	25	25	25	40	40
Ecocarb (g/L)	4	4	4	4	4	6	6
Sulfur (g/L)	3	3	3	3	3	6	6
Untreated	-	-	-	-	-	Sulfur	Sulfur
Rate (L/ha)	300	300	500	600	900	900	900
2004/2005	8/10/2004	18/10/2004	29/10/2004	10/11/2004	25/11/2004	22/12/2004	
Program 1	SH 3 ml/L Eco 5 g/L	SH 3 ml/L Eco 5 g/L	Whey 25 g/L	SH 3 ml/L Eco 5 g/L	Whey 25 g/L	SH 3 ml/L Eco 5 g/L	
Milk (dilution)	1:10	1:05	1:05	1:10	1:10	1:10	
Whey (g/L)	25	40	25	25	25	25	
Ecocarb (g/L)	5	5	5	5	5	5	
Synertrol (ml/L)	3	3	3	3	3	3	
Sulfur (g/L)	6	6	6	6	6	6	
Milk <i>Bacillus subtilis</i>	1:10 1x10 <sup>6</sup>	1:05 1x10 <sup>6</sup>	1:05 1x10 <sup>6</sup>	1:10 1x10 <sup>6</sup>	1:10 -	1:10 1x10 <sup>6</sup>	
Untreated	-	-	-	-	-	-	
Rate (L/ha)	300	300	500	600	900	900	
2005/2006	5/10/2005	19/10/2005	16/11/2050	5/12/2005	23/12/2005		
Program 1	SH 3 ml/L Eco 5 g/L	SH 3 ml/L Eco 5 g/L	Whey 25 g/L	SH 3 ml/L Eco 5 g/L	Whey 25 g/L		
Milk (dilution)	1:10	1:05	1:05	1:10	1:10		
Whey (g/L)	25	40	25	25	25		
Ecocarb (g/L)	5	5	5	5	5		
Synertrol (ml/L)	3	3	3	3	3		
Sulfur (g/L)	6	6	6	6	6		
Milk <i>Bacillus subtilis</i>	1:10 1x10 <sup>6</sup>	1:05 1x10 <sup>6</sup>	1:05 1x10 <sup>6</sup>	1:10 1x10 <sup>6</sup>	1:10 -		
Untreated	-	-	-	-	-		
Rate (L/ha)	300	300	500	600	600		

### Field trials: Lenswood

The Lenswood trials were established primarily to assess control of downy mildew, however, the spray program was maintained in such a way that the efficacy of the treatments for control of powdery mildew could be assessed also.

The trial was set out in a completely randomized design with four replicates. A two-row buffer was established between the trial rows and the rest of the vineyard, with two-vine buffers between plots. Water-sensitive papers were used to monitor spray drift, which was negligible. Plants were treated on a 10 to 14-day cycle until veraison and disease assessed throughout the season (Table 5). The treatments were applied using a Solo 475 backpack spray with a hand held wand; spray applications were made from both sides of each row, at a pressure of 400 Kpa, and a walking speed of approximately 3 km/h, delivering a total volume

of between 300 and 900 L per hectare. Vines sprayed with water were used as controls. As the vineyard had a history of powdery mildew vines, were not inoculated.

### **Evaluation of the Gubler-Thomas index**

Weather measurements were taken at the Langhorne Creek vineyard using a model T weather station (Western Digital, Bookpurnong, Australia) mounted at the edge of the trial site (Figure 3). Temperature, humidity, leaf wetness and light intensity were measured every 10 minutes, and rainfall was monitored constantly. The weather data were used to assess the potential of the Gubler-Thomas powdery mildew index (Gubler et al. 1999) as a forecasting tool for severity of powdery mildew and a guide to commencement of spray programs and spray intervals throughout the season. The Gubler-Thomas index is a temperature-based index used in California to predict the onset of powdery mildew in vineyards (Gubler et al. 1999). The recommendations from the Gubler-Thomas index were then compared with the actual sprays applied using a nominal 10-14 day interval adjusted following regular monitoring of the vineyard.



**Figure 3.** Model T weather station (Western Digital, Australia) at Langhorne Creek, South Australia.

### **Germination of *E. necator* conidia**

The effect of the test materials on spore germination and, hence, their potential in prevention of disease establishment, was assessed *in vitro*. Conidia of *E. necator*, collected from infected vines maintained in a greenhouse using a modified cyclone separator, were brushed onto either 90-mm Petri dishes containing 2% water agar or onto surface sterilised, disease-free detached leaves of Viognier on water agar in 90-mm Petri dishes (Evans et al. 1996). The effect of the following on germination was assessed: milk (1:5 and 1:10 dilutions of full cream milk), whey (30 g/L whey powder), lactoferrin (from bovine colostrum, 20 mg/L, Sigma Chemicals) and Synertril Horti-Oil® (2 ml/L); and Synertril Horti-Oil® (2 ml/L) mixed with potassium bicarbonate (4 g/L Ecocarb®, Organic Crop Protectants Pty Ltd) (Table 3). There were 10 replicate plates for each treatment. Sulfur was included as the industry standard and plates sprayed with water served as controls. Treatments were applied using an Atomizer reagent sprayer (Alltech Associates Inc., Belgium) 6 hours prior to inoculation.

The application rates of the test materials were selected to emulate rates used in previous greenhouse or field experiments (Crisp et al. 2006 a, b) and again no surfactants were used.

All Petri dishes were incubated at approximately 15/25°C night/day, in natural light and examined 24 and 48 h (agar) and 7 days (leaves) after treatment using a dissecting microscope. One hundred of the conidia applied directly to agar plates were counted and the proportion that had germinated was calculated. For the detached leaf assays the proportion of the leaf surface affected by powdery mildew was scored as a percentage.

**Table 3.** Materials applied to water agar (2%) and detached leaves of Viognier in 90-mm Petri dishes 6 hours prior to inoculation with *E. necator* conidia.

Material	Supplier	Concentration
Milk	Sunshine full cream powder	15 g/L
Milk	Sunshine full cream powder	30 g/L
Whey	Murray Goulburn Co-operative Co. Ltd	30 g/L
Lactoferrin	Sigma Chemicals	20 mg/L
Synertril Horti-Oil	Organic Crop Protectants	2 ml/L
Ecocarb	Organic Crop Protectants	2 g/L
Synertril Horti-Oil	Organic Crop Protectants	2 ml/L
Water	Reverse osmosis	
Sulfur	Yates Garden King	3 g/L

### Assessment of grape and wine quality

Fruit was harvested from Shiraz and Verdelho in 2004 and 2005 and from Verdelho only in 2006 and yield measured. Bunches with <5% of the surface area affected by powdery mildew were selected for assessment of quality parameters as described below.

Bunches collected for assessment of pH, titratable acidity (TA) and °Brix, approximately 250 g per plot, were placed in paper bags in cooled foam boxes for transport, stored at approximately 4° C for up to 24 h prior to analysis, then placed into separate plastic bags and squeezed by hand. The juice was then centrifuged at 3500 rpm at 20° C for 5 min, and pH, TA and °Brix of the supernatant were measured as described by Iland et al. (2000). °Brix was measured using an Atago hand-held refractometer, with readings adjusted for temperature. The pH was determined using a Metrohm 691 pH meter, calibrated at pH 4 and 7 prior to use.

For each treatment, 90 kg of fruit that was disease-free or with < 5% of the surface area of the bunch affected by powdery mildew was harvested and then divided into three 30 kg lots for processing. Fermentation and bottling were carried out by the University of Adelaide in 2004/05 and by Provisor Pty Ltd in 2005/06. Juice from Verdelho and must from Shiraz grapes were fermented to dryness, stabilised and bottled at least 3 months prior to sensory evaluation. The Shiraz wine was not subjected to malo-lactic fermentation as this process can alter wine characteristics and confound the assessment of treatment effects.

Duo-trio difference tests (Amerine et al. 1965) were carried out on samples of juice and wines to determine if the novel treatments produced any sensory effect. One of the sulfur replicates was used as a standard against which other samples are assessed.

### Effects of novel powdery mildew control on grapevine microbiota

The populations of bacteria, yeasts and filamentous fungi on leaves and berries, and in juice, from vines subjected to selected treatments for powdery mildew in the Langhorne Creek vineyard were assessed in terms of overall numbers and diversity of genera and species. Populations were estimated in terms of colony forming units on several laboratory culture



media and representative organisms were subcultured and identified on the basis of DNA sequences. Reference cultures of selected vine-associated bacteria, yeasts and filamentous fungi were obtained from Dr Belinda Stummer, University of Adelaide, Drs Ailsa Hocking and Su-lin Leong, Food Science Australia, and Ms Ai-lin Beh and Professor Graham Fleet, University of New South Wales, for comparison.

#### ***Microbial populations on leaves***

Leaves were collected from Verdelho and Shiraz vines at Langhorne Creek 2 days prior to each application of the test materials in 2003/04 and 2004/05, beginning when shoots were approximately 15 cm long (see Table 2). Fifteen leaves were collected from the six central vines in each of four replicate plots treated with milk, whey or sulfur or left untreated. Leaves were kept cool and transported to the laboratory, where microbial populations were estimated as described by Stummer et al. (2003). Briefly, the leaves were washed in sterile 0.1% magnesium sulfate, then serial dilutions of the resulting suspension were prepared and 0.1 ml aliquots were spread on agar in 90-mm diameter Petri dishes. Three culture media were used; nutrient agar plus Benlate fungicide (for bacteria), acidified potato dextrose agar (for filamentous fungi) and yeast peptone dextrose agar (for yeasts). Plates were incubated at 28°C for 5 days and the number of colony forming units was estimated. Representative colonies were subcultured to establish pure cultures and the morphological characteristics observed by light microscopy. Pure cultures of bacteria and yeasts were stored in glycerol at -80°C and those of fungi were stored in sterile water at room temperature for subsequent identification.

#### ***Microbial populations on berries and in juice***

Bunches were collected from Verdelho and Shiraz vines at veraison and just prior to harvest in 2003/04, 2004/05 and 2005/06. Bunches collected at veraison were obtained 2 days prior to the next spray application. Four to five bunches were collected at random from the six centre vines of each plot. Only bunches with <5% of the surface area affected by powdery mildew were collected, kept cool and transported to the laboratory for processing. Bunches from each treatment were pooled for analysis and approximately 60 g of berries were placed in beakers and washed in magnesium sulfate as described above. The colony forming units of bacteria, filamentous fungi and yeasts were estimated as described above. Representative colonies were purified, examined and stored as described above.

Samples of juice were removed from homogenised mature berries immediately prior to determination of pH, TA and °Brix. The number of colony forming units was determined as above and representative colonies were again selected, examined and stored.

#### ***Identification of representative microorganisms***

Representatives of the culturable bacteria, filamentous fungi and yeasts isolated from berry skins at veraison and commercial harvest and from berry juice at harvest in 2004 and 2006 and filamentous fungi in 2005 were identified using a molecular phylogenetic approach. Pure cultures of bacteria and yeast isolated in 2005 were lost due to mite-borne contamination that was beyond our control. This contamination resulted in the loss of 4 months of work.

Cultures were retrieved from storage and grown on appropriate culture media (as above) at 25°C until colonies were visible. Single colonies were transferred to buffer and DNA was extracted using a simple boiling lysis method proprietary to the CRC for Viticulture. The method was adapted from Hamelin et al. (2000). A fragment of the small-subunit RNA-encoding gene (SSU) was amplified from each isolate using the polymerase chain reaction (PCR). Amplified DNA was submitted to the Australian Genome Research Facility for sequence analysis. Sequences were compared with corresponding gene sequences held in public and private reference databases. Approximately 900 gene sequences were generated from the representative microorganisms studied. Isolates were then assigned to genus or species based on the closest match with the databases.

## Downy Mildew

### Greenhouse experiments

A range of materials was selected, following a literature search and communication with producers of crop protection products, for testing in the greenhouse. The experiment was conducted in 2004 and 2005 using 2-year old potted vines of Cabernet Sauvignon. Five fully expanded leaves were tagged on each plant, treated with one of the materials listed in Table 4 and inoculated by spraying to run-off with a suspension of *Plasmopara viticola* sporangia ( $10^6$ – $10^7$  per ml) in the evening 48 hours after treatment. The shoots were covered with plastic bags overnight to maintain leaf wetness. After 12 days, the leaves were removed from the vines and incubated overnight in moist conditions to induce sporulation. Leaves were then assessed using a dissecting microscope and the area of the leaf with sporulating colonies of downy mildew estimated. Incidence was assessed as presence or absence for each individual leaf. Severity was scored based on the percentage of leaf area infected. Copper (cupric hydroxide) was used as an industry standard and vines sprayed with water were used as untreated controls (Table 4).

**Table 4.** Treatments applied to Cabernet Sauvignon in the greenhouse for control of downy mildew.

Material	Supplier	Concentration
Copper (400 g/kg Cupric Hydroxide)	Yates copper fungicide	3 g/L
Ferrous sulfate	Ace Chemicals	9 g/L
Ferrous sulfate	Ace Chemicals	9 g/L
Synerscreen	Organic Crop Protectants	1 ml/L
Ferrous sulfate	Ace Chemicals	6 g/L
Synerscreen	Organic Crop Protectants	1 ml/L
Ferrous sulfate	Ace Chemicals	3 g/L
Synerscreen	Organic Crop Protectants	1 ml/L
Synerscreen	Organic Crop Protectants	1 ml/L
Ecocarb	Organic Crop Protectants	3 g/L
Synertril Horti-Oil	Organic Crop Protectants	3 ml/L
Chitosan	Organic Crop Protectants	3 ml/L
Vermiboost		10 ml/L
Water	Reverse osmosis	

### Field trials: Langhorne Creek

As the Langhorne Creek vineyard was a commercial property, vines were not inoculated with *P. viticola*. In 2003/04 and 2004/05 downy mildew was not detected despite several weather events conducive to both primary and secondary infection. Following a downy mildew outbreak on Verdelho vines in October 2005, a trial was established to evaluate products that had shown promise in greenhouse trials and at the Lenswood vineyard in 2004/05 (see below). However, several days of high temperature ( $>40^{\circ}\text{C}$ ) and low humidity ( $<30\%$ ) in late October, November and December prevented further disease development and confounded results.

### Field trials: Lenswood

The trials were set out in a completely randomized block design with four replicates, four vines per plot, and buffer rows were established to minimise spray drift from other parts of the vineyard. Vines at the ends of the rows that were shaded by adjacent trees were not included (Figure 4). The treatments described in Table 5 were applied using a Solo 475 backpack spray with a hand-held wand. Spray applications were made from both sides of each

row. Total water volumes of between 300 and 900 L/ha were applied, increasing as the vine canopy developed to ensure best possible coverage of leaf surfaces. Vines were artificially inoculated with *P. viticola* on two occasions in 2004/2005 and on three occasions in 2005/2006. Six shoots with five fully expanded leaves were tagged in each plot and inoculated with a suspension of *P. viticola* sporangia ( $10^6$ – $10^7$  per ml) in the evening 48 hours after treatment and the shoots were bagged overnight to maintain leaf wetness (Figure 5). After 12 days, six shoots from each plot were removed and incubated overnight in moist conditions to induce sporulation. Disease incidence and severity were assessed as for the greenhouse trials.

In 2005/2006 the shoots first inoculated with *P. viticola* were damaged by a severe storm while still bagged and >50% of inoculated shoots were broken. As a result, this trial was abandoned. The subsequent trials were confounded by widespread natural downy mildew infection and a severe powdery mildew outbreak and results considered unreliable.



**Figure 4.** Aerial view of the vineyard at Lenswood Research Centre, South Australia, with trial area indicated.

### Statistical analysis

Data for disease assessment were subjected to analysis of variance (ANOVA) using Genstat (Lawes Agricultural Trust) version 5 release 4.1 or version 6.1.0. The hypothesis tested was that there would be no significant difference in the mean disease score among test materials and the control treatments imposed. The 5% level of significance ( $P=0.05$ ) was used for all experiments. Likewise, data for pH, TA and °Brix were subjected to ANOVA using Genstat 5, with the null hypothesis that there would be no significant difference in the quality parameters assessed among test materials and the control treatments imposed.

Analysis of the microbial populations proved problematic. On the recommendation of consultants at BiometricsSA, original and transformed data were first analysed by ANOVA, however, the variable distribution undermined the validity of the analysis and non-parametric tests were then evaluated.

**Table 5.** Treatments applied to Nebbiolo in 2004/2005 at Lenswood for the control of downy mildew, using a Solo 475 backpack spray with a hand-held wand.

Treatment	Supplier	Date applied and rate				
		11/11/2004	25/11/2004	7/12/2004	21/12/2004	7/01/2005
<b>2004/2005</b>						
Copper (400 g/kg cupric hydroxide)	Yates copper fungicide	3 g/L	3 g/L	3 g/L	3 g/L	3 g/L
Acadian	Acadian Seaplants Limited	5 g/L	5 g/L	5 g/L	5 g/L	5 g/L
Acadian	Acadian Seaplants Limited	10 g/L	10 g/L	10 g/L	10 g/L	10 g/L
Synerscreen	Organic Crop Protectants	3 ml/L	3 ml/L	3 ml/L	3 ml/L	3 ml/L
Ecocarb + Synertrol Horti-Oil	Organic Crop Protectants	3 ml/L	3 ml/L	3 ml/L	3 ml/L	3 ml/L
Timorex	Biomor (Australia) Pty Ltd	7.5 ml/L	7.5 ml/L	7.5 ml/L	7.5 ml/L	7.5 ml/L
Ferrous sulfate + Synerscreen	Ace chemicals Organic Crop Protectants	3 g/L 3 ml/L	3 g/L 3 ml/L	3 g/L 3 ml/L	3 g/L 3 ml/L	3 g/L 3 ml/L
Brotomax	Agrometodos Biomor (Australia) Pty Ltd	5 ml/L	5 ml/L	5 ml/L	5 ml/L	5 ml/L
Timor		7.5 ml/L	7.5 ml/L	7.5 ml/L	7.5 ml/L	7.5 ml/L
Ferrous sulfate	Ace Chemicals	9 g/L	9 g/L	9 g/L	9 g/L	9 g/L
Control	Water					
Rate (L/Ha)		300	500	700	900	900



**Figure 5.** Nebbiolo vines at Lenswood, South Australia immediately after inoculation with *Plasmopara viticola*. Shoots were bagged overnight to maintain leaf wetness.

## 5.2. Research in Tasmania

Crop stage was identified using the modified E-L system described by Coombe (1995).

### **Small-plot field trials in southern Tasmania, 2003/04**

In October 2003 two small-plot field trials were established in the Coal River Valley of southern Tasmania. Details of site and treatments are given below. Treatments were applied to the crop canopy with a handgun connected to a hose reel and pump mounted on the flat tray of a utility vehicle. The spray was propelled with a pump pressure of 1,500-1,600 kPa, delivering approximately 63 ml/s. Spray coverage was checked using water-sensitive paper.

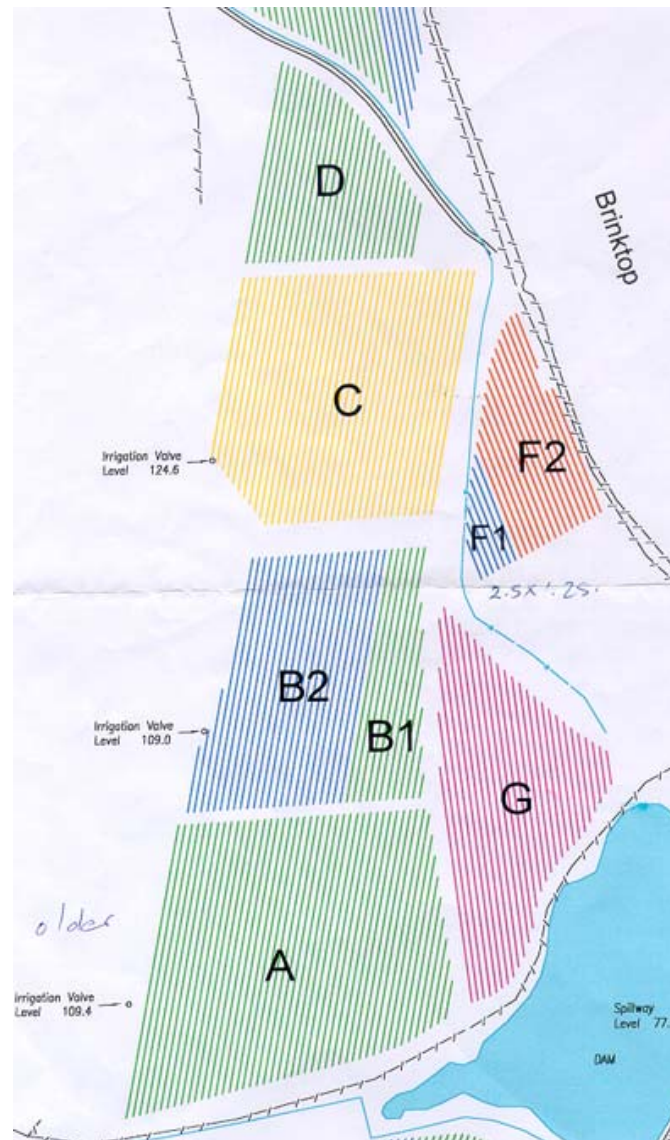
Each trial was inspected for powdery mildew at fortnightly intervals until early November and then at weekly intervals thereafter. Disease severity on leaves and bunches was assessed with the aid of standard area diagrams (R. Emmett pers. com.), for 40 leaves or 20-30 bunches per plot. Disease incidence was derived from the data for severity. From previous experience, assessment of powdery mildew close to veraison usually provides greater separation of treatments than assessment close to harvest. In Tasmania, the period from veraison to harvest can be more than 12 weeks. By harvest, active colonies of powdery mildew are difficult to detect and assessment of disease severity by the level of scarring and splitting becomes difficult, especially on red grapes. Therefore, emphasis was placed on the results of disease assessment at veraison.

ANOVA was used to identify treatments that were significantly different at  $P = 0.05$ . Homogeneity of variances of treatment means was checked by plotting fitted values against residuals. Data were transformed in some analyses, as indicated in tables of results, to reduce heterogeneity of variances among means. Analyses were performed using GenStat Release 8.1 (2005, Lawes Agricultural Trust, Rothamsted Experimental Station).

### **Frogmore Creek Chardonnay, 2003/04**

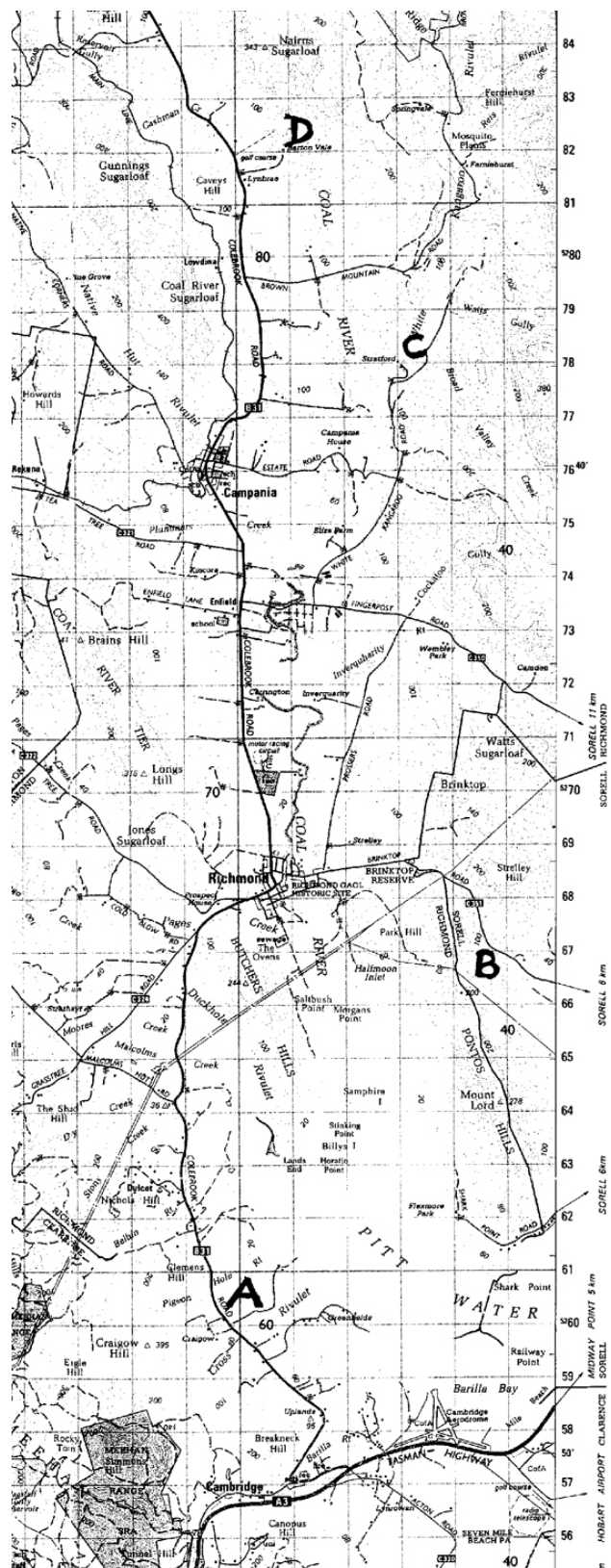
The first trial was located in Chardonnay at the south-west end of block F1 (Figure 6) at Frogmore Creek vineyard, near Richmond (Figure 7). This 26 ha vineyard had a range of soil types, including black cracking clay, and was managed organically. The ripening period was prolonged in 2003/04 and the Chardonnay was harvested on May 18, 2004. The vines, planted in 1999, were spaced at 1.25 m with 2.5 m between rows that were oriented north west to south east. The prevailing north-west winds flowed down the row and a sea breeze from the south often increased in strength in the afternoon. The open canopy had low vigour and was managed by vertical shoot positioning (VSP). The trial was designed as a randomised complete block with seven treatments in six blocks. Each plot had 5-9 vines, depending on patchiness in growth within sections of some vineyard rows.

Average air temperature, relative humidity, rainfall and other weather variables were recorded at 15 min intervals using a standard MEA automatic weather station (Measurement Engineering Australia) located at the south east corner of block B1 (Figure 6).



**Figure 6.** Layout of selected blocks at Frogmore Creek vineyard. The Measurement Engineering Australia automatic weather station was located at the south east corner of block B1.





**Figure 7.** Location of vineyards in the Coal River Valley, southern Tasmania (B) Frogmore Creek and (D) Cooinda Vale. Map reproduced from a section of Derwent 8312, Tasmania, 1:100,000 Land Tenure Index Series, Edition 1, 1985, Tasmap, Lands Department, Hobart.

**Treatments 2003/04**

1. untreated control
2. sulfur, Cosavet<sup>®</sup> DF (S present at 800 g/kg), 6 g/L
3. milk: bovine, 4% fat, pasteurised, diluted 1:10 with water
4. cheese whey (Lotus Foods Pty Ltd), non-hydroscopic powder (1090 mg sodium/100 g, 12.2 g protein/100 g), 25 g/L
5. potassium bicarbonate, Ecocarb<sup>®</sup> (Organic Crop Protectants Pty Ltd), 4 g/L
6. potassium bicarbonate plus riboflavin, Ecocarb SR (Organic Crop Protectants Pty Ltd), 4 g/L, or  
potassium bicarbonate plus riboflavin, Ecocarb SR, 3 g/L plus Protector (Organic Crop Protectants Pty Ltd), 10 ml/L
7. Program 1: canola-based oil, Synertrol Horti-oil<sup>®</sup> (Organic Crop Protectants Pty Ltd), 2ml/L plus Ecocarb<sup>®</sup>, 3 g/L, or cheese whey (as above), 15-25 g/L

Table 6 specifies the frequency of application, estimated spray volume and weather conditions during spraying. For Program 1, the mixture of Synertrol Horti-oil<sup>®</sup> and Ecocarb<sup>®</sup> was applied on 3 Oct., 23 Oct., 5 Nov., 17 Nov., 9 Dec. and 31 Dec. 2003. The whey treatment in Program 1 was applied at 25 g/L on Nov. 27 and at 15 g/L on Dec. 19 2003, Jan. 13 2004, Jan. 27 and 11 Feb.. Ecocarb SR was the only treatment applied beyond 11 Feb. 2004. After 11 Feb., Protector adjuvant was added to Ecocarb SR and the mixture was applied on 24 Feb., 11 Mar., 7 Apr. and 20 Apr.. Fulvic acid and kelp (Acadian<sup>®</sup>) were applied to the whole trial site on 5 Dec., 2003. Untreated plots and buffer rows surrounding the trial area were sprayed with sulfur (Cosavet<sup>®</sup> DF) at 6 g/L on 11 Feb. 2004.

**Table 6.** Schedule of treatments applied at Frogmore Creek vineyard Tasmania in 2003/04. A dash indicates missing information.

Date	Spray interval (days)	Crop Stage (modified E-L)	Estimated spray volume (L/ha)	Temperature °C	RH (%)
Oct 3	-	3-9	-	12.1-12.6	59-72
Oct 23	20	9-13, shoots 10-20 cm	560	8.6-11.7	49-71
Nov 5	13	12-13 (14), shoots 15-25 cm	500	15.1-19.1	44-68
Nov 17	12	13-15, flowers compact groups	900	-	-
Nov 27	10	15-16	-	11.1-13.4	62-70
Dec 9	12	19-20, 10% capfall	900	16.9-18.4	59-67
Dec 19	10	25, 90% capfall	-	18.3-18.7	72-75
Dec 31	12	26-27, berries 3-4 mm	900	16.4-20.8	33-57
Jan 13	13	29-30, berries 2-7 mm	1300	13.7-18.8	54- 74
Jan 27	14	29-31	1300	14.7-15.9	63-72
Feb 11	15	32-34	1300	14.2-15.1	58-61

**Cooinda Vale Cabernet Sauvignon, 2003/04**

The second trial was established in Cabernet Sauvignon, clone Laurel Bank 2217 from Granton, Tasmania, planted in 1997 at Cooinda Vale vineyard, 6 km north of Campania (Figure 7). The vines were spaced at 1 m with 2.3 m between rows that were oriented north north-east to south south-west. The canopy was managed conventionally by VSP and was vigorous relative to the Chardonnay canopy at the organic vineyard of Frogmore Creek. The



crop was harvested 31 May 2004. The trial was designed as a randomised complete block with seven treatments in six blocks and five vines per plot.

#### ***Treatments 2003/04***

1. untreated control
2. sulfur, Thiovit Jet<sup>®</sup> (S present at 800 g/kg), 12 g/L until 23 January 2004, then 6 g/L from 23 January, 2004
3. sulfur, Thiovit Jet<sup>®</sup>, 6 g/L
4. milk: bovine, 4% fat, pasteurised, diluted 1:10 with water until 23 January 2004, then 1: 5 with water from 23 January 2004
5. cheese whey (Lotus Foods Pty Ltd), non-hygroscopic powder (1090 mg sodium/100 g, 12.2 g protein/100 g), 25 g/L
6. potassium bicarbonate, Ecocarb<sup>®</sup> (Organic Crop Protectants Pty Ltd), 4 g/L
7. Program 1: canola-based oil, Synertril Horti-oil<sup>®</sup> (Organic Crop Protectants Pty Ltd), 2 ml/L plus Ecocarb<sup>®</sup>, 3 ml/L, or cheese whey (as above), 15-25 g/L

Table 7 specifies the frequency of application, estimated spray volume and weather conditions during spraying. For Program 1, the mixture of Synertril Horti-oil<sup>®</sup> and Ecocarb<sup>®</sup> was applied on 23 Oct., 5 Nov., 17 Nov., 8 Dec. and 30 Dec. 2003. The whey treatment in Program 1 was applied at 25 g/L on 26 Nov. and at 15 g/L on 19 Dec. 2003, 12 Jan. 2004, 23 Jan., 4 Feb. and 17 Feb.. On Nov. 26, block 5 of the Program 1 treatment was inadvertently sprayed with sulfur (Thiovit Jet<sup>®</sup>) at 12 g/L and block 5 of the 12 g/L sulfur treatment was sprayed with whey at 25 g/L (Program 1 treatment). These two plots were adjacent to each other. On Jan. 12, the west side of block 5 of the untreated control was inadvertently sprayed with sulfur (Thiovit Jet<sup>®</sup>) at 6 g/L. Untreated plots and buffer rows surrounding the trial area were sprayed with sulfur (Thiovit Jet<sup>®</sup>) at 6 g/L on Feb. 17, 2004. Between Feb. 24 and March 11, the whole trial area was sprayed by the grower cooperator with sulfur (Thiovit Jet<sup>®</sup>) at 12 g/L.

Total soluble solids content (°Brix) was estimated close to harvest by sampling five bunches per plot and extracting the juice using a domestic juice extractor. The juice from the five bunches was pooled and a 1-ml sub-sample placed on the well of a digital refractometer (Pocket PAL-1, Atago, Japan) for estimating °Brix.

#### ***Frogmore Creek, 2004/05***

In October 2004, two small-plot field trials were established in Pinot noir and Chardonnay in Blocks B1 and B2 (Figure 6), respectively, at Frogmore Creek vineyard. Vineyard details were as described previously and treatments are listed below. The Pinot noir canopy was trained to a Scott Henry trellis and the Chardonnay canopy had vertical shoot positioning with arched canes. The vines were spaced at 1.25 m with 2.5 m between rows that were oriented north to south. The Chardonnay vines in the trial area were harvested before berries were ripe on March 23, 2005, for production of verjuice. The Pinot noir vines were harvested in May, 2005.

Each trial was designed as a randomised complete block with six or seven treatments in six blocks. Each row that was treated was separated from other rows by a 'buffer' row for minimising spray drift (see details of treatment application below). The number of vines per plot was typically seven.

Weather data were collected from the MEA station as described previously. Disease assessment and data analyses were performed as described above.

**Table 7** Schedule of treatments applied at Cooinda Vale vineyard in 2003/04. A dash indicates missing information.

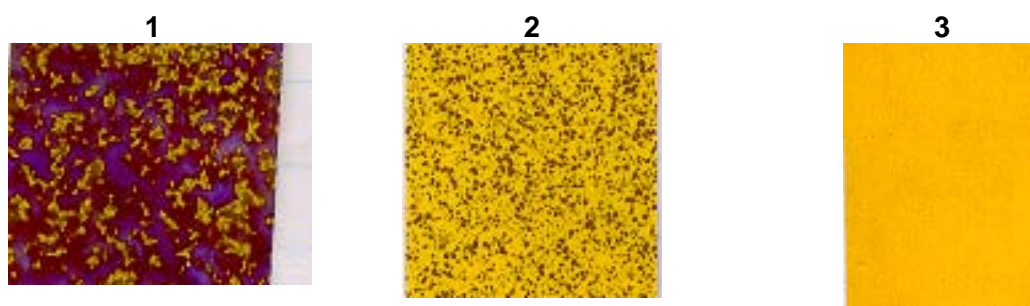
Date	Spray interval (days)	Crop Stage (modified E-L)	Estimated spray volume (L/ha)	Temperature °C	RH (%)
Oct 23	-	7-9	400	11.6-13	47-52
Nov 5	13	7-12, shoots 5-20 cm	800	10.5-11.7	71-83
Nov 17	12	13-15, flowers compact groups	1000	-	-
Nov 26	9	14-16-17	-	11-13.7	61-70
Dec 8	12	17-18	1000	15.9-17.4	59-63
Dec 19	11	21-23, 30-50% capfall	1000	16.5-19.9	68-83
Dec 30	11	25-26, 80-100% capfall	1000	21.4-30.5	23-42
Jan 12	13	27-29, berries 1-6 mm	1400	12.5-16	47-55
Jan 23	10	29-31, pre-bunch closure	1400	-	-
Feb 4	12	31-32, berries 7-10 mm, just beginning to close	1400	-	-
Feb 17	13	33-34, the odd berry was changing colour	1400	-	-

#### Treatment application and spray coverage

All treatments up to and including the spray date of 17 January 2005 were applied with a small plot fan-assisted sprayer. The fan, nozzles and pump were mounted on a trailer and connected to the 'power take off' of a small tractor (Figure 8a). The spray was propelled with a pump pressure of 550 kPa, delivering approximately 73 ml/s. The angle of the fan and orientation of the nozzles were adjusted to reduce overhead drift to adjacent plots. Water-sensitive papers showed that buffer rows received significant spray material, but the second row across from the sprayed row did not (Figure 8b).



**Figure 8.** Spray application in 2004. (a) Small plot air-blast sprayer in action on 18 October 2004. The sprayer was built and operated by Dr Gordon Brown, Scientific Horticulture Pty Ltd, Tasmania.



**Figure 8.** Spray application in 2004. (b) Water sensitive papers illustrating spray coverage (750 L/ha) on 29 October, 2004, in (1) a treated plot, (2) also in a treated plot, but also seen in a buffer row, and (3) in the second row across from the treated plot.

#### ***Treatments 2004/05***

Treatments 1 to 7 (listed below) were applied to Chardonnay and Pinot noir with the exception that treatment 5 was not applied to Pinot noir. For commercial reasons, it was necessary to minimise potential damage to the valuable Pinot noir grapes and so there was only one untreated plot in the Pinot noir, which was located in the centre of the trial area. Table 8 describes when each component of the spray program was applied. Table 9 lists the schedule of treatments.

1. untreated control
2. sulfur, Cosavet<sup>®</sup> DF (S present at 800 g/kg), 6-10 g/L
3. milk: bovine, 4% fat, pasteurised, diluted 1:10 or 1:5 with water
4. cheese whey (Lotus Foods Pty Ltd), non-hydroscopic powder (1090 mg sodium/100 g, 12.2 g protein/100 g), 25 g/L
5. Synertrol Horti-oil<sup>®</sup> (Organic Crop Protectants Pty Ltd), 3 ml/L plus Ecocarb<sup>®</sup>, 6 g/L, or sulfur, Cosavet<sup>®</sup> DF (S present at 800 g/kg), 6-10 g/L
6. canola-based oil, Synertrol Horti-oil<sup>®</sup> (Organic Crop Protectants Pty Ltd), 3 ml/L plus Ecocarb<sup>®</sup>, 6 g/L, or cheese whey (as above), 25 g/L, or sulfur, Cosavet<sup>®</sup> DF, 8 g/L

**Table 8.** Crop stage at which each component of the spray program was applied for treatments using more than one material or dose.

<b>Treat- ment</b>	<b>1. Pre-flowering E-L 12 to E-L 17</b>	<b>2. Flowering to fruit set E-L 19 to E-L 28</b>	<b>3. Fruit set to veraison E-L 29 to E-L 34</b>
3	milk 1:10	milk 1:5	milk 1:10
5	Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup>	whey	whey
6	Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup>	sulfur 6-10 g/L	whey
7	Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup>	whey	sulfur 8 g/L

**Table 9.** Schedule of treatments applied at Frogmore Creek vineyard in 2004/05. The number in parenthesis after the date is the crop stage as defined in Table 8. A dash indicates missing information.

Date	Spray interval (days)	Crop stage (modified E-L)	Estimated spray volume (L/ ha)	Temp. °C	RH (%)	Material/s applied to buffer rows <sup>e</sup>
Oct 18 (1)	-	11-12-13	900	12.7-16.5	45-61	No spray
Oct 29 (1)	11	12-14	750	10.5-13.9	54-66	No spray
Nov 9 (1)	11	15-16	750	12-13.9	66-78	No spray
Nov 24 (1)	15	17	700	11.9-15.9	61-93	Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup>
Dec 4 (2)	10	19-20, 10% caps off mainly on lower bunches	700	13.3-19.7	34-70	whey @ 25 g/L
Dec 16 (2)	12	25-26-27, 80% to capfall complete	700	14.1-17.5	48-61	sulfur @ 6 g/L
Dec 24 <sup>a</sup> (2)	8	27-29, fruit set to pepper-corn size berries	700	17.4-20.3	57-72	sulfur @ 10 g/L
Jan 3 <sup>b</sup> (3)	10	27-31, up to pea size	700	13.3-16.1	75-95	whey @ 25 g/L
Jan 17 <sup>c</sup> (3)	14	29-32, up to bunch closure	700	-	-	sulfur @ 8 g/L
Feb 11 <sup>d</sup> (3)	25	33-34, pre-veraison	600	-	-	

<sup>a</sup> Sulfur applied at 10 g/L

<sup>b</sup> Sulfur applied at 8 g/L

<sup>c</sup> Sulfur applied at 8 g/L and to all untreated control plots

<sup>d</sup> Grower sprayed the whole trial area with Cosavet sulfur at 1 kg/100 L plus Synertrol Horti-oil<sup>®</sup> at 250 ml/100 L

<sup>e</sup> Materials were applied to rows between treatment rows with the fan of the air-blast sprayer switched off

### Assessment of canopy density

The point quadrat method (Smart and Robinson 1991) was used to characterise the canopy density for each cultivar. The point quadrat is a thin metal rod, which is inserted into a canopy. It represents a beam of light that passes from the canopy exterior to the interior. Contact of the rod with parts of the canopy relates to their exposure to sunlight. The method is used to estimate the proportion of leaves and fruit that are exterior or interior in the canopy.

Measurements were taken on 13 January 2005, by selecting representative sections of the canopy within plots of the untreated and sulfur treatments in Chardonnay and the whey and sulfur treatments in the Pinot noir. A metre ruler was suspended in the fruiting zone from the top foliage wire. The metal rod was inserted into the canopy at 10-cm intervals using the ruler as a guide. Each leaf (L) and cluster (C) contact was recorded as the metal rod was inserted into the canopy. If there was no contact, it was recorded as a gap (G). Ten measurements were taken in each of six plots of each treatment. Therefore, there was a total of 120 measurements for each grapevine variety. In addition, the length and number of nodes on three upright shoots per plot near the 50-cm mark of the ruler was measured.

### Calculations

Percentage gaps: Total number of gaps (G) divided by number in insertions (60) multiplied by 100.

Leaf layer number (LLN): Total number of leaf contacts (L) divided by the number of insertions (60).

Percentage interior leaves: Number of interior leaves (L) (i.e. not at either surface), divided by the total number of leaves, multiplied by 100.

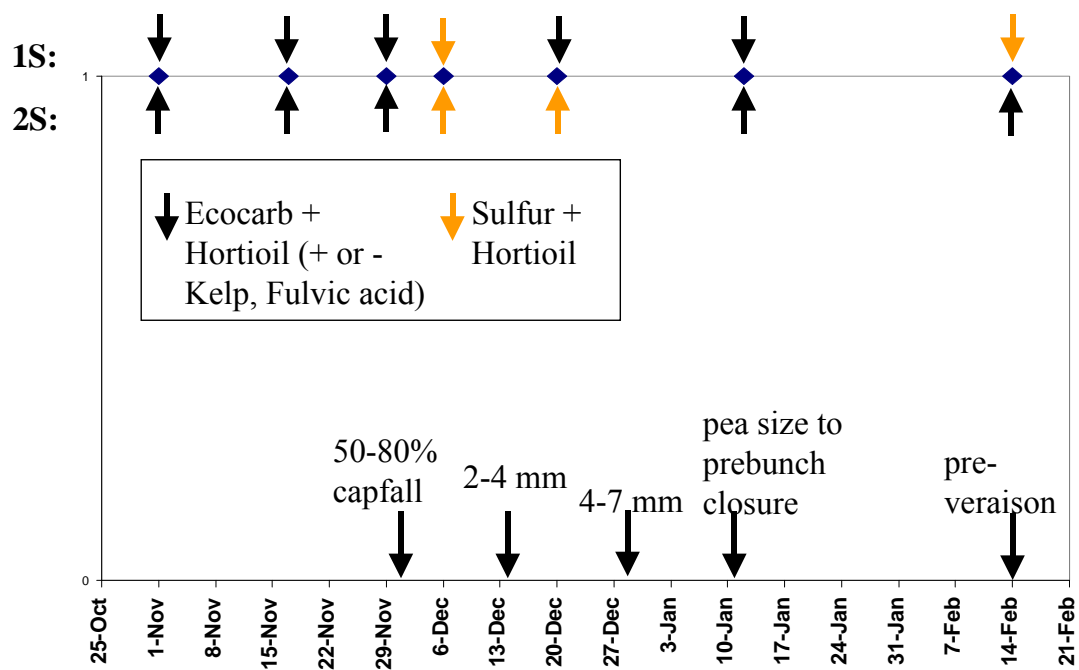
Percentage interior clusters: Number of interior clusters (C) (i.e. not at either surface), divided by the total number of clusters, multiplied by 100.

### **Evaluating organic spray programs in Tasmania using spatial information**

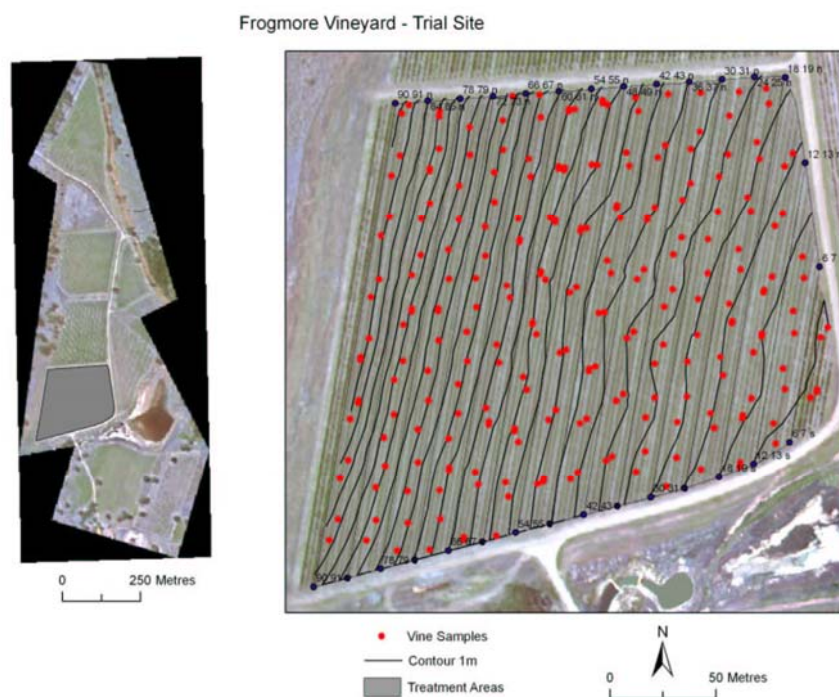
In 2005/06, each of two spray programs was applied using a commercial fan-assisted sprayer to a block of six rows, with each program applied in an alternating pattern across a 4.5 ha block of Pinot noir with 92 rows at Frogmore Creek (organic) vineyard. A mixture of Synertrol Horti-oil<sup>®</sup> (250 ml/100 L) plus Ecocarb<sup>®</sup> (400 g/100 L) was applied to the whole block from E-L stage 16 until the beginning of capfall. The next two applications of fungicide comprised a) Program S2: two applications of sulfur (800 g/100 L) mixed with Synertrol Horti-oil<sup>®</sup> (250 ml/100 L) or b) Program S1: one application of the sulfur/oil mixture followed by an application of the Ecocarb<sup>®</sup>/oil mixture. The Ecocarb<sup>®</sup>/oil mixture was then applied to the whole block for the remainder of the season until veraison (Figure 9).

Powdery mildew was assessed on at least 116 single vines per spray program. Single vines were sampled in the middle two rows of the six row 'block', using a regular grid but with sampling points removed at random from 15% of the grid nodes and reallocated to vines adjacent to other grid nodes (Bramley 2005, Figure 10). Each of the sample vines was geo-referenced using a real-time kinematic global positioning system (GPS). Single vines were monitored weekly until powdery mildew was detected. Once disease was evident, single vines were assessed weekly or fortnightly and scored for the presence or absence of powdery mildew on leaves or bunches. Disease incidence and severity on the abaxial surface of 20 leaves was also assessed pre-flowering according to methods described above. At veraison, mean disease severity per vine was scored for 20 bunches per vine.

Average air temperature, relative humidity, rainfall and other weather variables were recorded at 15 min intervals using a standard MEA automatic weather station (Measurement Engineering Australia) located at the north east corner of the block.



**Figure 9.** Schematic representation of the two spray programs applied to Pinot noir at Frogmore Creek vineyard in 2005/06.



**Figure 10.** Location of vines used for assessment of powdery mildew in 4.5 ha of Pinot noir, Frogmore Creek vineyard. Contour lines indicate the slope of the block.



### 5.3. Research in New South Wales

The Charles Sturt University Commercial Vineyard (Stage 1), Wagga Wagga, NSW (approximately 452 km SW of Sydney) was used for the trials. Wagga Wagga has a warm and dry climate with an annual mean rainfall of 572 mm. Maximum temperatures in summer average between 29°C and 32°C with a 3 pm average relative humidity of approximately 30% (Australian Bureau of Meteorology, [www.bom.gov.au](http://www.bom.gov.au)). Powdery mildew occurs every season when control measures are less than optimal.

Cultivars Chardonnay (Figure 11a) and Cabernet Sauvignon (Figure 11b) were used for the field trials conducted in the 2003/04 and 2004/05 growing seasons. In 2005/06 the trial was conducted on Chardonnay only. Both were trained to a single wire (“Aussie sprawl”) and were spur pruned. The spacing between rows was 3 m and the spacing between vines (within row) was 1.5 m. There were approximately 2200 vines per hectare, with rows orientated in a north-south direction.

All trials were established as a randomized complete block design.



**Figure 11.** (a) Chardonnay trial site and (b) Cabernet Sauvignon trial site

#### **Trial design**

##### **2003/04**

There were six replicates of six vines per treatment for Chardonnay and four replicates of six vines per treatment for Cabernet Sauvignon. Plots consisted of two panels containing six vines. Only the four middle vines for each plot were assessed for symptoms of disease. The total number of vines per treatment was 36 for Chardonnay and 24 for Cabernet Sauvignon. Each treatment within a row was separated by a buffer panel consisting of three vines.

##### **2004/05**

Four blocks were established for each of Chardonnay and Cabernet Sauvignon with six plots in each block. Each row contained six treatments. Each plot consisted of eight vines and the six middle vines were assessed for disease. The total number of vines per treatment for each variety was 32. Each treatment within a row was separated by one buffer vine.

##### **2005/06**

The Cabernet Sauvignon vines were removed at the end of the 2005. Five blocks were established for Chardonnay with six plots in each block. Each row contained six treatments. Each plot consisted of six vines and the middle four vines were assessed for disease. The total number of vines per treatment was 30. Each treatment row was separated by one buffer vine.

#### **Treatments in 2003/04**

At woolly bud (E-L 3) vines were sprayed with sulfur (5 g/L, Thiovit<sup>®</sup> Jet, Syngenta Australia) for the control of mites. The treatments included pasteurised bovine milk

(approximately 4% fat, 1:10 dilution), spray dried non-hygroscopic whey powder (25 g/L, Bonlac Foods Ltd, Victoria), Ecocarb<sup>®</sup> (4 g/L, activated potassium bicarbonate, Organic Crop Protectants Pty Ltd), and sulfur (3 g/L, Thiovit<sup>®</sup> Jet, Syngenta Australia). A program consisting of the first two sprays of a mixture of Ecocarb<sup>®</sup> (3 g/L) and Synertrol Horti-oil<sup>®</sup> (2 ml/L) followed by whey (15 g/L) and alternated with the Ecocarb<sup>®</sup> and Synertrol Horti-oil<sup>®</sup> mixture until veraison. Untreated vines acted as controls. The spray schedule is detailed in Table 10.

**Table 10.** Spray schedule for 2003/04.

	Day 1	Day 34	Day 42	Day 56	Day 86	Day 100
1. Untreated	-	-	-	-	-	-
2. Sulfur (g/L)	3	3	3	3	3	3
3. Milk (dilution)	1:10	1:10	1:10	1:10	1:10	1:10
4. Whey (g/L)	25	25	25	25	25	25
5. Ecocarb <sup>®</sup> (g/L)	4	4	4	4	4	4
6. Program 1 <sup>1</sup>	SH/E	SH/E	W	SH/E	W	SH/E

<sup>1</sup>A mixture of Synertrol Horti-Oil<sup>®</sup> (2ml/l) and Ecocarb<sup>®</sup> (3 g/l); W = whey (15 g/L).

### Treatments in 2004/05

At woolly bud (E-L 3) vines were sprayed with sulfur (5 g/L, Thiovit<sup>®</sup> Jet) for the control of mites. The treatments included pasteurised bovine milk (approximately 4% fat, 1:5 dilution from flowering to capfall and 1:10 dilution at all other times), spray dried non-hygroscopic whey powder (25 g/L), Surround<sup>™</sup> WP (first spray at 50 g/L, all others at 25 g/L, Engelhard, NJ, USA), a mixture of Ecocarb<sup>®</sup> (4 g/L, activated potassium bicarbonate) and Synertrol Horti-oil<sup>®</sup> (3 ml/L), and sulfur (3 g/L; Thiovit<sup>®</sup> Jet). Program 1, consisting of the first two sprays of a mixture of Ecocarb<sup>®</sup> (4 g/L) and Synertrol Horti-oil<sup>®</sup> (3 ml/L), followed by whey (15 g/L), then whey alternated with the Ecocarb<sup>®</sup> and Synertrol Horti-oil<sup>®</sup> mixture until veraison. Following advice from the CSU vineyard manager, control plots were sprayed with sulfur due to the high incidence of powdery mildew in unsprayed plots in the 2003/2004 trial. The spray schedule is detailed in Table 11.

### Treatments in 2005/06

Test materials were applied in programs targeted at the different phenological stages of the vine. At woolly bud (E-L 3) vines were sprayed with sulfur (5 g/L; Thiovit<sup>®</sup> Jet) for the control of mites. The treatments included pasteurised bovine milk (approximately 4% fat, 1:10 dilution), a mixture of Ecocarb<sup>®</sup> (4 g/L; activated potassium bicarbonate) and Synertrol Horti-oil<sup>®</sup> (3 ml/L), and sulfur (6 g/L; Thiovit<sup>®</sup> Jet). Control plots were sprayed with sulfur due to the high incidence of powdery mildew in unsprayed plots in the 2003/2004 trial. The spray schedule is detailed in Table 12.

### Application of treatments

The treatments were first applied using a spray wand attached to a 12V pump on the back of a four-wheel bike (Figure 12a). Treatments were delivered from 20 L containers. As the canopy increased in size, a Solo<sup>®</sup> 450 motorised mist blower was used to deliver a higher rate of spray (Figure 12b). The spray was propelled with a pump pressure of 300 to 600 KPa delivering 56 ml/sec. Spray rates varied from 300 L/ha at the beginning of the growing season to 900 L/ha at veraison. Water-sensitive papers (TeeJet<sup>®</sup>, Wheaton, IL USA) were used at the fourth application to monitor spray distribution. The distribution varied from 258 droplets/cm<sup>2</sup> to overdosing. Care was taken to ensure that optimum coverage of leaf and bunch surfaces was achieved thereafter.

The first spray application was when shoots were approximately 15 cm in length. Where possible (no rainfall predicted for 24 hours, winds less than 8.1 knots, and predicted



temperature not over 35°C) treatments were applied every 7 to 14 days until veraison. All sprays were applied between 6.00 am and 2.00 pm.

Vines beyond the trial block were sprayed for powdery mildew with a conventional program consisting of sulfur and demethylation inhibiting fungicides.



**Figure 12.** Spray application applied with (a) a spray wand and (b) a mist blower.

**Table 11.** Spray schedule for trials in the CSU vineyard in 2004/05.

Treatment	Day 1	Day 12	Day 22	Day 32	Day 43	Day 56	Day 78	Day 89	Day 113
1. Sulfur (g/L)	3	3	3	3	3	3	3	3	3
2. Milk (dilution)	1:10	1:10	1:10	1:10	1:10	1:5	1:5	1:10	1:10
3. Whey (g/L)	25	25	25	25	25	25	25	25	25
4. Ecocarb <sup>®</sup> (g/L) plus	4	4	4	4	4	4	4	4	4
Synertrrol Horti-oil <sup>®</sup> (ml/L)	3	3	3	3	3	3	3	3	3
5. Surround <sup>TM</sup> WP (g/L)	50	25	25	25	25	25	25	25	25
6. Program 1 <sup>1</sup>	SH/E	SH/E	W	SH/E	W	SH/E	W	SH/E	W

<sup>1</sup>A mixture of Synertrrol Horti-Oil<sup>®</sup> (3ml/L) and Ecocarb<sup>®</sup> (4 g/L); W = whey (25 g/L).

**Table 12.** Treatments applied to trial in the CSU vineyard in 2005/06.

	Phenological Stage									
	1 <sup>st</sup> leaf unfolded to flowering			Flowering to fruit-set			Fruit-set to veraison			
Treatment/Date	23/9	10/10	28/10	8/11	14/11	1/12	8/12	21/12	5/1	20/1
1. (S <sup>1</sup> S S)	S	S	S	S	S	S	S	S	S	S
2. (S SH/E <sup>2</sup> M <sup>3</sup> )	S	S	S	SH/E	SH/E	SH/E	M	M	M	M
3. (S M M)	S	S	S	M	M	M	M	M	M	M
4. (S S M)	S	S	S	S	S	S	M	M	M	M
5. (SH/E M S)	SH/E	SH/E	SH/E	M	M	M	S	S	S	S
6. (SH/E S M)	SH/E	SH/E	SH/E	S	S	S	M	M	M	M

<sup>1</sup>Sulfur (6 g/L; Thiovit<sup>®</sup> Jet)

<sup>2</sup>A mixture of Synertrrol Horti-Oil<sup>®</sup> (3 ml/L) and Ecocarb<sup>®</sup> (4 g/L)

<sup>3</sup>Pasteurised bovine milk (approximately 4% fat)

### Disease assessment

For 2003/04 and 2005/06, randomly selected leaves (10) and bunches (10) were assessed, non-destructively, from each of the four centre vines in each plot, five from each side of the vine on a 1 to 10 scale (Table 13) based on percentage of leaf or bunch area with sporulating colonies of *E. necator*. The method of assessment was the same in 2004/05 however six centre vines in each plot were assessed rather than four. Assessments, where possible, were made every fortnight in the two to three days prior to spray application, from bud burst until harvest. Vines were also monitored for the presence or absence of flag shoots. Table 14 shows the dates on which the incidence and severity of powdery mildew were monitored for each of the four centre vines in each plot.

**Table 13.** Disease assessment score table for powdery mildew in NSW.

Score	Percentage bunch/leaf area with sporulating colonies of <i>E. necator</i>
0	
1	1-10
2	11-20
3	21-30
4	31-40
5	41-50
6	51-60
7	61-70
8	71-80
9	81-90
10	91-100

### Assessment of humidity, temperature and dew point

In the 2004/05 and 2005/06 growing seasons, five Hobo<sup>®</sup> U12 Temperature and Relative Humidity Data Loggers were randomly placed in the canopy at budburst. Temperature, relative humidity and dew point were recorded every 20 mins from budburst to veraison. The temperature data were used to calculate the Gubler-Thomas powdery mildew risk index (Gubler et al. 1999).

### Analysis of canopy density

To measure the canopy density of Chardonnay and Cabernet Sauvignon a plant canopy analyzer (LAI-2000, LI-COR<sup>®</sup>, NE, USA) was used to determine the leaf area index at berries pea-sized in 2004. Measurements were taken for all treated vines.

### Assessment of berry quality

At harvest for 2003/04 and 2004/05, 50 berry samples (25 from each side of the vine row) were taken from each treatment and the mean berry weight, pH, total soluble solids (TSS) and TA of juices calculated. Chardonnay berries were not tested in 2004/05.

### Analysis of shoot length and leaf number

In 2004/2005, shoot length and leaf number for Chardonnay and Cabernet Sauvignon vines treated with Surround<sup>™</sup> WP and sulfur were measured at flowering, bunch closure and veraison. The length of two shoots per vine, one on each side (12 shoots per treatment) was measured. The number of leaves on each shoot (total number of nodes) was counted.

### Data analysis

Data were subjected to an analysis of variance (ANOVA) using the CoStat statistical package (version 6.303, CoHort Software, Monterey, CA, USA). Means were separated by the least significant difference test at  $P = 0.05$ .

**Table 14.** Disease monitoring schedule for Chardonnay and Cabernet Sauvignon in 2003/04 and 2004/05 and Chardonnay in 2005/06 in NSW.

Date	Phenological Stage
<b>2003/04<sup>1</sup></b>	
23 Oct	E-L 9-12, 3 leaves unfolded to inflorescence visible
5 Dec	E-L 29-31, 3mm to berries pea-sized
5 Jan	E-L 32-33, bunch closure
18 Jan	E-L 32-35, bunch closure to veraison
9 Feb	E-L 38, harvest Chardonnay
5 Mar	E-L 38, harvest cabernet Sauvignon
<b>2004/05<sup>1</sup></b>	
29 Sept	E-L 9-12, 3 leaves unfolded to inflorescence visible
6 Oct	E-L 9-15, 3 leaves unfolded to inflorescence swelling
15 Oct	E-L 9-17, 3 leaves unfolded to inflorescence fully developed
29 Oct	E-L 17-19, inflorescence fully developed to start of flowering
18 Nov	E-L 19, start of flowering
2 Dec	E-L 31, berries pea-sized
20 Dec <sup>2</sup>	E-L 32, bunch closure
4 Jan	E-L 32, bunch closure
18 Jan	E-L 35, veraison
4 Mar	E-L 38, harvest
<b>2005/06<sup>3</sup></b>	
23 Sept	E-L 7-9, 1 <sup>st</sup> to 3 leaves unfolded
5 Oct	E-L 12, inflorescence visible
17 Oct	E-L 15, inflorescence swelling
31 Oct	E-L 15, inflorescence swelling
16 Nov	E-L 19-23, start flowering to 50% cap fall
5 Dec	E-L 29-31, 3mm to pea size
12 Dec	E-L 31-32, pea size to begin bunch closure
23 Dec	E-L 31-32, pea size to begin bunch closure
9 Jan	E-L 32-33, bunch closure
23 Jan	E-L 32-35, bunch closure to veraison
7 Feb	E-L 38, harvest

<sup>1</sup>Monitoring schedule for Chardonnay and Cabernet Sauvignon.

<sup>2</sup>Chardonnay sprayed with Bayfidan® 250 EC due to high incidence of powdery mildew. All remaining dates are monitoring conducted for Cabernet Sauvignon.

<sup>3</sup>Monitoring schedule for Chardonnay.

## 6. Results and Discussion

### 6.1. South Australia

#### Powdery mildew

##### Field trials in Langhorne Creek

In the 2003/04 season, all materials applied to Verdelho vines at Langhorne Creek significantly reduced the severity of powdery mildew on treated vines when compared with untreated vines (Table 15). Disease was first detected on Verdelho leaves on 3 November 2003 and on fruit on 25 November 2003.

The severity of powdery mildew on foliage of vines sprayed with milk (1% of leaf surface affected), whey (2%), and Ecocarb plus Synertrol Horti-Oil (4%) was not significantly different from that on vines sprayed with sulfur (1%) (Table 15). Vines that received Program 1 had significantly more severe disease (9%) than those sprayed with other materials, including the components of the program. All treated vines had significantly less powdery mildew than untreated control vines (80%). On untreated vines 80% of the leaf area was affected by powdery mildew by mid-December and vines were sprayed with sulfur on 24 December to reduce the risk of contamination of other plots and the rest of the vineyard. Leaf results presented are for 24 December 2003 prior to application of sulfur.

The severity of powdery mildew on fruit of vines sprayed with milk (3% of bunch surface affected), whey (3%), and Ecocarb plus Synertrol Horti-Oil (7%) was not significantly different from that on vines sprayed with sulfur (2%) (Table 15). Vines that received Program 1 had significantly more severe disease than those sprayed with the other materials. Disease severity on the untreated control vines at harvest was 100%

No powdery mildew was detected during routine assessments of leaves and bunches in 2004/05 (Table 15). However, examination of bunches collected from vines sprayed with all materials, with a dissecting microscope, revealed occasional colonies of *E. necator* on the rachis and aborted berries.

In 2005/06, disease on the untreated controls was less severe than in 2003/04. The severity of powdery mildew on vines sprayed with milk (0.1% of leaf surface affected), whey (0.1%), and Ecocarb plus Synertrol Horti-Oil (0.2%) was not significantly different from that on vines sprayed with sulfur (0.1%) (Table 15). Vines that received Program 1 had significantly more severe disease (0.8%) than those sprayed with the other materials. All vines had significantly less powdery mildew than the untreated control vines (3%). The severity of powdery mildew on bunches followed a similar pattern; disease on vines treated with sulfur (0.2%), milk (0.3%), whey (0.1%), Ecocarb plus Synertrol (0.6%), and Program 1 (0.7%) was significantly less severe than on the untreated control vines (4.7%).

No detectable powdery mildew developed on the leaves or bunches of Shiraz vines, including the untreated control vines, at Langhorne Creek in any of the three seasons. These Shiraz vines were in the rows immediately adjacent to the Verdelho rows where in 2003/2004 the untreated vines exhibited 100% bunch infection.

The only phytotoxicity detected in the three years of the trial at Langhorne Creek was in Verdelho grape bunches in the 2005/2006 season on vines sprayed with sulfur (Figure 13). In this vineyard the alternative fungicides controlled powdery mildew to levels not significantly different from sulfur, except Program 1 where the two products applied in rotation were less effective than either product applied alone. However, in two of the three seasons disease pressure was low and incidence across the vineyard was less than in previous

seasons (D. Bruer pers. com.). While rapid development of powdery mildew in vines nearby suggested weather conditions were conducive to the development of powdery mildew, vines in that area appeared more vigorous than in the majority of the vineyard and more flag shoots were found in those vines.

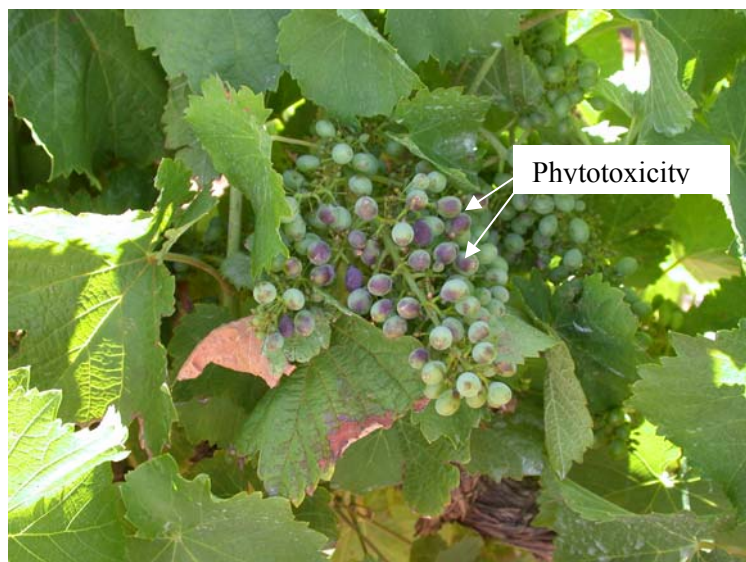
Powdery mildew is now controlled in the Langhorne Creek vineyard without sulfur except in the area mentioned above.

The difference in powdery mildew severity between cultivars clearly demonstrated the potential to vary spray programs between cultivars and reduce the number of spray applications and associated costs. The variation in susceptibility could also be used to select cultivars for planting depending on the disease pressure anticipated in a vineyard.

**Table 15.** Powdery mildew severity on leaves and bunches of Verdelho at harvest after five to seven applications of test materials, expressed as a percentage of surface area with sporulating colonies in three growing seasons. Results within columns with the same letters are not significantly different ( $P=0.05$ ).

Treatment	2003/04		2004/05		2005/06	
	Leaf	Bunch	Leaf	Bunch	Leaf	Bunch
Sulfur	1 a	2 a	0 a	0 a	0.1 a	0.2 a
Milk	1 a	3 a	0 a	0 a	0.1 a	0.3 a
Whey	2 a	3 a	0 a	0 a	0.1 a	0.1 a
Ecocarb plus Synertrol Horti-Oil	4 a	7 a	0 a	0 a	0.2 a	0.6 a
Program 1	9 b	13 b	0 a	0 a	0.8 b	0.7 a
Untreated	80 c*	100 c	0 a	0 a	3 c	4.7 b

\* Result for 24 December 2003, when control vines were sprayed with sulfur to limit risk to the vineyard.



**Figure 13.** Sulfur-treated Verdelho at Langhorne Creek vineyard, displaying phytotoxicity.

There was no significant difference in yield among treatments in any season except for Verdelho in 2004 when 100% yield loss was observed for the untreated control vines (Table 16).

**Table 16.** Average yield in kg of fruit harvested per panel of 4 vines of Verdelho and Shiraz in the Langhorne Creek vineyard for after 5 – 7 sprays of test materials. Results within columns with the same letters are not significantly different ( $P=0.05$ ).

<b>Verdelho</b>	<b>2004</b>	<b>2005</b>	<b>2006</b>	<b>3-year total</b>
Untreated	0 b	12 a	10 a	22 b
Ecocarb	12 a	12 a	12 a	36 a
Milk	16 a	12 a	13 a	41 a
Program 1	16 a	12 a	12 a	40 a
Sulfur	16 a	11 a	11 a	38 a
Whey	12 a	11 a	11a	34 a
<b>Shiraz</b>	<b>2004</b>	<b>2005</b>		<b>2 -ear total</b>
Untreated	9 a	7 a		16 a
Ecocarb	10 a	8 a		18 a
Milk	10 a	8 a		18 a
Program 1	11 a	8 a		19 a
Sulfur	9 a	7 a		16 a
Whey	8 a	8 a		16 a

### Field trials in Lenswood

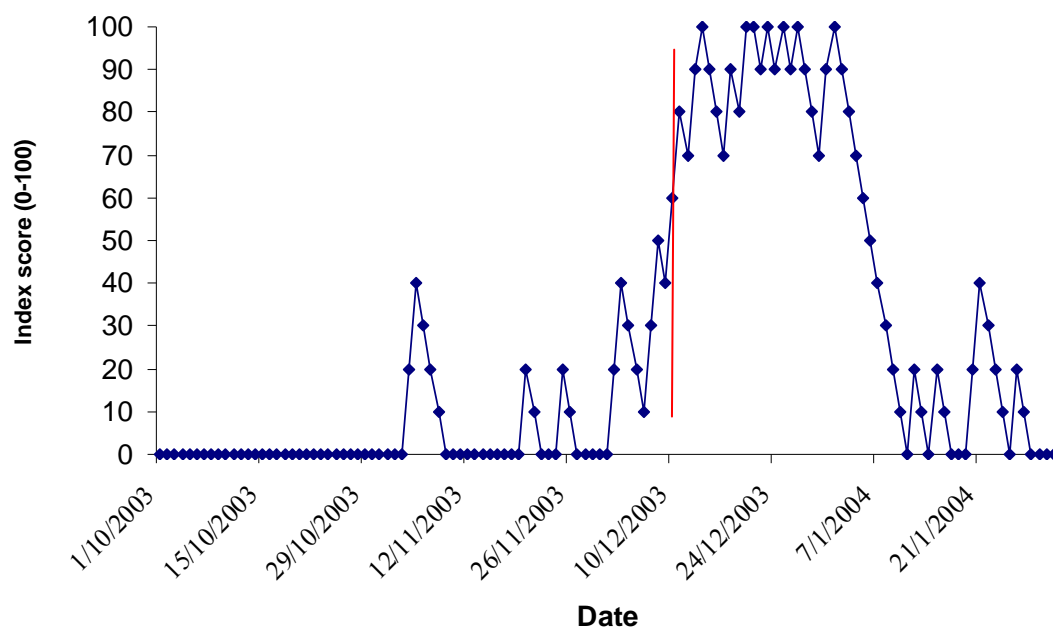
Minimal powdery mildew was detected on the Nebbiolo vines at Lenswood in 2004/2005 until more than 4 weeks after the last application of test materials. As such no conclusions can be drawn on the efficacy of the treatments applied for control of powdery mildew. Powdery mildew was detected on vines at Lenswood in January 2006, however, assessment of disease severity and incidence was confounded by the severe downy mildew that had established on most vines. Therefore, the effect of the materials on the severity or incidence of powdery mildew could not be assessed. The treatments had no obvious effect on downy mildew.

### Evaluation of the Gubler-Thomas index

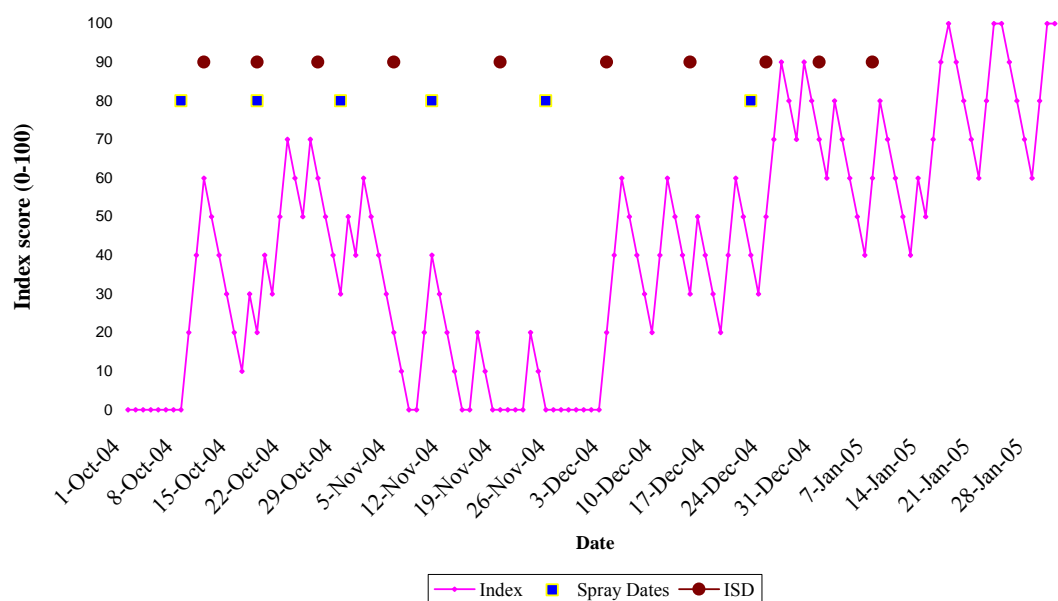
In 2003/2004 the Gubler-Thomas index did not reach 60, where spraying should commence, until 10 December 2003 by which time powdery mildew had affected 53% and 51% of the surface of leaves and bunches of untreated control Verdelho vines, respectively (Figure 14). In 2004/2005 the Gubler-Thomas index reached 60 on 12 October 2004 and “recommended” ten spray applications throughout the season. The spray program based on monitoring of powdery mildew included six spray applications; however, the last three were not required for disease control and were applied only to maintain spray conditions similar to 2003/2004 for the microbiology component of this research (Figure 15).

Table 17 summarises three possible spray programs for the Langhorne Creek vineyard: the actual spray program based on monitoring; a theoretical calendar-based schedule; outcome if the Gubler-Thomas index had been used. In 2003/04, use of the Gubler-Thomas index would have resulted in significant losses to powdery mildew in the Verdelho, whereas in 2004/05 and 2005/06 use of the index would have resulted in unnecessary spray applications and additional costs. The calendar-based spray program would have resulted in three unnecessary spray applications in 2004/05 and 2005/06, and would have resulted in additional cost.

The experience at the Langhorne Creek vineyard was that monitoring was the most cost effective and reliable method of planning spray programs. In large vineyards, disease “hot spots” can be monitored and, once disease has been detected, a wider survey can be conducted to establish the severity of the disease within the vineyard and determine suitable management strategies.



**Figure 14.** Gubler-Thomas index for Langhorne Creek vineyard (SA), 2003/04, spray program initiation point 60 indicated by vertical red line.



**Figure 15.** Gubler-Thomas index for Langhorne Creek vineyard (SA), 2004/05. Actual spray dates based on monitoring disease severity are indicated, Gubler-Thomas index spray dates (ISD) are also indicated.



**Table 17.** Spray application summary for Verdelho and Shiraz at the Langhorne Creek vineyard, 2003-2006.

Cultivar	Season	Bunch severity (untreated)	Flagshoots (per plot)	No. sprays actually applied	1st spray	Calendar sprays	Outcome if Gubler Thomas index applied
Verdelho	2003/04	100%	7	8	15-Oct	6	possible loss
	2004/05	0	0	3+3*	8-Oct	6	extra cost
	2005/06	4.7%	0	3+2*	5-Oct	6	extra cost
Shiraz	2003/04	0	0	8	15-Oct	6	extra cost
	2004/05	0	0	3+3*	8-Oct	6	extra cost
	2005/06	0	0	3+2*	5-Oct	6	extra cost

\* Applications not necessary for disease control but included for the microbiology research.

### Germination of *E. necator* conidia

All test materials reduced germination of conidia of *E. necator* on agar and disease severity on grape leaves when applied 6 hours before inoculation. Almost 90% of conidia germinated on 2% water agar (controls). All treatments reduced germination on agar compared with the controls (Table 18). Spores on plates treated with milk, sulfur or lactoferrin exhibited least germination, 3.3, 5.7 and 1.5% respectively. Approximately 85% of the leaf surface area on the water-treated controls was affected by powdery mildew. Disease severity was similar on leaves sprayed with sulfur, milk, lactoferrin or whey, viz. 1.0-4.2% of the leaf area affected.

The results showed that these products may protect vines from powdery mildew, in addition to the curative properties demonstrated in earlier research (Crisp et al. 2006 a, c).

**Table 18.** The percentage of conidia of *E. necator* that germinated on water agar and percentage of leaf area affected by powdery mildew on detached leaves of Viognier sprayed 6 hours before inoculation, assessed 24 hours and 7 days after inoculation, respectively. Results within columns with the same letter are not significantly different.

Material	% conidia germinated on agar	Percent leaf area infected
Milk (30 g/L)	3.3 a	1.0 a
Sulfur (3 g/L)	5.7 ab	1.1 a
Lactoferrin (20 mg/L)	1.5 a	1.3 a
Whey (30 g/L)	8.4 b	4.2 a
Ecocarb plus (4 g/L)		
Synertrol Horti-Oil (2 ml/L)	15 c	10 b
Milk (15 g/L)	7.9 b	11.8 b
Synertrol Horti-Oil (2 ml/L)	45.6 d	14.5 b
Water	89.4 e	85 c

### Effects on grape and wine quality

No significant differences in pH, TA and °Brix were detected in juice or wine from Verdelho or Shiraz grapes harvested from vines sprayed with alternative fungicides, sulfur or unsprayed controls. Duo-trio testing revealed no significant differences among Shiraz juices (2003/04) or wines (2004/05, 2005/06) nor Verdelho wines (2005/06) (Table 19). Significant differences in flavour were detected between reference juice and test juices of Verdelho. Both samples were then subjected to further evaluation by an expert tasting panel. The differences were considered to be due to a grape flavour character (whey 2003/2004) and pressing variation (2004/2005), rather than to the treatments applied in the field. The tasting panel also judged

that all juices they evaluated were suitable for wine making and the treatments would be unlikely to affect wine quality.

**Table 19.** Quality parameters and duo-trio results for grapes harvested by hand from Verdelho and Shiraz vines sprayed 5-7 times with test materials at Langhorne Creek vineyard. There was no significant difference in pH, titratable acidity or °Brix.

<b>2003/2004</b>				
<b>Verdelho</b>	<b>pH</b>	<b>Titratable acidity</b>	<b>°Brix</b>	<b>Duo/Trio Juice</b>
Sulfur	3.2	8.2	22.7	Reference
Milk	3.2	7.7	22.4	14/16
Whey	3.2	7.9	22.8	21/9 s <sup>1</sup>
Program 1	3.2	7.7	23.2	17/13
Ecocarb plus Synertrrol				
Horti-Oil	3.2	7.8	22.5	13/17
Untreated	N/A	N/A	N/A	N/A
<b>Shiraz</b>	<b>pH</b>	<b>Titratable acidity</b>	<b>°Brix</b>	<b>Duo/Trio Juice</b>
Sulfur	3.5	7.1	25.5	Reference
Milk	3.5	6.9	25.3	16/14
Whey	3.5	7.1	25.7	15/15
Program 1	3.5	6.7	25.8	N/A
Ecocarb plus Synertrrol				
Horti-Oil	3.4	6.8	25.6	18/12
Untreated	3.5	7.1	25.9	17/13
<b>2004/2005</b>				
<b>Verdelho</b>	<b>pH</b>	<b>Titratable acidity</b>	<b>°Brix</b>	<b>Duo/Trio Juice</b>
Sulfur	3.1	7.1	22.6	Reference
Milk	3.0	6.6	21.4	12/15
Whey	3.1	5.7	22.3	15/12
Program 1	3.0	7.7	22.3	13/14
Ecocarb plus Synertrrol				
Horti-Oil	3.0	7.5	21.7	18/9 s
Milk/B.s	3.1	6.0	21.8	18/9 s
Untreated	3.0	9.6	20.7	N/A
<b>Shiraz</b>	<b>pH</b>	<b>Titratable acidity</b>	<b>°Brix</b>	<b>Duo/Trio Wine</b>
Sulfur	3.5	5.0	27.5	Reference
Milk	3.6	4.9	26.6	18/12
Whey	3.6	5.1	27.1	17/13
Ecocarb plus Synertrrol				
Horti-Oil	3.6	4.9	27.0	11/19
<b>2005/2006</b>				
<b>Verdelho</b>	<b>pH</b>	<b>Titratable acidity</b>	<b>°Brix</b>	<b>Duo/Trio Wine</b>
Sulfur	3.2	8.2	22.7	Reference
Milk	3.2	7.7	22.4	17/13
Whey	3.2	7.9	22.8	18/12
Program 1	3.2	7.7	23.2	N/A
Ecocarb plus Synertrrol				
Horti-Oil	3.2	7.8	22.5	N/A
Untreated	N/A	N/A	N/A	N/A
<b>Shiraz</b>	<b>pH</b>	<b>Titratable acidity</b>	<b>°Brix</b>	<b>Duo/Trio Wine</b>
Sulfur	3.5	7.1	25.5	Reference
Milk	3.5	6.9	25.3	14/16
Whey	3.5	7.1	25.7	17/13
Program 1	3.5	6.7	25.8	N/A
Ecocarb plus Synertrrol				
Horti-Oil	3.4	6.8	25.6	N/A
Untreated	3.5	7.1	25.9	N/A

<sup>1</sup> s = significant difference detected between reference and test sample.

The similarity in yields between treatments applied to vines and the absence of differences in the quality parameters measured indicates that there would be no economic loss to producers

as a result of use of alternative fungicides. The wines produced using grapes from vines sprayed with alternative fungicides at the Langhorne Creek winery have shown no flaws that could be attributed to the treatments applied or the presence of powdery mildew.

### **Effects of novel powdery mildew control on grapevine microbiota**

The mean microbial populations obtained from leaves, berries and juice of Verdelho and Shiraz treated with milk, whey, sulfur or left untreated (controls) in seasons 2003/04, 2004/05 and 2005/06 are summarised below. The large variation among replicate blocks, a common feature of microbial populations, precluded comparison of these data using parametric statistics such as ANOVA. In consultation with BiometricsSA, Friedman's test (Dytham 2003), a non-parametric statistical test, was eventually identified as an appropriate means to analyse the highly variable data and was used to rank treatments according to microbial population.

#### **Microbial populations on leaves**

The colony forming units of bacteria, filamentous fungi and yeasts recovered from leaves in 2003/04 and 2004/05 generally increased with time, and populations were larger on vines treated with milk or whey than on those treated with sulfur or left untreated (see Appendix 5).

#### **Microbial populations on berries and in juice**

At veraison in 2003 to 2005, populations of bacteria, filamentous fungi and yeast were generally larger on berries collected from milk-or whey-treated plots than from sulfur-treated or untreated plots (data not shown). There were no obvious differences between cultivars.

At harvest in 2004, populations of bacteria and yeasts were generally larger on Verdelho grapes treated with milk or whey than sulfur, whereas those of filamentous fungi did not differ significantly among treatments (Tables 20 and 21). Trends for the corresponding juices were similar. For Shiraz, only bacterial populations in juice differed among treatments, in that populations were larger in blocks treated with milk or whey than with sulfur or untreated (Table 21).

At harvest in 2005, populations of yeasts and filamentous fungi were generally larger on Verdelho grapes treated with milk or whey than sulfur or untreated, whereas for juice samples, these rankings were observed for bacteria and filamentous fungi, and yeast populations were similar across the four treatments. For Shiraz, populations of bacteria and yeasts at harvest and in juice were larger for plots treated with milk or whey than sulfur or untreated and, generally, milk resulted in the largest populations.

At harvest in 2006, populations of yeasts and bacteria were larger on Verdelho grapes treated with milk or whey than sulfur or untreated, whereas for juice samples, similar rankings were observed for yeasts, bacteria and filamentous fungi (Table 21). For Shiraz, populations of bacteria and filamentous fungi were generally largest on berries treated with milk or whey, although one plot of sulfur-treated grapes yielded the second largest population of bacteria and another plot, the second largest population of filamentous fungi.

**Table 20.** Mean microbial populations on the surface of berries at harvest and in juice obtained from Verdelho and Shiraz vines treated with milk, whey, sulfur or left untreated (control). Data are colony forming units per g berries and per ml juice. There was no untreated control at vintage 2004, as vines had been sprayed with sulfur at veraison.

<b>2004</b>		<b>Yeast</b>	<b>Bacteria</b>	<b>Fungi</b>
<b>F Verdelho berries</b>	<b>milk</b>	385000±59909.1	327916.7±449014.1	8041.7±4342.7
	<b>sulfur</b>	21250±31389.3	55250±103167.3	3370.8±4447.4
	<b>whey</b>	158291.7±218928.8	54375±39154.1	3762.5±2856.7
<b>G Verdelho juice</b>	<b>milk</b>	18675±13159.9	12800±7842.2	2192.5±1252
	<b>sulfur</b>	920±930	1282.5±1166.1	1327.5±2515.1
	<b>whey</b>	13800±14214.5	11400±7025.2	1295±1485.2
<b>H Shiraz berries</b>	<b>milk</b>	75833.3±32044.5	61666.7±20321.5	5291.7±1342.8
	<b>control</b>	343375±637896.9	48000±68146.2	4125±1272
	<b>sulfur</b>	33291.7±42466	21166.7±7155.7	4875±2973.3
	<b>whey</b>	74166.7±27638.5	57500±17716.9	5250±1449.8
<b>I Shiraz juice</b>	<b>milk</b>	6525±2889.5	6550±3861.3	242.5±143.1
	<b>control</b>	2575±1017.8	2400±476.1	110±31.6
	<b>sulfur</b>	2100±81.6	2025±50	112.5±9.6
	<b>whey</b>	5125±2220.2	7700±4581.1	370±166.3

<b>2005</b>		<b>Yeast</b>	<b>Bacteria</b>	<b>Fungi</b>
<b>O Verdelho berries</b>	<b>milk</b>	102083.3±29670.3	78750±13220	1795.8±1776.9
	<b>control</b>	7708.3±3876.3	5216.7±3001	320.8±238.6
	<b>sulfur</b>	18916.7±16722.4	63116.7±113537.5	1000±1667.1
	<b>whey</b>	97083.3±52217.1	50250±18715	13841.7±18909.7
<b>P Verdelho juice</b>	<b>milk</b>	5700±2692	6475±3621.6	227.5±130
	<b>control</b>	532.5±158	420±173	27.5±15
	<b>sulfur</b>	2070±2151.6	885±654.8	40±40.8
	<b>whey</b>	3782.5±2554.7	2397.5±598.9	1105±1153.2
<b>Q Shiraz berries</b>	<b>milk</b>	66250±6718.5	77500±14688.1	116.7±88.2
	<b>control</b>	8625±1480.6	8666.7±304.3	33.3±13.6
	<b>sulfur</b>	15083.3±14467.4	13708.3±4957.9	91.7±117.5
	<b>whey</b>	72916.7±29576.5	62916.7±18726.8	108.3±142.4
<b>R Shiraz juice</b>	<b>milk</b>	3225±763.2	2007.3±887.1	72.5±101.8
	<b>control</b>	522.5±125.3	320±95.9	22.5±38.6
	<b>sulfur</b>	332.5±104.4	240±60.6	17.5±28.7
	<b>whey</b>	1670±756.5	1277.5±194.8	30±29.4

**Table 20** (continued)

<b>2006</b>		<b>Yeast</b>	<b>Bacteria</b>	<b>Fungi</b>
<b>T Verdelho berries</b>	<b>milk</b>	24541.7±19383.7	21708.3±6743.1	2204.2±2192.6
	<b>control</b>	7416.7±1641.5	5679.2±2666.4	304.2±258
	<b>sulfur</b>	6833.3±2211.1	4100±2809.7	404.2±387.1
	<b>whey</b>	60416.7±51521.6	36333.3±18014.4	3895.8±4362.2
<b>U Verdelho juice</b>	<b>milk</b>	18027.5±28065.8	4270±5395.7	867.5±339.8
	<b>control</b>	1950±3100	620±541.7	327.5±423.3
	<b>sulfur</b>	1465±2108	457.5±480.8	142.5±199.1
	<b>whey</b>	14675±18302.9	3752.5±3467.9	1222.5±412.1
<b>V Shiraz berries</b>	<b>milk</b>	59291.7±30998.3	64833.3±21630.7	4195.8±4612.7
	<b>control</b>	11375±3192.5	8250±4074.5	287.5±275
	<b>sulfur</b>	13125±3811.2	21750±7555.4	454.2±147.4
	<b>whey</b>	79625±77275.6	72208.3±38737.8	2187.5±3548.7
<b>W Shiraz juice</b>	<b>milk</b>	56125±37776.5	361.9±281.4	4085±3197
	<b>control</b>	5075±1493	36.4±37.9	267.5±163.6
	<b>sulfur</b>	5100±18927.5	220±177.7	2712.5±2750.7
	<b>whey</b>	60250±18927.5	220±177.7	2712.5±2750

**Table 21.** Ranking tables for the microbial populations on Verdelho and Shiraz grapes at harvest or in juice sampled from four replicate plots that received milk, whey or sulfur or were left untreated (control). Friedman's statistic was used to rank the treatments on a scale of 1 to 4, where 1 = smallest microbial population and 4 = largest microbial population. Where a cultivar, treatment or microbial group (bacteria, yeasts or filamentous fungi) is not shown, the corresponding population did not differ significantly among treatments. (For all tables, 5% point = 7.80, 1% point = 9.60).

#### 2004

<b>Verdelho Berries F</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Sulfur 1	Sulfur 1	Whey 1
Intermediate population	Whey 2	Whey 2	Whey 2	Sulfur 2
Largest population	Milk 3	Milk 3	Milk 3	Milk 3

Friedman's statistic = 6.50; P-value using chi-square approximation (2 d.f.) = 0.039.

Control omitted, as sprayed with sulfur at veraison.

<b>Verdelho Juice G</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Yeast</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Sulfur 1	Sulfur 1	Whey 1
Intermediate population	Whey 2	Whey 2.5	Whey 2	Sulfur 2
Largest population	Milk 3	Milk 2.5	Milk 3	Milk 3

Friedman's statistic = 6.12, P-value using chi-square approximation (2 d.f.) = 0.038

Control omitted, as sprayed with sulfur at veraison.

<b>Shiraz Juice I</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Sulfur 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Control 2	Control 2	Whey 2
Intermediate population	Whey 3	Milk 3	Milk 3	Control 3
Largest population	Milk 4	Whey 4	Whey 4	Milk 4

Friedman's statistic = 9.30, P-value using chi-square approximation (3 d.f.) = 0.026

#### 2005

<b>Verdelho Berries O</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Yeast</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Control 1	Control 1
Intermediate population	Control 2	Sulfur 2	Sulfur 2	Sulfur 2
Intermediate population	Whey 3	Whey 3	Milk 3	Whey 3
Largest population	Milk 4	Milk 4	Whey 4	Milk 4

Friedman's statistic = 10.20, P-value using chi-square approximation (3 d.f.) = 0.017

<b>Verdelho Berries O</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Filamentous fungi</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Control 1	Control 1
Intermediate population	Control 2	Milk 2	Sulfur 2	Milk 2
Intermediate population	Milk 3	Control 3	Whey 3	Sulfur 3
Largest population	Whey 4	Whey 4	Milk 4	Whey 4

Friedman's statistic = 8.10, P-value using chi-square approximation (3 d.f.) = 0.044

**Table 21** (continued)**2005 (continued)**

<b>Verdelho Juice P</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Control 1	Control 1	Control 1	Control 1
Intermediate population	Sulfur 2	Sulfur 2	Sulfur 2	Sulfur 2
Intermediate population	Whey 3	Whey 3	Whey 3	Whey 3
Largest population	Milk 4	Milk 4	Milk 4	Milk 4

Friedman's statistic = 12.00, P-value using chi-square approximation (3 d.f.) = 0.007

<b>Verdelho Juice P</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Filamentous fungi</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Control 1	Control 1
Intermediate population	Control 2	Sulfur 2	Sulfur 2	Sulfur 2
Intermediate population	Whey 3	Milk 3	Milk 3	Whey 3
Largest population	Milk 4	Whey 4	Whey 4	Milk 4

Friedman's statistic = 9.67, Adjusted for ties = 9.92, P-value using chi-square approximation (3 d.f.) = 0.019

<b>Shiraz berries Q</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Control 1	Control 1	Control 1	Sulfur 1
Intermediate population	Sulfur 2	Sulfur 2	Sulfur 2	Control 2
Intermediate population	Whey 3	Whey 3	Whey 3	Whey 3
Largest population	Milk 4	Milk 4	Milk 4	Milk 4

Friedman's statistic = 11.10, P-value using chi-square approximation (3 d.f.) = 0.011

<b>Shiraz berries Q</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Yeast</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Control 1	Sulfur 1
Intermediate population	Control 2	Sulfur 2	Sulfur 2	Control 2
Intermediate population	Whey 3	Milk 3	Milk 3	Whey 3
Largest population	Milk 4	Whey 4	Whey 4	Milk 4

Friedman's statistic = 9.60, P-value using chi-square approximation (3 d.f.) = 0.022

<b>Shiraz Juice R</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Sulfur 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Control 2	Control 2	Control 2
Intermediate population	Whey 3	Whey 3	Whey 3	Milk 3
Largest population	Milk 4	Milk 4	Milk 4	Whey 4

Friedman's statistic = 11.10, P-value using chi-square approximation (3 d.f.) = 0.011

<b>Shiraz Juice R</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Yeast</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Sulfur 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Control 2	Control 2	Control 2
Intermediate population	Whey 3	Whey 3	Milk 3	Whey 3
Largest population	Milk 4	Milk 4	Whey 4	Milk 4

Friedman's statistic = 11.10, P-value using chi-square approximation (3 d.f.) = 0.11

**Table 21** (continued)**2006**

<b>Verdelho Berries T</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Yeast</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Sulfur 1	Sulfur 1	Control 1
Intermediate population	Control 2	Control 2	Control 2	Sulfur 2
Intermediate population	Milk 3	Milk 3	Milk 3	Whey 3
Largest population	Whey 4	Whey 4	Whey 4	Milk 4

Friedman's statistic = 10.20, P-value using chi-square approximation (3 d.f.) = 0.017

<b>Verdelho Berries, T</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Sulfur 2	Control 2	Control 2
Intermediate population	Milk 3	Milk 3	Whey 3	Milk 3
Largest population	Whey 4	Whey 4	Milk 4	Whey 4

Friedman's statistic = 10.20, P-value using chi-square approximation (3 d.f.) = 0.017

<b>Verdelho Juice U</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Yeast</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Sulfur 2	Control 2	Control 2
Intermediate population	Milk 3	Whey 3	Milk 3	Whey 3
Largest population	Whey 4	Milk 4	Whey 4	Milk 4

Friedman's statistic = 9.90, P-value using chi-square approximation (3 d.f.) = 0.019

<b>Verdelho Juice U</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Sulfur 2	Control 2	Control 2
Intermediate population	Whey 3	Whey 3	Milk 3	Whey 3
Largest population	Milk 4	Milk 4	Whey 4	Milk 4

Friedman's statistic = 10.20, P-value using chi-square approximation (3 d.f.) = 0.017

<b>Verdelho Juice U</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Filamentous fungi</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Sulfur 1	Control 1	Sulfur 1	Sulfur 1
Intermediate population	Control 2	Sulfur 2	Control 2	Control 2
Intermediate population	Milk 3	Whey 3	Milk 3	Milk 3
Largest population	Whey 4	Milk 4	Whey 4	Whey 4

Friedman's statistic = 10.20, P-value using chi-square approximation (3 d.f.) = 0.017

<b>Shiraz berries V</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>	<b>Plot</b>
<b>Bacteria</b>	<b>I</b>	<b>III</b>	<b>IV</b>	<b>VI</b>
Smallest population	Control 1	Control 1	Control 1	Control 1
Intermediate population	Whey 2	Sulfur 2	Sulfur 2	Sulfur 2
Intermediate population	Sulfur 3	Whey 3	Milk 3	Milk 3
Largest population	Milk 4	Milk 4	Whey 4	Whey 4

Friedman's statistic = 9.30, P-value using chi-square approximation (3 d.f.) = 0.026



**Table 21** (continued)

Shiraz berries V	Plot	Plot	Plot	Plot
Filamentous fungi	I	III	IV	VI
Smallest population	Control 1	Control 1	Control 1	Sulfur 1
Intermediate population	Sulfur 2	Whey 2	Sulfur 2	Control 2
Intermediate population	Whey 3	Sulfur 3	Milk 3	Whey 3
Largest population	Milk 4	Milk 4	Whey 4	Milk 4

Friedman's statistic = 8.70, P-value using chi-square approximation (3 d.f.) = 0.034

The predominant trend observed over the three growing seasons was that milk and whey-treated vines supported larger microbial populations than sulfur-treated and untreated vines. This was more marked in 2004/05 and 2005/06 than in 2003/04, and bacterial populations tended to respond more consistently than yeasts or filamentous fungi. It is possible that milk and whey provide a nutrient source for the indigenous, epiphytic/saprophytic microbial populations on the leaves and berries. Many of the filamentous fungi are likely to be present as dormant spores, which may or may not germinate and grow on the plant surface to produce more spores, whereas the epiphytic bacteria and yeasts may be able to respond more quickly to the nutrients in milk or whey. *E. necator*, in contrast, obtains nutrients only from the living epidermal cells of the leaf or berry. Although milk and whey caused collapse of hyphae and rupture of conidia of *E. necator* when applied to colonized leaves exposed to light (Crisp et al. 2006c), if they had a similar effect on these indigenous organisms, it may have been temporary such that populations were able to re-establish in the 2 weeks between application and sampling. Epiphytic or saprophytic microorganisms may increase the competition for space on the leaf and berry surfaces, which could restrict the ability of *E. necator* to colonise these surfaces. This may contribute to the reduction in the severity of powdery mildew following treatment of vines with milk or whey.

#### Identification of representative microorganisms

The rapid DNA extraction protocol proved effective for bacteria, yeasts and filamentous fungi. Sequencing protocols for the rapid identification of large numbers of microorganisms were refined and successfully applied to the vine surface microbiota. In total, 927 microorganisms isolated from Verdelho and Shiraz vines treated with milk, whey and sulfur, and untreated control vines were selected for molecular identification. Of these, 438 isolates were filamentous fungi, 249 were bacteria and 240 were yeasts (Table 22).

Common filamentous fungi included *Alternaria*, *Aspergillus*, *Cladosporium* and *Penicillium* species. The distribution of these organisms varied considerably among replicate plots within cultivars and treatments (Tables 23 and 24), and the sampling strategy was such that data were not amenable to statistical analysis. However, some general trends were evident. A greater variety of genera and species were detected at veraison than at harvest, however, *Cladosporium cladosporoides* appeared most frequently, irrespective of sampling time and cultivar. *Aspergillus niger* was detected on Shiraz berries and in juice, although there was no obvious association between treatment and distribution, but was not found on Verdelho berries nor in juice. These organisms are common members of the phylloplane microbial community and are considered unlikely to persist during vinification.



**Table 24.** Distribution of filamentous fungi on Verdelho and Shiraz grapes at harvest or in juice sampled in 2004 from four replicate plots that received milk, whey or sulfur or were left untreated (control). Data represent number of plots, of 4, in which the fungus was detected.

**Verdelho berry skins at harvest**

Organism	Milk	Whey	Sulfur	Untreated
<i>Cladosporium cladosporoides</i>	4	4	4	n.a.
<i>Cladosporium tenuissimum</i>	4			n.a.
<i>Paraconiothyrium estuarinum</i>	1			n.a.
<i>Paraconiothyrium brasiliense</i>		4		n.a.
<i>Penicillium glabrum</i>		1		n.a.
<i>Alternaria alternata</i>		4		n.a.

**Verdelho juice**

Organism	Milk	Whey	Sulfur	Untreated
<i>Cladosporium cladosporoides</i>	4	4	4	n.a.
<i>Paraconiothyrium brasiliense</i>	3		1	n.a.
<i>Paraconiothyrium estuarinum</i>	2			n.a.
<i>Cladosporium tenuissimum</i>			4	n.a.
<i>Aspergillus foetidus</i> var. <i>acidus</i>			1	n.a.

**Shiraz berry skins at harvest**

Organism	Milk	Whey	Sulfur	Untreated
<i>Cladosporium cladosporoides</i>	4	4	4	
<i>Lewia infectoria</i>	3			
<i>Aspergillus niger</i>	4	4	4	4
<i>Alternaria alternata</i>			3	
<i>Penicillium glabrum</i>			4	3
<i>Pleospora herbarum</i>				*

**Shiraz juice**

Organism	Milk	Whey	Sulfur	Untreated
<i>Cladosporium cladosporoides</i>	4	4	4	4
<i>Aspergillus niger</i>	4		3	
<i>Aspergillus sydowii</i>			1	
<i>Alternaria triticina</i>			1	
<i>Penicillium glabrum</i>			1	
<i>Pleospora herbarum</i>			1	
<i>Eutypa lata</i>			4	
<i>Cryptovalsa ampelina</i>				1
<i>Paraconiothyrium brasiliense</i>		1		1

The most common yeasts identified were *Sporobolomyces roseus*, *Aureobasidium pullulans* and *Cryptococcus carnescens*. *Sporobolomyces*, a pink yeast, is one of the most commonly found on the leaf surface of a wide range of plants. *A. pullulans* was commonly isolated from leaves and berries at each sampling period, and from juice. At harvest, it was the predominant yeast isolated from berry surfaces. Likewise, Beh et al. (2004) reported populations of  $10^2$ - $10^5$  colony forming units per gram of berries, whether healthy or damaged, throughout the season. This growth pattern contrasts with findings in the Northern hemisphere, where populations of *A. pullulans* decreased in number from veraison, when it was superseded by fermentative yeasts, and by harvest the organism was undetectable (Renouf et al. 2005). Beh et al. (2004) reported that approximately 5% of the 195 strains of *A. pullulans* isolated in their study in

New South Wales were antagonistic to numerous vine-associated microorganisms, including *Saccharomyces cerevisiae*, and to various wine spoilage yeasts (Beh et al. 2004). Over half of the strains were antagonistic to the insect biocontrol agent, *Bacillus thuringiensis*. Although antagonism to *Erysiphe necator* was not studied in the present project, there was no obvious relationship between the population of *A. pullulans* present on the tissue and severity of powdery mildew in 2003/04, when disease was severe on the untreated control vines in the Langhorne Creek vineyard. Likewise, there was no obvious effect of *A. pullulans* on juice or wine quality.

Although leaves were collected for examination of arthropods, it was observed that opulations of arthropods with limited movement within the canopy were too small to yield meaningful results for the effects of the test materials. The majority of arthropods within the canopy were highly mobile and not suitable indicators of treatment effects in small-scale plots. Soil arthropod populations were highly variable and there was no significant difference in populations in soil under vines (data not shown).

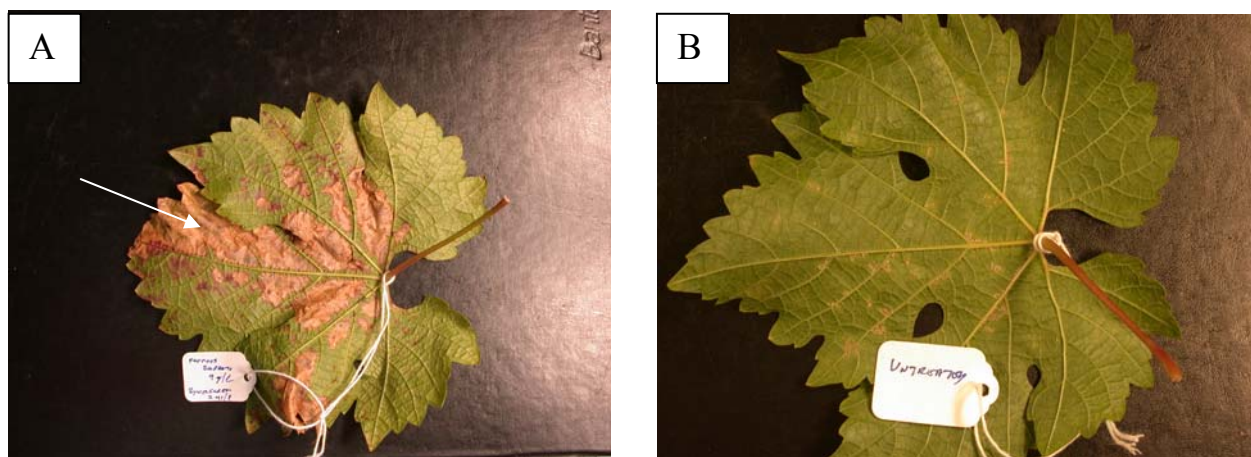
## Downy mildew

### Greenhouse trials

In the preliminary greenhouse experiment, all treatments except Vermiboost significantly reduced downy mildew on inoculated vines compared with vines sprayed with water (Table 25). Leaves on vines sprayed with ferrous sulfate displayed phytotoxic effects of the spray application (Figure 16) and, in some cases, leaves dropped from the vines before assessment. While the ferrous sulfate treatment limited the development of downy mildew as effectively as copper, it caused severe phytotoxicity, which would be expected to limit photosynthesis and, therefore, reduce yield and quality of grapes. The phytotoxicity may have artificially reduced the severity of disease on leaves treated with ferrous sulfate by greatly reducing the leaf area; however, the remaining green areas were largely free of downy mildew. Phytotoxicity was far less severe, but still evident, on leaves that were not kept in plastic bags overnight.

**Table 25.** Severity of downy mildew and phytotoxicity on Cabernet Sauvignon sprayed with test materials and inoculated with *Plasmopara viticola* in the greenhouse. Disease is expressed as percentage of leaf area affected. Results within columns with the same letter are not significantly different.

Treatment	Disease severity (%)	Phytotoxicity (%)
Ferrous sulfate 9 g/L		
Plus Synerscreen	0 a	70 d
Copper	1 a	3 a
Ferrous sulfate 6 g/L		
+ Synerscreen	1 a	59 c
Ferrous sulfate 9 g/L	2 a	19 b
Ferrous sulfate 3 g/L		
+ Synerscreen	2 a	23 b
Synerscreen	7 ab	0 a
Ecocarb plus Synertröl Horti-Oil	11 b	2 a
Chitosan	16 b	0 a
Untreated	40 c	0 a
Vermiboost	46 c	4 a



**Figure 16.** Leaves of Cabernet Sauvignon sprayed with A) 9 g/L ferrous sulfate or B) reverse osmosis water and inoculated with *Plasmopara viticola* 24 hours after application of test materials. Symptoms of phytotoxicity are arrowed.

### Field trials: Lenswood

On the first occasion in the 2004/2005 trials, inoculation was not successful, however, disease developed on inoculated shoots after the second inoculation. Despite weather conditions suitable for both primary and secondary downy mildew, natural infection was not detected.

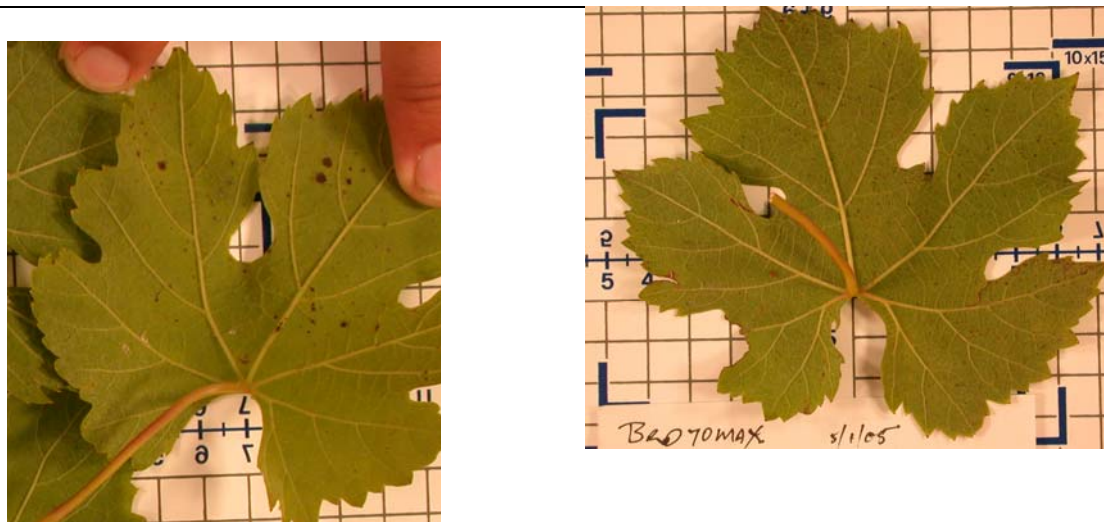
The severity of the downy mildew on leaves of Nebbiolo was reduced significantly ( $p < 0.001$ ) on vines treated with Timor<sup>®</sup>, Timorex<sup>®</sup>, Brotomax, ferrous sulfate plus SynerScreen and ferrous sulfate when compared with the untreated control plants and not significantly different from leaves sprayed with copper (Table 26). The materials also reduced the incidence of downy mildew compared with the untreated control and to a degree not significantly different from vines treated with copper at 3 g/L.

Ferrous sulfate, Timor, Timorex, Ecocarb plus SynerScreen Horti-Oil and Brotomax appeared to be suitable alternatives to copper for control of downy mildew. However, ferrous sulfate-treated leaves kept moist overnight after inoculation showed phytotoxicity similar to that seen in the greenhouse (Figure 17). Leaves on vines sprayed with Brotomax had many black spots.

Due to delays in the arrival of some of the materials and the loss of the first trial in 2005/06, downy mildew had developed on leaves prior to establishment of the second trial. Conditions suitable for downy mildew persisted throughout the season, which confounded results in that new growth developed disease symptoms regardless of treatment applied. While data collected were not useful for assessing the protective capacity of the products, none of the test materials had obvious curative effects. However, leaves on vines sprayed with copper and Timor (1%) remained on the vines longer than did leaves on vines that received lower rates of Timor.

**Table 26.** Severity (% of leaf surface with sporulating colonies) and incidence (of 5 leaves) of downy mildew on inoculated leaves of Nebbiolo at Lenswood Research Centre. Shoots were inoculated with *Plasmopara viticola* 24 hours after treatment. Results within columns with the same letter are not significantly different.

Treatment	Severity	Incidence
Untreated	28 d	4 a
Acadian	25d	4 a
Synerscreen	19 c	4 a
Acadian x2	17 bc	4 a
Ecocarb plus		
Synertril Horti-Oil	10 ab	3 a
Timorex	10 ab	3 a
Ferrous sulfate plus		
Synerscreen	9 a	3 a
Brotomax	8 a	3 a
Timor	8 a	3 a
Copper	7 a	3 a
Ferrous sulfate	6 a	3 a



**Figure 17.** Leaf of Nebbiolo 7 days after being sprayed with ferrous sulfate (left) or (b) Brotomax (right) showing brown spots indicative of phytotoxicity.

## 6.2 Research in Tasmania

### Results

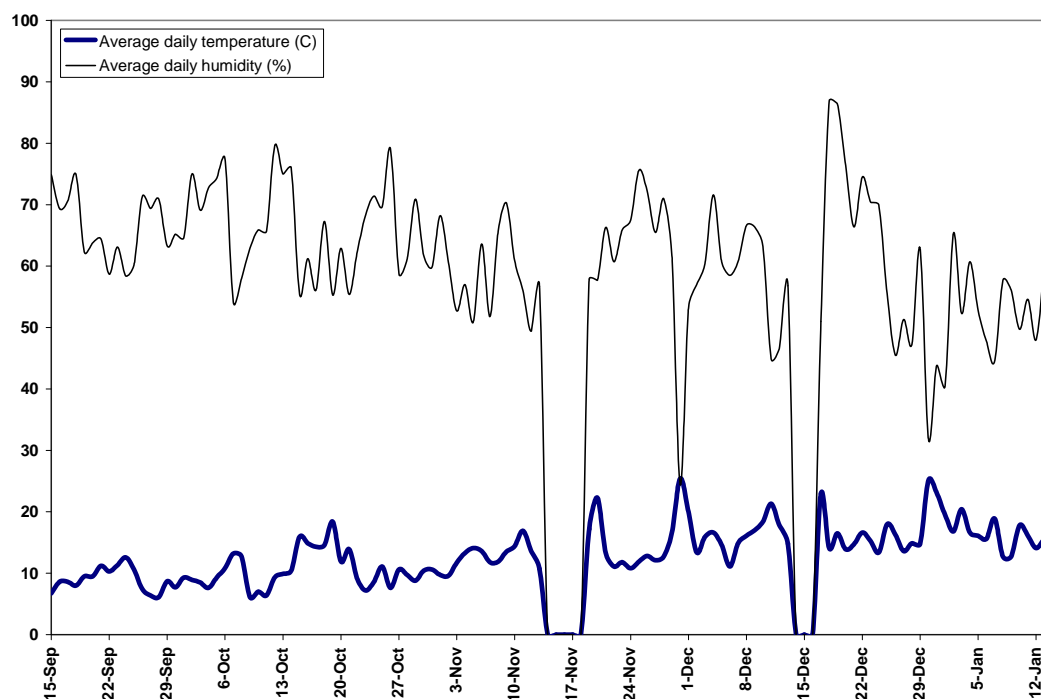
In all tables, means with the same letter are not significantly different at  $P = 0.05$ , lsd is the least significant difference and residual df is the residual degrees of freedom. The number in parenthesis for residual df is the number of missing values in cases where outliers were removed from the analysis. A dash next to the mean for the untreated control indicates that it was excluded from the analysis of variance to reduce heterogeneity in variance among the means.

#### Frogmore Creek Chardonnay, 2003/04

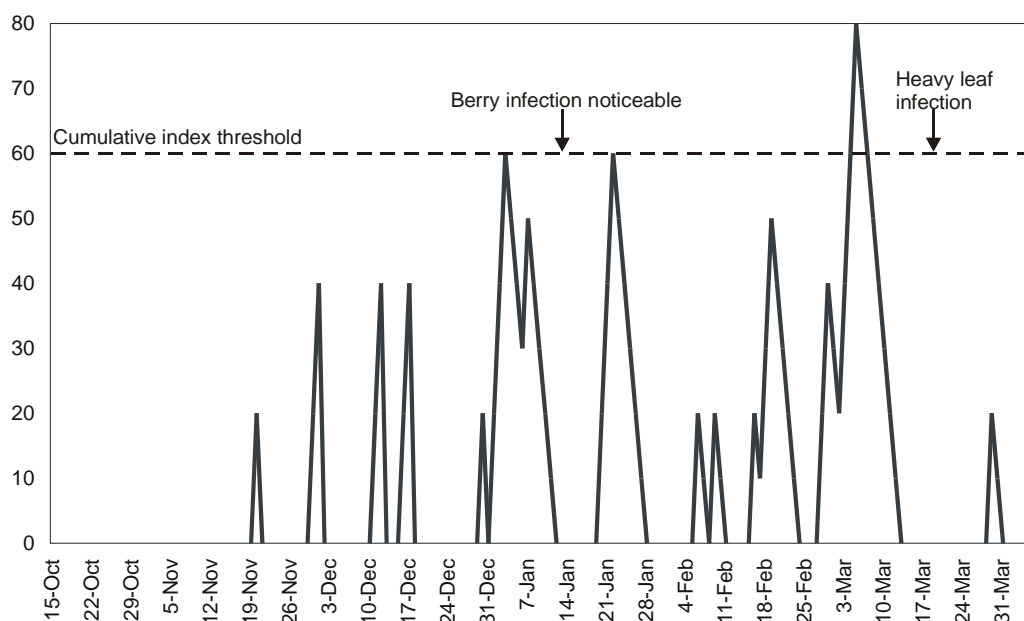
Weather conditions in the early stages of the powdery mildew epidemic are summarised in Figure 18. The first flag shoot was found in Block A (Pinot noir) on 4 November 2003 and more flag shoots were detected in this block on 7 Nov. 2003. One flag shoot was found in the trial site on 17 Nov. in an untreated control plot (replicate 3) and it was removed. On 17

Dec., two leaves sampled from separate untreated plots were found to have a low level of sporulation, evident on the underside of the leaves and associated with browning on the veins and faint chlorotic spots. On 30 Dec. leaves in all untreated plots had developed signs of powdery mildew on the abaxial surface. Berry infection was first recorded on 14 January 2004 (berries 2-8 mm). At this time berry infection was found only in the untreated control plots with the exception of one cluster in the Ecocarb®-only treatment. Disease incidence increased rapidly on bunches in January and mildew was noticeable on the upper surface of leaves in early February.

The threshold of 60 for the Gubler-Thomas risk index for powdery mildew was recorded on 3 January 2004, Figure 19), just after powdery mildew was detected on the abaxial surface of leaves on 30 Dec. 2003.



**Figure 18.** Average daily temperature (°C) and humidity (%) to 12 January at Frogmore Creek vineyard, 2003/04. The troughs around 17 Nov. and 15 Dec. represent missing data.



**Figure 19.** Gubler-Thomas risk index for powdery mildew at Frogmore Creek vineyard to 31 March 2004. Arrows refer to observations for the untreated control in Chardonnay. Powdery mildew on bunches was first recorded on 14 January, when the berries were 2-8 mm in diameter.

All treatments suppressed powdery mildew significantly when compared with the untreated control (Tables 27-30). The mean severity of scarring on berries was not significantly different among treatments in early April, more than one month before harvest (Table 31). Overall, sulfur applied at 6 g/L was the most effective treatment and provided commercially acceptable control of powdery mildew on bunches and leaves. The severity of powdery mildew on bunches following treatment with milk or whey was statistically equivalent to that observed for sulfur (Tables 29 and 30). Milk or whey were the only treatments that resulted in a mean severity of powdery mildew on bunches of less than 1% close to veraison (Table 29). By early April, prior to harvest, powdery mildew was not detected on bunches treated with sulfur, whereas the milk or whey treatments had a mean incidence of 20 and 32%, respectively.

Program 1 and the Ecocarb<sup>®</sup> treatment were not separated statistically for all measures of powdery mildew, except that the mean severity of leaf infection was higher for the Ecocarb<sup>®</sup> treatment on April 6 (Table 28). Prior to veraison, bunches treated with Ecocarb SR (3 g/L) plus the adjuvant Protector had a significantly higher severity of powdery mildew when compared with Program 1 or the Ecocarb<sup>®</sup> (4 g/L) treatment (Table 29). Unlike Ecocarb<sup>®</sup>, applications of Ecocarb SR plus Protector continued beyond veraison and the difference in severity between the two treatments was not evident at the later assessment (Table 30). The final application of all treatments, except Ecocarb SR plus Protector, was close to veraison on 11 February. Some 53 days later, sulfur that had been applied throughout the season and sulfur applied to the untreated control on 11 February, resulted in a mean severity of powdery mildew on leaves that was less than 6% (Table 28). All other treatments, except Ecocarb SR plus Protector, resulted in significantly higher mean severity and incidence of leaf infection.



**Table 27.** Mean incidence and severity of powdery mildew on the adaxial surface of Chardonnay leaves (n = 20 per plot), 10 February 2004, E-L 32-33 (bunch closure), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	52	c	17.5	-
sulfur 6 g/L	0	a	0	a
milk 1:10	19	ab	0.44	a
whey 25 g/L	21	b	0.9	ab
Ecocarb® 4 g/L	30	b	1.32	ab
Ecocarb SR 3 g/L ± Protector	25	b	0.96	ab
Program 1	31	b	1.47	b
lsd	19.5		8.1	
residual df	30		30	
P	<0.001		0.001	

**Table 28.** Mean incidence and severity of powdery mildew on the adaxial surface of Chardonnay leaves (n = 40 per plot), 6 April 2004, E-L 36 (intermediate Brix values), Frogmore Creek vineyard. Note that the untreated control was sprayed with sulfur on 11 February 2004.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	22	a	5.2	a
sulfur 6 g/L	5.4	a	2.0	a
milk 1:10	76	bc	37	b
whey 25 g/L	68	b	38	b
Ecocarb® 4 g/L	96	d	66	d
Ecocarb SR 3 g/L ± Protector	64	b	11	a
Program 1	90	cd	52	c
lsd	17.4		13	
residual df	30		30	
<b>P</b>	<0.001		<0.001	

**Table 29.** Mean incidence and severity of powdery mildew on Chardonnay bunches (n = 30 per plot), 10 February (untreated control) or 24 February (all other treatments), 2004, E-L 33 (34) (pre-veraison), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	59	d	14	-
sulfur 6 g/L	2.2	a	0.12	a
milk 1:10	9.4	ab	0.19	a
whey 25 g/L	9.4	ab	0.11	a
Ecocarb® 4 g/L	29	bc	1.3	b
Ecocarb SR 3 g/L ± Protector	46	cd	2.6	c
Program 1	34	c	0.93	ab
lsd	21.1		1.15	
residual df	29 (1)		24 (1)	
<b>P</b>	<0.001		<0.001	

**Table 30.** Mean incidence and severity of powdery mildew on Chardonnay bunches (n = 20 per plot), 5 April 2004, E-L 36 (intermediate Brix values), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	69	d	14.9	-
sulfur 6 g/L	0	a	0	a
milk 1:10	20	ab	0.4	ab
whey 25 g/L	32	bc	0.5	ab
Ecocarb® 4 g/L	48	cd	2.5	dc
Ecocarb SR 3 g/L ± Protector	55	cd	2.1	bc
Program 1	60	d	2.9	d
lsd	24.1		1.33	
residual df	29 (1)		24 (1)	
<b>P</b>	<0.001		<0.001	

**Table 31.** Mean severity of scarring on Chardonnay bunches (n = 20 per plot), 5 April 2004, E-L 36 (intermediate Brix values), Frogmore Creek vineyard. The mean incidence of scarring in each plot was 100%.

Treatment	Mean severity (%)
untreated control	4.6
sulfur 6 g/L	2.5
milk 1:10	1.2
whey 25 g/L	2.4
Ecocarb <sup>®</sup> 4 g/L	2.7
Ecocarb SR 3 g/L ± Protector	2.7
Program 1	3.2
lsd	2.9
residual df	29 (1)
<b>P</b>	0.405

Bunch rot was defined broadly as necrotic, split or shrivelled berries, with or without fungal sporulation. All treatments reduced the amount of bunch rot pre-harvest when compared with the untreated control (Table 32). Applications of sulfur, milk or whey resulted in equivalent incidence of severity of bunch rot before harvest. The Ecocarb<sup>®</sup> (4 g/L) treatment resulted in significantly more bunch rot than all other treatments except Program 1 (Table 32). When bunches were sampled and moist incubated on the laboratory bench, all treatments had significantly less bunch rot than the untreated control (Table 33). Fungi that sporulated after moist incubation included *Botrytis cinerea*, *Alternaria* and *Penicillium* spp. On 17 May, one day before harvest, there was no sign of *Botrytis cinerea* on bunches. A *Cladosporium* species was observed to have colonised damaged berries and old mildew colonies.

Treatments were not separated statistically in terms of the mean weight of 20 bunches per plot, measured on 29 April (data not shown). On 29 April 2004 the mean of °Brix of berries in plots treated with sulfur was 20.8.

**Table 32.** Mean incidence and severity of rot on Chardonnay bunches (n = 20 per plot), 5 April 2004, E-L 36 (intermediate Brix values), Frogmore Creek vineyard. Rot was defined as necrotic, split or shrivelled berries, with or without fungal sporulation.

Treatment	Mean incidence (%)		Mean severity (%)	
untreated control	32	-	2.1	-
sulfur 6 g/L	2.5	a	0.021	ab
milk 1:10	1.7	a	0.025	ab
whey 25 g/L	1.7	a	0.013	a
Ecocarb <sup>®</sup> 4 g/L	17	b	0.25	c
Ecocarb SR 3 g/L ± Protector	5	a	0.058	ab
Program 1	9.2	ab	0.18	cb
lsd	9.8		0.169	
residual df	24 (1)		24 (1)	
<b>P</b>	0.015		0.037	

**Table 33.** Mean severity of rot on Chardonnay bunches (n = 20 per plot) sampled from Frogmore Creek Vineyard on 29 April 2004, and moist incubated in plastic bags at room temperature for 8 days. Rot was defined as necrotic, split or shrivelled berries, with or without grey or white/grey sporulation. Scarring or blackening of the berry skin was not included in the assessment. The mean incidence of rot on these bunches was > 95% in all plots.

Treatment	Mean severity (%)	
untreated control	25	b
sulfur 6 g/L	5.9	a
milk 1:10	6.3	a
whey 25 g/L	9.6	a
Ecocarb <sup>®</sup> 4 g/L	7.2	a
Ecocarb SR 3 g/L ± Protector	11	a
Program 1	11	a
lsd	8.5	
residual df	30	
<b>P</b>	0.002	

#### **Cooinda Vale Cabernet Sauvignon, 2003/04**

All experimental treatments suppressed powdery mildew significantly when compared with the untreated control (Tables 34-36). At veraison, the mean severity of powdery mildew on bunches in all treatments was less than 3%.

Powdery mildew increased rapidly on bunches in early January. Disease increase on the upper surface of leaves was most noticeable around early February. All vines were tip pruned on February 10, 2004. The mean severity and incidence of powdery mildew on leaves at veraison was statistically equivalent for all treatments, except Program 1 (Table 34). Program 1 was also applied to 'buffer' rows at both ends of the trial area. Control of powdery in one of the buffer rows was commercially unacceptable, especially where vines were shaded by a large tree.

Commercially acceptable control of powdery mildew was achieved by application of sulfur at rates of 6 g/L (label rate) or 12 g/L (Tables 34-36). Although the difference in mean incidence and severity of powdery mildew between the two rates of sulfur was not statistically significant for most assessments, the 12 g/L rate appeared to provide marginally better control. At veraison, the mean severity of powdery mildew on bunches was significantly less in the sulfur treatments than the other treatments, although the milk and whey treatments resulted in a mean severity below 2% (Table 34).

Overall, treatments of Ecocarb<sup>®</sup>, milk or whey were not separated statistically for the mean incidence or severity of powdery mildew on bunches. Although whey, milk or Ecocarb<sup>®</sup> appeared to reduce the severity of powdery mildew to a level that approached the standard sulfur treatment, there was a high incidence of powdery mildew in these treatments. On 22 April 2004, there was visible infection on rachis of bunches in four out of six plots of the milk treatment, whereas rachis infection was not obvious in other treated plots, except block 2 of the Program 1 treatment. At veraison, all treatments except sulfur resulted in a mean incidence of powdery mildew on bunches that was greater than 50%. Restricted mildew colonies formed in plots treated with milk, whey or Program 1 and these colonies appeared to be maintained in a restricted state by these treatments.

All treatments reduced the severity of bunch rot and colonisation by bunch rot fungi, when compared with the untreated control (Tables 37 and 38). Sporulating colonies of *Botrytis cinerea* were present in all plots to some degree before harvest. The milk and whey treatments were equivalent statistically to the two sulfur treatments for the mean severity of colonisation by bunch rot fungi, whereas the mean severities for the Ecocarb<sup>®</sup> or Program 1 treatments were significantly higher (Table 38).

There were significant differences in the mean total soluble solids among treatments close to harvest (Table 39). The untreated control had a significantly higher mean °Brix than all treatments and the higher rate of sulfur, 12 g/L, resulted in a significantly lower mean °Brix than all treatments except the milk and Program 1 treatments.

**Table 34.** Mean incidence and severity of powdery mildew on the adaxial surface of Cabernet Sauvignon leaves (n = 40 per plot), 16 February 2004, E-L 34 (very early veraison), Cooinda Vale vineyard.

Treatment	Mean incidence (%)		Mean severity (%)	
untreated control	40	-	11.5	-
sulfur 12 g/L	0	a	0	a
sulfur 6 g/L	0	a	0	a
milk 1:10 or 1:5	2.1	a	0.08	a
whey 25 g/L	3.3	a	0.08	a
Ecocarb <sup>®</sup> 4 g/L	0.4	a	0.008	a
Program 1	19	b	1.3	b
	lsd	6.83	0.6012	
	df	24 (1)	24 (1)	
	<b>P</b>	<0.001	<0.001	

**Table 35.** Mean incidence and severity of powdery mildew on bunches of Cabernet Sauvignon (n = 20 per plot), 18 February 2004, E-L 34 (very early veraison), Cooinda Vale vineyard.

Treatment	Mean incidence (%)		Mean severity <sup>1</sup> (%)	
untreated control	95	c	1.071 (14)	d
sulfur 12 g/L	1	a	-5.09 (0.03)	a
sulfur 6 g/L	13	a	-1.081 (0.12)	b
milk 1:10 or 1:5	58	b	0.074 (1.8)	c
whey 25 g/L	52	b	-0.141 (0.84)	c
Ecocarb <sup>®</sup> 4 g/L	60	b	0.163 (2.5)	c
Program 1	59	b	0.140 (1.6)	c
	lsd	20	0.3863	
	df	28 (2)	27 (3)	
	<b>P</b>	<0.001	<0.001	

<sup>1</sup>Means transformed according to  $\text{LOG}_{10}(x+0.00001)$ . Numbers in parentheses are the arithmetic means for each treatment.

**Table 36.** Mean incidence and severity of powdery mildew on bunches of Cabernet Sauvignon (n = 20 per plot), 22 April 2004, E-L 36 (3-4 weeks preharvest), Cooina Vale vineyard. Note that the whole trial area was sprayed with sulfur 6-8 weeks previously.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	62	c	5.5	-
sulfur 12 g/L	0.8	a	0.01	a
sulfur 6 g/L	1.7	a	0.01	a
milk 1:10 or 1:5	13	ab	0.16	bc
whey 25 g/L	11	ab	0.09	ab
Ecocarb® 4 g/L	12	ab	0.11	bc
Program 1	19	b	0.3	c
	lsd	14.1	0.1451	
	df	28 (2)	24 (1)	
	<b>P</b>	<0.001	0.007	

**Table 37.** Mean incidence and severity of rot on bunches of Cabernet Sauvignon (n = 20 per plot), 22 April 2004, E-L 36 (3-4 weeks pre-harvest), Cooina Vale vineyard. Rot was defined as necrotic, split or shrivelled berries, with or without fungal sporulation.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	97	c	21	-
sulfur 12 g/L	46	a	0.6	a
sulfur 6 g/L	52	a	0.7	a
milk 1:10 or 1:5	90	bc	2.4	bc
whey 25 g/L	88	bc	1.9	b
Ecocarb® 4 g/L	85	b	2.3	bc
Program 1	90	bc	3.0	c
	lsd	11.8	1.09	
	df	28 (2)	24 (1)	
	<b>P</b>	< 0.001	< 0.001	

**Table 38.** Mean incidence and severity of bunch rot fungi on Cabernet Sauvignon (n = 20 per plot), 17 May 2004, E-L 37 (pre-harvest), Cooinda Vale vineyard. Rot was defined as necrotic, split or shrivelled berries, with or without sporulation. Severity was defined as the area of the bunch that was covered in mycelia and sporulating colonies. *Botrytis cinerea* was the predominant bunch rot fungus.

Treatment	Mean incidence (%)		Mean severity (%)	
untreated control	92	c	5.6 <sup>1</sup>	c
sulfur 12 g/L	53	a	1.5	a
sulfur 6 g/L	55	ab	1.3	a
milk 1:10 or 1:5	49	a	0.91	a
whey 25 g/L	58	ab	1.0	a
Ecocarb <sup>®</sup> 4 g/L	73	bc	3.3	b
Program 1	72	b	2.9	b
	lsd	18.8	1.35	
	df	28 (2)	27 (3)	
	<b>P</b>	<0.001	<0.001	

<sup>1</sup>Block 1 of the untreated control had a severity of 44% and this outlier was removed from the analysis.

**Table 39.** Mean incidence and severity of bunch rot fungi on Cabernet Sauvignon (n = 20 per plot), May 17, 2004, E-L 37 (pre-harvest), Cooinda Vale vineyard.

Treatment	Mean total soluble solids (°Brix)	
untreated control	23.6	d
sulfur 12 g/L	20.5	a
sulfur 6 g/L	21.7	bc
milk 1:10 or 1:5	20.8	ab
whey 25 g/L	21.7	bc
Ecocarb <sup>®</sup> 4 g/L	22.4	c
Program 1	21.4	abc
	lsd	1.15
	residual df	28 (2)
	<b>P</b>	< 0.001



### Frogmore Creek 2004/05

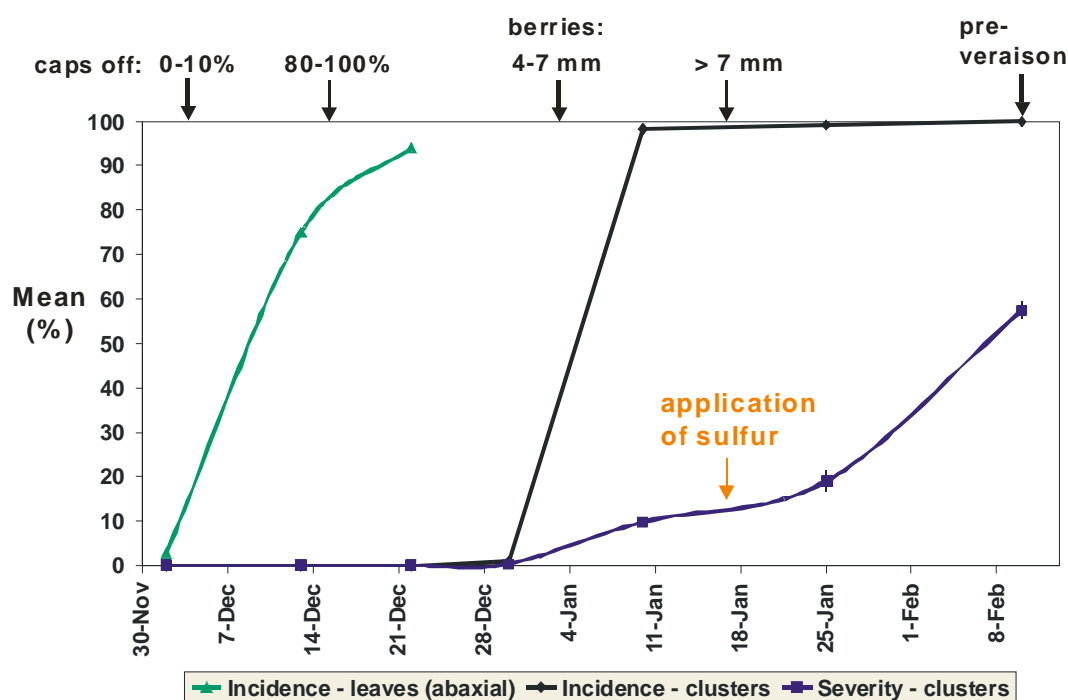
#### Disease progression in Chardonnay

High crop vigour (Table 40) and weather conditions were conducive to development of a severe powdery mildew epidemic (Figures 20 and 21). Flag shoots were not detected in the trial area, although they were detected in other blocks of the vineyard between 22 and 25 October 2004 and again on 10 November 2004. Powdery mildew was not detected anywhere in the trial area before 18 November and disease was first recorded on the underside of leaves (abaxial surface) on 3 December 2004, at the beginning of cap fall. Powdery mildew on bunches was first detected on 30 December 2004, albeit on one bunch only in an untreated plot in block 6. Incidence of disease on bunches then increased rapidly to almost 100% by 10 January 2005. Figure 20 illustrates disease progression and the increase of disease severity on bunches to a maximum of 58% by 10 February 2005. At this time, powdery mildew was evident on the upper surface (adaxial surface) of leaves. Disease severity on bunches continued to increase after sulfur was applied to untreated control plots on 17 January 2005, when berries were, on average, pea size.

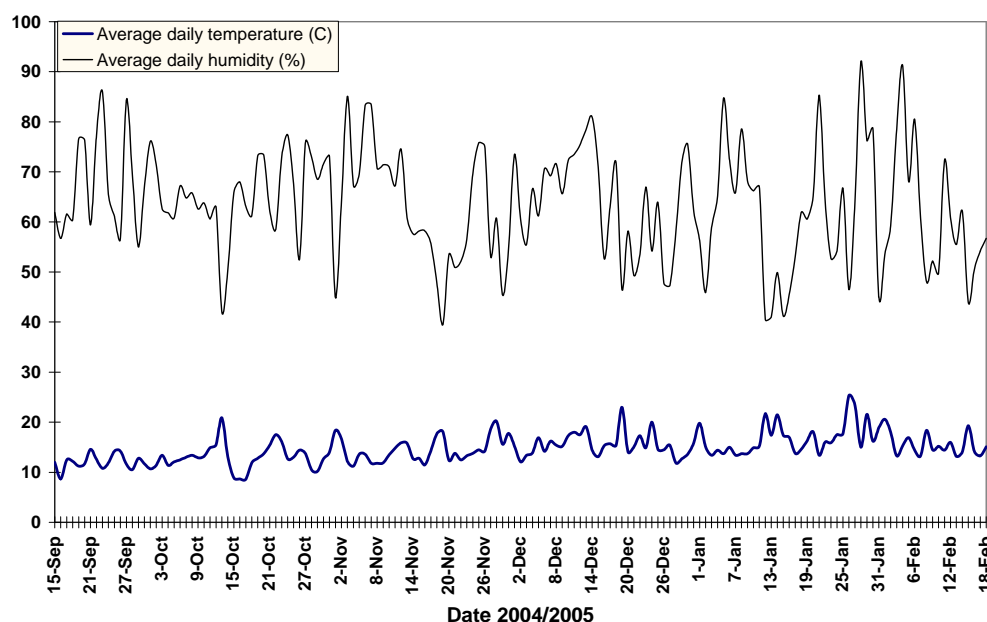
The threshold of 60 for the Gubler-Thomas risk index for powdery mildew was recorded on January 29, 2005, well after the powdery mildew epidemic had commenced in untreated plots (Figure 22).

#### Disease progression in untreated Pinot noir

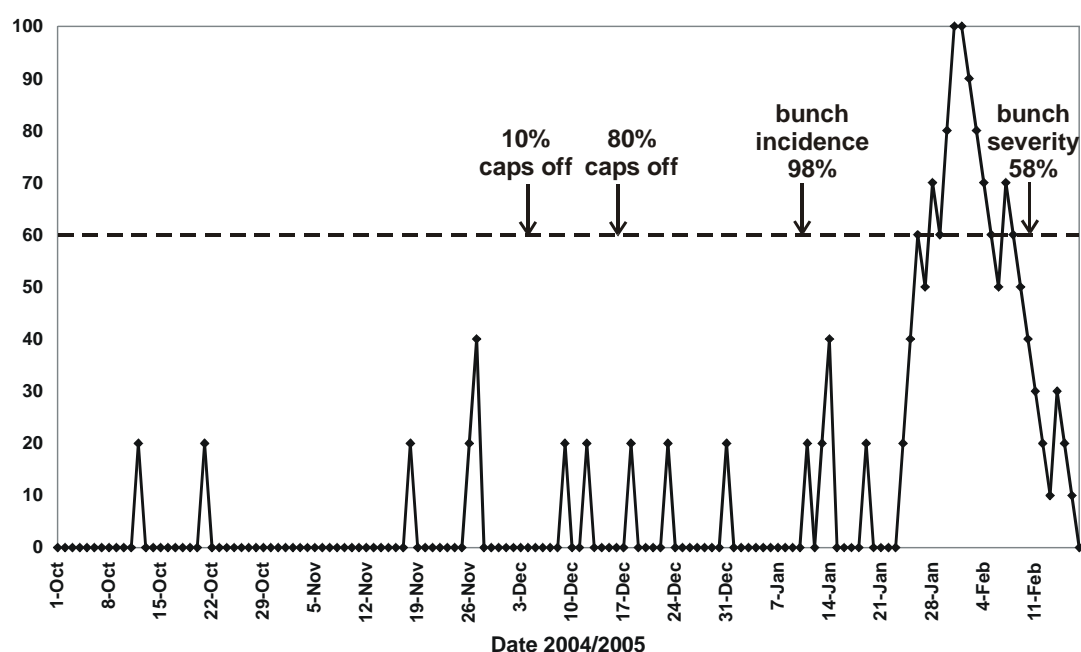
Disease incidence on the abaxial surface of leaves in the single untreated plot increased rapidly between 13 December (43% incidence) and 15 December (75% incidence). In general, disease was less severe in Pinot noir than Chardonnay.



**Figure 20.** Progression of powdery mildew in small plots of Chardonnay vines that were not treated with fungicide until pea-sized berries. Each point is the mean for six plots. Frogmore Creek vineyard, Coal River Valley, 2004/05.



**Figure 21.** Average daily temperature (°C, solid line) and humidity (%) at Frogmore Creek vineyard, 2004/2005.



**Figure 22.** Gubler-Thomas risk index for powdery mildew at Frogmore Creek vineyard to 18 February 2005. Bunch incidence and severity of powdery mildew refer to the mean for the untreated control in Chardonnay.

### Canopy vigour

The Chardonnay canopy, with vertical shoot positioning, had a higher percentage of interior leaves and clusters than the Pinot noir trained as a Scott Henry trellis (Table 40). When compared with the low vigour of block F1 Chardonnay used for the milk/whey trial in

2003/04, spring rainfall in 2004/05 year appeared to favour vigour, especially during November and the pre-flowering period.

**Table 40.** Canopy density for Chardonnay and Pinot noir, Frogmore Creek vineyard, 13 January 2005.

	<b>Gaps (%)</b>	<b>Leaf layer number (LLN)</b>	<b>Interior leaves (%)</b>	<b>Interior clusters (%)</b>	<b>Mean no. of nodes (n = 3 shoots)</b>	<b>Mean shoot length (cm) (n = 3 shoots)</b>
<b>Chardonnay</b>						
Untreated	12	1.5	22	26	21	110
Sulfur	5	2.0	27	40	21	104
<b>Pinot Noir</b>						
Whey	13	1.1	14	11	19	103
Sulfur	8	1.5	12	11	20	130
Optimum <sup>a</sup>	20 - 40%	1.0-1.5 or less	<10%	<40%	-	-

<sup>a</sup>according to Smart and Robinson (1991)

### Effectiveness of treatments

The epidemic of powdery mildew in the trial area was severe and would have reduced grape yield and quality if left unchecked. In addition to the untreated control plots, inoculum was spread to treated plots from the buffer rows where disease suppression was sub-optimal. The shoots extended well beyond the top wire in conditions of high crop vigour, and spray coverage in the shoot-tip region at maximum canopy development appeared to be inadequate. The Pinot noir was shoot tipped on 17 January 2005, to reduce both vigour and the amount of inoculum from diseased shoot tips.

When compared with the untreated control, all treatments reduced the mean severity of powdery mildew on leaves and bunches of Chardonnay and Pinot noir (Tables 41-52). The mean incidence of powdery mildew on leaves of Chardonnay and Pinot noir was also reduced by all treatments; whereas only the standard sulfur treatment reduced the mean incidence of powdery mildew on bunches at veraison.

If a mean severity of powdery mildew on bunches of greater than 3% is commercially unacceptable, then sulfur, applied at a rate of 6-10 g/L, was the only treatment that provided acceptable control of powdery mildew in both Chardonnay and Pinot noir (Tables 45 and 51). If up to 5% is acceptable, then sulfur and 'Horti-Oil+Ecocarb, whey, sulfur' provided effective control on Pinot noir and milk alone was marginal (5%). If the economic threshold for the severity of powdery mildew is increased to 10%, then all treatments applied to Pinot noir provided acceptable control, using this criterion. Tables 46 and 52 describe the mean number of bunches (per 20 sampled) per treatment with a severity of powdery mildew  $\geq 10\%$ .

### Chardonnay

In Chardonnay, treatments were first separated statistically during late flowering when the incidence of mildew on the abaxial surface of leaves was 75% in the untreated control (Table 41). A subsequent assessment of leaves (Table 42) confirmed that the two treatments that included at least three sulfur applications before berries were peppercorn to pea size had a significantly lower incidence and severity of powdery mildew on leaves than other treatments. However, after powdery mildew developed on Chardonnay berries, the difference between

these two treatments in mean disease severity and incidence became statistically significant (Tables 43-46), with the standard sulfur treatment providing the greatest disease suppression.

The milk or 'whey only' treatments were not separated statistically for any measure of disease incidence or severity on bunches (Tables 43-45). By the final assessment (Tables 45 and 46), the treatment 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then whey' resulted in a higher mean disease severity when compared with the 'whey only' treatment. This treatment was not separated statistically from the milk treatment for mean disease severity (Table 44) but the 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then whey' treatment had more bunches with a severity of powdery mildew that was greater than or equal to 10%, when compared with the milk or 'whey only' treatments (Table 46). On January 10, when the berries were pea size (Table 43), the 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then whey then sulfur' treatment, which had one application of sulfur on January 3, had a lower mean incidence of powdery mildew on bunches when compared with the 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then whey' treatment. However, from bunch closure onwards (Tables 44 and 45), these two treatments were equivalent in their level of disease suppression.

If sulfur was applied during flowering instead of whey, a higher level of disease suppression was observed at the pre-veraison assessment (Tables 45 and 46). While this treatment had a lower mean disease severity than the treatment where sulfur was applied after flowering, it was not separated statistically from the milk or 'whey only' treatments.

**Table 41.** Mean incidence and severity of powdery mildew on the abaxial surface of Chardonnay leaves (n = 40 per plot), 13 December 2004, E-L 25-26 (80-100% caps off), Frogmore Creek vineyard.

Treatment <sup>1</sup>	Mean incidence (%)		Mean severity (%)	
untreated control	75	c	0.37	e
sulfur 6 g/L	7	a	0.013	a
milk 1:10 then 1:5 (then 1:10)	49	b	0.072	bc
whey 25 g/L	44	b	0.090	cd
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey	41	b	0.076	bc
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey (then sulfur)	48	b	0.120	d
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur (then whey)	14	a	0.048	ab
lsd	9.4		0.039	
residual df	29 (1)		22 (3)	
<b>P</b>	< 0.001		<0.001	

<sup>1</sup>Text in parentheses indicates materials that were applied after the date of assessment.

**Table 42.** Mean incidence and severity of powdery mildew on the abaxial surface of Chardonnay leaves (n = 40 per plot), 30 December 2004, E-L 30 (berries peppercorn to pea size), Frogmore Creek vineyard.

Treatment <sup>1</sup>	Mean incidence (%)		Mean severity (%)	
untreated control	93	c	1.9	d
sulfur 6-10 g/L	13	a	0.04	a
milk 1:10 then 1:5 (then 1:10)	71	b	0.38	b
whey 25 g/L	71	b	0.31	b
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey	78	b	0.55	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey (then sulfur)	75	b	0.53	bc
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur (then whey)	21	a	0.04	a
lsd	12.3		0.152	
residual df	29 (1)		21 (4)	
<b>P</b>	<0.001		<0.001	

<sup>1</sup>Text in parentheses indicates materials that were applied after the date of assessment.

**Table 43.** Mean incidence and severity of powdery mildew on Chardonnay bunches (n = 20 per plot), 10 January 2004, E-L 31 (berries pea size), Frogmore Creek vineyard.

Treatment	Mean incidence (%)		Mean severity (%)	
untreated control	98	e	9.9	d
sulfur 6-10 g/L	16	a	0.13	a
milk 1:10 then 1:5 then 1:10	88	de	2.3	c
whey 25 g/L	79	cd	2.1	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey	84	d	2.1	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey then sulfur	68	bc	1.6	bc
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur then whey	64	b	1.1	b
lsd	12		0.89	
residual df	27 (3)		21 (4)	
<b>P</b>	<0.001		<0.001	

**Table 44** Mean incidence and severity of powdery mildew on Chardonnay bunches (n = 20 per plot), 25 January 2004, E-L 32 (beginning of bunch closure), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	99	d	19	d
sulfur 6-10 g/L	38	a	0.3	a
milk 1:10 then 1:5 then 1:10	92	bcd	5.1	c
whey 25 g/L	78	b	3.6	bc
Horti-oil® + Ecocarb® then whey	95	cd	5.1	c
Horti-oil® + Ecocarb® then whey then sulfur	80	bc	3.8	bc
Horti-oil® + Ecocarb® then sulfur then whey	79	b	2.6	b
lsd	15.5		2.0	
residual df	30		22 (3)	
<b>P</b>	<0.001		<0.001	

**Table 45.** Mean incidence and severity of powdery mildew on Chardonnay bunches (n = 20 per plot), 10 February 2004, E-L 33-34 (some berries beginning to soften), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	100	b	58	e
sulfur 6-10 g/L	79	a	2.3	a
milk 1:10 then 1:5 then 1:10	96	b	24	bcd
whey 25 g/L	97	b	18	bc
Horti-oil® + Ecocarb® then whey	98	b	32	d
Horti-oil® + Ecocarb® then whey then sulfur	98	b	25	cd
Horti-oil® + Ecocarb® then sulfur then whey	94	b	15	b
lsd	9.4		9.3	
residual df	30		29 (1)	
<b>P</b>	0.002		<0.001	

**Table 46.** Mean number of Chardonnay bunches with a severity of powdery mildew greater than or equal to 10% (n = 20 per plot), 10 February 2004, E-L 33-34 (some berries beginning to soften), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean number</b>	
untreated control	20	d
sulfur 6-10 g/L	1.7	a
milk 1:10 then 1:5 then 1:10	13	b
whey 25 g/L	12	b
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey	18	cd
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey then sulfur	14	bc
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur then whey	11	b
lsd	4.0	
residual df	30	
<b>P</b>	<0.001	

### **Pinot noir**

The results for Pinot noir were consistent with those observed for Chardonnay, although there was less separation of treatment effects. Again, the two treatments that included at least three sulfur applications before berries were peppercorn size resulted in significantly lower incidence of powdery mildew on leaves than other treatments (Tables 47 and 48). Mean severity of powdery mildew on the abaxial surface of Pinot noir leaves was very low in the untreated control, the two sulfur treatments had the lowest mean disease severity, although the 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then sulfur' treatment was not significantly different from the milk or whey treatment.

All treatments reduced the mean severity of powdery mildew on Pinot noir bunches, but only the standard sulfur treatment expressed a level of disease that was significantly less than all other treatments, except for the 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then sulfur then whey' treatment on 11 January (Table 49). At bunch closure and veraison, only the standard sulfur treatment reduced the mean incidence of powdery mildew on Pinot noir bunches, relative to the other treatments (Tables 50 and 51). The effectiveness of sulfur, when assessed at veraison, was also evident when results were expressed as the mean number of bunches with a severity of powdery mildew greater than or equal to 10% (Table 52).

**Table 47.** Mean incidence and severity of powdery mildew on the abaxial surface of Pinot noir leaves (n = 40 per plot), 15 December 2004, E-L 25-26 (80-100% caps off), Frogmore Creek vineyard.

<b>Treatment<sup>1</sup></b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	75	d	0.25	d
sulfur 6 g/L	12	a	0.02	a
milk 1:10 then 1:5 (then 1:10)	56	c	0.12	bc
whey 25 g/L	56	c	0.13	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey (then sulfur)	51	c	0.13	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur (then whey)	31	b	0.06	ab
lsd	14		0.053	
residual df	20		19 (1)	
<b>P</b>	< 0.001		<0.001	

<sup>1</sup>Text in parentheses indicate materials that were applied at a later date.

**Table 48.** Mean incidence and severity of powdery mildew on the abaxial surface of Pinot noir leaves (n = 40 per plot), 30 December, 2004, E-L 29 (berries peppercorn size), Frogmore Creek vineyard.

<b>Treatment<sup>1</sup></b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	50	c	0.14	d
sulfur 6-10 g/L	5	a	0.01	a
milk 1:10 then 1:5 (then 1:10)	31	b	0.05	bc
whey 25 g/L	33	b	0.07	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey (then sulfur)	35	b	0.06	c
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur (then whey)	9	a	0.02	ab
lsd	11		0.026	
residual df	20		19 (1)	
<b>P</b>	<0.001		<0.001	

<sup>1</sup>Text in parentheses indicate materials that were applied at a later date.



**Table 49.** Mean incidence and severity of powdery mildew on Pinot noir bunches (n = 20 per plot), 11 January 2004, E-L 30-31 (berries up to pea size), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	95	d	6	c
sulfur 6-10 g/L	8	a	0.05	a
milk 1:10 then 1:5 then 1:10	69	c	0.97	b
whey 25 g/L	66	bc	0.72	b
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey then sulfur	48	b	0.72	b
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur then whey	24	a	0.20	a
lsd	20		0.46	
residual df	20		18 (2)	
<b>P</b>	<0.001		0.003	

**Table 50.** Mean incidence and severity of powdery mildew on Pinot noir bunches (n = 20 per plot), 25 January 2004, E-L 32 (beginning of bunch closure), Frogmore Creek vineyard.

<b>Treatment</b>	<b>Mean incidence (%)</b>		<b>Mean severity (%)</b>	
untreated control	85		9.9	
sulfur 6-10 g/L	15	a	0.10	a
milk 1:10 then 1:5 then 1:10	77	c	1.6	b
whey 25 g/L	62	bc	1.5	b
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then whey then sulfur	71	c	1.8	b
Horti-oil <sup>®</sup> + Ecocarb <sup>®</sup> then sulfur then whey	47	b	1.1	b
lsd	23		0.94	
residual df	20		19 (1)	
<b>P</b>	<0.001		0.01	

**Table 51.** Mean incidence and severity of powdery mildew on Pinot noir bunches (n = 20 per plot), 10 February 2004, E-L 33-34 (some berries beginning to soften), Frogmore Creek vineyard.

Treatment	Mean incidence (%)		Mean severity (%)	
untreated control	90		24	
sulfur 6-10 g/L	39	a	0.60	a
milk 1:10 then 1:5 then 1:10	95	b	5.0	b
whey 25 g/L	83	b	5.4	b
Horti-oil® + Ecocarb® then whey then sulfur	88	b	6.2	b
Horti-oil® + Ecocarb® then sulfur then whey	79	b	4.0	b
lsd	18.5		3.9	
residual df	20		19 (1)	
<b>P</b>	<0.001		0.053	

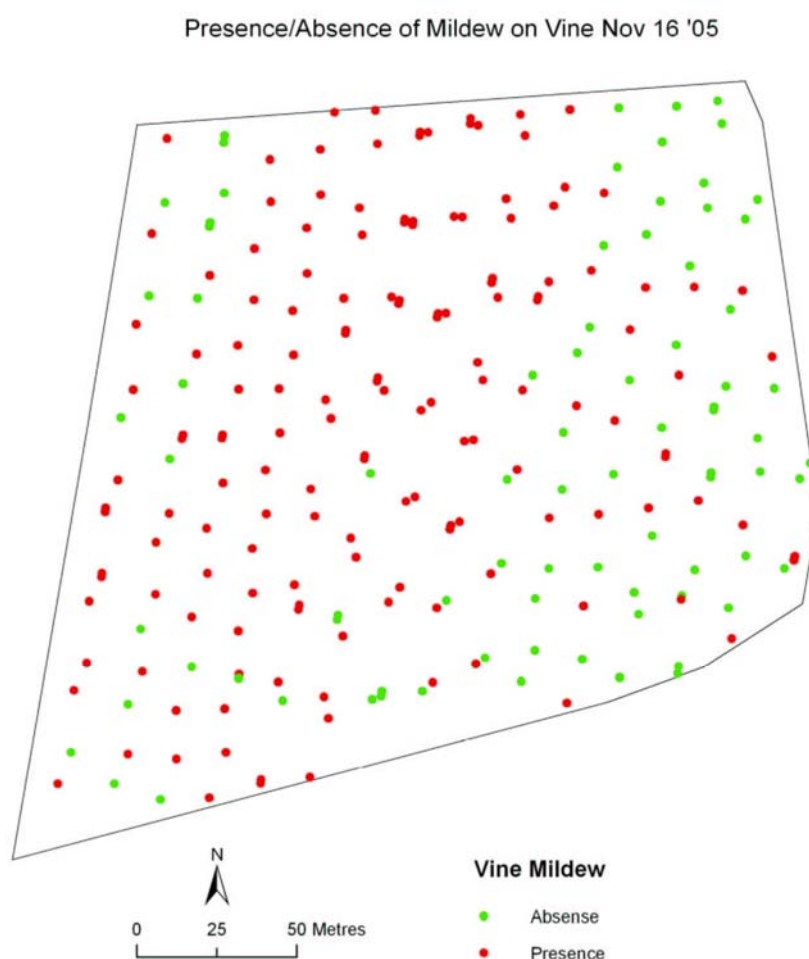
**Table 52.** Mean number of Pinot noir bunches with a severity of powdery mildew greater than or equal to 10% (n = 20 per plot), 10 February 2004, E-L 33-34 (some berries beginning to soften), Frogmore Creek vineyard.

Treatment	Mean number <sup>1</sup>	
untreated control	(15)	
sulfur 6-10 g/L	-3.50 (0.5)	b
milk 1:10 then 1:5 then 1:10	0.55 (4.2)	a
whey 25 g/L	-0.89 (5.7)	a
Horti-oil® + Ecocarb® then whey then sulfur	-0.14 (4.8)	a
Horti-oil® + Ecocarb® then sulfur then whey	-1.01 (4.3)	a
lsd	2.11	
residual df	20	
<b>P</b>	0.008	

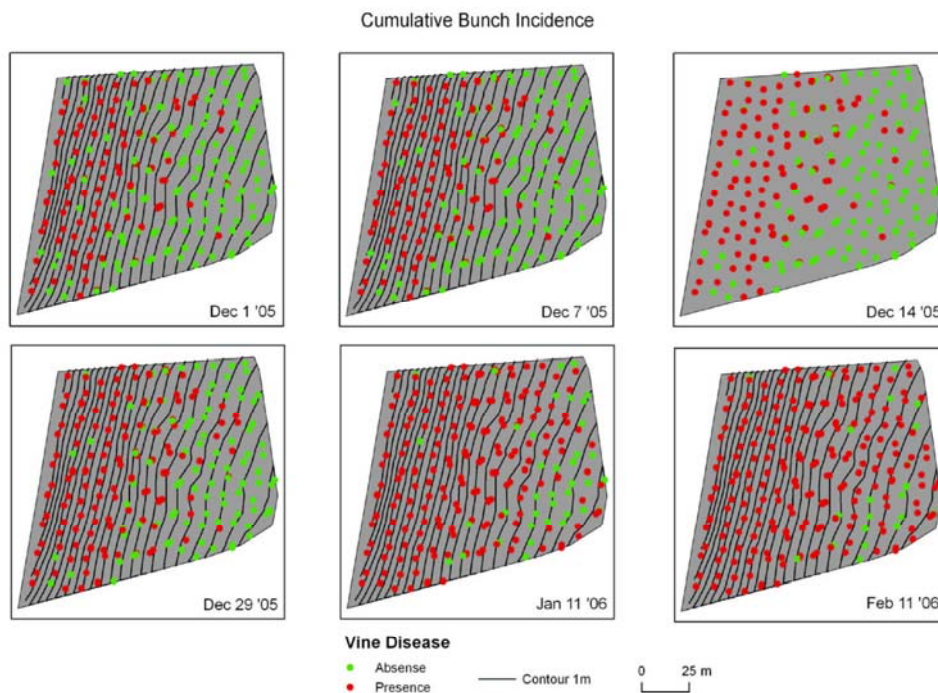
<sup>1</sup>Means transformed according to  $\text{LOG}_{10}(x+0.00005)$ . Numbers in parentheses are the arithmetic means for each treatment.

### Evaluating organic spray programs in Tasmania using spatial information

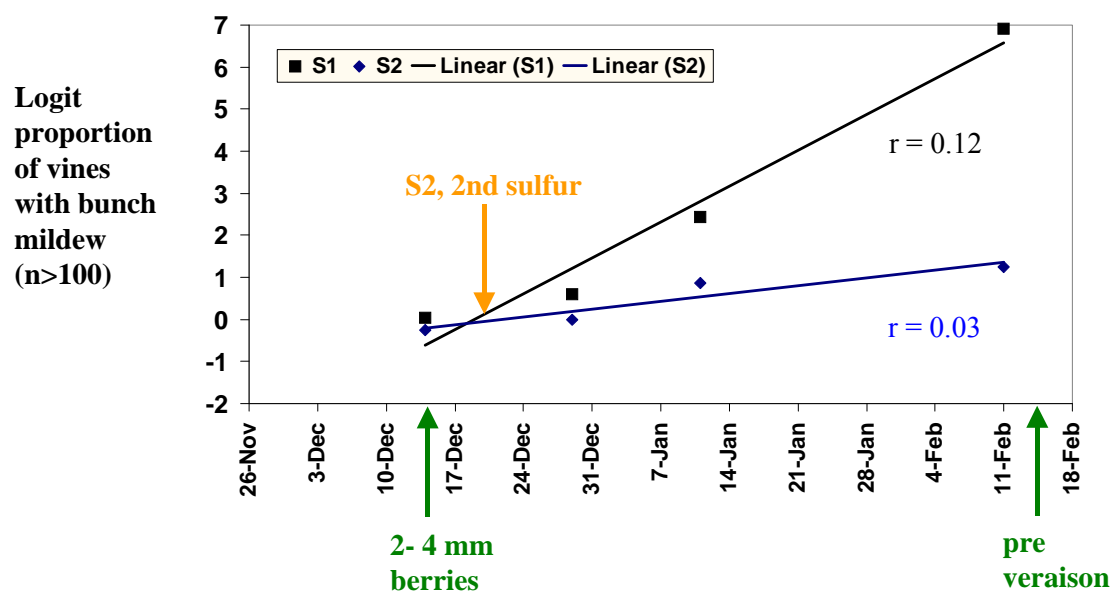
Powdery mildew was first detected within the block of Pinot noir in mid-November, before flowering. The disease appeared suddenly and extensively across the block (Figure 23). Powdery mildew progressed onto the bunches at 100% cap fall and incidence appeared to increase significantly from mid to late December, 2005, when berries were increasing in diameter from 2 to 7 mm (Figure 24).



**Figure 23.** Presence or absence of powdery mildew on the abaxial surface of Pinot noir leaves on November 16, 2005, pre-flowering.



**Figure 24** Presence or absence of powdery mildew on Pinot noir bunches from December 1, 2005 (100% capfall) to February 11, 2006 (pre-veraison).

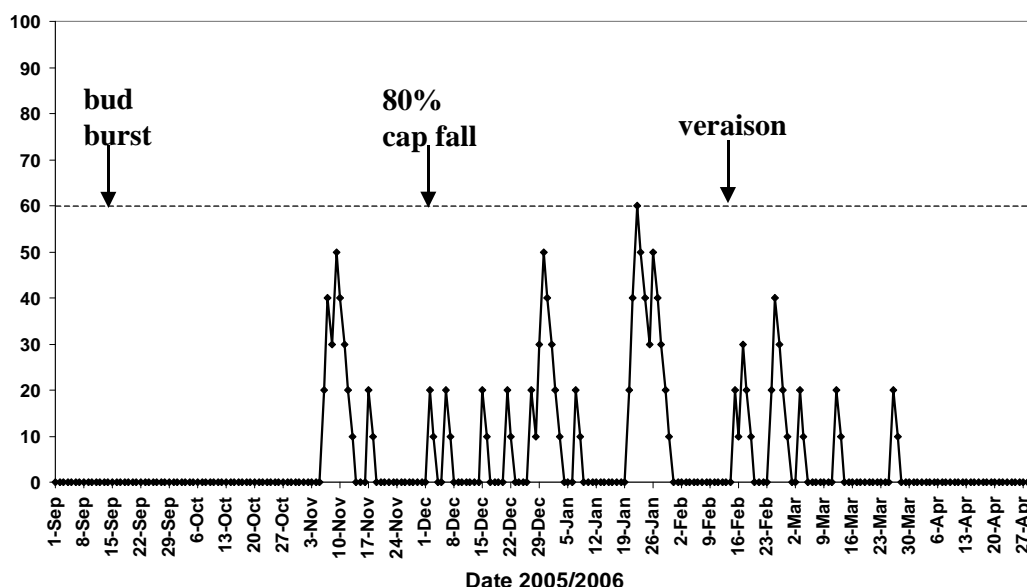


**Figure 25.** Disease incidence as a function of time. The value of  $r$  is the slope of the linear regression:  $\text{logit}(\text{disease incidence}) = r(\text{days}) + \text{intercept}$ .

At veraison, bunches of Program S2 vines had a mean disease severity of 1.5%, which was significantly less than the mean of 3.1% recorded for bunches of Program S1 vines ( $P < 0.001$ ,  $df = 230$ , one-sided  $t$  test of angular transformed data). The corresponding mean disease incidence was 31 and 61%, for Program S2 and S1 respectively. Program S2 had a slower rate of disease increase than Program S1 after the second application of sulfur had been applied to Program S2 (Figure 25).

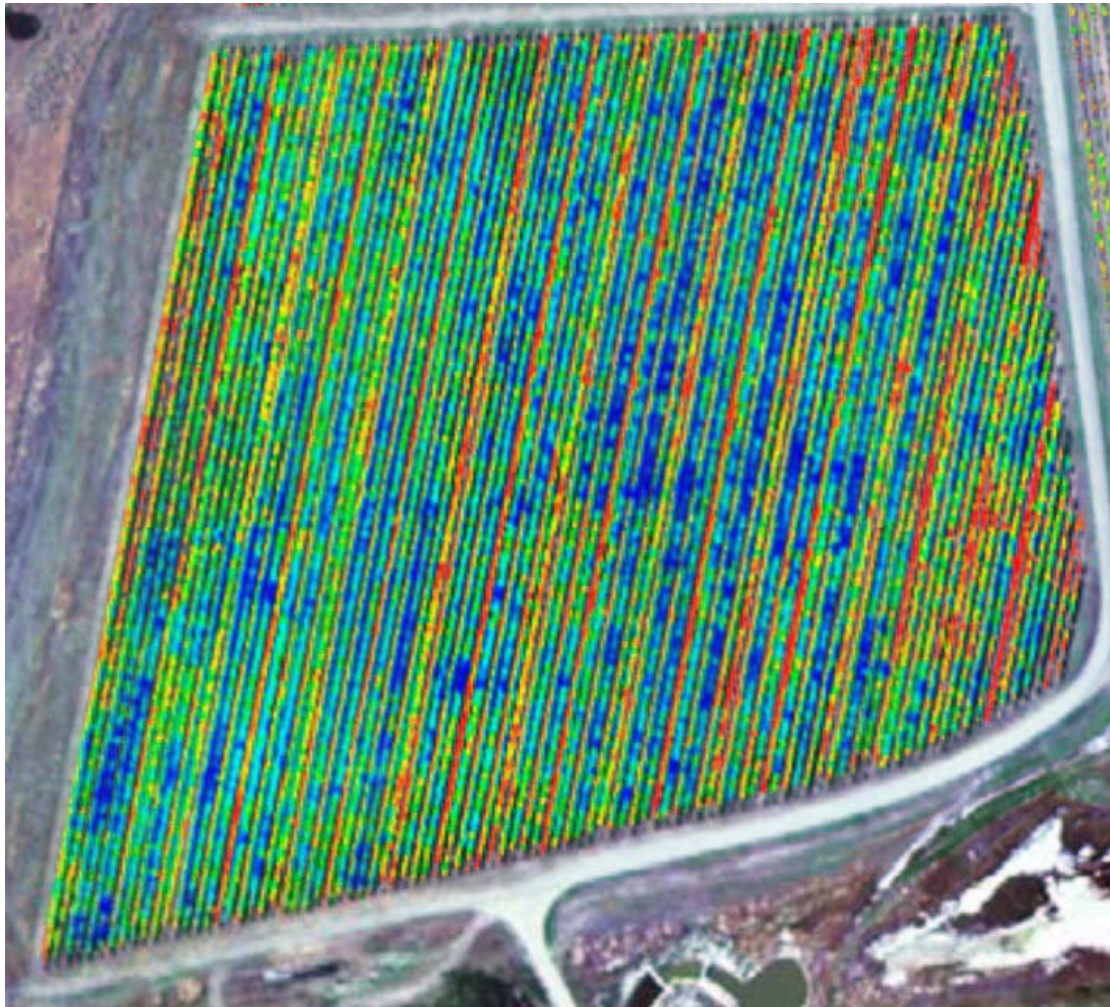
The cumulative Gubler-Thomas risk index for powdery mildew reached the threshold value of 60 once only on January 22, 2006 (Figure 26), at least 9 weeks after the epidemic of powdery mildew was first detected.

Disease severity was not assessed at harvest because colonies of powdery mildew were difficult to detect on the red grapes. At harvest our grower cooperator was pleased that the low severity of powdery mildew across the block was commercially acceptable. In October 2006, the block was monitored for the development of flag shoots. Although no flag shoots were observed in the sample vines, our grower cooperator removed seven flagshoots from the 4.5 ha block on October 11, 2006.



**Figure 26.** Cumulative Gubler-Thomas risk index for powdery mildew at Frogmore Creek vineyard, 2005/06.

We are working with Dr Rob Bramley, CSIRO Sustainable Ecosystems, in the application of geostatistical methods (Bramley 2005) to compare spatial patterns (variograms) in powdery mildew severity with elevation and patterns of variation in vine vigour across the 4.5 ha block (Figure 27).



**Figure 27.** Aerial image of the reflectance of the vine vegetation, specifically the plant cell density, at veraison in a 4.5 ha block of Pinot noir, Frogmore Creek vineyard. The dark blue shades represent high vine vigour relative to the orange and red shades that represent low vine vigour. Imagery flown and processed by SpecTerra Services, with base mapping imagery supplied by DPIWE LIS to GDA94 MGA55.

## Discussion

### Frogmore Creek Chardonnay, 2003/04

Milk or whey provided commercially acceptable control of powdery mildew on bunches, equivalent to sulfur, under conditions of high disease pressure but low vine vigour. Sulfur was the most effective treatment given the low incidence of powdery mildew on bunches relative to the test materials. The degree of powdery mildew control was evident 6 weeks before harvest, when the mean incidence and severity of bunches with berries that were necrotic, split or shrivelled was highest in the untreated control and lowest for treatments of sulfur, milk or whey.

The rate of Ecocarb<sup>®</sup> appeared to be important in disease control. Ecocarb<sup>®</sup> SR, applied at 3 g/L resulted in a higher mean disease severity on bunches pre-veraison than Ecocarb<sup>®</sup> applied at 4 g/L. Both treatments resulted in a mean disease severity on bunches pre-veraison that was between 1 and 3%, as did Program 1, in which the Ecocarb component was applied at 3 g/L.

Higher rates of Ecocarb may be needed for effective management of powdery mildew on Chardonnay bunches, but may be too costly for adoption.

Bunch rot, caused by *Botrytis cinerea* or other bunch rot fungi, was not expressed in this trial. However, moist incubation of Chardonnay bunches in late April revealed that the suppression of powdery mildew by all treatments reduced the mean severity of 'bunch rot' when compared with the untreated control.

The lack of a significant difference in bunch weights among treatments may have resulted from an interaction between the effect of the treatment prior to veraison and subsequent severity of powdery mildew infection on leaves.

All test materials and spray programs, except sulfur, failed to persist and provide late season control of powdery mildew on leaves when applications ceased close to veraison. Result illustrated the persistence of sulfur, up to 53 days in this study, in protecting leaves from infection by *E. necator*.

#### **Cooinda Vale Cabernet Sauvignon, 2003/04**

Unlike the results for the trial in Chardonnay (2003/04), milk or whey was not as effective as sulfur in reducing the mean incidence and severity of powdery mildew on Cabernet Sauvignon bunches. The difference in the results between the two trials might be explained by the greater vigour observed in the Cabernet Sauvignon vines, as this variety is unlikely to be more susceptible to powdery mildew than Chardonnay. The denser canopy may have reduced spray coverage when compared with the Chardonnay. Like the trial in Chardonnay, Ecocarb<sup>®</sup> reduced the severity of powdery mildew on the adaxial surface of leaves to a level equivalent to that observed for milk, whey or sulfur at pre-veraison.

Program 1 was the least effective treatment in the trial and resulted in unacceptable control of powdery mildew on a buffer row that experienced high disease pressure. Greater suppression of disease might have occurred if the rates of Ecocarb<sup>®</sup> and Synertril Horti-oil<sup>®</sup> were increased to 6 g/L and 4 ml/L respectively.

There appeared to be no significant advantage in applying sulfur at a rate greater than 6 g/L, when sulfur was the only component of the spray program. It is common practice for growers in Tasmania to apply sulfur at rates between 8 and 10 g/L. Had a commercial sprayer been used in this trial then the effective dose of sulfur on fruit and foliage may have been lower. However, application of high rates of sulfur in Tasmania should be reviewed in relation to timing sprays according to pathogen activity and other materials used in the spray program. Powdery mildew can predispose grape berries to colonisation by bunch rot fungi and this trial demonstrated that suppression of powdery mildew resulted in less bunch rot. The fact that the sulfur treatments had a bunch rot incidence of around 50% (Table 14), indicated that berry damage was caused not only by powdery mildew infection but also by environmental or physiological factors. The incidence of damaged berries was lowest in plots treated with sulfur (Table 37), whereas colonisation by bunch rot fungi was least for the sulfur treatments and the whey and milk treatments. This result suggested that milk or whey might have favoured a microflora on the berry surface that was suppressive to colonisation by *Botrytis cinerea* and other bunch rot fungi.

When compared with the treatments, the larger amount of soluble solids observed in grapes from the untreated plots close to harvest may reflect the greater level of colonisation by the bunch rot fungus, *B. cinerea*. In certain environmental conditions, this fungus is known to make the berry skin more permeable, leading to a loss of moisture and concentration of berry sugars (Donèche 1987 cited in Geny et al. 2003).



### **Frogmore Creek, Chardonnay and Pinot Noir, 2004/05**

Clearly, the Gubler-Thomas mildew risk index was unreliable in predicting the onset of the disease epidemic in this trial. Disease was more severe in the Chardonnay vines when compared with the Pinot noir vines, which may have reflected the greater inherent susceptibility of this variety to powdery mildew and/or the likely higher relative humidity and/or shading within the denser canopy of Chardonnay.

Given the timing of disease increase on leaves and bunches, it appeared that spray applications in October were unnecessary. The standard sulfur treatment provided commercially acceptable disease control, although mean incidence of powdery mildew on bunches was relatively high (79% in Chardonnay, Table 45), indicating that either spray coverage, timing and/or sulfur rate were sub-optimal. The presence of powdery mildew on shoot tips, presumably from inadequate spray coverage, would have increased the amount of inoculum available for bunch infection. The rate of sulfur applied during the flowering period was 6 g/L, which may have been sub-optimal for fan-assisted application based on anecdotal evidence from growers in Tasmania who claim that 8-10 g/L sulfur is needed for adequate powdery mildew control in this cool climate region.

The 'whey only' treatment provided greater suppression of powdery mildew on Chardonnay bunches than the 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then whey' treatment. Either whey was the more effective material pre-flowering or the Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> mixture had some direct or indirect effect on shoot vigour and/or leaf physiology. Powdery mildew remained undetected in the trial area until December 2 and the timing of the last application of the Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> mixture was November 24. This last application may have had some impact on an extremely low and undetectable incidence of powdery mildew, given that disease incidence on leaves increased rapidly between December 2 and 22. Alternatively, application of the Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> mixture had an insignificant effect on disease incidence at the times it was applied. Evidence for the latter is that the incidence of powdery mildew on Chardonnay leaves was similar for both the 'whey only' and 'Horti-oil<sup>®</sup> + Ecocarb<sup>®</sup> then whey' treatments. These results demonstrate the limits in effectiveness of milk or whey in controlling powdery mildew in Chardonnay when disease pressure is high and shoot growth is vigorous. Milk or whey can suppress disease to acceptable levels in less vigorous canopies, such as the Chardonnay block F1 tested in the 2003/2004 growing season. Milk or whey appears to provide a promising alternative to sulfur for a well-managed canopy of Pinot noir, depending on the amount of powdery mildew that will be tolerated in this premium grape variety.

### **Evaluating organic spray programs in Tasmania using spatial information**

This 'whole-of-block' evaluation of spray programs based on a mixture of potassium bicarbonate and a canola-based oil demonstrated that commercially acceptable control of powdery mildew was achieved in Pinot noir with one or two applications of the sulfur/oil mixture during fruit set. During consultation with our grower cooperator, we recommended that the sulfur mixture be applied immediately before and/or during flowering. In practice, the timing of applications was made by the grower, which meant that the first sulfur application was later than expected. In practice, the sulfur mixture appears to have worked as an eradicant, given that the second application in Program S2 reduced the rate of disease increase when disease incidence on bunches was already 50%.

Plant pathologists from the USA who attended the 5th International Workshop on Grapevine Downy and Powdery Mildew in 2006 were surprised that the mixture of sulfur and canola-based oil applied in this trial did not cause phytotoxicity. Although no damage was observed in this trial, this potential effect should be monitored in the future.



While small plot trials identified materials that effectively controlled powdery mildew, the ‘whole-of-block’ experiment allowed the performance of the test materials to be evaluated over a range in conditions of vine vigour in a single growing season. The need for close liaison with the grower cooperator and the researcher enabled the direct transfer of ‘commercial ready’ knowledge. The grower had control over decisions about timing fungicide applications and the results related directly to the capability of his or her commercial equipment. Indeed, the grower had no problem with giving the whole block over to experimentation and the ease of implementation meant that the grower did not need to understand the complex nature of the geostatistical design.

Geostatistical analysis, in progress, will allow ‘proof of concept’ of the whole-of-block approach in the development of disease management strategies.

### **6.3. New South Wales**

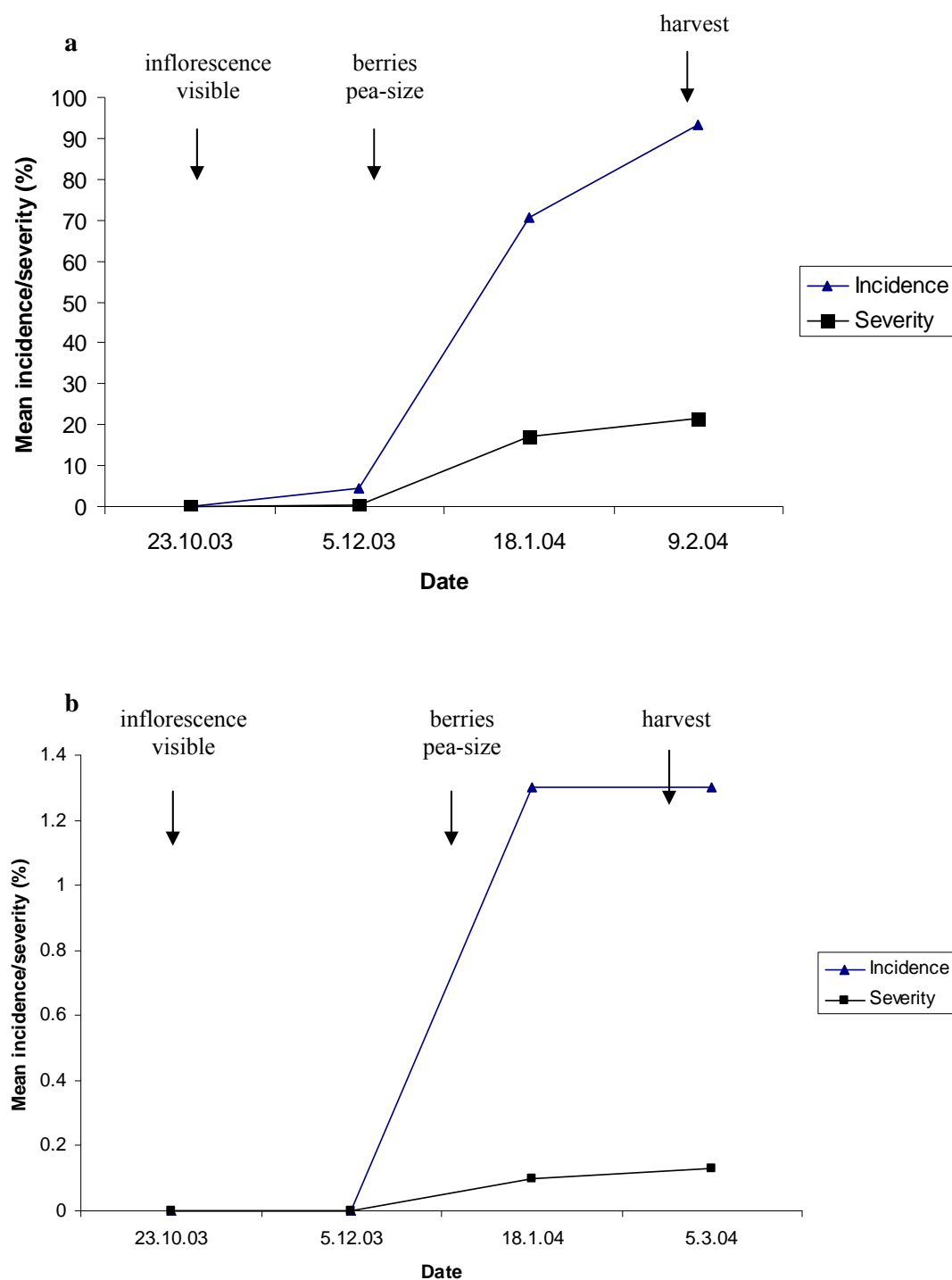
#### **Results of trials in New South Wales**

##### **2003/2004 Disease development**

No flag shoots were detected and powdery mildew was first observed on the leaves of Chardonnay and Cabernet Sauvignon on 23 October, 2003. Figure 28 (a and b) shows the progression of powdery mildew on the untreated vines for Chardonnay and Cabernet Sauvignon. For Chardonnay, powdery mildew increased from berries pea-sized to harvest. For Cabernet Sauvignon, powdery mildew also increased during berry development, however, remained constant from 18 January until harvest.

At inflorescence visible for Chardonnay the incidence and severity of powdery mildew on leaves was not significantly different between treatments ( $P = 0.24$  and  $0.13$ , respectively) (Table 53). The mean incidence ranged from 34.6% for treatment 4 (whey) to 47.9% for the untreated control. Severity ranged from 3.5% for treatment 2 (sulfur) to 5.0% for the untreated control. The incidence and severity of powdery mildew on Chardonnay bunches increased from berries pea-sized to harvest (Table 54). At berries pea-sized no significant differences in incidence and severity were recorded between treatments. At bunch closure the mean percentage incidence and severity ranged from 31.3% and 3.2%, respectively, for treatment 2 (sulfur) to 91.3% and 19.1% for treatment 5 (Ecocarb®). At harvest sulfur provided the best control with an incidence of 34.2% and severity of 4.2% (Table 54). There were no significant differences between the remaining treatments for incidence, however, milk and whey resulted in reduced severity of disease on bunches.

At inflorescence visible for Cabernet Sauvignon the incidence and severity of powdery mildew on leaves was not significantly different between treatments ( $P = 0.46$  and  $0.37$ , respectively) (Table 55). The mean percentage incidence ranged from 18.8% for treatments 5 and 6 (Ecocarb® and P1, respectively) to 31.9% for the untreated control. Disease severity ranged from 1.9% to 3.4%. No powdery mildew was recorded on Cabernet Sauvignon bunches from inflorescence visible to berries pea-sized (Table 56). At bunch closure slight powdery mildew was observed on untreated control bunches and those treated with milk. No significant differences between treatments were recorded at this phenological stage. At harvest slight powdery mildew remained on untreated control bunches. Bunches treated with Ecocarb® also showed slight disease, with an incidence of 0.6% and severity of 0.1%. Acceptable control was provided by all treatments applied to Cabernet Sauvignon.



**Figure 28.** Mean percentage incidence and severity of powdery mildew on (a) Chardonnay and (b) Cabernet Sauvignon bunches from inflorescences visible to harvest on untreated vines in the CSU vineyard in 2003/04.

**Table 53.** Mean percentage incidence and severity of powdery mildew on Chardonnay leaves at inflorescences visible for 2003/04<sup>1</sup>.

<b>Treatment<sup>2</sup></b>	<b>Incidence</b>	<b>Severity</b>
1. Untreated	47.9 (10.4)	5.0 (0.5)
2. Sulfur (g/L)	35.4 (9.8)	3.5 (1.1)
3. Milk (dilution)	39.6 (9.9)	4.0 (1.0)
4. Whey (g/L)	34.6 (9.3)	3.5 (0.9)
5. Ecocarb <sup>®</sup> (g/L)	38.3 (10.8)	3.8 (1.1)
6. Program 1	39.1 (7.8)	3.8 (0.8)
<i>P</i> value	0.24	0.13

<sup>1</sup>Standard deviations are presented in parentheses.<sup>2</sup>Treatments are according to Table 10.**Table 54.** Mean percentage incidence and severity of powdery mildew on Chardonnay bunches at berries pea-sized, bunch closure and harvest for 2003/04<sup>1</sup>.

<b>Treatment<sup>2</sup></b>	<b>Berries pea-sized</b>		<b>Bunch closure<sup>3</sup></b>		<b>Harvest</b>	
	<b>Incidence</b>	<b>Severity</b>	<b>Incidence</b>	<b>Severity</b>	<b>Incidence</b>	<b>Severity</b>
1. Untreated	4.6 (5.6)	0.5 (0.6)	70.6 (34.2) ab	17.0 (14.8) a	93.3 (11.8) a	21.5 (2.5) a
2. Sulfur (g/L)	0.8 (2.0)	0.1 (0.2)	31.3 (33.3) c	3.2 (3.4) b	34.2 (22) b	4.2 (0.6) e
3. Milk (dilution)	5.0 (7.9)	0.9 (1.4)	55.0 (23.3) bc	5.5 (3.7) b	80 (22.6) a	12.9 (1.7) c
4. Whey (g/L)	2.5 (4.2)	0.3 (0.5)	70.4 (31.2) ab	10.5 (7.6) ab	76.3 (19.4) a	9.8 (0.9) d
5. Ecocarb <sup>®</sup> (g/L)	3.3 (4.4)	0.4 (0.5)	91.3 (8.0) a	19.1 (8.8) a	80.4 (29.3) a	18.6 (2.1) b
6. Program 1	1.3 (3.1)	0.2 (0.4)	59.2 (28.0) bc	6.8 (3.7) b	79.6 (17.8) a	10.3 (0.8) cd
<i>P</i> value	0.61	0.44	0.04	0.01	0.007	0.001
LSD <sup>3</sup>	ns	ns	38.6	11.1	15.0	0.29

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.<sup>2</sup>Treatments are according to Table 10.<sup>3</sup>ns = not significant at  $P = 0.05$

**Table 55.** Mean percentage incidence and severity of powdery mildew on Cabernet Sauvignon leaves at inflorescences visible for 2003/04<sup>1</sup>.

<b>Treatment<sup>2</sup></b>	<b>Incidence</b>	<b>Severity</b>
1. Untreated	31.9 (20.1)	3.4 (2.0)
2. Sulfur (g/L)	22.5 (5.4)	2.3 (0.5)
3. Milk (dilution)	21.3 (4.8)	2.1 (0.5)
4. Whey (g/L)	23.8 (9.2)	1.9 (1.5)
5. Ecocarb <sup>®</sup> (g/L)	18.8 (3.2)	1.9 (0.3)
6. Program 1	18.8 (5.2)	2.0 (0.7)
<i>P</i> value	0.46	0.37

<sup>1</sup>Standard deviations are presented in parentheses.<sup>2</sup>Treatments are according to Table 10.**Table 56.** Mean percentage incidence and severity of powdery mildew on Cabernet Sauvignon bunches at berries pea-sized, bunch closure and harvest for 2003/04<sup>1</sup>.

<b>Treatment<sup>2</sup></b>	<b>Berries pea-sized</b>		<b>Bunch closure<sup>3</sup></b>		<b>Harvest</b>	
	<b>Incidence</b>	<b>Severity</b>	<b>Incidence</b>	<b>Severity</b>	<b>Incidence</b>	<b>Severity</b>
1. Untreated	0 (0)	0 (0)	1.3 (2.5)	0.1 (0.3)	1.3 (2.5)	0.13 (0.1)
2. Sulfur (g/L)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
3. Milk (dilution)	0 (0)	0 (0)	0.8 (1.5)	0.1 (0.2)	0 (0)	0 (0)
4. Whey (g/L)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
5. Ecocarb <sup>®</sup> (g/L)	0 (0)	0 (0)	0 (0)	0 (0)	0.6 (1.3)	0.1 (0.1)
6. Program 1	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
<i>P</i> value	ns	ns	0.55	0.55	0.52	0.22

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.<sup>2</sup>Treatments are according to Table 10.

### 2003/04 Berry quality assessments

The mean berry weight, pH, total soluble solids (TSS) and titratable acidity (TA) were measured at harvest for Chardonnay and Cabernet Sauvignon (Table 57). No significant differences were observed between treatments with respect to berry weight, pH and TA for Chardonnay or Cabernet Sauvignon. A significant difference was observed for the TSS between treatments ( $P = 0.004$ ) for Chardonnay, however, when the data from the control treatment (unsprayed) were removed from the statistical analysis no significant differences were observed.

**Table 57.** Mean pH, total soluble solids (TSS) and titratable acidity (TA) for Chardonnay and Cabernet Sauvignon at harvest in 2003/04<sup>1</sup>.

Treatment <sup>2</sup>	Berry Weight (g)	Chardonnay		
		pH	TSS (°Brix)	TA (ml/L)
1. Untreated	1.0 (0.1)	3.9 (0.05)	21.0 (1.2) c	5.4 (0.9)
2. Sulfur (g/L)	1.1 (0.1)	3.9 (0.1)	23.3 (0.8) a	5.0 (0.4)
3. Milk (dilution)	1.1 (0.1)	3.9 (0.04)	22.0 (1.6) bc	5.5 (0.5)
4. Whey (g/L)	1.0 (0.1)	3.9 (0.02)	22.0 (0.8) a	5.2 (0.4)
5. Ecocarb® (g/L)	1.0 (0.1)	3.9 (0.03)	23.0 (0.9) ab	4.9 (0.4)
6. Program 1	1.0 (0.1)	3.9 (0.02)	23.1 (0.7) a	5.2 (0.2)
<i>P</i> value	0.90	0.78	0.004	0.41
LSD <sup>3</sup>	ns	ns	1.2	ns
Cabernet Sauvignon				
1. Untreated	1.0 (0.1)	4.0 (0.04)	18.8 (0.7)	3.7 (0.1)
2. Sulfur (g/L)	1.0 (0.03)	4.0 (0.1)	20.2 (0.9)	3.5 (0.3)
3. Milk (dilution)	1.0 (0.03)	4.0 (0.1)	19.6 (1.1)	3.6 (0.1)
4. Whey (g/L)	1.0 (0.08)	4.0 (0.1)	19.5 (0.7)	3.4 (0.2)
5. Ecocarb® (g/L)	1.0 (0.05)	4.0 (0.1)	19.2 (0.8)	3.5 (0.1)
6. Program 1	1.0 (0.05)	4.0 (0.03)	18.7 (0.8)	3.6 (0.3)
<i>P</i> value	0.51	0.66	0.13	0.61
LSD <sup>3</sup>	ns	ns	ns	ns

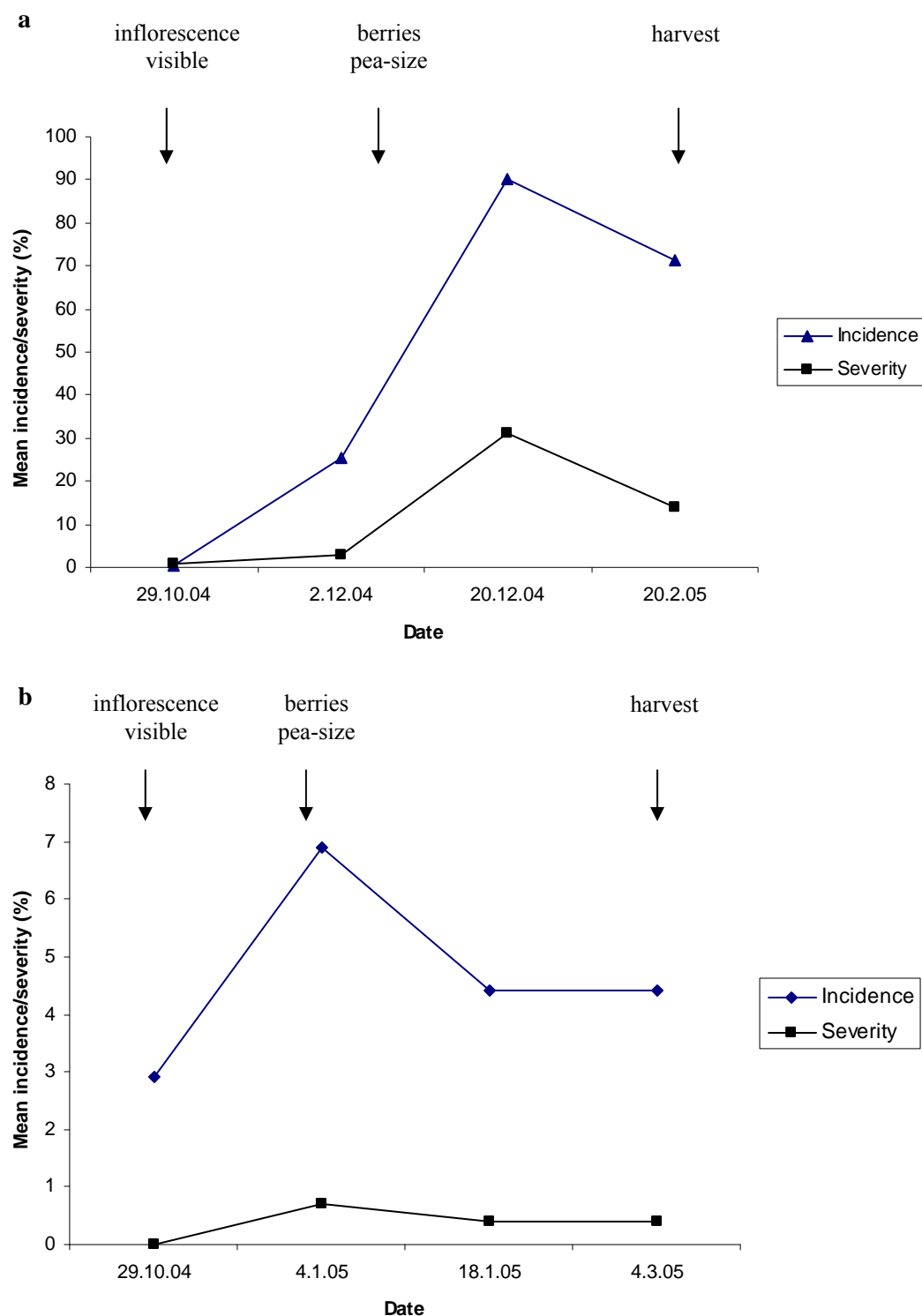
<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.

<sup>2</sup>Treatments are according to Table 10.

<sup>3</sup>ns = not significant at  $P = 0.05$

### 2004/2005 Disease development

No flag shoots were detected and powdery mildew was first observed on the leaves of Chardonnay on 29 September, 2004 at shoots 5-10 cm (E-L 9 -12) and on the leaves of Cabernet Sauvignon on 6 October, 2004 at inflorescences visible (E-L 12). Figure 29a and b shows the progression of powdery mildew for the sulfur only treatment for Chardonnay and Cabernet Sauvignon, respectively. For Chardonnay, powdery mildew increased from berries pea-size then decreased towards harvest. For Cabernet Sauvignon, powdery mildew also increased at berries pea-size, however, severity changed little towards harvest.



**Figure 29.** Percentage mean incidence and severity of powdery mildew on (a) Chardonnay and (b) Cabernet Sauvignon bunches from inflorescence visible to harvest for 2004/05.

At inflorescence visible for Chardonnay the incidence and severity of powdery mildew on leaves were not significantly different between treatments ( $P = 0.87$  and  $0.83$ , respectively) (Table 58). The mean percentage incidence on leaves ranged from 8.3% for treatment 5 (Surround<sup>TM</sup> WP) to 12.5% for treatment 3 (whey). Significant differences between treatments were recorded for the incidence and severity of powdery mildew on inflorescences

( $P = 0.005$  and  $0.0002$ , respectively). Treatments 2, 4 and 5 (milk, Ecocarb<sup>®</sup>/Synertrol Horti-oil<sup>®</sup>, Surround<sup>™</sup> WP) were not significantly different from the sulfur treatment. Disease incidence and severity increased from berries pea-sized to harvest. Despite a systemic fungicide being sprayed at bunch closure, disease incidence and severity were not significantly different between treatments (Table 59). For all treatments, the severity of powdery mildew was above commercially acceptable levels of 5%.

**Table 58.** Mean percentage incidence and severity of powdery mildew on Chardonnay leaves and bunches at inflorescences visible for 2004/05<sup>1</sup>.

Treatment <sup>2</sup>	Incidence		Severity	
	Leaf	Bunch	Leaf	Bunch
1. Sulfur (g/L)	11.7 (6.1) a	0.4 (0.9) b	1.2 (0.7) a	0.1 (0.2) b
2. Milk (dilution)	10.0 (3.0) a	0 (0) b	1.0 (1.3) a	0 (0) b
3. Whey (g/L)	12.5 (8.0) a	12.1 (5.2) a	1.3 (1.3) a	2.4 (1.0) a
4. Ecocarb <sup>®</sup> (g/L)/ Synertrol Horti-oil <sup>®</sup> (ml/L)	12.1 (7.0) a	0 (0) b	1.2 (0.7) a	0 (0) b
5. Surround <sup>™</sup> WP (g/L)	8.3 (6.7) a	0 (0) b	0.8 (0.7) a	0 (0) b
6. Program 1	11.7 (6.6) a	7.4 (6.6) a	1.1 (0.6) a	1.5 (1.4) a
<i>P</i> value	0.87	0.005	0.83	0.0002
LSD <sup>3</sup>	ns	0.65	ns	0.78

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.

<sup>2</sup>Treatments are according to Table 11.

<sup>3</sup>ns = not significant at  $P = 0.05$

The incidence and severity of powdery mildew on Cabernet Sauvignon was greater on leaves than on inflorescences. Powdery mildew was observed only on bunches treated with milk or whey. No significant differences in the incidence and severity of powdery mildew were recorded between treatments (Table 60). All treatments were as effective as the sulfur control. No powdery mildew was present on bunches at berries pea-sized. At veraison, all treatments except for treatment 6 (Program 1) provided commercially acceptable control (Table 61). The most effective control was sulfur, with a powdery mildew severity on bunches of 0.4 %. At harvest, the incidence and severity of powdery mildew remained at the same level for sulfur, however this increased for all other treatments except for treatment 5 (Surround<sup>™</sup> WP). Despite this, no significant differences between treatments were recorded at harvest ( $P = 0.293$  and  $0.408$ ).

**Table 59.** Mean percentage incidence and severity of powdery mildew on Chardonnay bunches at berries pea-sized, bunch closure and harvest for 2004/05<sup>1</sup>.

Treatment <sup>2</sup>	Berries pea-sized		Bunch closure <sup>3</sup>		Harvest	
	Incidence	Severity	Incidence	Severity	Incidence	Severity
1. Sulfur (g/L)	25.6 (12.5) c	2.9 (1.8) c	90 (10.2) b	31.1 (16.5) d	71.3 (33.6) a	14 (10.6) c
2. Milk (dilution)	48.8 (12.0) b	5.3 (1.7) c	99.4 (1.3) a	58.3 (14.4) c	98.8 (1.8) a	66.1 (5.5) b
3. Whey (g/L)	70.0 (24.0) a	11.1 (6.9) a	100 (0) a	73.9 (10.9) ab	100 (0) a	69.6 (11.3) b
4. Ecocarb <sup>®</sup> (g/L)/ Synertrol Horti-oil <sup>®</sup> (ml/L)	45.6 (8.5) b	4.8 (1.0) c	99.4 (1.3) a	60.3 (19.7) bc	100 (0) a	67.9 (18.2) b
5. Surround <sup>TM</sup> WP (g/L)	72.5 (7.4) a	10.6 (2.5) ab	100 (0) a	77.9 (7.9) a	100 (0) a	98.6 (0.5) a
6. Program 1	58.8 (7.2) ab	6.4 (1.3) bc	100 (0) a	68.3 (13.7) abc	100 (0) a	74.8 (12.9) b
<i>P</i> value	0.0003	0.007	0.02	0.00001	0.36	0.003
LSD <sup>4</sup>	16.9	4.4	6.2	13.7	ns	22.5

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.

<sup>2</sup>Treatments are according to Table 11.

<sup>3</sup>Chardonnay sprayed with Bayfidan<sup>®</sup> 250 EC due to high incidence of disease and risk of spread to other commercial blocks.

<sup>4</sup>ns = not significant at  $P = 0.05$



**Table 60.** Mean percentage incidence and severity of powdery mildew on Cabernet Sauvignon leaves and bunches at inflorescences visible for 2004/05<sup>1</sup>.

Treatment <sup>2</sup>	Incidence		Severity	
	Leaf	Bunch	Leaf	Bunch
1. Sulfur (g/L)	2.9 (3.4)	0 (0)	0.3 (0.4)	0 (0)
2. Milk (dilution)	2.1 (2.1)	0.8 (1.7)	0.2 (0.2)	0.08 (0.2)
3. Whey (g/L)	1.5 (3.0)	0.1 (0.1)	0.2 (0.4)	0.2 (0.4)
4. Ecocarb <sup>®</sup> (g/L)/ Synertrol Horti-oil <sup>®</sup> (ml/L)	4.6 (2.8)	0 (0)	0.4 (0.3)	0 (0)
5. Surround <sup>TM</sup> WP (g/L)	3.3 (1.3)	0 (0)	0.3 (0.1)	0 (0)
6. Program 1	7.1 (3.2)	0 (0)	0.7 (0.3)	0 (0)
<i>P</i> value	0.10	0.45	0.14	0.45

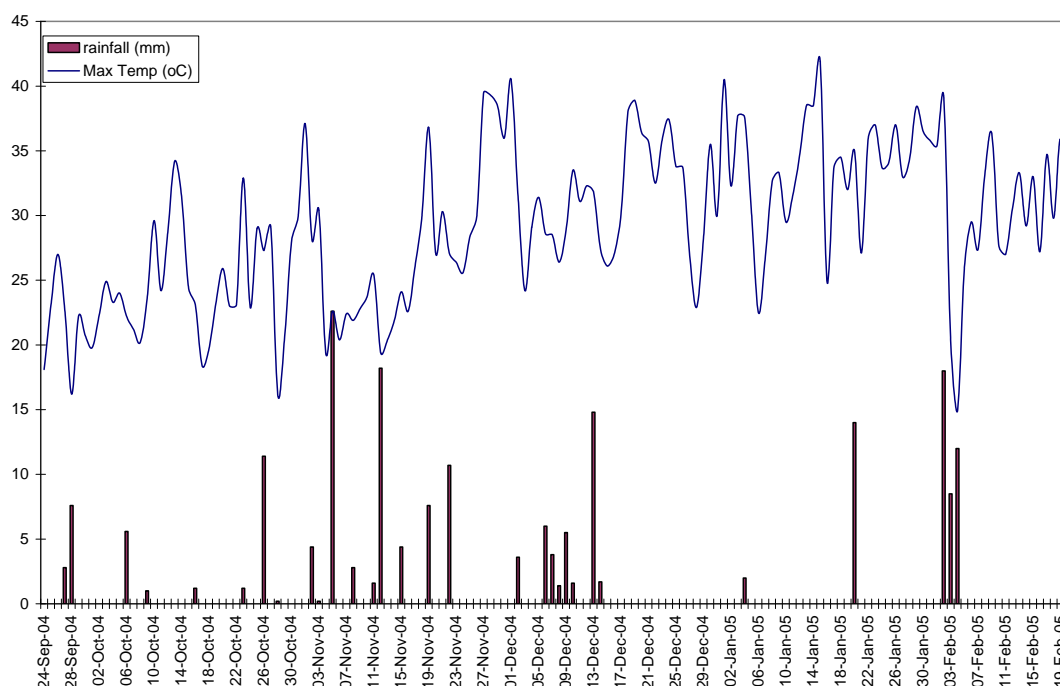
<sup>1</sup>Standard deviations are presented in parentheses.<sup>2</sup>Treatments are according to Table 11.**Table 61.** Mean percentage incidence and severity of powdery mildew on Cabernet Sauvignon bunches at veraison and harvest for 2004/05<sup>1</sup>.

Treatment <sup>2</sup>	Veraison		Harvest	
	Incidence	Severity	Incidence	Severity
1. Sulfur (g/L)	4.4 (2.4) c	0.4 (0.2) a	4.4 (5.5)	0.4 (0.6)
2. Milk (dilution)	10.6 (3.4) bc	1.2 (0.5) a	19.4 (25.7)	2.3 (3.3)
3. Whey (g/L)	11.9 (7.2) bc	1.4 (1.1) a	22.5 (19)	2.5 (2.2)
4. Ecocarb <sup>®</sup> (g/L)/ Synertrol Horti-oil <sup>®</sup> (ml/L)	12.5 (9.6) bc	1.3 (1.0) a	31.3 (35.9)	3.3 (3.8)
5. Surround <sup>TM</sup> WP (g/L)	17.5 (4.1) ab	2.4 (1.2) a	28.8 (14.5)	3.0 (1.6)
6. Program 1	29.4 (19.8) a	5.8 (5.6) b	28.1 (20.7)	2.9 (2.2)
<i>P</i> value	0.015	0.041	0.293	0.408
LSD <sup>3</sup>	12.6	3.3	ns	ns

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.<sup>2</sup>Treatments are according to Table 11.<sup>3</sup>ns = not significant at  $P = 0.05$

### Assessment of rainfall and temperature

Figure 30 illustrates the rainfall and maximum daily temperature for the 2004/05 growing season.



**Figure 30.** Rainfall (mm) and maximum daily temperatures recorded in Wagga Wagga for 2004/05.

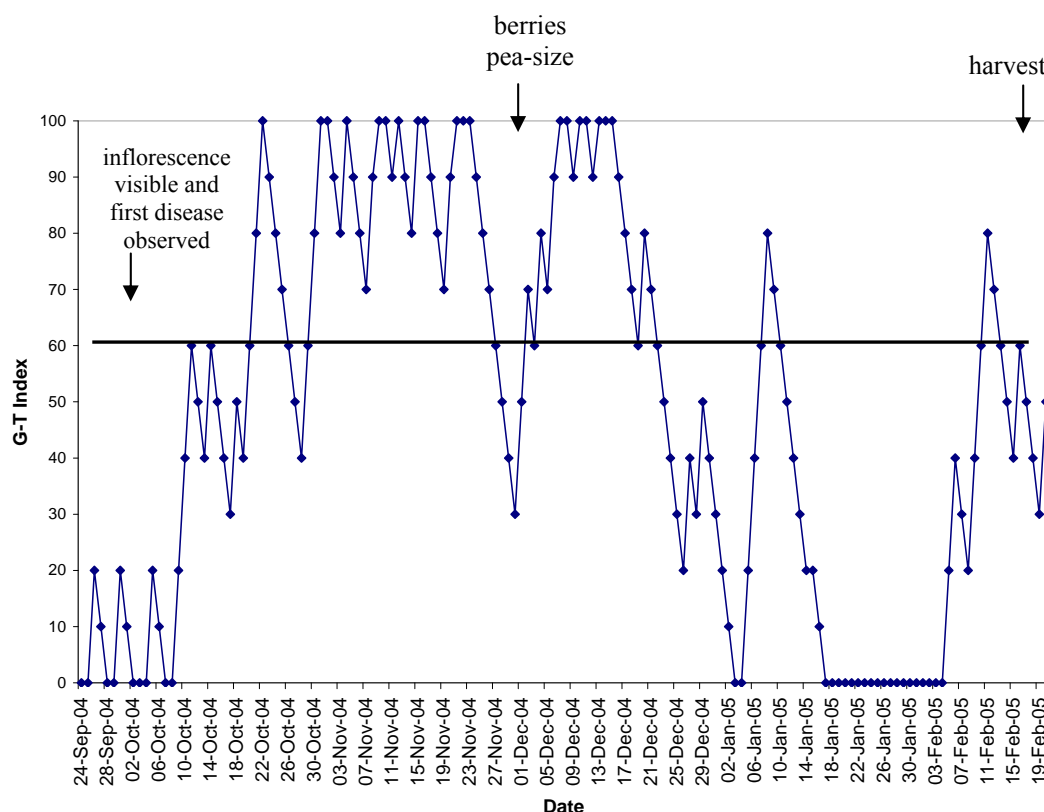
The data presented in Figure 30 were used to calculate the cumulative Gubler-Thomas risk index for powdery mildew (Figure 31). From budburst to harvest, a total of 140 days out of 149 were recorded as having a temperature above 20.5 °C. The index first reached 60 points on 11 October, 2004 and the first symptoms of powdery mildew were observed on the leaves of Chardonnay and Cabernet Sauvignon on 29 September, 2004 and 6 October, 2004, respectively. A total of 70 days above 60 points was recorded.

### Analysis of canopy density

The mean leaf area index did not vary significantly between vines (Table 62), therefore the effect of canopy density was disregarded when analysing the incidence and severity of powdery mildew.

### Berry quality assessments

No significant differences were observed between treatments for Cabernet Sauvignon in terms of berry weight, pH, TSS and TA (Table 63).



**Figure 31.** Cumulative Gubler-Thomas risk index for powdery mildew from budburst until harvest in 2004/05. The conditions for the commencement of a powdery mildew epidemic occur at a threshold of 60 points (indicated by the solid line).

**Table 62.** Mean leaf area index of Chardonnay and Cabernet Sauvignon recorded at berries pea-sized<sup>1</sup> in CSU vineyard for 2004/05.

Treatment <sup>2</sup>	Chardonnay	Cabernet Sauvignon
1. Sulfur (g/L)	4.9 (0.5)	5.2 (0.7)
2. Milk (dilution)	5.0 (0.3)	5.6 (0.6)
3. Whey (g/L)	5.0 (0.3)	5.1 (0.4)
4. Ecocarb <sup>®</sup> (g/L)/ Synertrol Horti-oil <sup>®</sup> (ml/L)	5.2 (0.3)	5.3 (0.5)
5. Surround <sup>™</sup> WP (g/L)	4.9 (0.1)	4.8 (0.3)
6. Program 1	5.0 (0.4)	5.2 (0.6)
<i>P</i> value	0.50	0.50

<sup>1</sup>Standard deviations are presented in parentheses.

<sup>2</sup>Treatments are according to Table 11.

**Table 63.** Mean pH total soluble solids (TSS) and titratable acidity (TA) for Cabernet Sauvignon at harvest<sup>1</sup> in 2004/05.

Treatment <sup>2</sup>	Cabernet Sauvignon			
	Berry Weight (g)	pH	TSS (°Brix)	TA (ml/L)
1. Sulfur (g/L)	1.1 (0.04)	4.0 (0.1)	23.5 (0.3)	3.0 (0.1)
2. Milk (dilution)	1.1 (0.1)	4.0 (0.1)	23.4 (1.1)	3.0 (0.1)
3. Whey (g/L)	1.0 (0.1)	4.0 (0.1)	23.8 (0.7)	3.0 (0.1)
4. Ecocarb <sup>®</sup> (g/L)/ Synertrol Horti-oil <sup>®</sup> (ml/L)	1.1 (0.1)	4.0 (0.05)	23.7 (0.3)	3.0 (0.2)
5. Surround <sup>™</sup> WP (g/L)	1.1 (0.1)	4.0 (0.04)	23.7 (0.4)	2.9 (0.1)
6. Program 1	1.0 (0.1)	4.0 (0.03)	23.2 (0.4)	3.0 (0.04)
<i>P</i> value	0.47	0.66	0.06	0.12

<sup>1</sup>Standard deviations are presented in parentheses.

<sup>2</sup>Treatments are according to Table 11.

#### Analysis of shoot length and leaf number

Figure 32 shows the white film deposited by the application of Surround<sup>™</sup> WP to Chardonnay. No significant differences were observed between sulfur and Surround<sup>™</sup> WP for the mean leaf number or shoot length of Chardonnay (Table 64). A decrease in the number of leaves was recorded at veraison for both treatments. For Cabernet Sauvignon, a similar trend was observed with no significant difference recorded between treatments (Table 65).



**Figure 32.** Chardonnay vines sprayed with Surround<sup>™</sup> WP. A white film layer covering the leaves was apparent on the vines.

**Table 64.** Mean leaf number and shoot length at flowering, bunch closure and veraison for Chardonnay treated with Sulfur and Surround<sup>TM</sup> WP in 2004/05.

Treatment <sup>2</sup>	Flowering		Bunch closure <sup>3</sup>		Veraison	
	Leaf no.	Shoot Length (cm)	Leaf no.	Shoot Length (cm)	Leaf no.	Shoot Length (cm)
Sulfur	12.5 (5.2)	55.8 (22.1)	24.4 (13.0)	73.5 (33.4)	16.9 (7.0)	85.4 (42.9)
Surround <sup>TM</sup> WP	12.2 (4.5)	52.5 (18.2)	24.2 (13.6)	73.3 (27.8)	18.0 (7.5)	87.2 (38.6)
<i>P</i> value	0.72	0.43	0.91	0.99	0.44	0.85

<sup>1</sup>Standard deviations are presented in parentheses.

<sup>2</sup>Treatments are according to Table 11.

<sup>3</sup>Chardonnay sprayed with Bayfidan<sup>®</sup> 250 EC due to high incidence of disease and risk of spread to other commercial blocks.

**Table 65.** Mean leaf number and shoot length at flowering, bunch closure and veraison for Cabernet Sauvignon treated with Sulfur and Surround<sup>TM</sup> WP in 2004/05.

Treatment <sup>2</sup>	Flowering		Bunch closure <sup>3</sup>		Veraison	
	Leaf no.	Shoot Length (cm)	Leaf no.	Shoot Length (cm)	Leaf no.	Shoot Length (cm)
Sulfur	12.0 (4.0)	57.4 (18.3)	17.6 (4.4)	97.1 (36.4)	16.8 (4.1)	103.6 (37.2)
Surround <sup>TM</sup> WP	12.8 (4.2)	56.1 (16.4)	17.4 (7.1)	96.0 (46.9)	17.1 (6.9)	97.3 (41.2)
<i>P</i> value	0.30	0.70	0.90	0.90	0.78	0.39

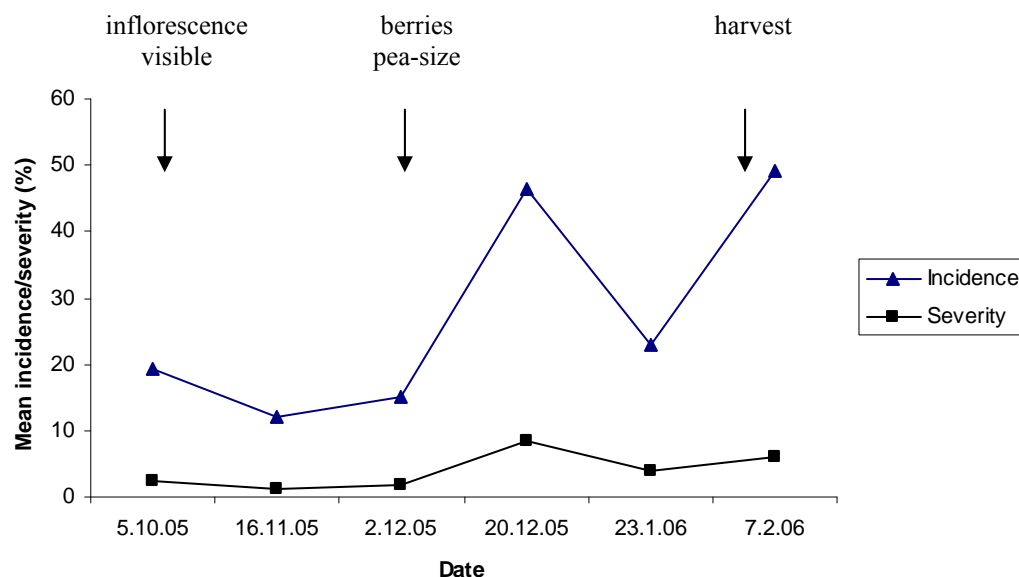
<sup>1</sup>Standard deviations are presented in parentheses.

<sup>2</sup>Treatments are according to Table 11.

<sup>3</sup>Chardonnay sprayed with Bayfidan<sup>®</sup> 250 EC due to high incidence of disease and risk of spread to other commercial blocks.

### 2005/2006 Disease development

No flag shoots were detected and powdery mildew was first observed on the leaves of Chardonnay on 5 October, 2005 at inflorescences visible (E-L 12). Figure 33 shows the progression of powdery mildew for the sulfur only treatment.



**Figure 33.** Percentage mean incidence and severity of powdery mildew on Chardonnay leaves on 5 October 2005 and on bunches from 16 November 2005 to harvest for vines treated with sulfur only.

No disease was detected on the inflorescences. At this phenological stage, the incidence and severity of powdery mildew were not significantly different ( $P = 0.31$  and  $0.44$  respectively) between the treatments. The incidence of powdery mildew ranged from 14% for treatment 5 (SH/E-M-S) and 6 (SH/E-S-M) to 19% for treatment 1 (sulfur only) (Table 66). The severity ranged from 1.8% for treatment 6 (SH/E-S-M) to 2.4 % for treatment 3 (S-M-M) (Table 66).

**Table 66.** Mean percentage incidence and severity of powdery mildew on Chardonnay leaves at inflorescences visible for 2005/06<sup>1</sup>.

Treatment <sup>2</sup>	Incidence	Severity
1. (S S S)	19.0 (4.6)	2.3 (0.8)
2. (S SH/E M)	17.2 (5.2)	2.3 (0.8)
3. (S M M)	18.8 (6.5)	2.4 (0.5)
4. (S S M)	15.2 (5.5)	2.0 (0.7)
5. (SH/E M S)	14.4 (6.7)	1.9 (0.9)
6. (SH/E S M)	14.4 (5.4)	1.8 (0.7)
<i>P</i> value	0.31	0.44

<sup>1</sup>Standard deviations are presented in parentheses.

<sup>2</sup>Treatments are according to Table 12.

At berries pea-sized, powdery mildew was observed on both leaves and bunches. A decrease in the incidence and severity of the disease was observed on leaves from inflorescences visible to berries pea-sized. However, control on bunches was unacceptable with disease

incidence ranging from 6% for treatment 4 (S-S-M) to 51.6% for treatment 5 (SH/E-M-S) (Table 67).

**Table 67.** Mean percentage incidence and severity of powdery mildew on Chardonnay leaves and bunches at berries pea-sized for 2005/06<sup>1</sup>.

Treatment <sup>2</sup>	Leaves		Bunches	
	Incidence	Severity	Incidence	Severity
1. (S <sup>1</sup> S S)	4.0 (3.2)	0.6 (0.4)	15.2 (7.2) bc	2.0 (0.9) b
2. (S SH/E <sup>2</sup> M <sup>3</sup> )	3.6 (4.1)	0.5 (0.5)	14.2 (12.3) bc	2.0 (1.4) b
3. (S M M)	2.8 (2.3)	0.4 (0.3)	17.6 (6.7) bc	2.7 (0.7) b
4. (S S M)	2.4 (0.9)	0.3 (0.1)	6.0 (4.7) c	0.9 (0.7) b
5. (SH/E M S)	4.4 (2.6)	0.6 (0.3)	51.6 (14.4) a	10.6 (6.7) a
6. (SH/E S M)	6.0 (4.7)	0.8 (0.6)	31.6 (26.2) b	11.0 (14) a
<i>P</i> value	0.63	0.63	0.0005	0.0335
LSD <sup>3</sup>	ns	ns	18.0	0.76

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.

<sup>2</sup>Treatments are according to Table 12.

<sup>3</sup>ns = not significant at  $P = 0.05$ .

The incidence and severity of powdery mildew increased on bunches from berries pea-sized to veraison. Treatments 1 (sulfur) and 4 (S-S-M) provided the best control however disease severity was still above those required by industry. At harvest, sulfur provided the most effective control, however, it did not significantly reduce the incidence of disease when compared with treatment 3 (S-M-M) and treatment 4 (S-S-M). For severity, sulfur was not significantly different from programs using sulfur from flowering to fruit-set and then combinations of SH/E and milk from flowering to fruit-set and fruit-set to veraison (Table 68).

**Table 68.** Mean percentage incidence and severity of powdery mildew on Chardonnay bunches at veraison and harvest for 2005/06<sup>1</sup>.

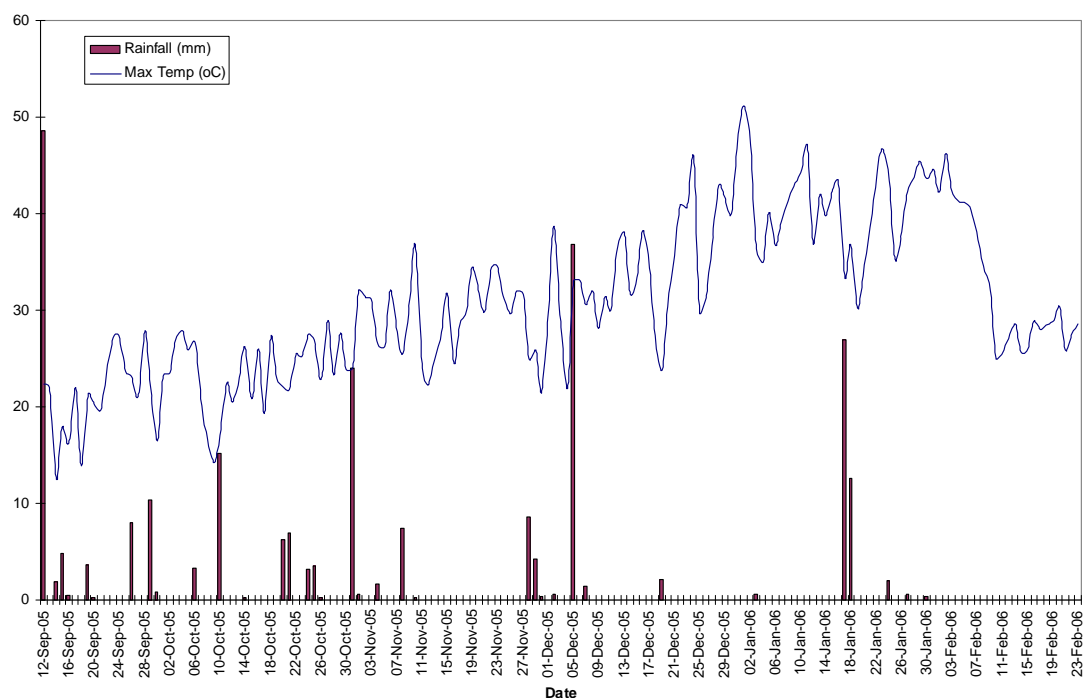
Treatment <sup>2</sup>	Veraison		Harvest	
	Incidence	Severity	Incidence	Severity
1. (S <sup>1</sup> S S)	22.8 (15.3) b	3.9 (2.8) b	49.0 (23.3) b	5.9 (3.2) b
2. (S SH/E <sup>2</sup> M <sup>3</sup> )	46.8 (23) b	6.0 (2.5) b	65.5 (21.6) ab	7.9 (2.7) b
3. (S M M)	34.4 (11.4) b	6.2 (3.0) b	51.0 (20.0) b	7.4 (3.5) b
4. (S S M)	22 (9.9) b	3.2 (1.7) b	53.0 (29.1) b	6.6 (4.1) b
5. (SH/E M S)	84 (15.1) a	18.0 (10.0) a	90.5 (8.7) a	21.7 (10.0) a
6. (SH/E S M)	73.6 (24.6) a	18.7 (14.5) a	87.0 (10.2) a	22.3 (16.9) a
<i>P</i> value	0.0001	0.0045	0.014	0.004
LSD	25.3	0.94	28.2	1.04

<sup>1</sup>Standard deviations are presented in parentheses. Means followed by different letters are significantly different ( $P = 0.05$ ) based on least significant difference.

<sup>2</sup>Treatments are according to Table 12.

### Rainfall and temperature assessments

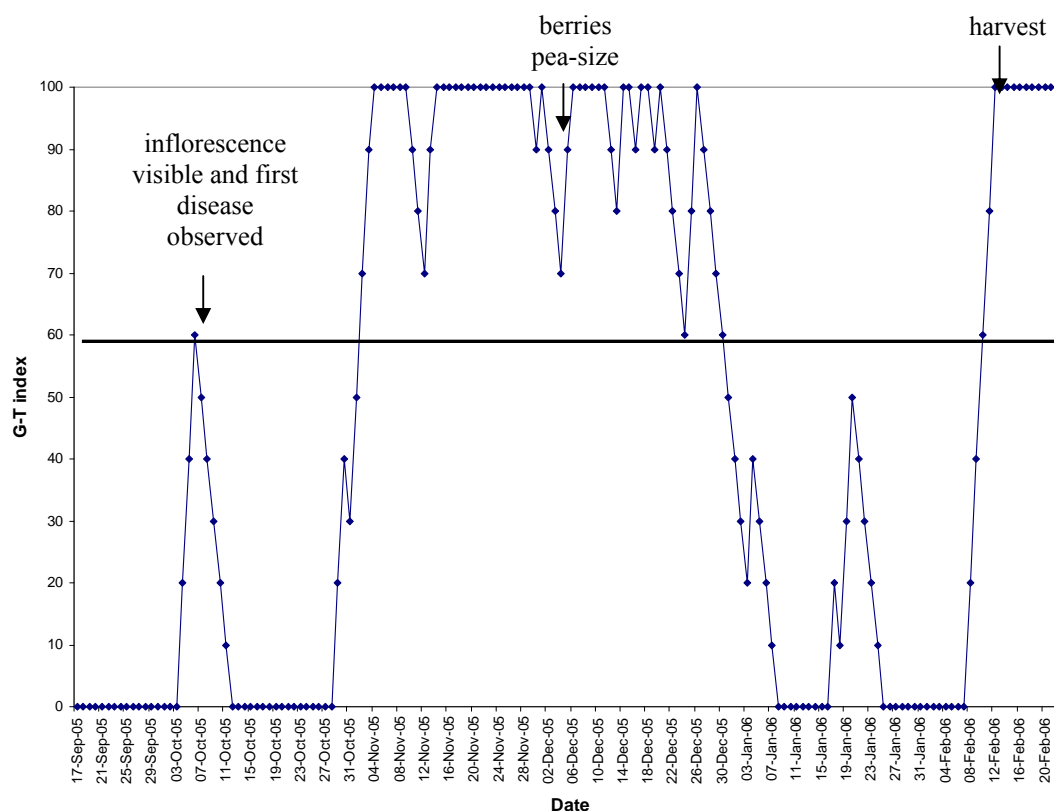
Figure 34 illustrates the rainfall and maximum daily temperature for the 2005/06 growing season. At budburst (approximately 12 December 2006) 49 mm of rain was recorded.



**Figure 34.** Rainfall (mm) and maximum daily temperatures recorded in Wagga Wagga for 2005/06.

The data presented in Figure 34 were used to calculate the cumulative Gubler-Thomas risk index for powdery mildew (Figure 35). From budburst to harvest, a total of 125 days out of 147 was recorded as having a temperature above 20.5 °C. The index first reached 60 points on 5 October, 2005 and the first symptoms of powdery mildew on leaves were also recorded on this date. A total of 74 days above 60 points was recorded.





**Figure 35.** Cumulative Gubler-Thomas risk index for powdery mildew from budburst to harvest in 2005/06. The conditions for the commencement of a powdery mildew epidemic occurs at a threshold of 60 points (indicated by the solid line).

## Discussion

Hot dry weather occurred during the growing seasons when the trial was conducted. Powdery mildew occurred in all seasons and was first observed between three leaves unfolded to “inflorescences visible” in both Chardonnay and Cabernet Sauvignon. No flag shoots were observed throughout the trial. In all three seasons, treatments were applied according to the industry standard of the first application at shoots 10 to 15 cm or from budburst and were continued until veraison. In 2003/04 and 2005/06 disease incidence and severity on Chardonnay leaves (data not shown) and bunches continued to increase after veraison despite reports that berries should become resistant to infection after this time (Ficke et al. 2002). However, a decrease in incidence and severity was observed over this period in 2004/05. For Cabernet Sauvignon, powdery mildew remained the same from veraison to harvest in each trial. For highly susceptible cultivars such as Chardonnay, the berries may remain susceptible to infection even after veraison. Sprays may need to be continued after veraison and post-harvest to minimise inoculum in the next growing season. This aspect requires further investigation.

The Gubler-Thomas index was calculated for the 2004/05 and 2005/06 growing seasons. In 2004/05, powdery mildew was first observed on the leaves on 29 September 2004, however, the Gubler-Thomas threshold of 60 points was not triggered until 12 days later on 11 October, suggesting that the model was not applicable in that season. In contrast, in 2005/06 the appearance of disease correlated well with the model and powdery mildew was observed on 5 October, 2005, the day on which the index reached 60 points. The

number of days recorded above 60 points suggests that powdery mildew was a threat to the vines in this region throughout the growing season. The use of the Gubler-Thomas index to predict outbreaks of powdery mildew for warm inland sites such as Wagga Wagga appeared promising, however, further seasons would need to be validated before this could be recommended as a disease prediction tool. Similar promising results were obtained from monitoring powdery mildew and applying the Gubler-Thomas index to vineyards in McLaren Vale, Coonawarra, Margaret River and Sunraysia (Luckhurst and Pettigrew, 2002). In the Coonawarra cost benefit of \$81/ha was identified when applying the model to inform spraying. However, the above mentioned study was conducted over one season only, therefore, further data are required over a number of seasons to ascertain if this is a viable option for predicting and informing spray schedules for control of powdery mildew.

In 2003/04, control of powdery mildew on Chardonnay generally was not commercially acceptable. A high incidence of disease was recorded on leaves at inflorescences visible and this continued onto bunches. Sulfur provided the best control, with an incidence of 34.2% and severity of 4.2%. There were no significant differences between the remaining treatments for incidence, however, milk and whey reduced severity of disease on bunches. In contrast, acceptable control was provided by all treatments on Cabernet Sauvignon even though a high incidence of disease was recorded on leaves at “inflorescences visible”. At harvest, disease incidence of 1.3% and 0.6% was recorded on bunches for the untreated control and the Ecocarb<sup>®</sup> treatment, respectively. According to the distributors, Ecocarb<sup>®</sup> should be mixed with Synertrol Horti-oil<sup>®</sup> for maximum effectiveness (J. Gardner, pers. com.), an approach that was adopted for the 2004/05 and 2005/06 trials.

To reduce the overall incidence of powdery mildew in the vineyard, the untreated control treatment was omitted from the 2004/05 and 2005/06 trials. Sulfur was used as the standard control in both seasons. The 2004/05 season was particularly conducive to powdery mildew and incidence reached 100% for Chardonnay with all treatments but sulfur (71.3%) and milk (98.8%). To minimize spread of inoculum to neighbouring grapes used for commercial winemaking, the Chardonnay trial was sprayed with a systemic fungicide on 21 December 2004. At harvest, there were no significant differences between treatments in terms of the incidence of powdery mildew. However, severity was less for the sulfur treatment (14%). For all other treatments, the severity of powdery mildew ranged between 66% and 99%. Surround<sup>™</sup> WP was least effective at controlling powdery mildew, however, it was also difficult to monitor infection due to the thin white layer formed on leaves and berries. Surround<sup>™</sup> WP had no obvious effect on the growth of the vine in terms of shoot length and leaf number for Chardonnay and Cabernet Sauvignon. As in 2003/04, acceptable control was achieved for Cabernet Sauvignon, with no significant differences between treatments for incidence and severity. Due to the unacceptable control of powdery mildew on Chardonnay from 2003 to 2005, it was decided to incorporate the test materials into programs rather than spraying one material continuously. The aim was to reduce the overall number of sulfur applications during the 2005/06 season. Sulfur was applied at 6 g/L rather than 3 g/L and sprays were targeted at the most mildew-susceptible stages of vine growth. At harvest, the sulfur-only treatment produced the best results, however, it did not significantly reduce the incidence and severity of powdery mildew when compared with treatment 2 (sulfur, Synertrol Horti-oil<sup>®</sup>/Ecocarb<sup>®</sup>, milk), treatment 3 (sulfur, milk, milk) and treatment 4 (sulfur, sulfur, milk). The severity of powdery mildew ranged from 5.9% for the sulfur-only treatment to 7.9% for the sulfur, Synertrol Horti-oil<sup>®</sup>/Ecocarb<sup>®</sup>, milk treatment both of which were above commercially acceptable level for the disease.

Analysis of juice from Cabernet Sauvignon and Chardonnay from 2003 to 2005 revealed no significant differences between berry weight, pH, TSS and TA for all treatments.

In all three seasons, wind speed was a major limitation in entering the vineyard for spraying. At the time of spraying wind speed often exceeded the 8.1 knots allowable for spray application. Therefore, the high incidence of powdery mildew in the vineyard may have been due to the large window between sprays from budburst to flowering. For example, in 2003/04 there were 34 days between the first and second spray. In 2005/06 there were two periods of 17 and 18 days when sprays could not be applied. For future experiments, consideration should be given to spraying during the night when wind speeds appear to ease.

## 7. Outcomes and conclusions

The application of milk, whey and Ecocarb® plus Synertrol Horti-Oil® controlled powdery mildew to levels not significantly different from those provided by sulfur on Shiraz and Verdelho in South Australia and on Cabernet Sauvignon in NSW. These materials did not provide adequate control of powdery mildew on Pinot noir in Tasmania, although they did reduce disease severity compared with untreated controls. They have generally failed to control powdery mildew on Chardonnay, and further development is required. Tea tree oil products assessed in 2004/05 showed promise in control of both powdery and downy mildew, however, further evaluation trials in 2005/2006 were inconclusive due to lack of powdery mildew and widespread development of downy mildew prior to receipt of products.

In general, the novel treatments were effective on less susceptible cultivars and when disease pressure was moderate to low. In all three states, coverage was a critical factor in the control of powdery mildew achieved when using the novel treatments, which act as contact fungicides. As such, the novel treatments appear to be best suited to open, sparse canopies, which facilitate good coverage and are not conducive to powdery mildew.

In South Australia, where most trials were conducted on a vineyard managed organically for 12 years, powdery mildew developed to a significant extent only in 2003/04, in spite of conducive environmental conditions in 2004/06. The reasons for this are not clear. In the warm, dry environment of the Charles Sturt University vineyard in NSW, powdery mildew control proved challenging, largely due to long intervals between spray applications and even the standard sulfur treatment failed to provide adequate control on occasions. The trials in southern Tasmania provided an opportunity to study disease progression and management in this cold climate region where flowering occurs over an extended period and harvest may extend to May-June. A 'best bet' spray program for management of powdery mildew in Pinot noir grown organically in southern Tasmania was developed. A 'whole-of-block' experiment demonstrated that a spray program beginning at E-L stage 16 and based on a mixture of Synertrol Horti-oil® and Ecocarb® with two applications of sulfur plus Horti-oil® during fruit set resulted in a mean maximum disease severity at veraison of 1.5%. By harvest, the grower cooperator judged the powdery mildew control to be commercially acceptable. The spray programs evaluated in Tasmania did not appear to prevent colonization of buds by *E. necator* and, hence, subsequent development of flag shoots.

The Gubler-Thomas powdery mildew risk index allowed prediction of onset of powdery mildew in NSW in 2005/06, but was not informative in South Australia or Tasmania. The index was less reliable than regular monitoring and growers would have incurred increased costs and potential crop loss had they relied on the index alone.

The test materials suppressed powdery mildew on leaves but their lack of persistence would make them less useful than sulfur for late season protection of leaves in cool climate regions where the last application of fungicide for powdery mildew is usually at veraison, before nets are applied (in Tasmania).

Populations of bacteria, filamentous fungi and yeasts were generally larger on leaves and berries from Verdelho and Shiraz vines sprayed with milk and whey than with sulfur or untreated, although there was considerable variation among replicate plots. Similar trends were evident for the corresponding juices. However, there were no obvious differences among treatments in terms of the diversity of microbial species detected. It is likely that the nutrients provided by milk and whey support growth of the vine surface microbiota, and this may play a role in reducing colonisation by *E. necator* and *B. cinerea*. These

changes in berry surface microbial population had no obvious effect on juice or wine quality. At harvest in 2004, 2005 and 2006 there was no effect of treatment with milk, whey or oil plus bicarbonate on pH, Brix and TA of Verdelho and Shiraz juices. Duo-trio testing of juices revealed differences in juices from Verdelho vines treated with sulfur and whey, sulfur and milk, and sulfur and Ecocarb® plus Synertrrol Horti-Oil®. Informal sensory evaluation showed differences among Verdelho juice samples, however these were attributed to differences in grape flavour characters or pressing variation. There were no detectable differences in wines made from grapes from Verdelho or Shiraz vines sprayed with milk, whey, Ecocarb® plus Synertrrol Horti-Oil® or sulfur. Likewise, the novel treatments had no detectable effect on pH, Brix or TA of Chardonnay and Cabernet Sauvignon grapes in New South Wales.

All materials evaluated for prevention of downy mildew on inoculated vines reduced disease incidence and severity compared with untreated, inoculated controls. Ferrous formulations, Timor® and Timorex® (tea tree oil-based products), Brotomax® and Ecocarb® plus Synertrrol Horti-Oil®, performed as well as the standard copper fungicide treatment. Application of ferrous formulations resulted in moderate phytotoxicity in 2004/2005; these have since have been re-formulated to reduce phytotoxic effects.

PhD student Carol Walker has not yet completed her thesis. Her progress was delayed for several reasons and she took leave of absence from July to December 2007 to take up a position in Tasmania. The aim is to submit the thesis in 2008.

## 8. Recommendations

For growers or vineyard managers considering using milk, whey, Ecocarb plus Synertril Horti-Oil or the mixed programs in commercial vineyards, it is suggested that a small trial be established first to ensure that the treatments are suited to the cultivars and canopy architecture on their vineyard. Furthermore, in the case of highly susceptible cultivars or high disease pressure other products, such as sulfur or conventional fungicides, should be included from bud burst until berries reach pea-size. If the fruit is contracted to an external winemaker the grower first should confirm that the winemaker will accept the fruit.

Further research is needed to modify the Gubler-Thomas powdery mildew index for use in Australia, particularly with respect to initiation of spray programs and inoculum early in the season, or to develop an alternative model. The index alone should not be relied upon to initiate and maintain spray programs, particularly in South Australia and Tasmania.

Any measure that reduces powdery mildew pressure in the vineyard will facilitate disease control using contact fungicides such as milk, whey or bicarbonate plus oil. These measures include removing flag shoots as soon as they are detected and managing canopies to reduce vigour and density.

Effective evaluation of any novel approach to managing powdery mildew depends on a sound understanding of disease epidemiology. The timing of materials evaluated in this study could be optimised further with improved knowledge of the development flag shoots, the timing of release of ascospores from cleistothecia and the relative importance of ascospores and conidia as sources of primary inoculum. Nevertheless, removal of flag shoots as soon as they are detected may assist in reducing disease pressure in the vineyard. Further research on the relative contribution of flag shoots and cleistothecia to powdery mildew outbreaks in Australian grape growing areas will allow refinement of modelling and management strategies, given that ascospore release requires rain, whereas conidia are released independent of rainfall events. Models that integrate the biology of the pathogen (inoculum), plant host (e.g. organ development, cultivar) and environmental conditions (e.g. weather) will facilitate prediction of disease severity and deployment of novel control methods among sites and seasons.

Further research is required to identify and develop the active components in milk into a reliable commercial product suited to use in viticulture and other horticultural crops.

## Appendix 1. Communication

### Conference presentations

- Bramley, R.G.V., Evans, K.J., Gobbett, D.I., Panten, K. and Scott, E.S. (2007) Optimising strategies for control of powdery mildew through whole of block experimentation. Poster presented at the 13<sup>th</sup> Australian Wine Industry Technical Conference, 28 July - 2 August, Adelaide, South Australia.
- Bramley, R.G.V., Evans, K.J., Gobbett, D.L., Panten, K., Scott, E.S. (2007) Optimising strategies for control of grapevine powdery mildew through whole of block experimentation. Proceedings of the 16th Biennial Australasian Plant Pathology Society Conference, Adelaide, South Australia, p 149.
- Crisp, P., Evans, K.J., Savocchia, S., Grbin, P.R., Wicks, T.J. and Scott, E.S. (2007) Reducing sulfur and synthetic fungicides in Australian vineyards. Poster presented at the 13<sup>th</sup> Australian Wine Industry Technical Conference, 28 July - 2 August, Adelaide, South Australia.
- Tan, M., Grbin, P.R. and Scott, E.S. (2007) The use of milk and whey in viticulture: impact on wine quality. Poster presented at the 13<sup>th</sup> Australian Wine Industry Technical Conference, 28 July - 2 August, Adelaide, South Australia.
- Crisp, P., Scott, E.S. and Wicks, T.J. (2006) Evaluation of novel controls of grapevine downy mildew, *Plasmopara viticola*. In Proceedings of the 5<sup>th</sup> International Workshop on Grapevine Downy and Powdery Mildew, Pertot, I., Gessler, C. Gadoury, D.M., Gubler, W.D., Kassemeyer, H-H. and Magarey, P.A. (eds), SafeCrop, Istituto Agrario di San Michele all'Adige, Trentino, Italy, pp 195-196.
- Crisp, P., Scott, E.S., Wicks, T.J. and Grbin, P.R. (2006) Novel control of grapevine powdery mildew on a commercial vineyard in South Australia: effects on disease and quality. In Proceedings of the 5<sup>th</sup> International Workshop on Grapevine Downy and Powdery Mildew, Pertot, I., Gessler, C. Gadoury, D.M., Gubler, W.D., Kassemeyer, H-H. and Magarey, P.A. (eds), SafeCrop, Istituto Agrario di San Michele all'Adige, Trentino, Italy, pp 185-186.
- Evans, K.J., Crisp, P. and Scott, E.S. (2006) Applying spatial information in a whole-of-block experiment to evaluate spray programs for powdery mildew in organic vineyards. In Proceedings of the 5<sup>th</sup> International Workshop on Grapevine Downy and Powdery Mildew, Pertot, I., Gessler, C. Gadoury, D.M., Gubler, W.D., Kassemeyer, H-H. and Magarey, P.A. (eds), SafeCrop, Istituto Agrario di San Michele all'Adige, Trentino, Italy, pp 169-171.
- Savocchia, S., Mandel, R., Crisp, P. and Scott, E.S. (2006). Organic control of grapevine powdery mildew in eastern Australia. In Proceedings of the 5<sup>th</sup> International Workshop on Grapevine Downy and Powdery Mildew, Pertot, I., Gessler, C. Gadoury, D.M., Gubler, W.D., Kassemeyer, H-H. and Magarey, P.A. (eds), SafeCrop, Istituto Agrario di San Michele all'Adige, Trentino, Italy, pp 191-192.
- Crisp, P., Evans, K., Savocchia S., Mandel, R., Wicks, T., and Scott, E. (2005). Novel powdery mildew control in Australian vineyards. 15th International Federation Of Agriculture Movements Organic World Congress, 20-23 September. Adelaide, South Australia.
- Crisp, P., Scott, E. and Wicks, T. (2003) Mode of action of potential novel controls of grapevine powdery mildew, *Uncinula necator*. 2<sup>nd</sup> National Organic Conference, Adelaide, South Australia, 2-3 October, pp 133-136.
- Crisp, P., Scott, E., Wicks, T. and Palmer, L. (2004) Sustainable control of grapevine powdery mildew (*Uncinula necator*). 1<sup>st</sup> International Symposium for Organic Wine Growing, Stuttgart, Germany, pp 47-52.
- Crisp, P., Evans, K., Savocchia S., Wicks, T.J. and Scott, E.S. (2005) Novel powdery mildew control in Australian vineyards. 8<sup>th</sup> International Federation Of Agriculture Movements Organic Viticulture and Wine conference. 20-23 September, Adelaide Convention Centre, Adelaide, Australia, pp 2-3.

- Crisp, P., Scott, E.S., and Wicks, T.J. (2005) Evaluation of novel controls of grapevine downy mildew, *Plasmopara viticola*. 15<sup>th</sup> Biennial Australasian Plant Pathology Society Conference, 26-29 September, Deakin University, Geelong, Australia, pp174
- Crisp, P., Scott, E.S. and Wicks, T.J. (2005) Evaluation of biological and novel controls of grapevine powdery mildew. Proceedings of the 15<sup>th</sup> Biennial Australasian Plant Pathology Society Conference, 26-29 September, Deakin University, Geelong, Australia p 92.
- Evans, K.J., Crisp, P. and Scott, E.S. (2005) Evaluation to alternatives to sulfur for managing grapevine powdery mildew in Tasmania. Proceedings of the 15<sup>th</sup> Biennial Australasian Plant Pathology Society Conference, 26-29 September, Deakin University, Geelong, Australia p 173.
- Walker, C., Grbin, P., Stephen, J., Crisp, P., Wicks, T. and Scott, E. (2005) Microbial population growth responses to novel powdery mildew controls in an organic vineyard. 8<sup>th</sup> International Federation Of Agriculture Movements Organic Viticulture and Wine Conference, Adelaide, Australia, 20-23 September, CD Proceedings.
- Crisp, P. and Scott, E.S. (2006) Poster display, University of Adelaide, School of Agriculture Food and Wine research day, November.
- Crisp, P. and Scott, E.S. (2006) Poster display, Waite Festival, November, staffed display, poster and handouts (approximately 75 distributed).

### **Presentations to industry**

- Crisp, P. (2006) Fosters Wines, Viticulturists and Field Managers Information Day, Adelaide, 21 August.
- Savocchia, S., Mandel, R., Crisp, P. and Scott, E.S. (2006). Organic control of grapevine powdery mildew in eastern Australia. National Wine and Grape Industry Centre Industry Symposium, Wagga Wagga, NSW, 22 June.
- Crisp, P (2005) McLaren Vale and Fleurieu Peninsula Winegrowers information day, McLaren Vale, December.
- Savocchia, S. and Mandel, R. (2004). Organic control of grapevine powdery mildew in NSW: 03/04 trials. National Wine and Grape Industry Centre Industry Symposium, Wagga Wagga, NSW, 24 June.
- Crisp, P. (2004) Temple Bruer Wines field day, Langhorne Creek, SA, November
- Evans, K.J., Crisp, P. and Scott, E.S. (2004) Tthe State Pinot Group with Organic Coalition of Tasmania and DPIWE Field Day, Coal River Valley, Tasmania, November

### **Technical reports and industry publications**

- Crisp, P., Scott, E., Savocchia, S. and Evans, K. (2006) Managing powdery mildew in organic vineyards. In: Grapevine Management Guide 2006-2007, Quirk, L. and Somers, T. (eds). p.71.
- Savocchia, S. (2006). Overview of research: Organic control of powdery mildew. In: Grapevine Management Guide 2006-2007, Quirk, L. and Somers, T. (eds). p.15.
- Savocchia, S. (2004). Overview of Research, Organic control of powdery mildew. In: Grapevine Management Guide 2004-2005, Quirk, L. and Somers, T. (eds). p. 16.
- Crisp, P., Scott, E. and Wicks, T. (2003) Sulfur-free control of powdery mildew in organic viticulture: successes, strategies and suggestions. Australian and New Zealand Grapegrower and Winemaker **473a**, 123-124.



## **Appendix 2. Intellectual Property**

The research has resulted in recommendations for management of powdery mildew with reduced sulfur applications. These have been made available to industry (see Appendix 1).

Background IP relating to this matter resides with the University of Adelaide, the University of Tasmania and Tasmanian Institute of Agricultural Research, the National Wine and Grape Industry Centre, Charles Sturt University, the South Australian Research and Development Institute, Organic Crop Protectants Pty Ltd, Temple Bruer Wines and the grower cooperator of Frogmore Creek vineyard.

Any matters likely to have bearing on IP would be discussed with stakeholders, as appropriate, prior to any public disclosure of the information.

### Appendix 3. References

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## **Appendix 4. Staff**

*The University of Adelaide, School of Agriculture, Food and Wine, PMB 1, Glen Osmond, South Australia 5064, Australia*

Associate Professor Eileen Scott  
Dr Peter Crisp  
Ms Carol Walker  
Dr Paul Grbin

*Australian Genome Research Facility Ltd, Plant Genomics Centre, University of Adelaide, PMB1 Glen Osmond, South Australia 5064*

Dr John Stephen

*University of Tasmania and Tasmanian Institute of Agricultural Research, 13 St John's Avenue, New Town, Tasmania 7008, Australia*

Dr Katherine Evans

*National Wine and Grape Industry Centre, Charles Sturt University, Locked Bag 588, Wagga Wagga, New South Wales 2650, Australia*

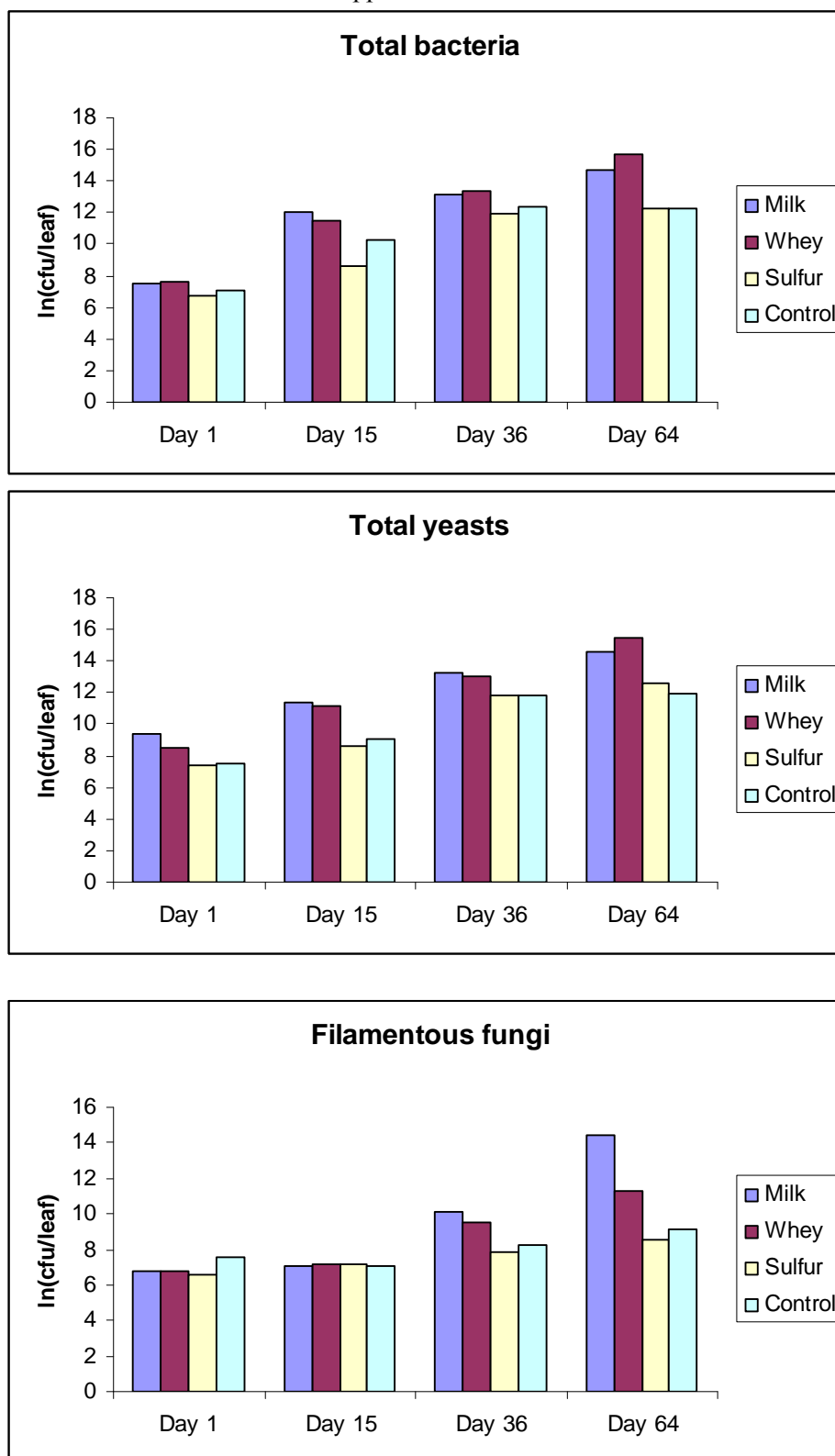
Dr Sandra Savocchia and Dr Roger Mandel

*South Australian Research and Development Institute, GPO Box 397, Adelaide, South Australia 5001, Australia*

Dr Trevor Wicks

## Appendix 5. Microbial population data

**Figure 1.** Summary of mean microbial populations (colony forming units per leaf) on leaves of Verdelho and Shiraz vines (data combined) treated with milk, whey or sulfur and untreated control in 2003. Day 1 was 13 October 2003. Due to variation among plots, ANOVA and error bars are not applicable.



**Appendix 6. Budget reconciliation**