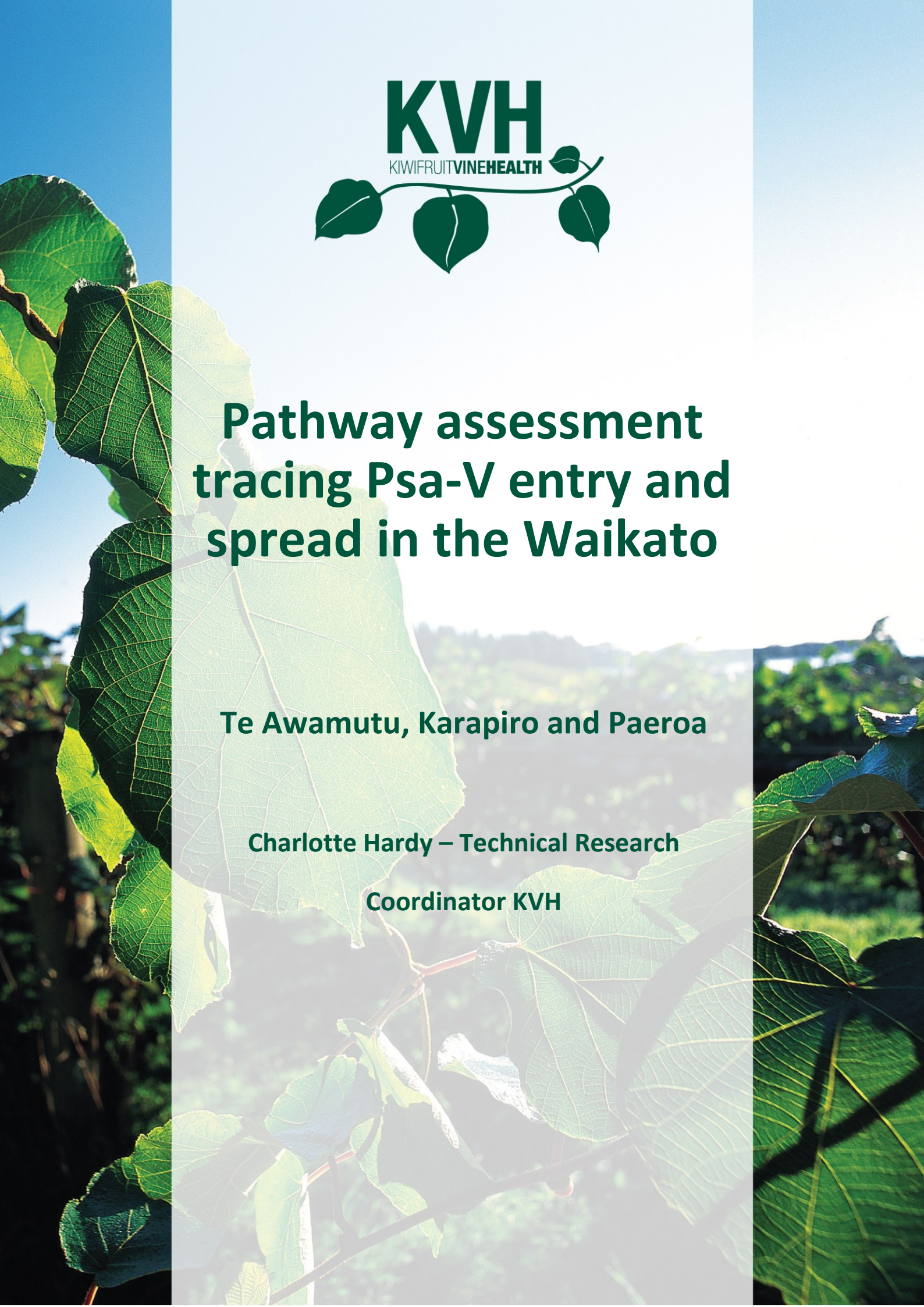


Pathway assessment tracing Psa-V entry and spread in the Waikato

Te Awamutu, Karapiro and Paeroa

Charlotte Hardy – Technical Research

Coordinator KVH

Contents

Executive summary

On 20 August 2012, KVH was notified of a positive Psa-V result in Te Awamutu in the South Waikato region. Over the previous weekend (18-19 August) a second neighbouring orchard sent in a suspect sample which was confirmed positive on 21 August 2012. At the first notification of a positive result, KVH senior technical staff visited the affected orchard to confirm symptoms. KVH initiated a pathway assessment into the Waikato region incursions.

Since the initial incursions, 20 sites have become Psa-V positive in the Waikato region. This assessment covers the first seven orchards to be identified with Psa-V, and an eighth orchard with close connections to one of the first seven orchards, which became Psa-V positive in late November 2012.

The following summarises key considerations in this report:

- Orchard susceptibility will determine how vulnerable an orchard is to Psa-V infection following an infection period or under local 'inoculum pressure'. Susceptible variety (Hort16A), predisposition to other diseases, vine juvenility, environmental risk factors such as waterlogging, and the linkage of orchards via shared contractors, equipment and machinery increased the risk profile for all orchards assessed.
- There were three main groupings of linked orchards – Group 1 (Orchards A, B and H), Group 2 (Orchards C and D) and Group 3 (Orchards E, F and G). Links were via budwood, contractors, and orchard machinery. Therefore none of the infection points are independent of other infected orchards.
- No single infection event leading to Psa-V entry into the Waikato has been identified. However, possible pathways and risk factors for infection include poor weather at harvest, risky orchard management practices, shared heavy machinery and equipment between infected regions and the Waikato, and budwood coming from Psa-V positive regions.
- Budwood movements were recorded for Orchards B, C, D and F during 2011-2012. Only Orchard F imported budwood from a high risk area (M33 Male budwood from Coromandel which is a containment region with 50% of orchards infected). All other budwood movements into the Waikato were from low risk areas such as Kerikeri and Kaikohe (containment region but with only one confirmed case of Psa-V).
- Weather is not thought to have been a pathway for spread in this assessment. Neither is transmission via bees during pollination. It is unlikely standardised hygiene practices and protocols were implemented early enough in 2011 to prevent the entry and spread of Psa-V into and throughout the Waikato. Spray programmes for all orchards were also assessed as insufficient to provide protection against Psa-V infection.
- A list of 16 'at risk' KPINs was gathered based on contractor and machinery movements from affected orchards to other parts of the Waikato, to Coromandel and Bay of Plenty. From this list, an additional four KPINs with a connection to heavy machinery used on the eight orchards assessed were also found to be positive; one KPIN in the Waikato, one in Bay of Plenty and two in Coromandel. Five of the remaining orchards had symptomatic vines, but received 'not detected' results.
- An analysis looked at the likelihood of a first 'not detected' result being followed by a positive result within six months. In all areas surveyed, it's highly likely with subsequent testing, that orchards with 'not detected' results will become Psa-V positive within six months.
- At this time the investigation has been unable to determine a primary pathway for the incursion into the Waikato region. However, its subsequent spread through three areas: Te Awamutu, Karapiro, and Paeroa is considered most likely associated with the movement of heavy machinery and equipment from orchard to orchard from mid-2011 to mid-2012.

Definition of terms

Primary symptoms: Symptoms expressed externally, ie leaf and bud spots indicating bacterial presence on the surface of the plant or in the environment.

Secondary symptoms: Symptoms expressed internally, ie shoot wilt, shoot die back, cane die back and/or red exudate, fruit shrivel indicating the bacteria is in the plants vascular system.

Infection periods: Defined as prolonged periods of leaf wetness, which is generally understood to be two to three days. From field observations, primary symptoms appear to typically express approximately seven to 14 days following a significant weather event, depending on local inoculum pressure.

Tolerance: Used to imply a plant is able, at some level, to handle or better manage the presence of Psa-V in its system or on its surface. It does not mean the plant is unable to be affected or infected by Psa-V.

Inoculum: The amount of bacteria that a plant may be exposed to which may result in infection.

Inoculum pressure: The local environmental bacterial pressure from infected orchards.

Graftwood: Small sections of kiwifruit canes grafted onto existing kiwifruit stumps or canes to establish a fruiting canopy or to replace a failed graft.

1.0 Pathway Assessment for New Incursions

1.1 Purpose of Assessment

The purpose of this assessment is to understand potential sources of infection, how infection could have entered a new region, and where it might spread to. KVH's primary objective is to minimise the impact of Psa-V on the New Zealand kiwifruit industry by identifying key understandings from the assessment including implications for future risk management. KVH will use biosecurity measures to minimise the further spread of Psa-V. This may involve the sanctioned:

- destruction of plant material that has been recently sourced from a known Psa-V environment and moved into a Psa-V 'free' environment; and
- destruction of 'suspect' plant material moved locally from orchard-to-orchard.

In conjunction with the above, KVH's containment strategies in existing/new regions focus on the following.

- Establishing a record of plant movement from any given infected orchard/s.
- Advising managers of orchards deemed to be 'high-risk' (ie any orchard directly associated with a Psa-V infected orchard through close proximity, shared plant material or contractor movements) to frequently monitor their orchards (or blocks) that may be susceptible to contamination.
- Providing regional advice (through grower meetings) about industry best practice in light of a Psa-V environment.
- Informing and implementing biosecurity compliance protocols, eg budwood supply, pollen supply etc relevant to a new incursion area.

1.2 Key Areas Considered in an Assessment

KVH will interview affected growers and packhouse Psa-V managers to obtain information on the following key areas.

- General orchard set-up—vine variety, rootstock, age of both scion and rootstock, growing system etc.
- The nature of the infection—number of infected vines, symptoms and spread.
- Records of plant movement/supply—on and off orchards.
- Orchard practices and associated contractor movements—with initial focus on high risk activities.

Three primary pathways are explored with regards to the trace back of Psa-V in a new region.

1. Weather-related events/infection periods against protective spray programmes in place on orchard/s.
2. Movement of plant material and/or orchard materials and equipment.

3. Work history and movement of orchard contractors.

In aid of this investigation the following questions need to be considered.

- Location—is the first orchard a true expression of the original point of contamination?
- Time—does the level/severity of infection indicate an association with a particular pathway, event or activity?
- Disease expression—do the symptoms indicate an association with a particular pathway, event or activity?
- Plausibility—is the pathway under investigation knowingly able to spread Psa-V?
- Are the incursions to be treated as individual cases; or is one of the orchards a primary-source orchard with a common link existing between the other incursions, ie linking them together?
- Are there any materials/contractors connected to known Psa-V regions?

The answers to these questions will rule some pathways in and others out. Ultimately, it can provide growers and KVH with the necessary information to help to manage the known risks to the wider region.

1.3 Limitations to a Trace Back Investigation

Trace back assessments are prepared by KVH to aid in the assistance and understanding of how to effectively manage a new Psa-V region. In known Psa-V environments it may be obvious how and when Psa-V has been transferred. However, the same is not necessarily true for new incursions outside of existing Psa-V regions.

In many cases:

- there will be a range of possible pathways;
- key background orchard information—plant movement, orchard work history and contractor whereabouts may be incomplete; and
- the undetected presence of the disease in a new region is a possibility.

For this reason this report can only assess the most probable pathways and provide an assessment of the likelihood of each of these as being a primary pathway. Gaps in understanding the lifecycle of Psa-V limit the interpretation of the information gathered. Therefore, KVH may be unable to determine the primary pathway responsible for the introduction of Psa-V into a new region.

Trace Back Investigation Time Line

- **20 August 2012**—Positive result received for Te Awamutu. Trace back process initiated.
- **21 August 2012**—Second positive result received for Te Awamutu. KVH field staff on site.
- **22 August 2012** – KVH field staff map / monitor infected orchards in controlled area.
- **9 October 2012** – Positive result received for Orchard C in Karapiro, and visual positive made for Orchard G in Paeroa
- **19 October 2012** – Positive result received for Orchard D in Karapiro
- **16 November 2012** – Positive result received for Orchards E and F in Paeroa
- **30 November 2012** – Positive result received for Orchard H in Te Awamutu
- **21 December 2012** – Draft of Pathway Assessment completed.
- **January 2012** – Internal review completed

2.0 Orchards in the pathway assessment – links and commonalities

2.1 Objective: Identify the primary orchards involved and assess orchard practice and management, and if possible, determine whether these incursions are to be treated as linked or individual cases.

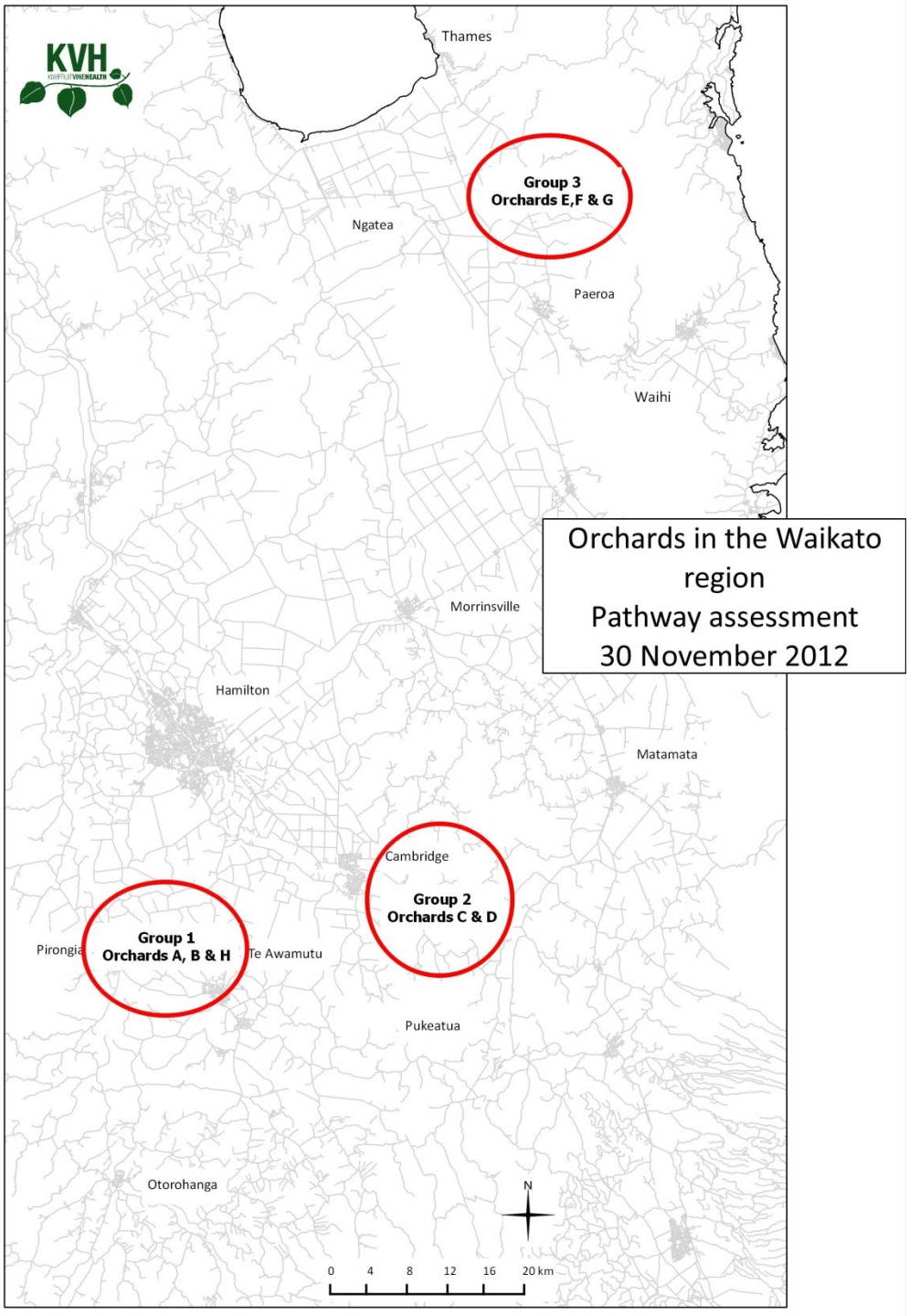
Table 1 ***Summary of timing of Psa-V confirmation, symptoms and orchard grouping***

Orchard	Date Psa-V confirmed	First infected variety	Incursion Grouping	Infected vines	Dates for test results Male/ Female	Symptoms
A	20 Aug 2012	Hort16A	Group 1	22 Females 46-60 Males	M series male H16A female (20th Aug)	Red exudate
					HW visual confirmation (12th Oct)	Leaf Spot
B	21 Aug 2012	Hort16A	Group 1	15 Females 15 Males	H16A female CK1 male (21st Aug)	Red Exudate
					HW Male and Female	Red Exudate Leaf spot
C	9 Oct 2012	Hort16A	Group 2	3 Females Adjacent Males and Females	H16A female (9th Oct)	Red exudate Dieback Leaf spot
					HW female & male (visual confirmation)	Leaf spot
D	19 Oct 2012	Hort16A	Group 2	Several Females Adjacent Males	Bruce male (11th Oct)	Leaf spot
					H16A female (19th Oct)	Red exudate Dieback
E	16 Nov 2012	HW	Group 3	Widespread Females Widespread Males	HW females and	Leaf spot
					undetermined male (16th Nov)	Leaf spot
F	16 Nov 2012	HW	Group 3	15-17 Females 1 Male	HW female	Leaf spot
					Chieftan male (16th Nov)	Dieback
G	9 Oct 2012	HW	Group 3	1 Male	HW male visual confirmation	Red exudate
H	30 Nov 2012	HW	Group 1	2 Females 1 Male	HW females (30th Nov)	Leaf spot Dieback

2.2 Linked Orchards

There are three distinct incursion groupings in this assessment. Group 1 which consists of Orchards A, B and H in Te Awamutu. These orchards have close geographic proximity (within 2km), and Orchards B and H share equipment and machinery.

Figure 1: ***Waikato region Psa-V map: Identifying groupings of linked orchards***



Group 2 consists of Orchards C and D, with small shared amounts of budwood material, beekeeper equipment and a close geographic proximity (within 800m) in Karapiro area. Orchard C was the only orchard in this assessment which did not use the same shelter trimmer in either 2011 or 2012.

Group 3 consists of Orchards E, F and G in Paeroa, which are managed concurrently and share machinery and equipment between them (Figure 1). Male budwood on Orchard F came from Whenuakite in the Coromandel, in winter 2012, a location which subsequently became Psa-V positive in September 2012. The same shelter trimmer was used in Orchards E-G, in Whenuakite, BOP and another Waikato location in 2011. All locations are now Psa-V positive.

Table 2: ***Summary table noting key information on Psa-V positive orchards***

KPIN	Date Psa-V +ve	Variety	Shared Plant material 2011-12	Shared orchard machinery 2011-12	Packhouse 2011-12	Pollination 2011-12	Shared contractors 2011-12
A	20 Aug 2012	Hort16A males + females	No	B,D,E,F,G,H	Packhouse L	Beekeeper 1	B
B	21 Aug 2012	Hort16A females + males	No	A,D,E,F,G,H	Packhouses M & N	Beekeeper 2	A & H
C	9 Oct 2012	Hort16A females + males	Yes – D	C	Packhouse L	Beekeeper 3	No
D	19 Oct 2012	Hort16A females + males	Yes – C linked via packhouse	A,B,E,F,G,H via linked orchards	Packhouse O	Beekeeper 3	No
E	16 Nov 2012	HW females + males	No	A,B,D,F,G,H	Packhouse P	Beekeeper 4	F & G
F	16 Nov 2012	HW females + 1 male	No	A,B,D,E,G,H,	Non-producing orchard	No Beekeeper	E & G
G	9 Oct 2012	HW visual positive	No	A,B,D,E,F,H	Packhouse R	Beekeeper 4	E & F
H	30 Nov 2012	HW females + 1 male	No	A,B,D,E,F,G	Packhouse S	Beekeeper 1	B

2.3 Commonalities between orchards

Postharvest facilities

During 2011 and 2012 only two orchards (A and C) shared a postharvest facility. Six of the seven packhouses have facilities in the Bay of Plenty, and transport companies are used during harvest to transport fruit between the two regions. All facilities have procedures and protocols in place for the cleaning and sanitisation of equipment—ie, bins, packing bags, fruit sorting machines etc. At bigger packhouses all hygiene systems tended to be automated, and at smaller pack houses a mix of automated and manual hygiene systems were present. It was noted that with automated systems, there is not always a manual check for remaining plant material in bins. One orchard contractor that was interviewed reported half a dozen fruit turned up in a couple of bins at the beginning of harvest in 2011. Fruit were put in the rubbish at the orchard when they were discovered, but fruit bins were not re-sanitised.

Vehicle transport and hygiene

Trucks used to transport equipment for harvest or hives for pollination were also subject to hygiene standards. No orchards used artificial pollination in either 2011 or 2010. Four hive suppliers were utilised. Not all vehicles run by packhouse facilities or beekeepers were brushed down (truck decks) for plant material to the same extent, although all were apparently water blasted and sanitised.

This usually included hosing or water blasting in 2011 and more regularly a combination of hosing/water blasting and sanitisers used in 2012. The combination of a large number of vehicles arriving on orchards during harvest in 2011 and inclement weather including heavy rain during harvest 2011 combined to make conditions muddy and messier than usual for some orchards interviewed.

Pollination

Most orchard owners restricted beehive placement to headlands and some beekeepers chose to have automated feeding devices in hives to reduce the normal two to three-day manual feeding of bees as was standard practice before the advent of Psa-V.

Once hives are taken off orchards, it is not possible to sanitise them, and organic material may remain. Because of the placement of hives on headlands, and several metres from vines, it is unlikely that infected plant material would directly contact hives. Some hives were placed on corrugated metal sheets on the ground to prevent organic material contamination on the base. Studies undertaken by Plant and Food Research in Te Puke, has shown that no Psa was isolated from any of the honeycomb samples tested from hives in infected orchards or from the bees tested. The limit of detection in this experiment was 40 colony forming units per sample (Anderson et al. 2011).

Further research is being conducted this season looking at whether bees can transfer Psa-V during pollination. Preliminary results indicate that when hives are placed in infected orchards then removed, Psa was still detected in hives several days later (David Pattemore pers. comm. Mar 2013).

Contractor movements and sharing

There was little sharing of independent contractors for canopy management work—eg pruning, fruit thinning or vine spraying. Most orchards had either their own permanent staff, did the majority of orchard work themselves or employed contractors through their packhouse. Only orchards B and H and Orchards E, F and G had substantial contractor links for regular orchard work. Orchard D did not have direct links to the other orchards assessed. It did have concurrent management with nine other KPINs which may have increased the risk of contaminated equipment or contractors between these orchards spreading Psa-V. No contractors came from the Bay of Plenty during 2011-2012.

In general, contractors and some affected growers in the Waikato did not have an accurate picture of when Psa-V was first identified in the Bay of Plenty. Many thought it was in mid-summer 2011, up to six months after the first case of Psa-V was actually identified in Te Puke in early November 2010. For some contractors a systematic and consistent hygiene programme was not implemented until July or November 2011.

At risk KPIN monitoring

Initially, orchards A and B were the first to be interviewed, and associated contractors, beekeepers, post-harvest facilities, machinery and equipment and their movements were compiled. Based on records of people and machinery movement from January 2011 to spring 2012, a list of ‘at risk’ KPINs was drafted (Table 3). Postharvest facilities were alerted, and monitoring of ‘at risk’ orchards in locations outside of the 10km controlled area was undertaken.

From the list of 16 ‘at risk’ KPINs, to date six have not been tested, because there was no symptomatic material to sample. Of the 11 KPINs tested, three did not detect Psa-V in the samples taken (ND=Not Detected), and five KPINs tested positive. The remaining KPIN has had a visual confirmation of Psa-V but no laboratory confirmation. An initial sample from this orchard came back with a ‘not detected’ result. Clear Psa-V symptoms would suggest that an adequate sample has not been able to be taken, but a future positive result is considered likely. The grower is confident there is Psa-V in the orchard, and is treating all orchards with similar hygiene protocols and spray programmes.

Table 3. ***List of at risk KPINs and risk categories. Shared Contractor and Shared Machinery, refers to orchards which have employed the same contractors or used the same heavy machinery in the last 2 years.***

KPIN	Region	Shared Contractor	Shared Machinery	Orchard Status	Date Tested
1	Coromandel		√	Not tested	
2	Coromandel		√	ND	1/11/12 H16A (ND)
3	Coromandel		√	Not tested	
4	Coromandel		√	Psa-V +ve	6/9/12 H16A
5	Waikato		√	Psa-V +ve	16/11/12 HW
6	Waikato		√	Visual Psa-V	16/10/12 HW (ND)
7	Waikato		√	Psa-V +ve	1/11/12 G3
8	Waikato		√	Psa-V +ve	30/11/12 HW
9	Waikato	√		Not Tested	
10	Waikato	√		Not Tested	
11	Waikato	√		Not Tested	
12	Waikato	√		ND	2/11/12 G3 & M33 (ND)
13	Waikato	√		Not Tested	Last test 3/11/11 (ND)
14	Waikato	√		ND	19/12/12 HW (ND)
15	Waikato	√		Not tested	
16	BOP		√	Psa-V +ve	26/11/12 HW

Several issues have been identified from the review and are relevant for the Waikato pathway assessment. Growers from two orchards in the Waikato incursion saw what they thought were Psa-V symptoms prior to a positive test result. One orchard took a sample in spring 2011 and got a 'not-detected' result. The next test was not taken until spring 2012. The other orchard saw symptoms six to eight months prior to the first positive test result, but did not get a test at the time of first symptom appearance. If a second sample had been taken earlier, Psa-V might have been identified in the region much earlier.

The visual confirmation on a linked orchard is evidence that even with symptoms present (cankers on leaders) it can be difficult to get a good sample to get a positive result. Wood is a particularly difficult plant part to sample, and cankers may not return a positive result, as easily as cane dieback or leaf spot. Once an orchard is confirmed positive for Psa-V in a new region with testing results from two laboratory service providers, there is less value in submitting repeated samples for testing. Orchard hygiene, vine protection and sanitation practices should include all areas of the orchard, for all varieties.

Growers from two Waikato orchards saw symptom expression in spring 2012. They took samples and received a 'not detected' result. Subsequently, on re-testing they were confirmed positive. The importance of taking good samples and re-testing within six months (see analysis of prior test results in Appendix 3) of a 'not-detected' result, are paramount for new regions being able to contain and control Psa-V early and rapidly.

Risks and links between orchards: Fruit and potentially other plant material in bins at 2011 harvest. Not all vehicle decks were brushed down and sanitised. Contractors and some growers had an inaccurate perception of when Psa-V was first identified. Psa-V like symptoms seen earlier but not tested. Sharing of equipment, budwood and contractors, link orchards assessed in the Waikato and also Waikato orchards with orchards in Coromandel and Bay of Plenty.

3.0 Nature of the infection on the affected orchards

Foreknowledge: *The location of infection within an orchard, spread throughout block and severity of disease expression can give direction as to where, when and how the initial infection occurred. In-field observations have seen weather-related contamination typically result in primary infection as leaf spot initially. Contamination by plant material and/or contractor work typically results in secondary infection as shoot wilt, cane dieback or orange exudate.*

Aim: Visit the affected orchards and observe the type and extent of symptom expression to determine focus for the trace-back investigation. These observations are made within the context of what is already known about Psa-V infection in orchards.

Recent research (within the last two years) has given us an understanding of basic biology and epidemiology of Psa-V within kiwifruit vines (Appendix 1). However, some unknowns remain—including what the threshold for infection by Psa is in an orchard, ie, what numbers or concentration of the bacterium are needed to enable infection to occur in the kiwifruit vine.

3.1 Infection patterns in Waikato orchards

In the case of the Waikato incursion, a combination of factors around symptom expression, provide evidence of infection timeframe and movement of Psa-V in the region. Psa-V-like symptoms were seen in the Waikato as early as spring 2011 in Te Awamutu, and late summer/early autumn 2012 in Karapiro. During a monitoring round in November 2010 when Psa-V was first confirmed in Te Puke, photos were taken on Orchard B that showed black irregular leaf spots with yellow haloes. However, test results were either negative for material sampled at these times, or no samples were taken when symptoms were seen.

In the orchards assessed, the most common combination of symptoms in Hort16A were secondary symptoms, including exudate and cane dieback. Leaf spot was also seen in some cases. In Hayward, symptoms were predominantly leaf spot with occasional cane dieback and cankering. This was a feature on orchards with only one variety and also orchards where both Hort16A and Hayward occurred together.

Symptom progression and expression in vines on orchards A and B advanced quickly with budbreak and cold spring weather. Cankers and ooze were seen in Hort16A blocks. Later, leaf spotting was observed in Hayward vines.

Symptom progression on orchards C and D included leaf spotting in some plants in a circular distribution around vines with secondary infection, including cane dieback, and in one instance whole vine death. Symptom progression on orchards E, F and G in Paeroa has been variable, cankers being the first evidence of Psa-V seen on Orchard G. Leaf spot and a small amount of dieback were seen on Orchards E and F. It's likely Orchard G, which has waterlogging issues and Armillaria in some blocks, may have been expressing symptoms earlier. However, they may not have been differentiated from other disease and stress symptoms.

From field observations, leaf spot has typically been expressed approximately seven to 14 days outside of a significant weather event—depending on local inoculum pressure. While one grower (Paeroa) thought it quite likely that weather could have spread Psa-V to his orchards, other growers did not believe that weather-related dispersal of Psa-V was likely. There were few observations made by growers that could pin down expression of symptoms to a particular weather event. More commonly, growers, orchard managers and contractors became aware of infection and guessed it could have been present for a longer time than they had been aware of.

There was no obvious symptom expression after budwood was grafted into Orchards C and F over the last two years. Kiwifruit graft tissue can remain asymptomatic for an unknown length of time, and infection from graftwood, versus infection entering the plant at the time of grafting or when grafted plants are very young, is difficult to distinguish. At this point, it is unknown whether graftwood contributed to Psa-V infection in these orchards. Symptom expression may become clearer as grafts age. However, other mitigating factors, such as increased inoculum pressure in the Waikato region, could prevent a conclusive connection being made.

Hayward tends to express symptoms more slowly than gold varieties, and Hort16A does not always produce primary infection such as leaf spot before secondary symptoms develop. It would appear that infection may have been in some orchards six to eight months prior to detection and on others, up to a year or more prior to detection. In the case of Orchard B, Psa-V-like symptoms were first seen as far back as November 2010. There is no certainty on timing. However, the range of symptoms seen in both Hayward and H16A blocks could imply various infection periods, some more recent than others, with the possibility of an initial infection event much earlier.

Infection expression: First Psa-V like symptoms seen in Te Awamutu in November 2010. Symptoms tested in spring 2011, not detected. Leaf spot seen six to eight months prior to first positive test result in October 2012 in Karapiro. Hort16A symptoms manifested as both primary and secondary symptoms. Hayward symptom expression more commonly leaf spot. No obvious symptoms from potentially infected budwood. Infected vines near natural and artificial shelters in several orchards.

Entry date into the Waikato: Between May-June 2011 and February-March 2012.

3.2 Key information gathered during field visits to assessed orchards.

Orchard A, confirmed Psa-V positive on 20 August 2012 from female and male cane sample in a Hort16A block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 20 August,2012 for the following: Hort16A block male and female	Secondary symptoms including rust and cankers. Approximately 40-60 infected male vines and 22 female vines. Central patch in one block and spread down several rows in the centre of a second adjacent block. Hayward vines visual confirmation of leaf spot in September 2012. Estimate of overall block infection was approximately 30 percent. 	The whole orchard was sprayed with copper immediately. Initially, infected vines were cut back and secondary infection was removed where it was found. H16A blocks were removed as soon as was practical (approximately one month after Psa-V positive confirmation). These blocks were subsequently re-grafted to G3. Cut out material was disposed of into a dug pit and buried.	Other diseases such as Armillaria were present in blocks where Psa-V was found. The vines were not in good shape before they got Psa-V. Some areas of the orchard were weak, and vines had never really thrived. In late spring the previous year (2011), infected material was sent away for testing by packhouse representatives. Symptoms seen at the time were rust and exudate. Tests came back 'not detected'.

Table 1. ***Key information on infection expression on Orchard A***

Orchard B, confirmed Psa-V positive on 21 August 2012 from male cane sample in a Hort16A block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 21 August,2012 for the following: Hort16A block Male	This orchard is in close proximity (2km) to Orchard A. A male plant in a Hort16A block was first confirmed with Psa-V in the form of rust and red exudate. Red exudate was seen more strongly on female vines but also appeared on male vines. Approximately 30 vines were expressing symptoms with even numbers of males and females. Hayward vines in adjacent blocks were confirmed positive via lab tests in September 2012. Widespread secondary symptoms were seen in both male and female Hayward plants. The photo below was taken on 22 August 2012 and shows infection in Hort16A. Estimate of overall block infection was approximately 20 percent. 	The whole orchard was sprayed with copper immediately. All Hort16A blocks were removed several weeks after initial confirmation of Psa-V on the orchard. Cut out material was put into a dug pit, then buried. They have not been re-grafted to new varieties. Hayward symptomatic vines were managed by cutting out infection in small sections and cankers burned with a butane torch.	Initial thoughts were that infection had blown in from a neighbouring property. Worst infected areas tended to be in the shadiest, coldest parts of the orchard. The photo below shows results from symptom monitoring sheets for Orchard B which were submitted on 12 November 2010 by packhouse representatives. It is likely the camera date on the photo is incorrect by a month. The symptoms below are on Hayward leaves seen on the orchard in November 2010. 

Table 2. ***Key information on infection expression on Orchard B***

Orchard C, confirmed Psa-V positive on 9 October 2012 from male cane sample in a Hort16A block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 9 October,2012 for the following: Hort16A block Female	Secondary symptoms were first seen in female Hort16A vines. These were wilting canes, and floppy terminal growth on shoots. Cankers and red ooze were seen on the canes further back from dieback and wilting. Leaf spot was seen on male vines around the most heavily infected females. Infection was concentrated in the north western edge of the block closest to a shelter belt. Estimate of overall infection in the block was approximately 20 percent. Photos show cane dieback (top right), typical black spots with yellow halo (bottom right) and yellow halo with no irregular black spot (bottom left). Photos were taken on 12 October 12. 	Copper was sprayed on the infected area of plants in October, but not on the wider block. Males were removed from the block after pollination in November and the block was completely cut over in December 2012. Cut out material was burned and buried. It has since been grafted to G3.	In March 2012 leaf spot symptoms were observed. However a sample was not taken at that time. Symptoms appeared first in female vines.  

Table 3. ***Key information on infection expression on Orchard C***

Orchard D, confirmed Psa-V positive on 19 October 2012 from male cane sample in a Hort16A block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 19 October,2012 for the following: Hort16A block Male	Expressed as primary and secondary infection in Hort16A. One male vine was observed with rust and ooze on the leader, and two female vines had dead canes and dead buds. Other surrounding vines had leaf spot, not always with a halo, frequently the dark irregular spots. Infection was concentrated in two areas of the block, under overhead shelter. One site was on the western side of the block and the other near the northern boundary. Approximately 46 vines exhibited primary leaf spot symptoms. Estimate of overall infection is about 25 percent of the block. Photos were taken on 17 October 2012.  	Copper was sprayed onto infected blocks, and infected secondary symptoms cut out as they appeared. At the time of KVH staff visiting the orchard, cut out infected material was being stored in plastic bags. Subsequent discovery of more infected orchards managed by the same owner/manager has led to a decision to cut out whole sections of affected blocks (December 2012).	The orchards were managed organically aside from a couple of inputs that are not BioGro certified. The owners/managers of this orchard manage a group of orchards in the same area. Contractors are shared across the grouping, and various orchard equipment and machinery are also shared between all orchards in the grouping.

Table 4: ***Key information on infection expression on Orchard D***

Orchard E, confirmed Psa-V positive on 16 November 2012 from male cane sample in a Hayward block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 16 November, 2012 for the following: Hayward block	Orchard E expressed as widespread leaf spotting throughout the whole block on Hayward vines, in both male and female plants. Photo below is leaf spot on Hayward vines and was taken on 14 November 12. Estimate of overall infection is approximately 50-80 percent.	Copper and Actigard™ were sprayed before flowering. No plant material has been cut out at this point. If secondary symptoms are seen infected material will be removed.	The owner of this orchard also manages and works on Orchards F and G. Staff, equipment and machinery such as sprayers, mulchers and shelter trimmers are shared between orchards. A sample had been previously taken in this orchard (Sept 2012) and returned a 'not detected' result. Orchard vine configuration is opposing female with internal skirts and three metre row width. This orchard is lighter and more open than orchard G.

Table 5. ***Key information on infection expression on Orchard E***

Orchard F, confirmed Psa-V positive on 16 November 2012 from male cane sample in a Hayward block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 16 November,2012 for the following: Hayward block	Expressed as leaf spot, in a semi-circle of Hayward vines, next to a shelter belt. Infected plants included approximately 15-17 females and one male with dieback on the main cane. Photo below shows leaf spot on mature Hayward leaves taken on 14 November 2012. Estimate of overall infection in the block is approximately three percent.	The block has been sprayed with copper and Actigard™. The dieback on the male was cut out. Infected material has been burnt and buried.	This block is a newer planting with vines about five years old. This orchard is managed quite differently to Orchards E and G, which are older and more established. There is a strip male configuration on Orchard F rather than opposing females as in Orchard G, and rows are four metres apart. This orchard also experiences cooler temperatures (2°C colder) than Orchards E and G.

Table 6. ***Key information on infection expression on Orchard F***

Orchard G, confirmed Psa-V positive on 9 October 2012 from male cane sample in a Hayward block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 9 October,2012 for the following: Hayward block	Expressed as cankers and rust on leaders in several Hayward vines which had a visual confirmation of Psa-V in August 2012 and were then removed and buried without samples being taken or lab tested. A sample was taken of rust and exudate on a leader behind a piece of wire on 9 October 2012 (Photo bottom right). The sample came back not detected. A visual confirmation has since been given for Orchard G. Estimate of overall infection in the block is approximately one percent.	The block was sprayed with copper and Actigard™. The rust and cankers seen were cut out. Infected material has been burnt and buried.	Orchard has opposing female vine configuration. Stressed plants which suffered waterlogging and Armillaria were worst affected by Psa-V. THis orchard has a denser canopy and bigger rows than orchards E or F.

Table 7. ***Key information on infection expression on Orchard G***

Orchard H, confirmed Psa-V positive on 30 November 2012 from female leaf sample in a Hayward block

Orchard profile:	KVH field observations:	Course of action:	Growers'/managers' comments:
Positive result 30 November,2012 for the following: Hayward block Female	Initially, infection expressed as a rust stain on a single cane on a single Hayward male. This male plant material was tested in mid-September and came back 'not detected'. Further leaf spot symptoms were seen on two female vines in late November and early December and one vine was confirmed positive for Psa on 30 November 2012. When the grower was initially interviewed, no symptoms were present in the orchard and budburst had not occurred. The orchard itself was not viewed by KVH staff.	Infected male was cut out and the orchard sprayed with Copper. No Picture Available	This orchard is linked to Orchard B. Staff, equipment and machinery such as sprayer, tractor and shelter trimmer are shared between the two properties.

Table 8. ***Key information on infection expression on Orchard H***

4.0 Assessing pathway 1: weather related events/infection periods

Foreknowledge*: Bacteria can be spread through a range of means. The most common method of bacterial spread is through wind and rain. Rain drops hitting a leaf causes 'splashing' which can redistribute the bacteria locally to neighbouring leaves. 'Free' water is required for the proliferation of Psa-V. Infection periods are defined as prolonged periods of leaf wetness; which is generally understood to be two to three days. From field observations, leaf spot has typically been expressed approximately seven to 14 days outside of a significant weather event—depending on local inoculum pressure.*

Aim: Identify key infection periods prior to the positive result for each of the orchards, and/or during key orchard activities, eg grafting. Once potential infection periods have been identified, assess the protective spray programme in place for each of the orchards to determine an orchard's expected protection against a potential weather related incursion.

4.1 Long distance dispersal potential of Psa-V

The exact distance Psa-V can travel via weather dispersal has not been quantified. A 10km radius is used to set controlled area limits by KVH during new incursions in regions where Psa-V has previously not been detected. In this assessment, the assumption is made that wind dispersal is most likely to occur over a maximum of 10km. However, is much less likely to occur up to 15km.

Estimates were made using the Google earth measuring tool of the distance from infected orchards in the Waikato to the next nearest infected orchard in neighbouring regions. Figures 2 and 3 below show the distance from an infected orchard in Paeroa to the next closest infection point in the Coromandel (17.59km), and the distance from an infected orchard in Te Awamutu to the next closest infection point in the Bay of Plenty (61.22km).

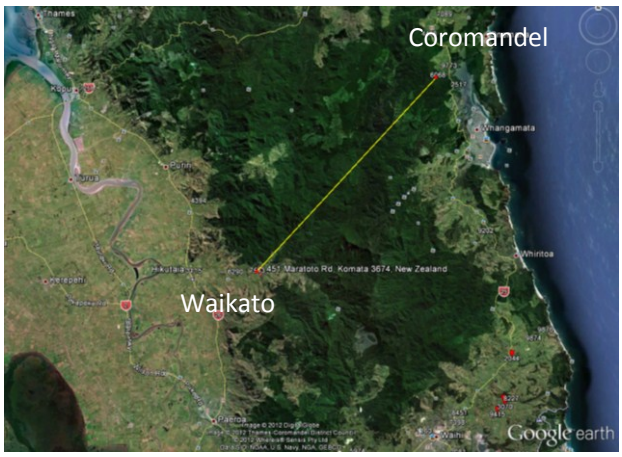


Figure 2. ***Paeroa to Coromandel (17.59km)***

Figure 3. ***Te Awamutu to Bay of Plenty (61.22km)***

The Group 3 Orchards in Paeroa are relatively sheltered in a valley, and two have tall shelter belts of Cryptomeria. Orchard F has shorter, less developed Cryptomeria shelters that are six years old. Wind-carried inoculum would have to make it over a significant geographic boundary, in the form of the Kaimai ranges. The Paeroa orchards are the closest sites in this assessment to another infected orchard. Group 1 and 2 Orchards in the Karapiro and Te Awamutu areas are at least 50-60km from the next closest infection point in the Bay of Plenty region (Figure 3 above). The further inoculum is taken by wind, the more dispersed it becomes and the lower the likelihood of bacterium landing on kiwifruit vines. It is considered unlikely that weather, and particularly wind dispersal, was responsible for entry of Psa-V into the Waikato.

4.2 Weather as a contributing factor to Psa-V infection

There are several time periods during 2011 and 2012 when weather could have contributed to Psa-V infection if inoculum was present. May and June 2011, were time periods in Te Awamutu when rainfall was high, and high-risk activities were carried out during poor weather conditions.

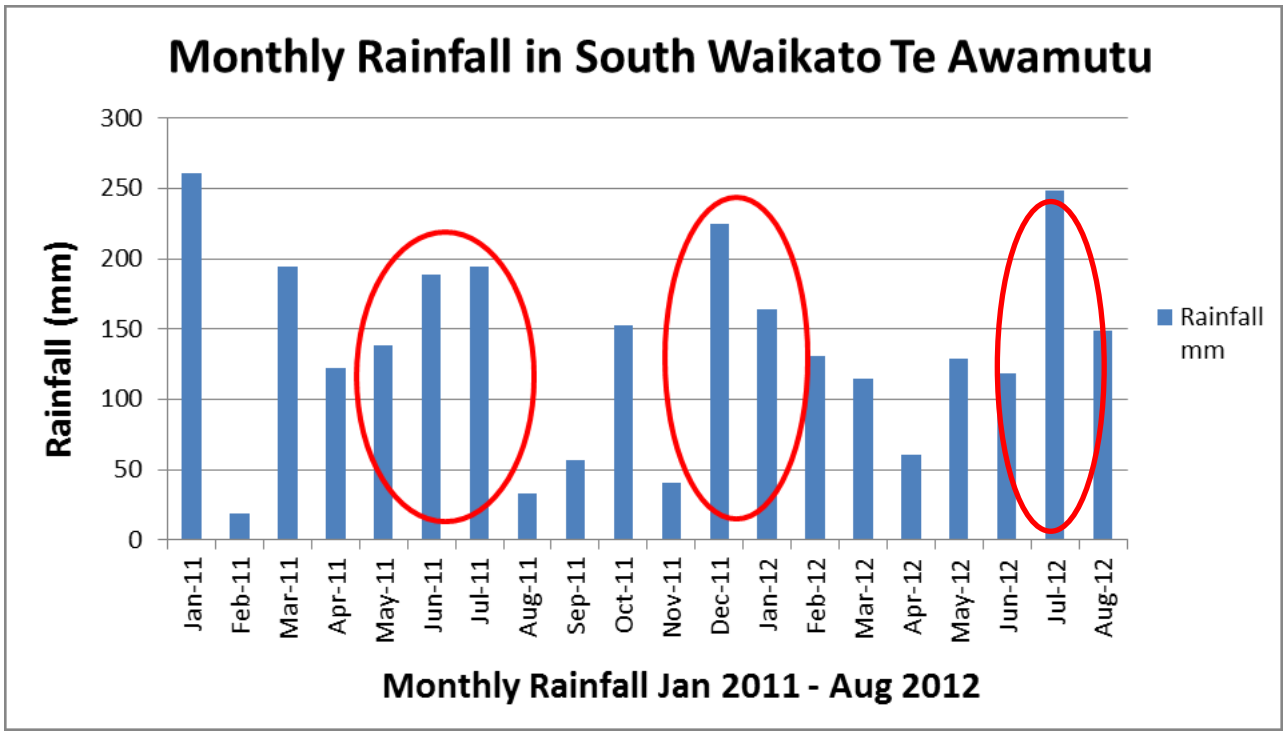


Figure 4: ***Rainfall data from a weather station on Orchard B in Te Awamutu***

Orchard A had heavy machinery break-down and it was subsequently repaired during the May-June period in 2011 during shelter trimming. If Psa-V was present on machinery at this time, there would have been an increased risk of it spreading. On Orchard B, harvest was disrupted with heavy rain and showers, and fruit was packed with two packhouses, resulting in a large number of fruit bins and trucks coming on and off the property. Pruning and shelter trimming were undertaken very close to harvest on this orchard, and again, both activities were frequently disrupted by rainfall. Low temperatures and frequent frosts from autumn to spring 2012 (Table 9) could have hastened symptom expression during 2012 when Psa was first identified.

Table 9. ***Frost events on Orchard B during 2011 and 2012***

2011	Temperature	2012	Temperature
July	-2.7	May	-1.3
Aug	-4.1	June	-6.1
Sep	-1.3	July	-4.5
		Sep	-1.3

In Karapiro, the first Psa-V symptoms were seen in March 2012 on Orchard C, and were thought likely to have appeared in autumn 2012 in Orchard D. However, no samples were collected or tests taken at this time. Higher rainfall in December 2011, coinciding with canopy management work could have increased the likelihood of infection occurring during the summer/autumn in 2012. Secondary symptoms were seen on both orchards by October 2012 when infection was confirmed by lab tests in the Karapiro area. Therefore, it seems unlikely that weather events in July and August 2012 (where heavy rain is indicated in the graph below) contributed to initial infection on Orchards C and D.

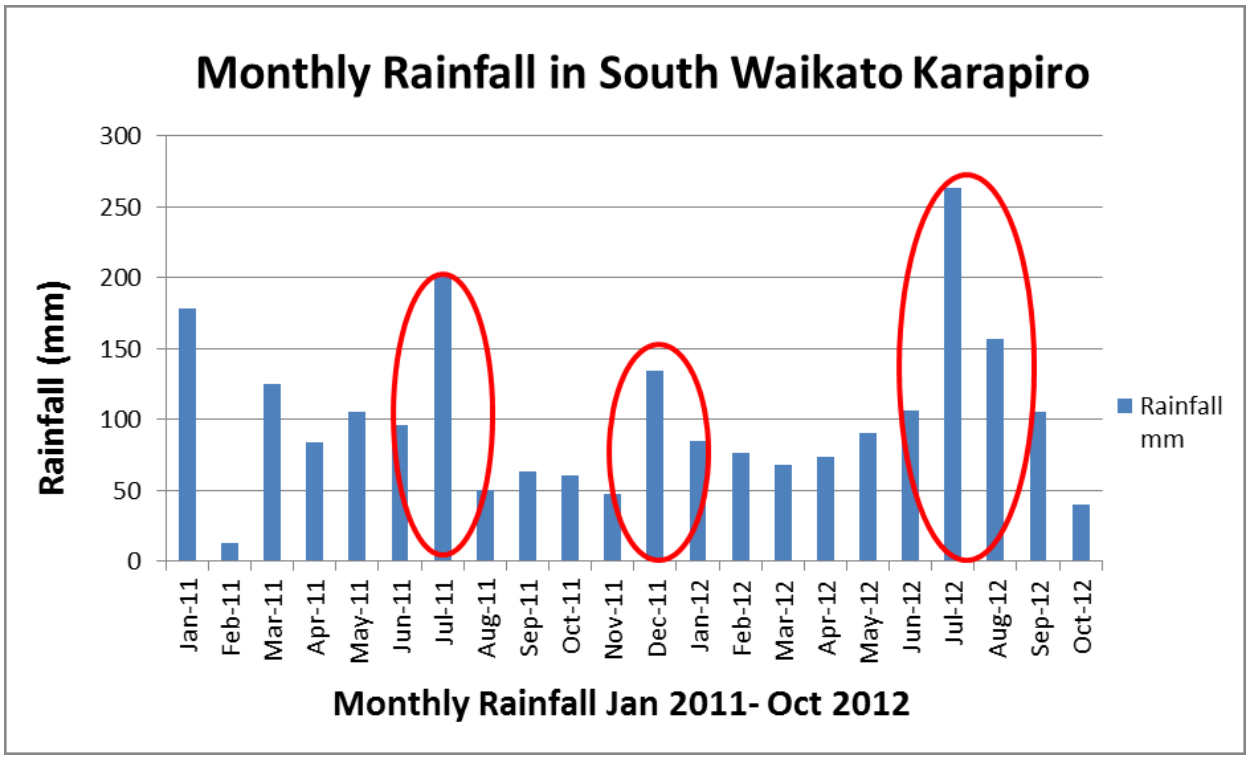


Figure 5: ***Rainfall data from a weather station on Orchard D in Karapiro***

There is no weather data specific to the Paeroa area available for this analysis. Secondary Psa- V symptoms, including cankers and rust, were seen at Orchard G in spring 2012. However, a 'not detected' test result was returned after samples were taken. A visual confirmation of Psa-V has been made for this orchard. Orchards E and F linked to this site through geographic proximity, shared staff and equipment, are Psa-V positive—confirmed by lab testing. These factors and others, such as budwood movement from an infected area into one of the three orchards at Paeroa, and concurrent management of those three orchards are seen as a more likely cause of initial infection than the possibility of wind borne inoculum.

4.3 Spray diary analysis

A spray diary analysis was undertaken for orchards in the pathway assessment (Figures 6-8). Where spray lines were not entered into Spray Diary for the 2011-2012 harvest season, information was augmented by discussions with growers/managers about number of sprays applied. The resulting graphs were available for six of the eight orchards assessed. It is clear that the majority did not have sufficient spray coverage to protect vines from weather events that could have contributed to significant Psa-V infection. Either in May-July 2011, December 2011 or in July-August 2012.

The general trend was for growers to have applied some type of protectant spray in winter or spring 2011, and then with the exception of Orchard B apply few protectant sprays until the following spring 2012. The most commonly sprayed Psa-V protectant was copper, followed by Actigard™. The assessment revealed that spray coverage during the 2011-2012 period could have been insufficient to protect orchards from either wind dispersed Psa-V infection or repeat localised infection once orchards had become infected. It is highly likely some local dispersal of Psa-V has occurred.

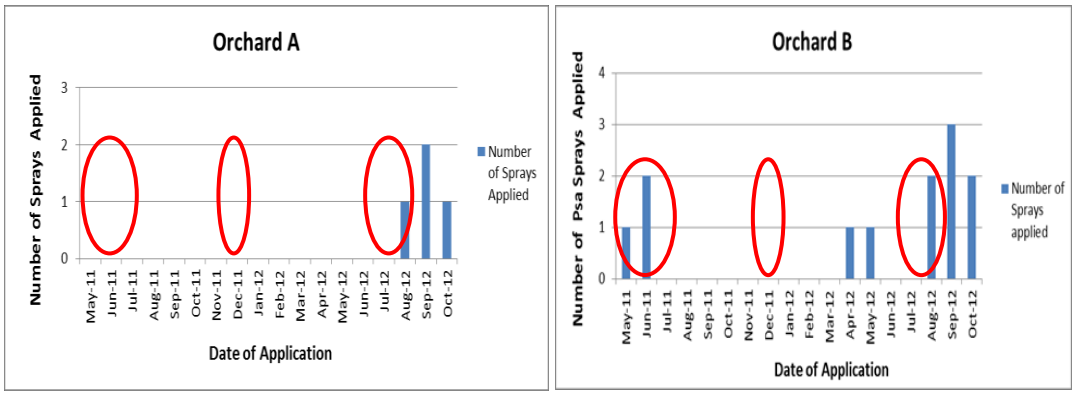


Figure 6: ***Group 1 Orchards A and B in Te Awamutu***

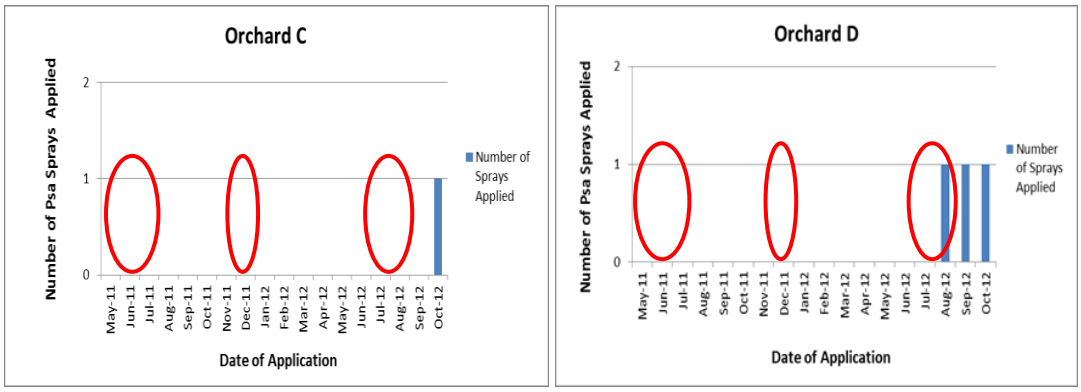


Figure 7: ***Group 2 Orchards C and D in Karapiro***

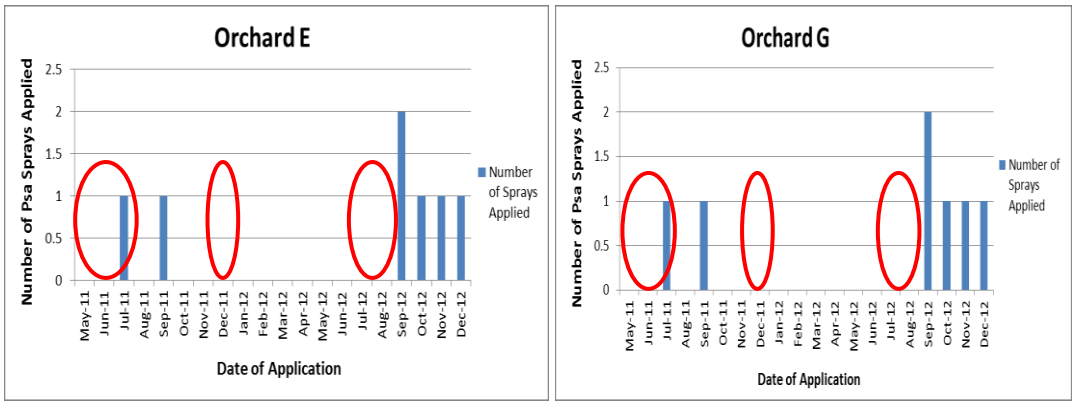


Figure 8: ***Group 3 Orchards E and G in Paeroa. Red circles denote main weather risk periods.***

Likelihood of wind dispersal: Assumption is made that wind dispersal is most likely up to 10km. Distances between Waikato orchards and the next nearest infected orchard are between 14-64km. Therefore, likelihood of wind dispersal is unlikely. Weather would have contributed to Psa-V spread in 2011, with prolonged rainfall and low temperatures. Spray programmes were insufficient to provide protection from weather dispersed Psa-V in Waikato region in 2011 or 2012.

5.0 Assessing pathway 2: plant material/movement

Foreknowledge: *Infected (knowingly or unknowingly) plant material moved from an infected orchard onto an uninfected orchard is one of the strongest known pathways for the spread of Psa-V. However, tracking this is difficult.*

Aim: Collect a full record of plant material that has moved onto, and off, any of the affected orchards—identifying the source orchard and its current Psa-V status to determine the risks associated with shared plant material.

5.1 Symptom expression in budwood plant material

There has been little research looking at the Psa-V symptom expression of infected budwood after it has been grafted. Infection in budwood is most commonly vascular in nature and likely asymptomatic at the time of grafting. There have been cases of stored budwood exhibiting severe Psa-V symptoms, with white toothpaste-like ooze seen in refrigerated scions in Te Puke.

Observations in orchards with suspected asymptomatic budwood show it can lead to cane dieback, cankers and bud rot, stunting of new growth and overall less vigorous growth of new shoots and canes.

Some of these symptoms, particularly stunting of new growth, and less vigorous growth of shoots and canes, can also be associated with poor graft take. It can be difficult to distinguish between effects in the early stages after grafting. Difficulty also exists distinguishing between budwood infection, and infection occurring from inadequate hygiene during grafting through tools. Insufficiently protected new grafts would also be susceptible to infection, particularly when weather events wash off waxes and pruning paints—again, making it difficult to distinguish between latent infection in plant material and infection via large grafting wounds and exposure to aerosol Psa-V.

Future research needs include quantifying the symptom expression and speed at which symptoms occur from infected budwood.

5.2 Inward and outward budwood movements on assessed orchards

The following diagram (Figure 9 below) shows inward budwood movements for the eight orchards assessed. A blue colour indicates no budwood movements occurred onto these orchards over the past two years (Spring 2010-Spring 2012). Yellow indicates budwood was received from a low-risk region, and red indicates budwood was received from a high-risk region.

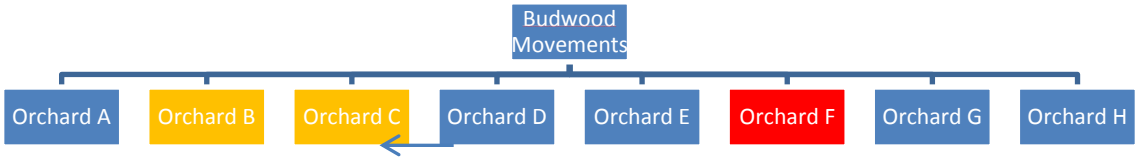


Figure 9: ***Budwood and plant movements in Orchards A-H from spring 2010-spring 2012.***

Orchard B received 10 Hort16A seedlings from Kerikeri in 2011 to replace vines which had been previously cut out due to other diseases. The seedlings did not come from the single infected orchard in Kerikeri. Therefore, the likelihood of infection from Hort16A seedlings is considered low. At the time of interview there were no symptoms in these seedlings, and none have been reported since.

Budwood movements were recorded for Orchard F in 2011 and 2012. Gold3 budwood was supplied from Zespri in winter 2011. The red colour indicates movement of non-Zespri male M33 budwood from Coromandel region in winter 2012. Currently 44 percent of hectares in Coromandel region are identified as Psa-V positive (based on each KPIN with a positive result). The likelihood of infection from male M33 budwood moved, and grafted into Orchard F in winter 2012 is considered high. However, there is currently no symptom expression on the Gold3 block on Orchard F. This block will continue to be monitored and progression of symptoms followed if/when this block becomes infected.

Orchard C received three sticks of male Russell budwood from Kerikeri region in August 2012 and 40 Bruno seedlings from a nursery in Kaikohe, in 2011. Some male M33 budwood came from Orchard D in July 2012 and some G14 budwood, and four G3 scions from another orchard in the Waikato in 2011. Currently two percent of hectares in the Kerikeri region are identified as Psa-V positive. The budwood did not come from the single infected orchard in Kerikeri. Therefore, the likelihood of infection from male Russell budwood moved and grafted into Orchard C in winter 2012, and Bruno seedlings planted in 2011 is considered low.

Table 10. ***Inward budwood/plant material movement to orchards in Waikato assessment***

	Budwood	**Place of Origin**	**Psa-V status Place of Origin**	**Observations on recipient orchard**	**Risk Rating**
Orchard B					
2011	10x H16A seedlings	Kerikeri	Not Tested	No symptoms	Low
Orchard C					
2011	40x Bruno seedlings	Kaikohe	Not Tested	No symptoms	Low
2011	G14+ (4xG3) budwood	Waikato	Psa-V +ve	Symptoms after H16A +ve	High
July 2012	20x M33 budwood	Waikato	Psa-V +ve	Symptoms after H16A +ve	High
August 2012	3x Russell budwood	Kerikeri	Not Detected	No symptoms	Low
Orchard F					
2011	G3 budwood	Zespri (?)	Not Detected?	No symptoms	Medium
July 2012	M33 budwood	Whenuakite	Psa-V +ve	No symptoms	High

Around 20 sticks of male M33 budwood came from Orchard D onto Orchard C for grafting into G3, and G9 blocks in July 2012. These blocks started to show symptom expression with leaf spot subsequent to infection expression in Hort16A blocks.

Table 11. ***Outward budwood/plant material movement from Waikato assessment orchards***

	Budwood	**Psa-V status Place of Origin**	**Region receiving budwood**	**Psa-V status recipient orchards**	**Risk Rating**
Orchard C		Psa-V +ve			
July 2012	G3		Waihi	Psa-V +ve	Low
July 2012	M91		Waikato A	Not Detected	High
July 2012	M91		Waikato B	Psa-V +ve	Low
July 2012	Red budwood		Zespri		
Orchard D		Psa-V +ve			
July 2012	M91 & M33		Waikato C	Not Detected	High
July 2012	M33		Coolstore	?	High
July 2012	M33		Waihi	Psa-V +ve	Low
July 2012	M33		BOP	Psa-V +ve	Low

Monitoring observations in these blocks have not indicated a difference between M33 male infection and other male cultivars—ie, M91. Monitoring will continue where M33 wood has been grafted and observation of symptom expression if/when infection occurs will be recorded.

G14 budwood and four sticks of G3 budwood were received from another orchard in the Waikato in winter 2011. Since the initiation of this assessment, that orchard has become Psa-V positive.

No other inward budwood movements are known. Budwood had been moved from Orchards C and D to other orchards in both Waikato and Bay of Plenty. Tables 10 and 11 above show the links of both inward and outward budwood movements. Risk ratings are given in at the right hand side of each table. High risk is indicated in red, medium risk in yellow and low risk in blue.

Because of the lack of understanding around the timing and progression of Psa-V symptoms from infected budwood, and the current lack of Psa-V symptoms in budwood grafted within the last two years on the affected orchards, it is difficult to establish whether any budwood has led directly to entry and infection of Psa-V into the Waikato. Time and monitoring of grafted vines, may answer this question at some point in the future.

Likelihood of dispersal through plant material: Infection in budwood most commonly vascular in nature and likely asymptomatic at time of grafting. Difficult to distinguish latent infection from infection during, or after grafting. Most significant budwood risk M33 males from Whenuakite in Coromandel. Monitoring has not revealed evidence of budwood Psa-V link. Likelihood of dispersal via plant material is unknown.

6.0 Assessing pathway 3: contractor movements

Foreknowledge*: Any work on vines that creates wounds, provides an entry point for Psa-V—either by means of air-borne inoculum or through direct contact with contaminated tools. Good orchard/contractor hygiene practices may reduce this risk. Contractors who have previously worked in regions infected with Psa-V, such as the Bay of Plenty, then work in a region where Psa-V has not been detected, pose a higher potential risk, and were subjected to a more vigorous follow-up process; in contrast to contractors who have been sourced locally within a Psa-V 'detection free' region.*

Aim*:* Identify key contractors and/or materials that have been supplied locally or from the Bay of Plenty. Account for their involvements in the affected orchards, specifically addressing the following.

- Where have they come from and what tools have they brought with them?
- What were their hygiene practices?
- What block did they start in on the orchard?
- Did they ever work in wet conditions?
- Where did they go after working in an (now known to be) infected orchard?
- What block did they start in following work done in a known Psa-V affected orchard?

6.1 Contractor links to Psa-V regions

There were only two contractors interviewed or recorded who had any links to regions outside of the Waikato. One group of contractors came specifically into the Waikato for a four to six month period each year to work on one orchard exclusively. They were based in South Auckland, but did not work in kiwifruit orchards in that region. The second contractor with links to other regions worked in the Waikato, Bay of Plenty and Coromandel.

Several contractor groups worked widely within the Waikato and were contracted to multiple orchards—through packhouses, or contracting companies. Orchard D had a contractor group which worked within 10 orchards, in a wider grouping of 35 orchards, in the Waikato, owned and managed by the same people. Hygiene protocols specific for Psa management were implemented at different times by different contractors, which reflected varying understanding of when Psa-V was first detected in Te Puke.

6.2 Risk factors in contractor practice and movement

Hygiene protocols and practices

Implementation of standard and consistent hygiene protocols ranged from December 2010, early 2011, June 2011 and Oct 2011. There was no vine to vine hygiene practiced by any contractors during winter or summer pruning, thinning or harvest. Tools were either 1) put in a bucket of steriliser every 2 hours, 2) at every break, or 3) every morning before work began.

Some orchard owners worked in Hort16A blocks and had contractors working only in Hayward blocks. Hygiene in these cases on Hort16A blocks was similar to, or less than standard contractor practices.

In the case of mulchers, tractors and spray equipment shared between orchards in this assessment, and other secondarily linked orchards, equipment was hosed down and water blasted but not sanitised in some cases until as late as spring 2012. It is highly likely that hygiene practices and protocols were not implemented early enough, and not followed as strictly as they could have been, to prevent the further dispersal of Psa-V in the Waikato region via human movement.

Of the seven orchards in the 'at risk' KPIN list compiled based on contractor movements, four orchards have not been tested for Psa-V, because no symptomatic material has been seen. Of the three orchards that have been tested, all had 'not detected' results. It is seen as likely that 'not detected' results could become positive within the next six months as symptom progression continues. Those 'at risk' orchards will be monitored and growers, managers and packhouse representatives are encouraged to test as soon as possible from first Psa-V symptom expression.

Weather conditions

Several contractors made the comment that it was very difficult to operate in the Waikato during autumn through to spring without occasionally having to work in rainy or damp conditions. The high proportion of fog, showers and heavy rain in this region meant that all contractors had worked in wet conditions—particularly during 2011 which was a wetter than normal year, and in 2012.

Some weather events during 2011 may have increased the likelihood of spreading Psa-V within the region once it had become established. A key risk period for the Te Awamutu area was harvest during May 2011. Harvest was disrupted multiple times on Orchard B due to rain. After harvest in May 2011 a shelter was trimmed on Orchard B at the same time that pruning was being undertaken. During this period, rain also interrupted work and would have increased the likelihood of Psa-V being transmitted to and from orchard equipment and heavy machinery, through grass soil and plant material sticking to the machinery.

Heavy orchard machinery

The shelter trimmer mentioned above then went from Orchard B to Orchard A within 24hrs and began work. The trimmer was cleaned by air blasting and water washed down prior to shifting orchards. There were no sanitising products used at this time. On Orchard A there was significant rainfall and the machinery broke down. The machinery stayed on the orchard for several days in rainy and windy conditions before a part was replaced and work resumed.

In the days during and following trimming activity on Orchards A and B, Orchard H, Orchards E, F and G and one of the linked orchards associated with Orchard D in Karapiro used the same contracted machinery. Orchards also linked to the assessment via budwood movements (Table 10) in the Waikato and an 'at risk' KPIN in the Bay of Plenty also used the same shelter trimmer within a five week period during May-June 2011. In June 2011 the contractor purchased a water blaster and sanitising chemicals and used them from this date onwards.

The hardest spots on the machinery to clean were between the wheels and underneath. Before the purchase of cleaning tools and chemicals, the shelter trimmer would not have been cleaned adequately to mitigate the risk of Psa-V being transported via soil, or plant material in hard to reach places.

In 2012, the same contractor worked at Orchard B on 19 April, then went to Orchard H on the 20 April. Orchards in Karapiro, Cambridge and Whenuakite in the Coromandel were also trimmed during May and June 2012. All those orchards have subsequently become Psa-V positive. It may not be possible to clean such a large piece of equipment to the extent needed to reduce risk to negligible. In the future, it is recommended protectant sprays be applied to vines before the movement, and use, of heavy machinery and equipment on orchards where Psa-V has not been identified.

Likelihood of dispersal via contractors and or orchard machinery: Only two contractors had links with other orchards outside the Waikato. Links were to South Auckland, Coromandel and Bay of Plenty. Hygiene protocols were not implemented early enough to have prevented transmission of the bacteria. High risk orchard activities were carried out during wet weather. Movement of orchard machinery is considered the most likely pathway for spread of Psa-V within the Waikato.

7.0 Conclusions

- Given the appearance of Psa-V like symptoms from as far back as 2010 in Te Awamutu, it is likely Psa-V has been present in the Waikato region for at least a year.
- There are three groupings of linked orchards where plant material, machinery or orchard management were shared between orchards within groups. Therefore, none of the infection points appear to be independent of each other.
- No single infection event has been identified, but a series of events and risk factors have highlighted possible pathways of infection.
- It is considered unlikely that any of the orchards assessed became Psa-V positive following weather events dispersing Psa-V inoculum from other adjacent regions.
- The dispersal of Psa-V from bees during pollination is not considered a contributing factor to the entry or spread of Psa-V in the Waikato.
- Contractor movements, with heavy machinery and equipment are likely to have aided in the dispersal of Psa-V through the Waikato.
- Concurrent management of linked orchards and use of high risk equipment between orchards is an important factor in risk and spread of infection.
- Budwood movement may have played a part in the dispersal of the bacterium into or across the Waikato, but evidence to support this is likely to be confounded by mitigating factors in symptom expression—ie, difficulty distinguishing Psa-V entry timing into grafted vines.
- Standard orchard hygiene practices and protocols were not implemented early enough.
- Protective sprays and preventative programmes were not adequate to have prevented spread of Psa once it had arrived in the Waikato.

8.0 Bibliography

Anderson, M. Cornish, D, Curtis, C. DeAstarloa, D. Everett, K. Gea, L. Goodwin, M. Hoeata, K. Horner, I, Manning, M. Martin, P. Pushparajah I.P.S. Rees-George, J. Taylor, M. Tyson, J. Vanneste, J, and Yu, J. Edited by Kay, S.J. & Dunn, J. (2011). Management of Psa short term – progress report September 2011. VI1175

EPPO (2011) Express pest risk analysis for *Pseudomonas syringae* pv. *actinidiae* 11-17321. European and Mediterranean Plant Protection organisation.

Hirano, S.S. & Upper, C.D. (2000) Bacteria in the leaf ecosystem with emphasis on *Pseudomonas syringae* – a pathogen, ice nucleus, and epiphyte. Microbiology and Molecular biology reviews 64: 624-653

Horner, I.J; Everett, K.R; Manning, M.A; Tyson, J.L; Curtis, C.L; Fullerton, R.A; Larsen, N.J; Pushparajah, I.P.S; Rees-George, J; Vergara, M.J; Vanneste, J.L; Cornish, D.A; Moffat, B.J; Oldham, J; Yu, J (2011) Survival of Psa in the orchard environment. A confidential report prepared for Zespri Group Limited. Ref VI 1257

Stefani, E. & Giovanardi, D. (2011) Dissemination of *Pseudomonas syringae* pv. *actinidiae* through pollen and its epiphytic life on leaves and fruits. Phytopathologia Mediterranea 50: 489-496

Vanneste J.L; Moffat, B.J; Yu, J; & Cornish, D.A. (2011) Survival of *Pseudomonas syringae* pv. *actinidiae* on evergreen shelter belts. New Zealand Kiwifruit Journal, Psa Scientific Edition October 2011 Pp 14-15

Vanneste, J.L; Yu, J; Cornish, D.A; Max, S; Clark, G (2011) Presence of *Pseudomonas syringae* pv. *actinidiae*, the causal agent of bacterial canker of kiwifruit, on symptomatic and asymptomatic tissues of kiwifruit. New Zealand Plant Protection 64: 241-245

VLS (2012) Mulching Trials: To identify an effective means of reducing the Psa-V inoculum source from cut out vine material. VLS Project No E2012-09/11 Verified Lab Services

9.0 Acknowledgements

The author would like to acknowledge the following people for their contribution towards this report:

- Affected growers for their co-operation and permission to visit and assess orchards.
- Packhouse representatives of affected orchards.
- Contractors, beekeepers and orchard managers who participated in interviews.
- KVH regional managers for Waikato.
- KVH staff for field assessments, maps, and review of the document.

10.0 Appendix 1

What we understand about Psa-V infection in orchards:

- Live Psa-V has been found in the canes, leaders, trunks and roots of female Hort16A vines and in the canes, leaders and trunks of Hayward and *A. deliciosa* male vines.
- The graft does not impede progress of the bacterium. There is potential for resident populations to be present in remaining rootstock and stumps prior to re-grafting.
- Psa-V has also been detected in vines with no visible symptoms and can be present in asymptomatic tissues quite remote from diseased tissues.
- The bacterium appears to be able to colonise the entire vine relatively rapidly. Alternatively, it may survive at populations below the limits of detection, established in asymptomatic tissues. A rapid increase in bacteria can occur after a pre-disposing event, plant growth stage, or when a critical population size is reached.
- The distribution of the pathogen within the tissues across any cross section of the plant is not regular and it may be isolated from some samples and not others taken from the same cross section.
- Psa-V survives in the leaf litter on the orchard floor for up to 15 weeks. It also appears to survive in the cane prunings from leaf fall and winter pruning for at least eleven weeks. It is a potential source of inoculum within the orchard the following spring.
- It is unknown if Psa-V survives in summer pruning debris for a similar length of time.
- Laboratory experiments have shown that Psa-V is able to survive epiphytically on the leaf surface of both its kiwifruit host Hort16A and a non-host plant—tobacco.
- Shelter species *Cryptomeria japonica*, *Pinus radiata*, *Casuarina cunninghamiana*, *Salix* sp. *Poplus* sp. and under canopy species *Crepis* sp and *Carex* sp were inoculated with Psa-V and tested for Psa-V survival over a series of days. Psa-V survived epiphytically on the various species from a few hours to a few days.
- The survival of Psa-V on other species was dependent on the initial bacterial population present on the plant. It seems that Psa-V populations always decline on these species, suggesting that Psa-V is most probably surviving and not multiplying
- A mulcher was swabbed 18 days after use in an infected Hort16A orchard and Psa-V was detected by PCR on all swabs. However, none produced viable colonies. This was after water blasting and sterilisation and storage in a dry shed. Given the duration since use and the dry conditions the lack of viability is not surprising. However, there is clearly a very significant risk of infection from recently used equipment.
- At this point in time it has not been determined what the threshold for infection by Psa-V is in an orchard—ie, what numbers or concentration of the bacterium are needed to enable infection to occur in the kiwifruit vine.

10.1 Appendix 2

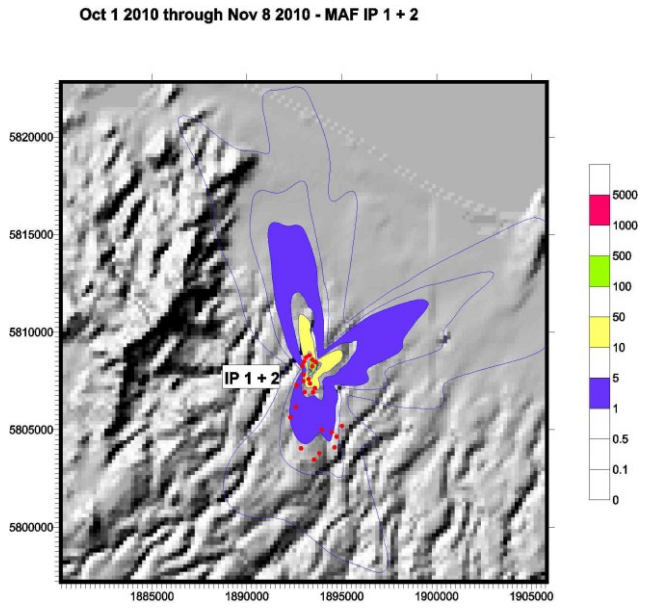
The likelihood of weather dispersal as a vector for Psa-V into the Waikato:

Research undertaken in Emilia Romagna (Italy) by Francisco Spinelli, showed Psa-V was capable of being dispersed by wind and rain over a distance of up to 1km. A further study performed in Italy on the progression of the bacterium in an area with gold kiwifruit orchards (Vanneste *et al.* 2011b in press) indicated a dispersion capacity of around 10km from the initial infected orchards in May 2009 and June 2010 (the authors note that Spring 2010 was characterised by cold wet weather and comment that the spread was rapid) EPPO 2011.

In November 2010 NIWA took three early Psa-V infected properties in Te Puke as base locations from which wind plume model scenarios were built. The Plume Dispersion Emergency Modelling System (PDEMS) which was part of MAF Biosecurity's Expert System for Foot and Mouth response was utilised. Assumptions made for the modelling included the threshold wind speed required for dispersal of Psa-V as an aerosol, the concentration of bacterial inoculum in the orchards and the type of weather conditions in combination required for its dispersal (i.e. wind and rain).

A time period in September where prolonged wind was experienced in the Bay of Plenty was used as the most likely initial dispersal weather event, and models were built based on this data. More models were put together using data from October to November 2010 (pictured below), and a January weather event in 2011. The graph below depicts the likely dispersal pattern of Psa from a combination of two locations IP1 and IP2.

Concentration of bacteria along the right hand axis of the graph on a log scale is indicated with colour bars, where blue is the least dense concentration and pink at the top is the most dense concentration. Red dots indicate known infected orchards. The scale is based on the NZ Map grids, NZTM with northing and easting coordinates on the vertical and horizontal axes respectively, and is in 5 kilometre increments. The likely dispersal of Psa-V covers a distance of between 5-10km, and not more than 15km at 1-5 concentration values, but up to 20km for small concentration values of 0.1-0.5.



10.2 Appendix 3

Analysis of 'not detected' Psa-V test results:

There have been a number of reports in several regions of 'not detected' results for vines that have visual Psa-V symptoms. These 'not detected' results are often followed by a subsequent re-test and positive result within a short time frame. A smaller number of reports have also been made of asymptomatic plant material returning a positive result. The smaller number reported in this category is likely due to tests being taken only when symptoms are apparent, and the cost of testing.

Because of this trend in other regions, a review of Psa-V test results was undertaken. Psa-V testing results from August 2011 to July 2012 were analysed for four localities within the Bay of Plenty. Katikati/Waihi, which were combined as one area, Tauranga, Opotiki and Whakatane. The analysis looked at the number of samples where retests within two months, three months and six months confirmed a positive result after an initial 'not detected' result. Over the course of the incursion and within the previous year and a half in Waikato, several 'not detected' results were also recorded, where a subsequent positive was found within six months. These results are also recorded (See Table 12).

Table 12. ***Testing for Psa-V—assessment of testing and retesting within regions. All results in subsequent months after an initial 'Not Detected' result are Psa-V positive.***

August 2011-July 2012	Not Detected	Re-test within 2 months	Re-test within 3 months	Re-test within 6 months
Katikati /Waihi	10	4	0	6
Tauranga	28	11	3	14
Opotiki	18	12	2	4
Whakatane	21	14	2	5
August 2011-Nov 2012				
Waikato	3	2	0	1

Several issues have been identified from the review and are relevant for the Waikato pathway assessment. Two orchards in the Waikato incursion saw what they thought were Psa-V symptoms prior to a positive test result. One orchard took a sample in spring 2011 and got a not-detected result. The next test was not taken until spring 2012. The other orchard saw symptoms six to eight months prior to the first positive test result, but did not get a test at the time of first symptom appearance. If a second sample had been taken earlier, Psa-V might have been identified in the region much earlier.