

Leafroll virus in vineyards: modelling the spread and economic impact

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July 2004

Report to New Zealand Winegrowers Limited

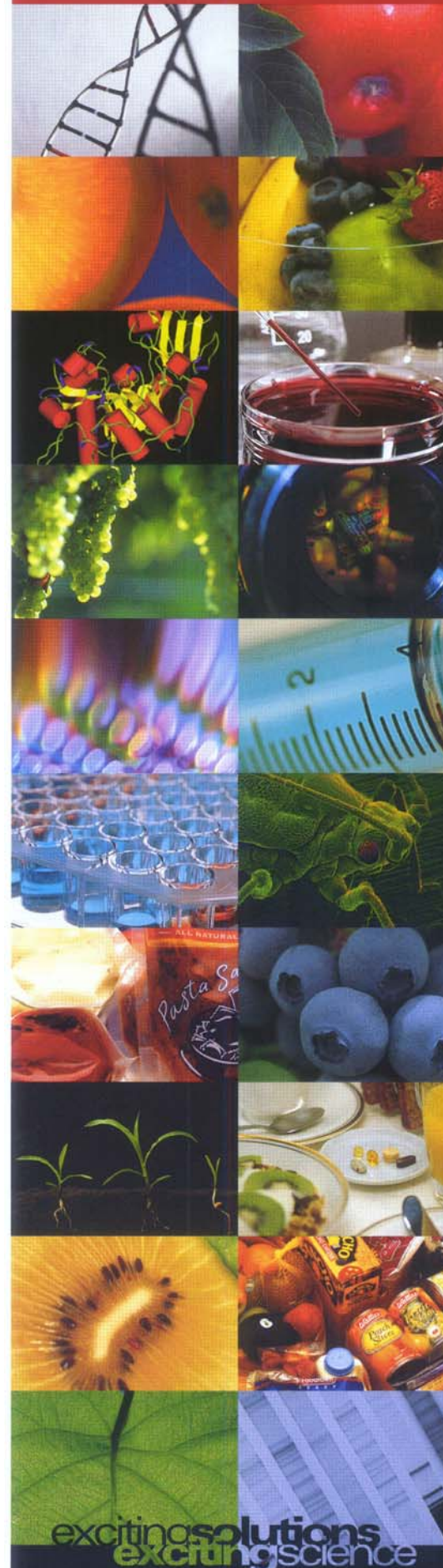
HortResearch Client Report No. 12795

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Date: 21 December 2004

Date: 21 December 2004

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EXECUTIVE SUMMARY

Leafroll virus in vineyards: modelling the spread and economic impact

J.T.S. Walker, J.G. Charles, K.J. Froud and P. Connolly

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Grape leafroll virus is the most significant virus disease found in vineyards in all grape producing regions of the world and consists of a complex of viruses that belong to the closterovirus group. Within this complex, grapevine leafroll associated virus type III (GLRaV-3), is one of the most economically damaging and widespread viral diseases of winegrapes world-wide. It is also one of the biggest threats to the production of high quality wines in New Zealand; it is now widespread in vineyards in the North Island and has become increasingly common in upper South Island vineyards. The spread of GLRaV-3 is rapid and, in the presence of mealybugs that act as vectors, a newly planted GLRaV-3 negative indexed block can become 100% infected within 6 years.

Charles et al. (in prep.) monitored the annual spread of GLRaV-3 and the associated mealybug populations between 1998 and 2003 within a re-planted Waikato block of virus-indexed plants that was established in 1996. Our study used their data to examine the potential for modelling spread of GLRaV-3 in this vineyard. We used this information on virus spread to investigate the economic impact of virus infection to vineyard profitability and longevity.

We examined the use of tree-based and probabilistic models to describe the relationship between mealybug activity and the nature of virus spread through the vineyard. While there was a macro-spatial trend in the spread of GLRaV-3, it was difficult to show this mathematically because there were highly variable results between the seasons. Mealybugs have been implicated in transfer of virus infection and this has been shown by the strong relationship between mealybug numbers and infection levels in the following season (Charles et al. in prep). Correlations between mealybug incidence and subsequent virus infection lack the sophistication of a model and are ultimately less useful in setting mealybug spray thresholds. While the use of this data to model the relationship between mealybugs and the spread of virus was not possible we were able to model the changes in the distribution of the virus itself for the period 1998-2003. Three scenarios of virus spread (low, intermediate and high infection rates) within this vineyard were modelled over the six seasons for which data was available. Virus infection was predicted to spread rapidly within the vineyard over time with 50% infection predicted to occur in years 6, 8 and 11 for high, intermediate and low infection rates respectively. All three scenarios are predicted to reach 90% vine infection in years 11, 12 and 15 respectively.

We used this information on the predicted spread of virus in this vineyard as the basis for an economic analysis of likely costs of virus spread in New Zealand vineyards where susceptible varieties are exposed to the risk of GLRaV-3 infection. A vineyard producing premium wine grapes could be expected to have a marked decline in profitability as infection increases. The decline in revenue was almost linear in the highest rate of infection so that by year 15, with all vines infected, net revenue per ha was reduced by 70%. A 50% reduction in net revenue was predicted to occur in years 8, 10 and 11 in high, intermediate and low virus spread scenarios respectively. The three virus spread scenarios were used to predict the cumulative cost of GLRaV-3 infection. This cost exceeded \$10,000 per ha by years 7, 9 and 12 and \$30,000 per ha by years 12, 15 and 17 in high, intermediate and low spread scenarios

respectively. In practical terms this means that profitability of blocks infected at these rates would be sufficiently impacted (financially) around year 11 to justify re-planting of the vineyard.

Once virus infection exceeds about 20% of vines, this annual cost exceeded \$1000 per hectare by year 3, 6 and 9 for the three infection scenarios and increased rapidly thereafter. It is economically justifiable to invest at least 50% of this sum annually to prevent this scenario developing. This includes preventative measures i.e. mealybug control and replacement of infected vines. Given difficulties in achieving high levels of mealybug control in vineyards and the cost of replacing infected vines, considerably more should be invested in reducing or eliminating the sources of initial infection. This not only includes comprehensive virus indexing of plant material but also additional measures to reduce infection in existing vineyards. More effort must go into identifying and managing the sources of initial infection in new plantings because once GLRaV-3 has established within a vineyard its rapid progression is almost inevitable, threatening the economic life of both infected block and adjacent plantings.

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INTRODUCTION

Leafroll virus is the most significant virus disease found in vineyards in all grape growing regions of the world and consists of a complex of viruses that belong to the closterovirus group. Within this complex, grapevine leafroll associated virus type III (GLRaV-3), is one of the most economically damaging and widespread viral diseases of winegrapes world-wide (Bovey et al. 1980). It is also one of the biggest threats to the production of high quality wines in New Zealand (Petersen and Charles 1997). This virus is common in vineyards in the North Island but has become increasingly common in upper South Island vineyards (Charles and Jordan 1993). The spread of GLRaV-3 in New Zealand is rapid and a newly planted GLRaV-3 negative indexed block can become 100% infected within 6 years (Jordan et al. 1993). The initial presence of GLRaV-3 in New Zealand vineyards was due to infected propagative material, however, the major means of GLRaV-3 spread in New Zealand currently is by vector transmission involving three species of mealybug; *Pseudococcus longispinus*, *P. calceolariae* and *P. viburni* (Petersen and Charles 1997, Cabaleiro and Segura 1997a). It is unknown if scale insects that are present in New Zealand vineyards (e.g. the soft scale insect, *Pulvinaria vitis*) are capable of spreading GLRaV-3.

GLRaV-3 does not destroy infected vines but does cause a decline in both yield and quality of grapes with symptoms such as delayed ripening, reduced yield and depressed berry sugar content that reduce wine quality (Mannini and Credi 2000). Crop reductions of 40-60% are commonly reported with fewer and smaller bunches from infected vines. Leafroll virus lowers the quality of the grapes by delaying the accumulation of sugars and lowering the production of anthocyanins and causing up to a 50% loss of pigment concentration in red wine varieties (Over de Linden and Chamberlain 1970). GLRaV-3 is therefore a particularly serious problem for red wines. The disease affects all grape varieties in the same manner but is most easily seen in red varieties and Chardonnay. It is less apparent in Sauvignon Blanc and is virtually symptom-less in Chenin Blanc. It can therefore be difficult to recognise the extent of potential virus infection risk to new planting presented by apparently symptom-less varieties in adjacent blocks within an established vineyard.



Figure 1. Grape leafroll virus symptoms in grape foliage

Propagation material and GLRaV-3 spread within vineyards

Closteroviruses such as GLRaV-3 are restricted to phloem tissue so are easily transmitted by grafting and propagation material has been recognised as an important means of spreading GLRaV-3. Rootstocks are symptom-less carriers of leafroll virus and have probably contributed significantly to current levels of virus infection in established plantings. Infected rootstock and scion plant material in a new block can lead to infection through the entire vineyard and spread to adjacent vineyards. Initial virus levels can be minimised through the use of certified virus-free rootstocks and scion material, and virus indexing programmes are increasingly used to ensure that new vineyards are established using high health plant material. Despite best attempts to supply growers with high health vines through virus indexing, nursery plants may not be 100% free of virus infection. This low incidence of residual infection may provide the primer for re-infection of new plantings and the subsequent spread of GLRaV-3. Any vines in new plantings showing symptoms within the first few years should be replaced as soon as symptoms become apparent to prevent further spread.

The role of mealybugs in the re-infection of vineyards

GLRaV-3, in the presence of mealybugs as vectors, can spread from a low incidence to almost completely infect a new vineyard within the 6 years (Jordan et al. 1993). Ensuring that planting material is free of the mealybugs, or separated from adjacent sources of mealybugs and infected vines is also important to prevent the rapid infection of new plantings. Mealybugs can survive for some weeks on residual vine roots and shoots once infected vines have been removed from a vineyard, but also commonly live on a number of other weedy host plant species that are present within most New Zealand vineyards. Cultivation and hygiene prior to planting are therefore important to minimise the risk of re-infection within a new vineyard.

The spread of GLRaV-3 within a re-planted vineyard has been documented in Charles et al. (in prep.). Their study monitored the annual spread of GLRaV-3 and the associated mealybug populations between 1998 and 2003 within a re-planted block of virus-indexed grafted plants that was established in 1996. In our study we used their data to examine the nature of GLRaV-3 spread in the vineyard. We used this information to investigate the potential in this data for modelling mealybug activity and the associated spread of GLRaV-3, and the economic impact of virus infection on vineyard profitability and longevity.

METHODS

The spread of GLRaV-3 within the re-planted vineyard and the associated mealybug populations between 1998 and 2003, documented by Charles et al. (in prep.), is illustrated in Figure 2. The 26 row block, re-planted in 1996 was centred within a large 60 row block of mealybug (predominantly *Pseudococcus longispinus*) infested, diseased vines. It was hence surrounded by both a source of GLRaV-3 infection and mealybugs as potential vectors for virus transmission.

This report provides data from two plots established in the vineyard. Plot A (West) consisted of vines within one row (22) of old, diseased Sauvignon Blanc and 4 rows of newly planted, disease-free Merlot (rows 23-26). Plot B (East) consisted of vines within 4 rows of newly planted, disease-free Chardonnay (rows 45-48) and one row (49) of old, infected Breidecker. The intervening rows between Plots A and B (rows 27 - 44) consisted of Merlot and are referred to as block C; there was no regular mealybug monitoring data from these disease-free vines but virus status data was collected annually within this area. Within each row, each plot contained ~50 vines within 10 post-post 'bays'. The ten bays contained approximately 1/3 of the vines in the row and were located approximately in the centre of the block. All vines in the block were grown on post-and-wire trellis, and were cane pruned.

Monitoring GLRaV-3 spread and mealybug activity

Presence of GLRaV-3 in each newly planted vine in Plots A and B was determined by double antibody sandwich ELISA (DAS-ELISA) (Clark and Adams 1997) using antibodies supplied by Bioreba AG (Basel, Switzerland). Either two mature leaves or two mature cane samples per vine were collected on each sampling occasion. The 3 main leaf veins (from leaf samples) or phloem scrapings (from canes) were ground in grape extraction buffer (500mM Tris (pH8.2), 1% PEG (MW 8,000), 2% PVP (MW 40,000), 140mM NaCl, 2% Tween-20, and sodium azide 0.2%), as described by Gugerli et al. (1984), but modified by Cohen and van den Brink (2000) to increase the Tween-20 concentration to 2%). Plates were read at 405nm using a plate reader (Labsystems multiscan bichromatic, Finland) and analysed kinetically over 2 hours.

The virus status of each new vine from Plots A and B was determined annually in late summer (March-May) from 1998-2003 and in Plot C from 1999-2003. Some young vines died during drought conditions in the early years of the trial and were replaced. Replacement vines were sampled as soon as they were large enough to remove foliage or cane material without hindering growth (when they were approximately two years old). In the final season (March 2003) an additional five bays of vines in each of rows 42-44 were sampled for ELISA testing.

Mealybugs on foliage were sampled approximately fortnightly during each season from 1996 to 2003. On each occasion two leaves were collected from the crown of each mature vine in rows 22 and 49 from Plots A and B respectively, and from the new vines when possible. Mean numbers of mealybugs per leaf were calculated for vines, bays and rows.

Modelling the relationship between mealybugs and the spread of virus

We considered two potential approaches to modelling the stochastic relationship between mealybug activity and virus spread:

1. The tree-based model

With this approach a classification tree is developed to predict the probability of a vine being infected or clean using a model of the form:

Response ~ Cultivar + Intra-row + Inter-row + Mealybug

where:

- Response takes the value 0 for a clean vine, and 1 for a newly infected vine. Infected vines stay infected, and thus a vine which is infected, but not newly so, is non-informative;
- Cultivar is either ‘Merlot’ or ‘Chardonnay’;
- Intra-row is the minimum distance within the row from the vine in question to a vine which was infected the previous year;
- Inter-row is the minimum distance across the rows from the vine in question to a vine which was infected the previous year;
- Mealybug is some measure of mealybug abundance – such as the maximum population in the previous year.

2. The probabilistic model

Two possible scenarios of leafroll virus transfer were considered with this approach: within row and between row. It was felt that the mechanism, and hence probability, of transfer would be different for each of these factors. Both probabilities are likely to be distance functions, with a decreasing probability of transfer as the distance from the infection source increased. We used Ordinary Runs Analysis (Campbell and Madden, 1990) to test for clustering of the distribution of virus-infected plants within the vineyard.

The economic impact of GLRaV-3 infection

The perception that GLRaV-3 is increasing in vineyards may in part be due to the restructuring of the wine industry and the increase in the red wine varieties that express visible symptoms. Delayed ripening, reduced yield and vineyard longevity are the primary impacts of GLRaV-3. While there is still some debate over the reduction in quality caused by GLRaV-3, any reduction in red pigmentation, brix and tannins are all important factors affecting the quality of red wines. In this assessment of the economic impact GLRaV-3 spread in a vineyard we have assumed that grapes from virus free vines are selected for premium wines and that grapes from infected vines are harvested separately for bulk wine production.

It is difficult to establish the market price for wine grapes as over the last four vintages price has been affected by weather, supply and quality. We made assumptions on prices and yields knowing that prices for grapes have been variable but higher than for 2000 as a consequence of the frosts that affected the 2001 and 2003 vintages. In this analysis of the economic impact of virus infection we used a 2002 Gross Margin analysis (Table 1, from the MAF Horticultural Monitoring Report 2002). For premium red wine grapes \$1800 per tonne was used in this analysis that is \$200-500 per tonne more than the 2000 vintage. Bulk varieties at \$500 per tonne were somewhat lower than the 2000 vintage.

Table 1. A gross margin analysis of premium and bulk wine varieties based on prices in the 2002 vintage.

Gross Margin Analysis for Wine Grapes (\$/ha)	
2002 Premium Wine Varieties	2002 Bulk Wine Varieties
Income: 6.3 tonnes/ha @ \$1,800/tonne	Income: 11.3 tonnes/ha @ \$500/tonne
11,340	5,700
Expenditure:	Expenditure:
Pruning	Pruning
1,400	400
Sprays/herbicides	Sprays/herbicides
1,210	1,210
Training/desuckering/shoot thinning	Training/desuckering/shoot thinning
2,080	1,600
Trimming	Trimming
300	300
Harvesting	Harvesting
378	684
Leaf plucking	Leaf plucking
400	150
Gross Margin	Gross Margin
5,470	1,356

Source: MAF Monitoring Report – July 2002

In this example we chose to use a vineyard with an established planting of a GLRaV-3 sensitive red variety that was fully producing at the outset of infection and then followed the infection levels predicted for Plot A (West), Plot B (East) and Plot C (middle), as discussed above. We assumed that sensitivity to GLRaV-3 infection would result in a 50% reduction in yield and that grapes from infected vines in premium wine grape vineyards would have a value equivalent to bulk wine grapes. We used the income and expenditure data for the 2002 vintage to determine the impact of virus on gross margins (Table 1) and examined the three infection scenarios for their impact on vineyard profitability. In this analysis we did not consider the return on capital investment. The price paid for land suitable for vineyards is dependent on location and is highly variable (~300%) while the costs of vineyard establishment are similar at ~\$27,000 per ha (MAF Horticultural Monitoring Report 2002).

RESULTS AND DISCUSSION

Most mealybugs in the vineyard were *P. longispinus*, although a few *P. calceolariae* were sometimes found. Mealybug numbers were always higher in the older, virus-source vines than in the young vines of Plots A, B and C. All mealybug data reported are for *P. longispinus*.

By 1998 virus had spread into 10% of the vines in the first row of Chardonnay (row 48), by 1999 infection in this row had increased to 56% and after 7 years to 93% (Figure 2). In comparison, early infection was more sporadic in rows 45-47; just 4% and 10% of vines were infected in row 45 by 1998 and 1999 respectively. By 2001 just 17% of plants in this row were infected but this jumped to 68% by 2002 following a 'high' mealybug season in 2000-2001 (Figure 3). The early and sporadic appearance of infected plants within rows 45-47 may have been a consequence of residual GLRaV-3 infection levels amongst the batch-tested, virus indexed plants. In contrast the movement of virus infection in rows 23-44 of the Merlot was slower with only 13-21% of vines infected in these rows after 7 seasons. These differences in the initial spread of the virus into the new planting could not be explained by either the direction of the prevailing wind or the counts of mealybugs (Figure 3) that were consistently greater in row 22 adjacent to the re-planted Merlot, than in row 49 nearest the newly planted Chardonnay.

The results showed a greater infection rate in the Chardonnay vines compared to the Merlot vines planted at the same time and suggest a significant difference in relative susceptibility of these two varieties to either GLRaV-3 transmission by mealybugs and/or susceptibility to mealybug infestation. Regardless of relative differences in varietal susceptibility, strong positive correlations were found between seasons with high mealybug activity (1997/98 and 2000/01) and the rate of virus infection in following seasons. The relationship between mealybug activity and the spread of virus infection within the vineyard is described in Charles et al. (in prep.), but the time delay of one season between mealybug infection of a vine and its detection by ELISA screening should be noted (Charles et al. in prep.). The study also revealed that initial GLRaV-3 colonisation of the new planting was slow and may have been linked to the relatively low numbers of mealybugs collected from the Merlot and Chardonnay vines over the first three years. During this period the vines had low numbers of leaves in summer and very little shelter in winter for adult females, which over-winter in crevices and under loose bark.

Wind may be an important means of GLRaV-3 infection spread. This virus is most effectively spread by first instar mealybugs and these crawler stages may also dispersed naturally by wind. Under certain conditions they are reported to deliberately 'launch' themselves into a wind current and may be carried considerable distances before coming to rest on a new host plant (or vineyard). The juvenile crawlers are also readily distributed on wind-blown leaves in autumn, by birds and on pruning and harvesting equipment, so that movement of people and equipment should also be from the most recent planting to older plantings with higher infection risks. This, together with closer vine-vine contact along rows, may have helped to facilitate the rapid spread of GLRaV-3 along rows (Figure 2, row 48) between 1999 and 2002.

Figure 2. Annual virus indexing for GLRaV-3 presence or absence from 1998 to 2003 in Merlot (rows 23-26) and Chardonnay grapes (rows 44-48) and final season indexing of rows 42,43 (Merlot) and 44 (Chardonnay). Circles represent a positive virus infection status of vines over the study period.

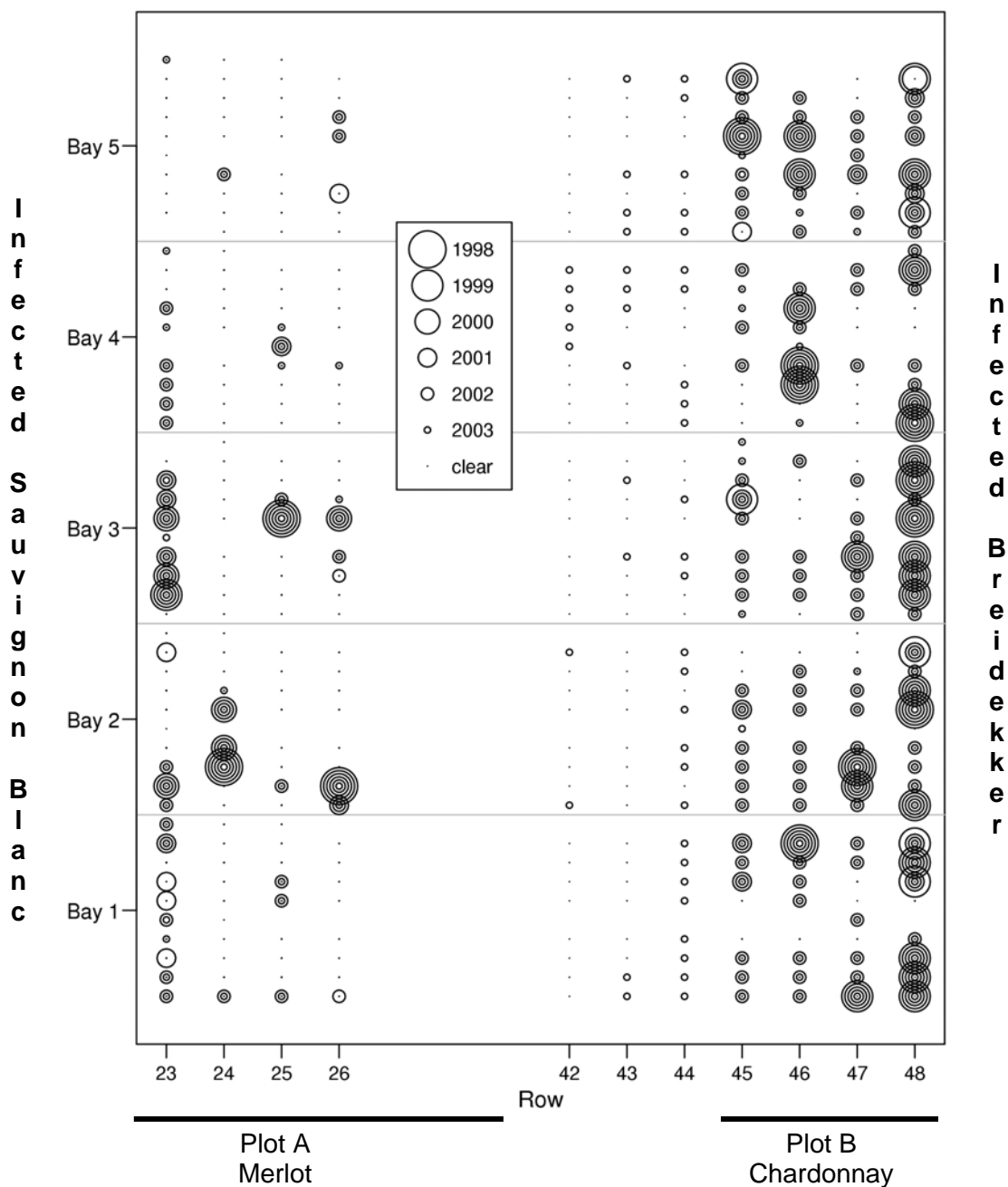
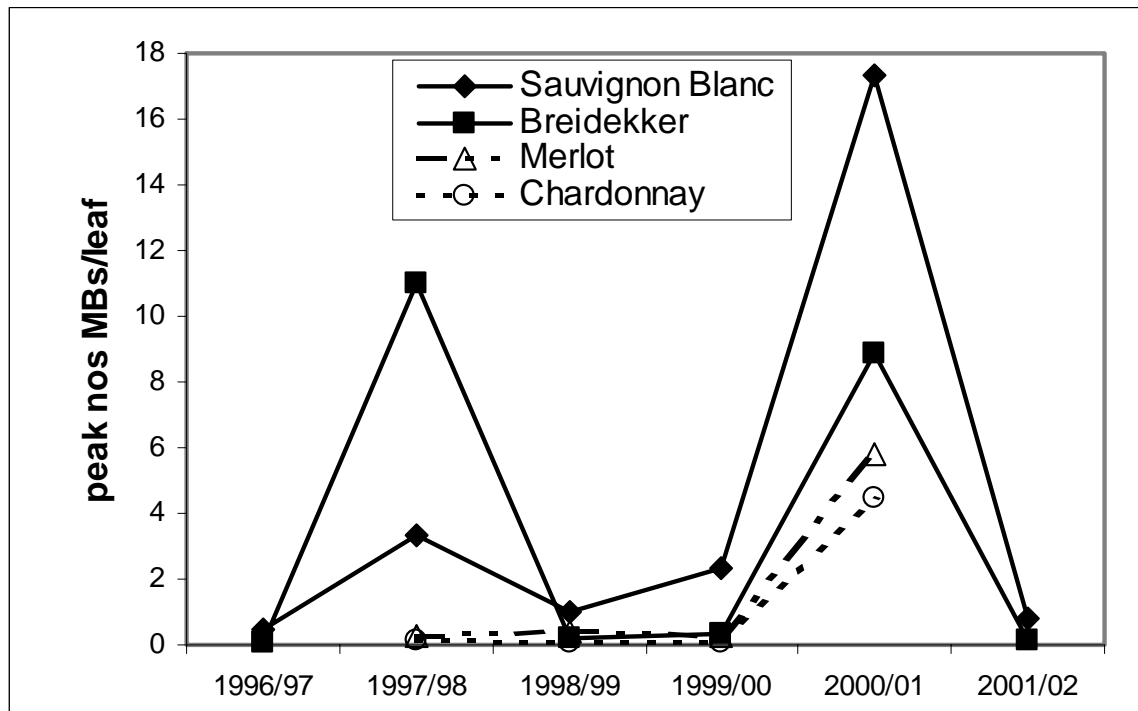


Figure 3. The maximum mealybug numbers per leaf each season: 1997-2002.



Issues in modelling the relationship between mealybugs and the spread of virus

The extent to which the data gathered by Charles et al. (in prep.) could be used to build a model of mealybug activity and GLRaV-3 spread was evaluated. Three approaches to modelling spread were considered, but none showed a good mathematical relationship between mealybug presence and the appearance of virus infection.

1. The tree-based model

With this non-parametric approach, a classification tree was developed to predict the probability of a vine being infected or clean using a model:

$$\text{Response} \sim \text{Cultivar} + \text{Intra-row} + \text{Inter-row} + \text{Mealybug}$$

Numerous methods of defining the intra-row measures were tried such as the number of infected vines within a range of numbers of adjacent vines. Inter-row measures were similarly extensive. Any other information that could have any possible effect on the response (such as number of days over 20°C) was tried in the model. Variables that had no effect on the response (i.e. virus spread) were eliminated. Grape cultivar was shown to have a substantial effect on the response, but the classification-tree approach was not successful in establishing a relationship between the response (i.e. virus spread) and any other variable that had been measured (or calculated from the observed ones).

2. The probabilistic model

Two possible scenarios of leaf roll virus transfer were considered with this approach as mentioned above:

(a) *Inter-row*

This model may be independent of the mealybug population (e.g. if the transfer was facilitated by machinery), and may differ depending upon whether the infection source was on the right or left (e.g. if machinery moved from right to left, or if wind transfer were important and the prevailing wind came from the left). It was felt that the inter-row distance function d_I could be estimated from the 1997/98 data assuming that rows ≤ 22 and ≥ 49 were fully infected. Note that three vines of rows 23-26 were infected (from rows ≤ 22) and nine vines of rows 45-48 were infected (from rows ≥ 49) (the nine included one pair of adjacent vines which could have been a single infection incident). It was tempting to regard this as evidence that vines in rows 45-48 were more likely to become infected than vines in rows 23-26. While this might be true (either due to a difference in the probability of inter-row transfer from the left and right, or because they were different cultivars) the evidence was not convincing (two-tailed binomial probabilities were 0.144 and 0.226 depending upon whether the pair of infected vines in row 46 were regarded as separate infection incidences or not).

In estimating d_I we needed to consider the possibility that the infection may not have come from the closest row with infected vines and thus the functions could sum. We could then account for progress across the block by assuming that d_I is proportional to the proportion of vines in the row that are infected. Given that the probability of inter-row transfer appeared to be low (from the data in Figure 2) we ignored the possibility of the same vine getting infected by multiple infection incidences.

(b) *Intra-row transfer*

This model is dependent on estimates of the mealybug population. Given that the mealybug populations in 1997/98 were very high (Figure 3), the increase in infection from 1997/98 to 1998/99 could be used to provide an estimate of the maximal distance function for virus transfer. Similar reasoning and logic to the inter-row transfer could be used, except that there would be a need to account for multiple transfers; i.e. a new infection may represent more than one infection incident. The possibility of successive virus transfers within a season needs to be accommodated; i.e. if new infections were found on the fifth and sixth plants from a previously infected plant, it could be that the sixth was infected from the fifth rather than from the original plant.

Since the only variable that showed an effect on the response in the classification tree was Cultivar, it became evident that the spread of virus was due substantially to factors which were not, or could not, be measured. It was considered fruitless to attempt to develop the probabilistic approach further.

Prospects for modelling the virus and mealybug vector relationship

The data set of Charles et al. (in prep.) is not ideal for either of the modelling approaches outlined above:

- There were missing plants in the rows, which would initially leave a gap (reducing the rate of transfer) but would eventually close up (increasing the rate of transfer);
- Some infected plants were removed and replaced with virus-free vines;
- The mealybug counts did not occur on the same rows as those measured for virus transmission;

- The virus and mealybug status of vines outside the study area is unknown.

These factors, particularly the latter, make the chances of using this data to successfully model the relationship between mealybug activity and virus transmission very small. There is clearly a macro-spatial trend in virus spread and point-wise clustering within the rows that requires no statistical verification but it was difficult to show this mathematically because there were highly variable results between the seasons. Mealybugs were probably the most likely means of virus transfer within this vineyard and there are some strong positive correlations between mealybug numbers and virus infection levels in the following season that are discussed in Charles et al. (in prep.). While correlation establishes the relationship between mealybug incidence and subsequent virus infection, it lacks the sophistication and potential value of a model. We therefore made a pragmatic choice of using this data to model the gradual spread in leafroll virus from year to year within this vineyard that could then be used to make predictions of spread over time.

Modelling the spread of virus infection

The use of these data to model the relationship between mealybugs and the spread of virus was not possible but we were able to model the changes in distribution of the virus itself. Using Ordinary Runs Analysis we found that there was little evidence to support a non-random distribution along rows of vines so a binomial model was used to describe the relationship between the levels of virus infection for the period 1998-2003. There was some suggestion of non-random spread in rows 23 and 24 after the 2000 season and in row 45 after 2002. It is probable that if the grape cultivar is highly susceptible then the initial virus transmission is by random processes but that once the level is high enough then transmission is associated with neighbouring infected vines.

The percentage of infected vines in each plot (Plots A and B) was calculated and the coefficients for the fitted lines were determined for the six seasons that data was available (Figure 4). Coefficients were also determined for the central vines (Plot C) based on five seasons of infection data; 1999-2003. Since these coefficients were calculated using a non-linear scale the fitted lines appear to be a poorer than expected fit to the plotted points.

Figure 4. The relationship between the percentage of infected vines each season over the assessment period 1998-2003.

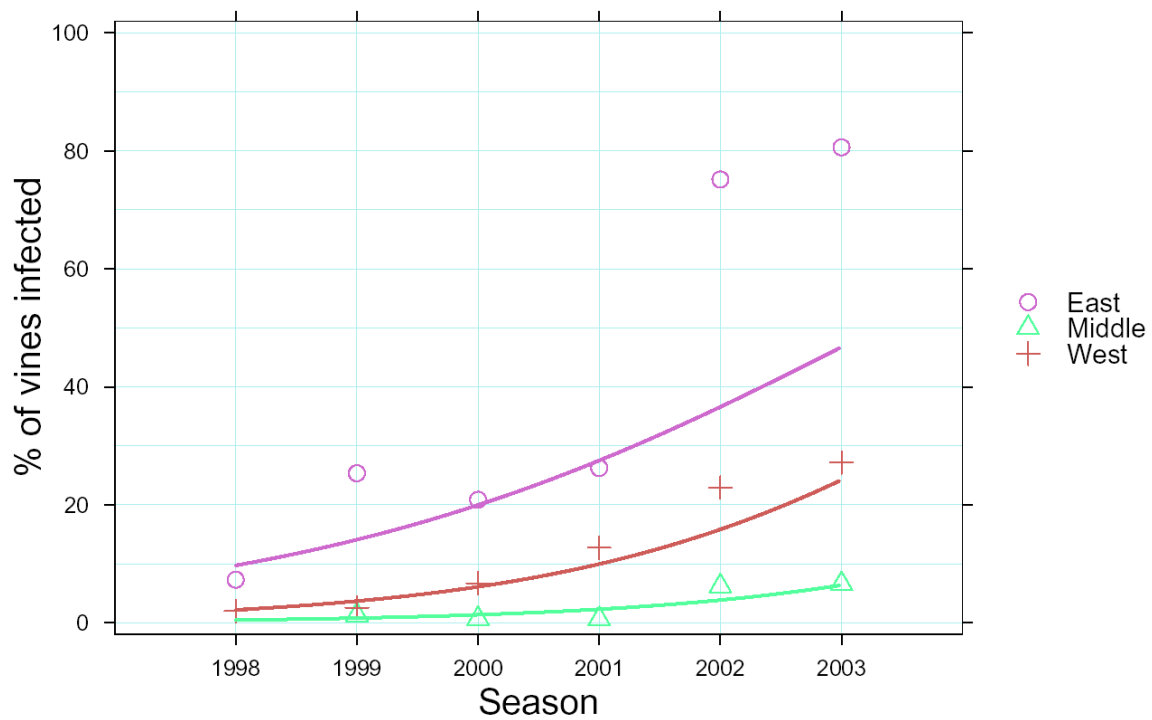
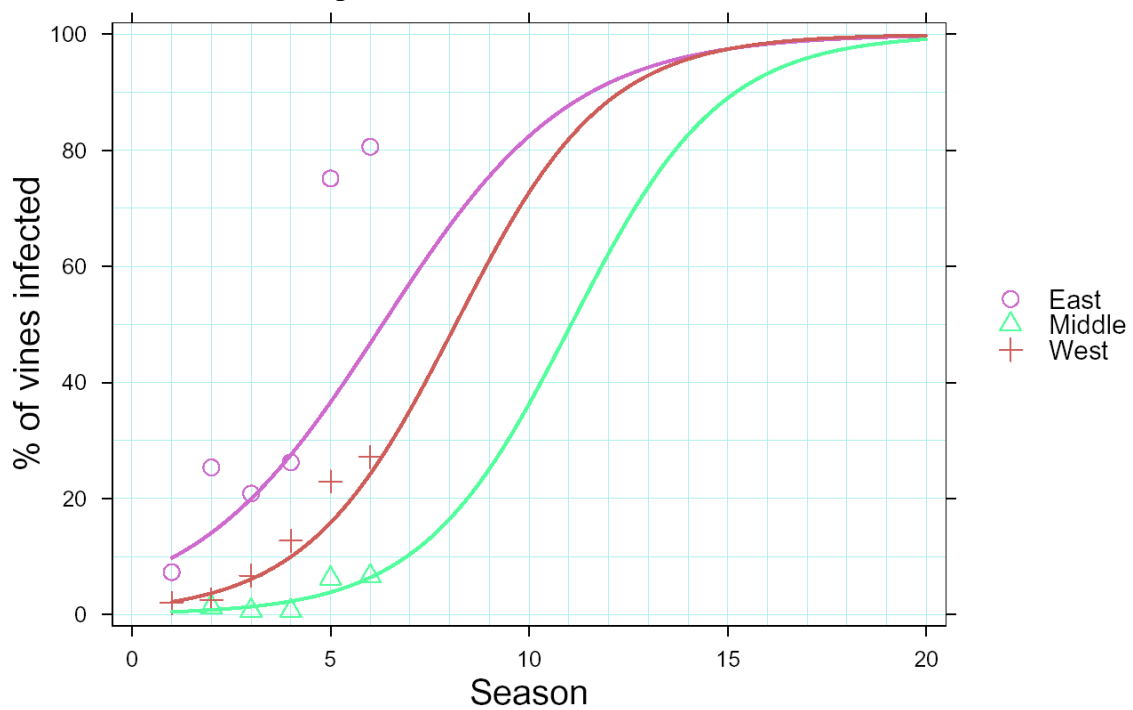


Figure 5. The predicted levels of virus infection over a 20 year period based on the relationship between the percentage of infected vines each season over the assessment period 1998-2003.



The coefficients were also used to fit plotted lines which are extrapolated for a further 14 seasons (Figure 5). As these extrapolations are based on just six seasons of data care should be taken in drawing conclusions from them; e.g. the mealybug populations involved with the transfer of virus during these seasons may not be representative of future populations and the practices that might be implemented to reduce their spread. The model has to assume that these effects are negligible but the lines could change significantly with the inclusion of additional data (i.e. more seasons).

Virus infection is predicted to spread rapidly within the vineyard over time with 50% infection occurring in years 6, 8 and 11 for Plots B, A and C respectively. All plots are predicted to reach 90% vine infection in years 11, 12 and 15 for Plots B, A and C respectively. We used this information on the predicted spread virus in this vineyard as the basis for the following economic analysis of likely costs of virus spread in New Zealand vineyards with susceptible varieties that are exposed to the risk of GLRaV-3 infection.

The economic impact of GLRaV-3 infection

A vineyard producing premium wine grapes that are sensitive to GLRaV-3 infection could be expected to show a marked decline in profitability as the level of infection increases. Lower initial productivity of premium wine grapes is offset by the increasing production coming from GLRaV-3 infected grapes within the vineyard. The decline in revenue was almost linear in the highest rate of infection (Figure 6, Plot B) so that by year 8, with about 70% of vines infected, the net revenue per ha was reduced by \$2000 per hectare over Plot C vines with about 19% infection. This period (year 8) represented the greatest difference between these two scenarios and the steepest decline in net revenue and so represents a likely decision point for vineyard removal and replacement. At this point there was only perhaps four years difference in the financial outcomes for the block with the highest infection risk (Plot B) against the lowest risk scenario (Plot C). As the virus spread increases the differences in vineyard profitability and virus costs diminish until year 15 at which point all three infection scenarios predict infection approaching 100% and a production that would be limited to low value grapes suitable for only bulk wine production.

The annual cost of virus infection changed markedly under the three infection scenarios (Figure 7). Once virus infection exceeded about 20% of vines, this cost (or productivity and quality loss) exceeded \$1000 per hectare by year 3, 6 and 9 for Plots B, A and C respectively and increased rapidly thereafter. It is therefore economically justifiable to invest at least 50% of this sum annually to prevent this scenario developing. This includes preventative measures i.e. mealybug control and replacement of infected vines. Given difficulties in achieving high levels of mealybug control in vineyards (i.e. >70-80% reduction in bunch infestation) and the cost of replacing infected vines, considerably more should be invested in reducing or eliminating the sources of initial infection. This not only includes virus-indexed plant material but the measures that might be taken (or developed) to reduce infection in existing vineyards. Considerably more effort must go into identifying the sources of initial infection of new plantings because once GLRaV-3 has established within a vineyard its rapid progression threatens the economic life of both the infected block and adjacent plantings.

Figure 6. The change over 20 years in the net revenue per hectare for a GLRaV-3 infected vineyard, based on observed scenarios for infection spread.

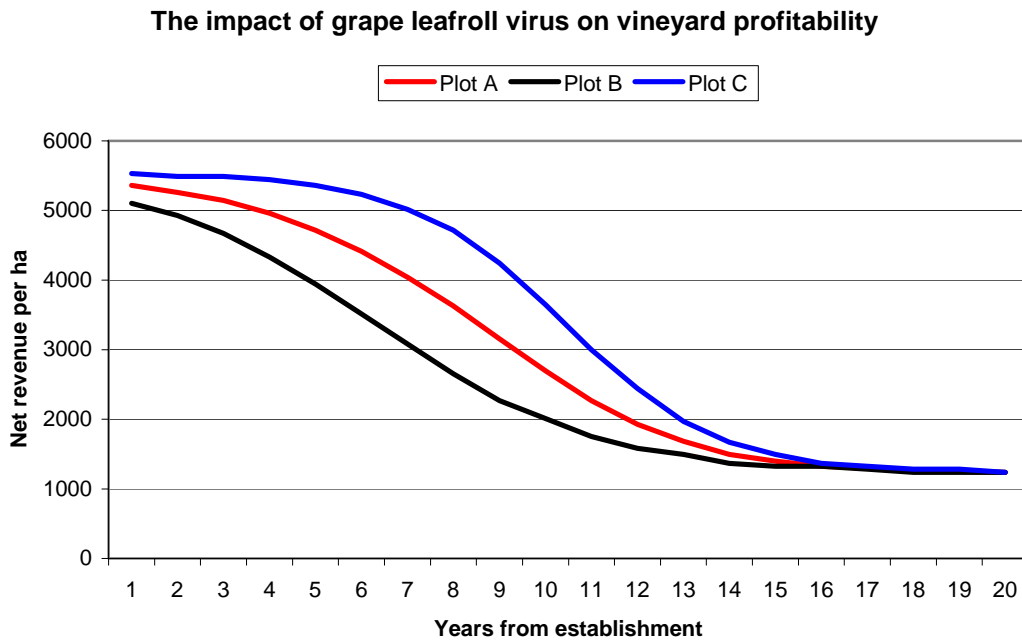


Figure 7. The annual cost of virus infection per hectare over 20 years for a GLRaV-3 infected vineyard, based on observed scenarios for infection spread.

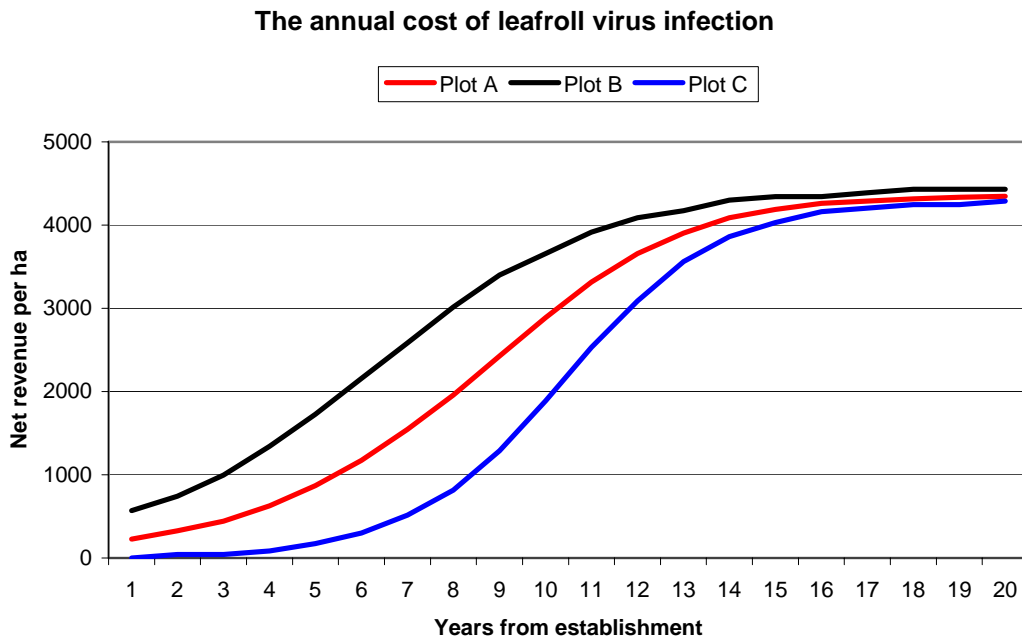
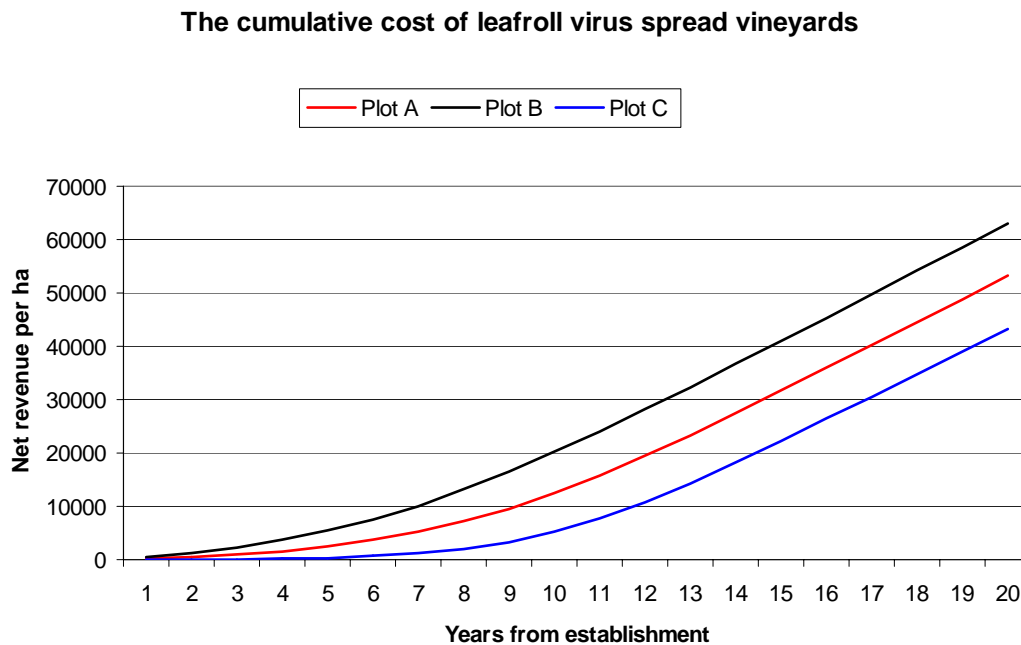


Figure 8. The cumulative cost per hectare over 20 years for GLRaV-3 infection within a vineyard based on three predicted infection scenarios.



The three virus spread scenarios were used to predict the cumulative cost of GLRaV-3 infection (Figure 8). This cost exceeded \$10,000 per ha by years 7, 9 and 12 and \$30,000 per ha by years 12, 15 and 17 in Plots B, A and C respectively as the rate of initial virus infection increased. In practical terms this means that the profitability of many infected blocks might be sufficiently impacted around year 11 to justify re-planting of the vineyard; i.e. the time at which the cumulative cost of virus infection exceeds the cost of new vineyard establishment.

CONCLUSIONS

There was a macro-spatial trend in GLRaV-3 spread in the data but it was difficult to show this mathematically because there were highly variable results between the seasons. Apart from grafting, mealybugs are the only known means of GLRaV-3 transmission and this has been shown by the strong positive correlations between mealybug numbers and infection levels in the following season. Three scenarios of virus spread (high, moderate and low) were modelled over the six seasons for which data was available. Virus infection was predicted to spread rapidly within the vineyard over time with 50% infection occurring in years 6, 8 and 11 for high, intermediate and low infection rates respectively. The three scenarios were predicted to reach 90% vine infection in years 11, 12 and 15 respectively. The economic impact of GLRaV-3 infection rates in sensitive varieties exceeded \$10,000 per ha by years 7, 9 and 12 (using the observed infection rates) and profitability was sufficiently impacted to justify re-planting by year 11 (depending on infection rates).

Once virus infection exceeds about 20% of vines, this annual cost exceeded \$1000 per hectare by year 3, 6 and 9 for the three infection scenarios and increased rapidly thereafter. It is economically justifiable to invest at least 50% of this sum annually to prevent this scenario developing including preventative measures i.e. mealybug control and replacement of infected vines. More should be invested in reducing or eliminating the sources of initial infection including comprehensive virus indexing of plant material and managing the sources of initial infection in new plantings.

Strategies that seek to eliminate mealybugs from within existing vineyards are impractical, if not impossible. All three species of mealybug are vectors of GLRaV-3. All are cryptic with high reproductive capacities, ensuring that pesticides alone will not provide complete control. In addition, immigration from many alternate host plant species ensures a constant, if unpredictable, threat of new infestation. An integrated approach to mealybug control is highly recommended, based on selective pest management and biological control together with strategies to manage any defensive interactions with ants. Agrichemicals must therefore be active against mealybugs while being selective for beneficial natural enemies. Thorough spray coverage is required for mealybug control and any technology (sprayer technology, calibration and adjuvants) that facilitates improved coverage and improved mealybug control will inevitably help to delay the spread of GLRaV-3 in vineyards.

The profitability of premium red wine production within New Zealand viticulture is more seriously affected by GLRaV-3 infection than in some countries where harvest can be delayed to compensate for virus impacts of delayed maturity and depressed berry sugar content. With our cool climate and often relatively abundant late summer rainfall, growers of premium red wine grapes must take every practical measure to ensure that GLRaV-3 infection risk is minimised by the use of virus-indexed planting material, early removal of infected vines. The gradual replacement of vineyards with virus-free plant material, together with control of mealybugs should have a major impact on GLRaV-3 spread, especially if the relationship between GLRaV-3 and different grape varieties was understood. However, success is dependent on an integrated approach between researchers, virus indexing services and vineyard nursery producers, and growers' identification and management of virus infection and mealybug infestation risks between their established and new plantings.

ACKNOWLEDGEMENTS

The authors would like to thank AgMardt for the provision of funding support for the completion of this analysis of the economic impact of leafroll virus on vineyard profitability. We would also like to express our sincere thanks to Peter Alspach (HortResearch, Riwaka) for his assistance with statistical and mathematical modelling.

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