Autumn/Winter Spray Protection from Psa
PROJECT CODE Zespri 18
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Confidential Report Prepared For Zespri International Limited

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1.0 Executive Summary

Zespri engaged HortEvaluation Ltd to undertake a trial to identify best practice agrichemical use for the autumn/winter period to protect kiwifruit vines from Psa infection and/or re-infection. In addition the trial explored whether forcing leaf fall by the application of copper sulphate, to shorten the leaf fall period, reduced the risk of infection during leaf fall compared with natural leaf drop.

The trial was carried out on a site near Te Puke on a commercial producing Gold 3 orchard which had Psa present. The trial was carried out as intended. Vines received one of six spray programmes. Treatments one to five were designed to test for the effects of different components within each of these programmes, with treatment one being essentially a standard programme. Treatment six was the minimally sprayed control.

Treatments were replicated eighteen times and were applied with a commercial tractor driven airblast sprayer where many plots were sprayed with the same treatment at the same time, or a motorized knapsack sprayer where a treatment was to be applied to some rather than all plots. Applications were made at defined growth and management stages.

Vines were assessed for Psa symptoms in early October and mid November 2013 for leaf spot and secondary symptoms such as cane dieback, cane and leader cankers prior to the orchard manager removing Psa symptoms evident at each time.

Return bloom was monitored in spring. Assessments were carried out by AgFirst, doing components of yield bud, shoot and flower counts for ten canes on each vine.

Leaf samples were collected from treatments one and three vines, to allow for analysis of the effects of Actigard (treatment one) versus no Actigard (treatment three) on vine metabolite profiles.

Weather data recorded at both Plant and Food Research Ltd, Te Puke site and DMS, Golf Course, were reviewed in conjunction with the historical KVH Psa infection risk prediction model for July and August 2013.

Psa symptoms were generally low level. Leaf spot was barely seen, which is not unusual for Gold 3. Cankers were observed to a greater extent in leaders and to a lesser extent in canes.

There were insufficient symptoms observed in the first spring assessment to separate any treatment effects.

In the second spring assessment, for the total amount of total dieback (shoots plus canes), per vine, treatment six (control) had significantly more dieback than all other treatments, apart from treatment four.

Treatment one using Actigard plus copper once post harvest, copper at late leaf fall and copper immediately after winter pruning demonstrated a trend for the least number of Psa symptoms, although not significant.

Results indicate that the immediate post pruning period was an important period for protection, which both treatment one and three covered with copper. A second post pruning copper application was made five weeks not three weeks after the post pruning treatments were applied.

There were no significant effects on yield as reflected by an analysis of components of yield.

The trial shows the challenge of relying on the real distribution of Psa on the vines we used because of the unknown and difficult to quantify nature of inoculum distribution.

2.0 Introduction

Autumn is considered to be a high risk period for Psa infection, with wetter environmental conditions and the presence of multiple wounds including exposed fruit stalks post harvest and leaf scars post leaf fall.

There are important questions about protection through this period, including;

- is the protection of fruit stalks important
- is forced or natural leaf drop a higher risk
- what is the best protection for leaf scars and winter pruning wounds

The trial outlined here aims to investigate these questions with an on-orchard trial comparing different autumn through to winter spray regimes.

3.0 Objective

This study has two aims.

Firstly, to identify best practice agrichemical use for the autumn/winter period to protect kiwifruit vines from Psa infection and /or re-infection. During this period multiple infection risks are present, including fruit stalks, leaf scars and pruning wounds, and environmental conditions are considered favourable to infection.

Secondly, to determine whether forcing leaf fall by the application of copper sulphate, to shorten the leaf fall period, reduces the risk of infection during leaf fall compared with natural leaf drop.

4.0 Materials and Methods

Treatments
Vines received one of six spray programmes.

Treatments one to five were designed to test for the effects of different components within each of these programmes.

Treatment six was the minimally sprayed control.

Treated vines received only the treatments outlined below, including the grower copper, from the start of the trial until three weeks after the post-pruning treatment.

Table 1: Treatments

Treatment	Post-harvest	Leaf fall	Late leaf fall	Post-prune	Post-prune + 3 weeks
Date of application	**6 June 2013**	**24 June 2013**	**1 July 2013**	**26 July 2013**	**30 August 2013**
1	Actigard + Nordox		Grower Cu	Nordox	Grower Cu
2	Actigard + Nordox	$CuSo_4$	Grower Cu	Nordox	Grower Cu
3	No spray		Grower Cu	Nordox	Grower Cu
4	Actigard + Nordox		Grower Cu	KeyStrepto + Engulf	Grower Cu
5	Actigard + Nordox		Grower Cu + KeyStrepto + Engulf	KeyStrepto + Engulf	Grower Cu
6	No spray		Grower Cu	No spray	Grower Cu

Treatment one was established as the standard programme using a copper protectant post harvest, plus an elicitor, followed by a copper protectant at late leaf fall, followed by a copper protectant immediately after pruning and a follow up copper protectant in late winter.

Treatment two was the same as treatment one except for the inclusion of copper sulphate, to force leaf fall rather than allowing leaves to drop naturally.

Treatment three removed post harvest sprays until late leaf fall.

Treatment four substituted Key Strepto and Engulf immediately after winter pruning, instead of copper. This treatment showed some potential for reduced Psa symptoms in a previous trial.

Treatment five substituted Key Strepto and Engulf at late leaf fall and immediately after winter pruning, instead of copper.

For both treatment four and five, ACVM permission was obtained to apply Key Strepto with Engulf. Part of the ACVM permission required collection of fruit for residue testing in 2014. However, this requirement was obviated by grower use of Key Strepto as part of their spring programme, after the trial had been completed.

Engulf is a super penetrant surfactant, designed to be used to promote agrichemicals into difficult to penetrate situations. Engulf was selected in this trial as the best option to enhance penetration of Key Strepto into wounds such as leaf scars and pruning wounds.

Treatment six received no Psa protectant sprays during the period from post-harvest to post-winter pruning, apart from the grower’s own copper sprays at late-leaf fall and post-pruning.

Site
Trials were carried out in a Gold 3 orchard in the Te Puke area of the Bay of Plenty region. The block selected for the trial had vines showing Psa symptoms but the vines used in the study did not show Psa symptoms at trial commencement.

Table 2: Site Information

Location	Maketu
Site Details	Block C Post-harvest service supplier is DMS
Plants	Conventional Gold 3 Grafted 2010 Full canopy Pergola trained
Spacing	Bays are 5.0m between rows and 6.0m between posts, double planted Plots are individual vines two bays wide
Water Rate	Post-harvest Copper at 600 litres/ha, All other treatments at 1000 litres/ha
Treatments	Refer Table 2
Equipment	New Holland TN75VA tractor & Fantini Eco 2000 sprayer for other products refer **Appendix 1** Solo Motorized Knapsack sprayer for Key Strepto + Engulf

Layout
Each treatment was replicated eighteen times. As treatments were mainly applied with a commercial tractor driven airblast sprayer, a large number of guard plants were required to allow for overspray, without drift onto adjacent treated vines. A blocking design was selected to reduce the number of guard vines required.

Refer **Appendix 2** Gold 3 Trial Layout.

Applications
Harvest was completed on 28 April 2013.

Winter pruning was undertaken on 20 to 22 July 2013.

Treatments were applied by HortEvaluation Ltd and Primo Grow Ltd spray contractors.

Applications were made at targeted times with treatment dates as summarised below.

All applications were made in suitable conditions as dilute sprays at the following rates.

Table 3: Product Application Rates

Product	Rate per 100L	Water Volume (l/ha)
Nordox post harvest & pre prune	70g	1000
Actigard	20g	1000
Copper sulphate	600g	1000
Key Strepto	60g	500
Engulf	150ml	500
Nordox post prune	70g	600

Figure 1: Canopy Stages 2013 from top right clockwise; 4 June before first application; 19 June before Copper Sulphate application, 24 June late leaf fall, 27 July after winter pruning

5.0 Assessments

5.1 Psa

Vines were assessed twice in spring for symptoms of Psa, recording any leaf spot and secondary symptoms such as cane dieback, cane and leader cankers.

First assessment was undertaken on 4 October 2013, followed by a second assessment about five weeks later on 14 November 2013. Each assessment was undertaken prior to the removal of Psa symptoms at that time.

5.2 Growth

Return bloom was monitored in spring. Assessments were carried out by AgFirst, doing components of yield bud, shoot and flower counts for ten canes on each vine.

5.3 Metabolites

Samples of leaves were taken from each of four treatment one vines and four treatment three vines, to allow for later analysis of the effects of Actigard (treatment 1) versus no Actigard (treatment three) on vine metabolite profiles.

5.4 Weather Data

Weather data recorded at both Plant and Food Research Ltd, Te Puke site and DMS, Golf Course, were reviewed in conjunction with the historical KVH Psa infection risk prediction model.

5.5 Data Analysis

All plot data were analysed as a split plot analysis i.e. plots within big plots, with a plot being a single vine and a big plot being where a treatment was applied to six vines in the same area.

For canker and dieback data, plots and big plots were of similar variability. Data was analysed for both big plots and individual vine plots, but there were no differences to the results.

For components of yield data, the six vine plots within a big plot were much less variable than the big plots were, so could not be treated as independent.

There were sufficient big plots in the trial to get a powerful test and increasing the number of plots made little difference.

Results are presented for both canker and dieback data and components of yield data big plot analyses of variance.

6.0 Results

6.1 Psa

Leaf spot was barely seen, which is not unusual for Gold 3.

Cankers were observed to a greater extent in leaders and to a lesser extent in canes. Total cankers are the sum of leader and cane cankers.

In the first spring assessment in early October, dieback was not observed in current season shoots. Dieback was observed in canes only at this time.

In the second spring assessment in mid – November, dieback was observed in both canes and current season shoots. Where shoots were dying on canes which were affected by dieback, only the cane dieback was counted. Shoot dieback was counted where only the current season shoot was affected.

Total dieback is the sum of dieback shoots and dieback canes.

There were insufficient symptoms observed in the first spring assessment to separate any treatment effects.

In the second spring assessment, for the total amount of total dieback (shoots plus canes), per vine, treatment six had significantly more dieback than all other treatments, apart from treatment four, and treatment four had significantly more dieback than treatment one.

The results of the analysis are presented in Table 4 below.

Table 4: Psa Disease Means

G3	Total Cankers Spring 1	Dieback Canes Spring 1	Leader Cankers Spring 2	Dieback Canes Spring 2	Total Dieback Spring 2*
residual d.f.	10	10	10	10	10
Treatment 1	0.24	0.18	0.36	0.18	0.30 a
Treatment 2	0.33	0.44	0.72	0.50	0.89 a
Treatment 3	0.50	0.33	0.72	0.56	0.67 a
Treatment 4	0.11	0.61	0.72	0.72	1.17 b
Treatment 5	0.17	0.28	0.39	0.33	0.44 a
Treatment 6	0.28	0.72	1.33	0.78	1.67 b
Standard Error of the Difference (s.e.d)	0.180	0.211	0.340	0.266	0.316
Least Significant Difference 5%	0.400	0.471	0.758	0.594	0.704
P-value	0.392	0.178	0.143	0.283	0.014
Treatment Significance	NS	NS	NS	NS	S

*For Total Dieback in the second spring assessment, different letters denote significant difference.

The spring assessment data are summarised in the graph below.

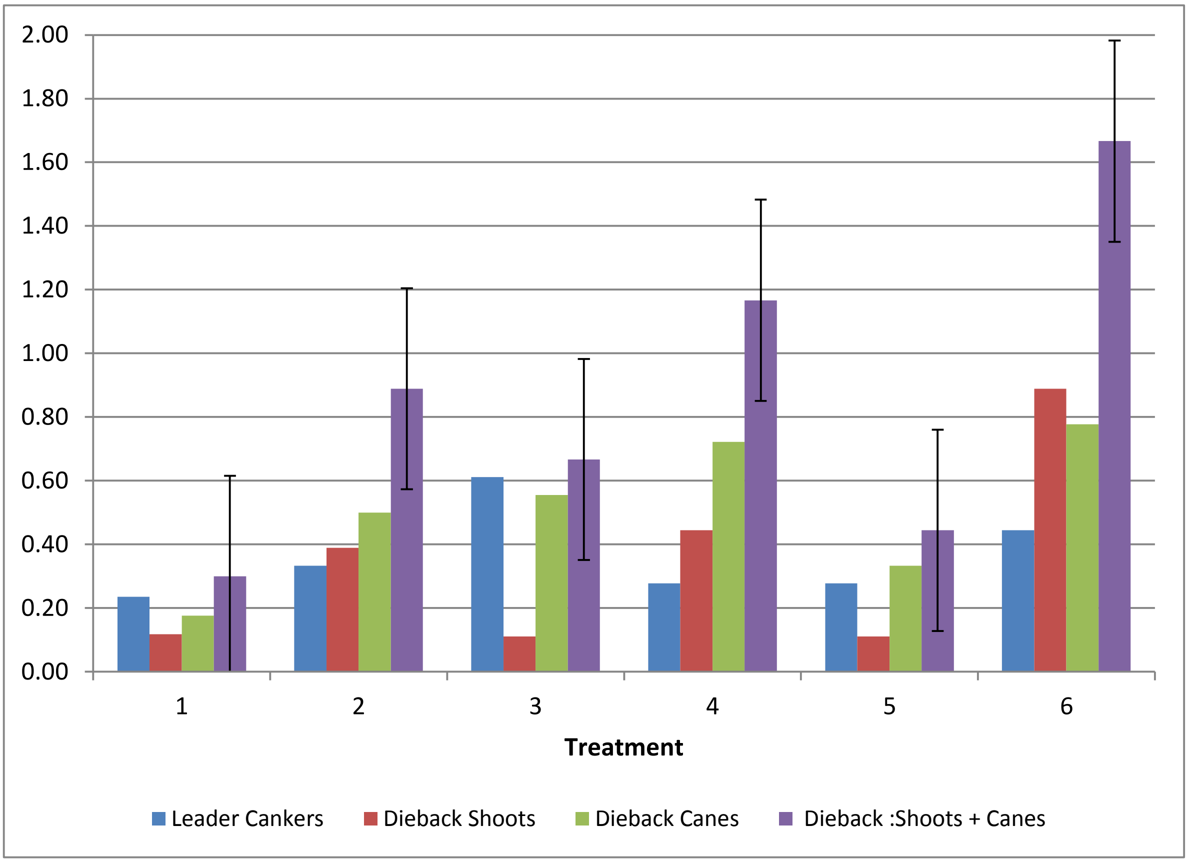

Figure 2: Treatment Programmes Psa Symptoms

6.2 Growth

There were no significant differences between the treatments for

- percentage bud break
- percentage fruitful bud break
- king flowers per shoot
- total flowers per/shoot
- king flowers per bud
- total flowers per bud

The results of the analyses are presented in Table 5 below.

Table 5: Gold 3 Components of Yield

G3	**% Bud Break**	**% Fruitful Bud Break**	**King Flowers /shoot**	**Side Flowers/ shoot**	**All Flowers/ shoot**	**King Flowers/ winter bud**	**All Flowers/ winter bud**
residual degrees of freedom	10	10	10	10	10	10	10
Trt 1	56.1	37.8	2.02	0.55	2.38	1.14	1.35
Trt 2	54.7	39.5	2.20	0.59	2.67	1.18	1.42
Trt 3	56.8	47.0	2.73	0.84	3.49	1.56	1.99
Trt 4	63.0	48.5	2.40	0.73	2.94	1.52	1.87
Trt 5	61.9	51.6	2.69	0.82	3.37	1.65	2.04
Trt 6	56.3	47.2	2.83	0.89	3.58	1.59	2.02
Standard Error of the Difference (s.e.d)	5.75	5.58	0.601	0.289	0.813	0.306	0.415
Least Significant Difference 5%	12.81	12.43	1.340	0.644	1.811	0.681	0.924
P-value	0.629	0.184	0.712	0.788	0.626	0.440	0.400
Treatment Significance	NS	NS	NS	NS	NS	NS	NS

6.3 Infection Conditions

Harvest was completed on 28 April 2013. A small amount of leaf fall had already occurred by the time of the first Actigard plus Copper application on 6 June 2013.

All post-harvest trial applications were made in dry conditions and were timely in relation to leaf fall and pruning targets.

There was significant rain in May and June 2013 of 195 mm occurring over 13 and 12 days respectively. In July 107mm rain fell over 7 days. No rain fell in the latter part of July from 21 -31 July, with winter pruning undertaken on 20 - 22 July 2013. This dry period was then followed by wet conditions in August 2013, when 167 mm rain fell on 17 days.

Refer **Appendices 3 and 4** for Risk Model predictions in July and August 2013. The model shows that there was no infection risk around the period of winter pruning, but there were frequent moderate infection risk periods through the month of August 2013.

7.0 Discussion

Natural inoculum pressure was sufficient for Psa disease symptoms to be expressed, although not sufficient to obtain a high level of symptom expression.

Treatment one was considered the standard, using Actigard plus copper once post harvest, copper at late leaf fall and copper immediately after winter pruning, against which the other treatments were compared.

Treatment one demonstrated a trend for the least number of Psa symptoms, although not significant.

Treatment two was the same as treatment one, except that copper sulphate was applied once during leaf fall, to hasten leaf fall. Although not statistically significant, treatment two had a higher level of Psa symptoms, indicating that artificially hastening leaf fall did not assist in Psa disease control in this trial.

Treatment three, the programme which had no post-harvest treatments applied, might have been expected to have a similar level of Psa symptoms as for treatment six, the key difference being that treatment three included a post pruning copper whereas treatment six did not. In fact, treatment three was not statistically different from treatment one.

Results indicate that the immediate post pruning period was an important period for protection, which both treatment one and three covered with copper. A second post pruning copper application was made five weeks not three weeks after the post pruning treatments were applied.

Treatment four was the same programme as treatment one, except that the immediate post winter pruning protectant was Key Strepto plus Engulf rather than copper. The level of symptoms in treatment four was not statistically different from treatment six, the programme which had the least protection and the highest level of disease symptoms. This seems surprising, given the known efficacy of Key Strepto against Psa.

There were twelve days when moderate infection risk was predicted in the period between treatment applications on 26 July 2013 and the next grower copper application of Nordox at 55g/100L on 30 August 2013. Compared with copper applied in the other treatments, Key Strepto applied on 26 July 2013 may not have persisted for long enough to provide protection.

The level of Psa symptoms were higher in treatment four than treatment five, yet both programmes received the same post winter pruning treatment of Key Strepto plus Engulf and treatment five also received the late leaf fall copper plus a further application of Key Strepto plus Engulf.

Although symptom expression was low, treatment six, the control treatment only receiving the grower copper applications had significantly more dieback than all other treatments apart from treatment four, underlining the value in applying an autumn/winter protection programme including a post pruning copper spray, against Psa. Treatment six also had the highest number of leader cankers (cankers were barely present on canes) although this result was not significantly significant.

The low level of symptom expression overall meant that there were no significant effects on yield as reflected by an analysis of components of yield.

The trial shows the challenge of relying on the real distribution of Psa on the vines we used because of the unknown and difficult to quantify nature of inoculum distribution.

Reliance on natural infection periods to test the relative efficacy of different treatments at different times is also challenging as conditions may or may not prevail at times treatments are applied, to actually test efficacy.

8.0 Acknowledgements

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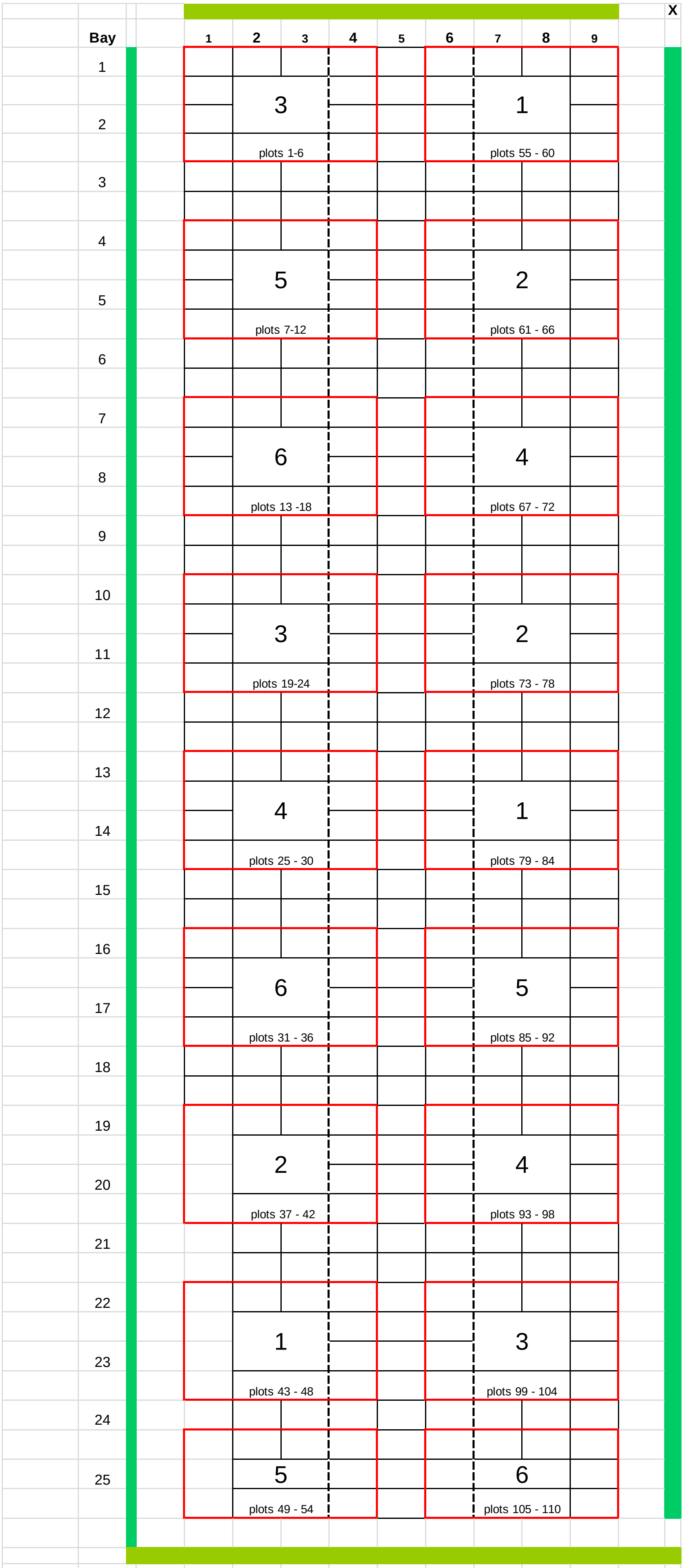
- Catherine Cameron, AgResearch Ltd, for statistical analysis.
- Dr Mary Black, Zespri for project leadership and management.

Appendix 1: Spray Equipment

Appendix 2: Gold 3 Trial Layout, Maketu



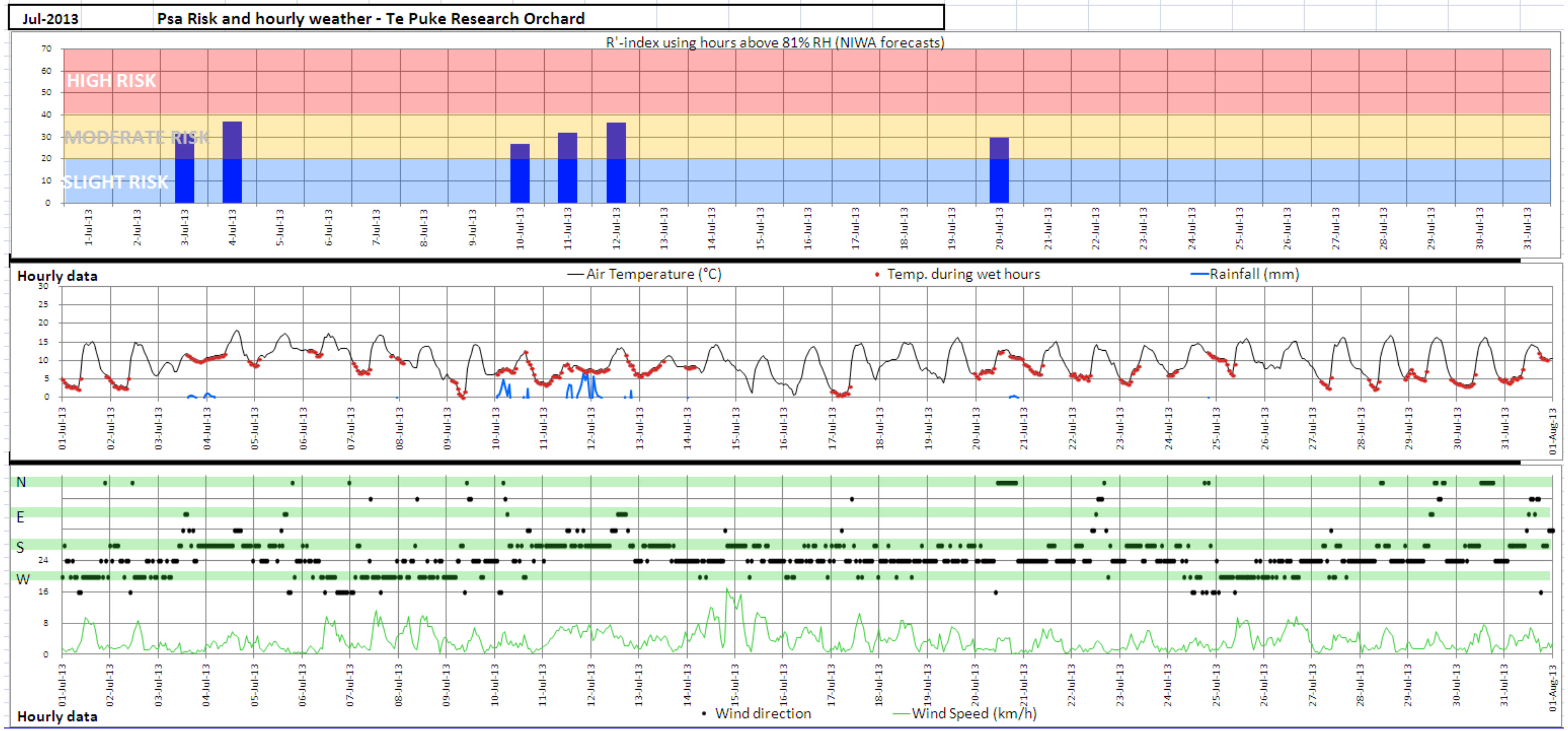
X Frost fan

Cryptomeria shelter

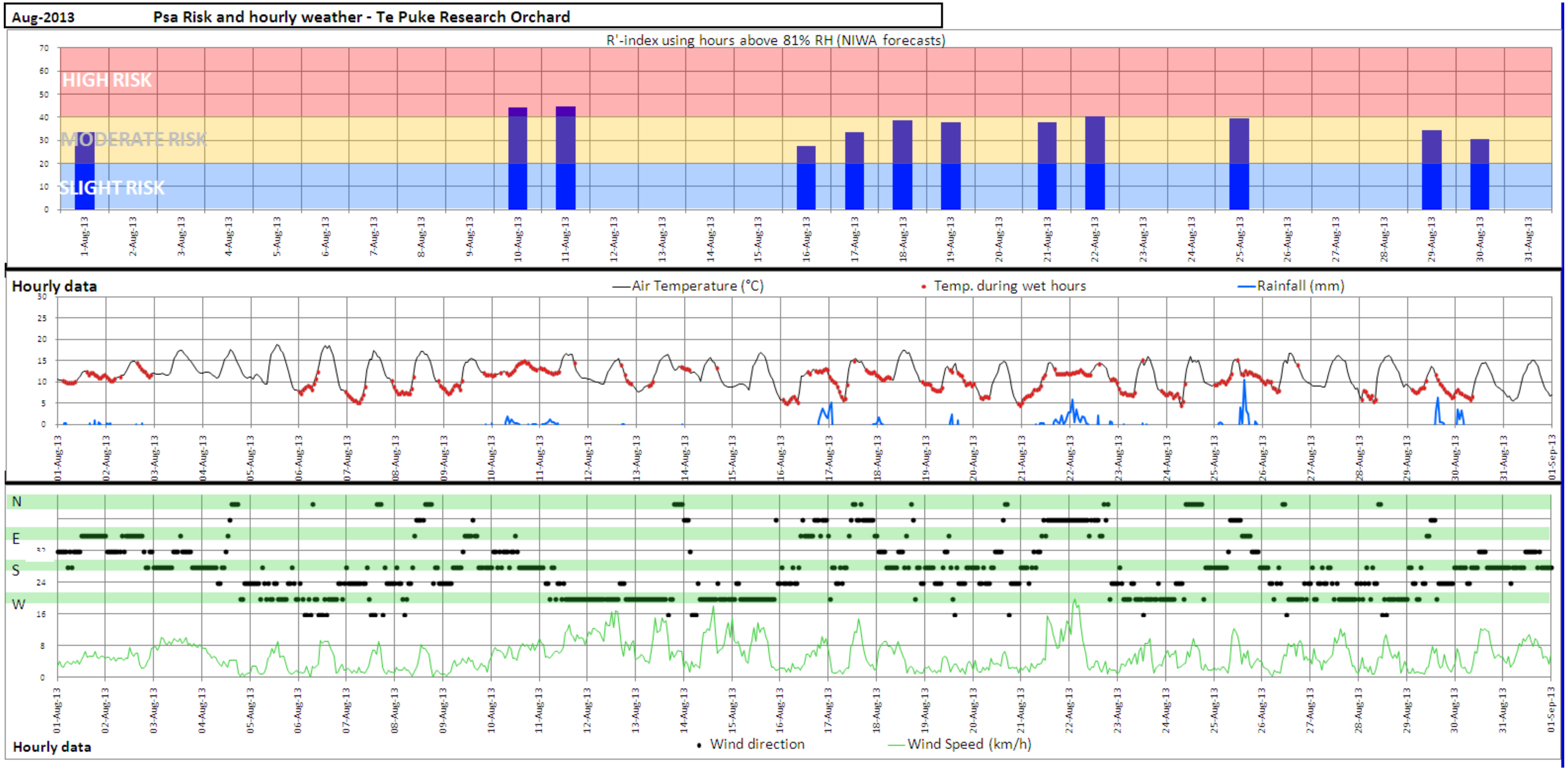
Casuarina shelter

undervine shelter; rows 4 and 7 are planted on eastern side of this shelter

Appendix 3


Jul-2013
Psa Risk and hourly weather - Te Puke Research Orchard
R'-index using hours above 81% RH (NIWA forecasts)
HIGH RISK
MODERATE RISK
SLIGHT RISK
Hourly data
Air Temperature (°C)
Temp. during wet hours
Rainfall (mm)
N
E
S
W
Hourly data
Wind direction
Wind Speed (km/h)

Appendix 4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Wind direction
Wind Speed (km/h)