RIVERSUN technical bulletin

Issue No.5 August 2001

Grapevine Leafroll Viruses in New Zealand Viticulture

Fact Sheet and Research Up-Date

Dr Roderick Bonfiglioli, Nick Hoskins and Fran Edwards, Riversun Nursery, Gisborne, New Zealand

Viruses of Grapevines, an overview

Grapevine viruses have presented serious and well recognised problems for grape-growers and winemakers for many years. They are known to have a wide range of effects from causing reduced yields, low quality fruit, incompatibility problems at the graft unions, declining health and early death of grapevines. Currently there are more than 56 known viruses of grapevines but only 13 of them are considered to be economically important, the rest only cause very minor problems, or are rarely found. Of those 13, only 3 are commonly found in New Zealand. These three viruses are all types of virus known as Grapevine Leafroll associated viruses.

Grapevine Leafroll associated Viruses have for many years been recognised as a major problem affecting quality and quantity of production in many of the older wine producing countries. In South Africa, for instance, Grapevine Leafroll disease was, and still is, seen as the single biggest obstacle to the development of a high quality export wine industry.

If allowed to get out of control in New Zealand, Grapevine Leafroll disease could become the single biggest hurdle our wine industry will face in its quest to become a producer of great red wines, especially for Pinot Noir. It may even critically affect the quality of our fine white wines.

Grapevine Leafroll viruses

Types present in New Zealand

Grapevine Leafroll associated Virus Type 3

GLRaV-3 is certainly the most destructive virus in this group, and is also the most common. In some varieties, especially reds, it will cause a serious loss of performance and a loss of vine health which may lead to affected vines being eventually removed and replaced. In those varieties it also causes a reduction in fruit quality and consequently a decline in wine quality. In the less sensitive varieties, which are mostly white, it can still have some effect on the fruit-set. It may also cause a reduction in flavour compounds and result in declining and variable yields. Some white varieties show virtually no visual symptoms, and the only way to know if they are infected is to test them.

Grapevine Leafroll associated Virus Type 1

GLRaV-1 is usually a less destructive virus which causes a milder form of Leafroll disease. It seems to mostly affect the red varieties, especially Pinot Noir and Merlot. Its effect upon white varieties is not fully understood, though it is thought to cause some problems with fruit set and may also cause a reduction of flavours. In New Zealand and Australia it is in all Chardonnay Mendoza.

Grapevine Leafroll associated Virus Type 2

GLRaV-2 is only known in New Zealand in the new Bordeaux clones of Sauvignon Blanc, BDX316 and 317 and its effect, while thought to be quite mild, is not properly known at this point.

Leafroll Virus testing in New Zealand,

the National situation

Over the last three years in New Zealand there has been a sharp increase in the number of vines tested for Grapevine Leafroll viruses, especially GLRaV-3. Our market research shows that there were approximately 32,000 composite tests for GLRaV-3 done last year nationally, we did 23,000 of those tests in our own laboratory, of which over 20,000 were for our own material. We are therefore doing more than 70% of the national GLRaV-3 testing.

Riversun accounts for around 25% of the national production of grafted grapevines, yet we are doing more than 65% of the national GLRaV-3 testing to ensure the high-health of our own source material.

We consider that this clearly demonstrates our commitment to the industry with respect to this issue of freedom from Grapevine Leafroll viruses.

Riversun GLRaV-3 testing

Nationwide protocol

At Riversun we have been extensively testing all our source vineyards of both vinifera bud-wood and root stock vines for the last three years. During this time we have tested nearly a quarter of a million vines for Leafroll viruses and we have compiled a huge amount of information on the spread of Grapevine Leafroll disease in New Zealand.

This season we have tested over 82,000 vines from 106 source vineyards across most viticultural regions. 74 of these vineyards had been tested in previous years, of those 23 had significant virus spread, and we abandoned 4 because the virus levels had become too high. Of the remaining 51, many were infected

Figure Two:
White variety (Pinot Gris) infected with GLRaV-3
Note rolled-back leaves

How are Grapevine Leafroll Viruses spread?

Grapevine Leafroll viruses are spread by the use of infected propagation material and by some specific sap-sucking insects in the vineyards, including all types of Mealybugs and (certainly in Europe) by Vinescale.

Infected bud-wood propagation material

Taking cuttings of vinifera bud-wood from vines that are infected with Grapevine leafroll viruses is the most efficient way to spread the infection. These cuttings will grow to become infected adult vines which may be mildly or more severely affected.

Infection by root stock vines

Root stock vines rarely show any visual symptoms of infection with any of the Grapevine Leafroll Viruses and many viticulturists do not seem to realise that the infection can be passed on just as easily through infected root stock as it can through infected scion material. We have abundant records of root stock vines infected with both GLRaV-3 and GLRaV-1 and, as mentioned earlier, we have rejected some root stock blocks for this reason.

Mealybugs

Most species of Mealybugs (*Psuedococcus* species) have been shown to spread all grapevine leafroll viruses in the field. Mealybugs feed on vine leaves by sucking the sap, and during this process they become infected with the virus and once the Mealybug has itself become infected, then it can transmit the infection. Infection of the Mealybugs is not permanent, the infection is not passed on through the eggs to the next generation. Mealybugs spend most of their life cycle crawling on the leaves, but over-winter under the old bark of the vines, often on the lower part of the trunks or even just below the surface of the soil. They also live on many other plants including several common weed species.

Our mapping studies of infection spread in vineyards shows that the infection spreads more often along the rows than it does across the rows. This is because it is much easier for the Mealybugs to crawl through the canopies from one adjacent vine to the next, than it is for them to crawl across rows. Commonly infection is spread to the adjacent vines, but it can be carried some distance in the vineyard on some occasions. Wind, vineyard machinery, leaf-plucking, pruning and traffic through the vineyard will all assist in the longer-distance spread within and between vineyards.

Mealybugs are known in most viticultural areas of New Zealand in both the North and South Islands. They were

very common early in 2001 in the regions of Hawkes Bay and Gisborne, they were also reported as being very common in Marlborough this year. Nearly every vineyard we examined late season in those regions was shown to have quite high numbers of Mealybugs present. Many vineyard managers we spoke with were monitoring for Mealybugs, but they stopped monitoring well before harvest. The high numbers of Mealybugs we saw this year were mostly well after harvest. Late season is the time when the concentration of virus is highest in the vine, and it is reasonable to think that this time is when feeding Mealybugs could most easily become infected and therefore most efficiently transmit the infection. Our ELISA testing of Mealybug samples confirms that many of them are infected with GLRaV-3. It is also interesting to note that nearly all the vineyards that we visited and found Mealybugs in were considered by the vineyard managers to be free of Mealybugs.

Grapevine scale insects

Scale insects are another group of sap-sucking insects that often live on grapevines. Grapevine scales (*Pulvinarium vitis* and other types) have been shown in Europe to be able to transmit and spread Grapevine Leafroll virus infections in the vineyard. It is now thought Grapevine scales can transmit all of the Grapevine Leafroll viruses. Grapevine scales are generally found on the trunk and cordons or on other older wood of the vines. They are often found by lifting off the outer bark. The juveniles are extremely small and very difficult to see, they are known to move onto to the fresh leaves. We have a type of Grapevine scale insect here in New Zealand called *Parthenolecanium persicae* that is a likely candidate for spreading Grapevine Leafroll Viruses here. There is no direct evidence yet that this scale insect is a vector of the viruses here in New Zealand, but it is considered quite likely and some virus transmission trials will be started this coming season.

Effects on Wine Quality

What is the effect of infection with Grapevine Leafroll viruses on the quality of the grapes and the wine?

It is well known that red grapevine varieties, especially Merlot, Pinot Noir, Cabernet Sauvignon, Cabernet Franc and Shiraz are more negatively affected by Grapevine Leafroll Viruses than are the white varieties.

The general effect of GLRaV-3 is to delay ripening, and reduce the accumulation and development of colour and flavour compounds. In the red varieties yields may also be adversely affected, and fruit set can also be sharply reduced. Red varieties, especially Pinot noir can suffer a serious loss of vigour as a result of infection with GLRaV-3. There are several recent examples within New Zealand of Reserve quality vineyards planted to red varieties that have increasing levels of GLRaV-3 in them and have subsequently been down-graded to standard or even bulk-wine quality. There are many studies in the viticultural journals concerning the negative effect of GLRaV-3 infection on fruit and wine quality in red varieties, but little research has been done on white varieties where the symptoms of infection are far less obvious.

The effect of GLRaV-3 on Sauvignon Blanc is not a subject that has yet been researched. To address the question we have begun a research project with some noted Marlborough winemakers and viticulturists. We expect the first round of results at the end of this coming vintage.

Some epidemiological considerations

Epidemiology is the study of epidemics, it helps us to understand and describe how diseases behave and how they move through populations. Research has shown that for many disease situations there is a critical level of infection that must be reached in a population before the disease incidence starts to increase sharply. This point of change in the disease behaviour is where it changes from being a small or even a hidden problem, to becoming an epidemic. Time, numbers of infected host plants, suitability of conditions, especially climate and the concentration and numbers of available insect vectors are all important factors that will interact to determine how quickly a disease becomes established in a region. Once properly established in a population, a disease may be very hard to eradicate.

In the Adelaide Hills in South Australia an experimental Pinot Noir vineyard planted with many different clones was noted to have one bay containing 3 Leafroll infected vines. Leafroll was not a known or well understood problem at the time. The vines were left in place and no newly infected vines were noticed for several years. Some years later it was noticed that the vines next to the original three with the leafroll disease were suddenly starting to show symptoms as well. The numbers of newly infected vines then began to increase very quickly over the next three years. A population of vector insects had obviously established themselves in and around the vineyard and had sufficient time and exposure to become infected. Emergency management procedures were put into place, but it is now expected to be difficult to contain the problem.

A paper presented by David Jordan in 1993 details how an infection of Grapevine Leafroll virus Type 3 spread rapidly through a Cabernet Sauvignon block in the Auckland region.

but our screening processes determined that it was at a low enough level to be manageable and we could safely exclude the virus affected vines. We also tested 32 new source vineyards this year and found that 22 of them had significant levels of virus in them, and 7 of those source vineyards were rejected for use by us because the levels of virus were too high. Of equal significance, this year we have also abandoned two root stock blocks due to unacceptably high levels of virus. One is a North Island 3306 block with GLRaV-1 and the other, a privately owned block of Riparia Gloire in Gisborne with GLRaV-3.

In a large number of vineyards, and in all major viticultural regions of New Zealand, we have seen significant increases in the incidence of GLRaV-3.

The rate of virus spread on a national level is surprisingly high, many vineyards where virus spread has been documented are showing increases around the 10% to 12% range over last years figures.

In response to this developing Leafroll disease problem, Riversun has begun a program of developing specialised source vineyard plantings of certified high-health material virus tested by the more stringent PCR method. These source vineyards are being planted in isolated locations specifically to reduce the risk of disease spread and will be used to provide the New Zealand wine industry with high-health quality material with an exact and precisely known clonal lineage.

Virus spread in vineyards, results of field studies

Using computer aided statistical analysis we can get a clear understanding of how the virus distribution is occurring within each vineyard, and what the rate of spread is. It is now also possible for us to quickly determine if the virus has been introduced with the original planting material, or if it has been spread naturally into a vineyard by virus infected insects. Some examples of our vineyard mapping studies are presented in the examples below.

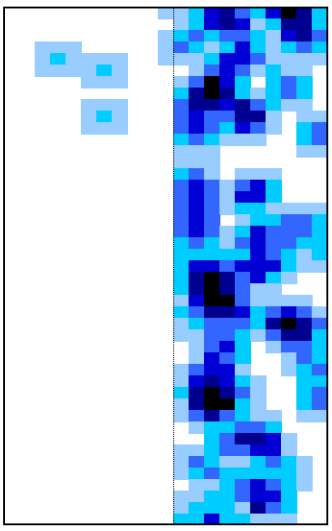
Vineyard examples

VINEYARD 1

The effect of different sources of propagation material on virus infection levels

This vineyard in Marlborough is planted to Sauvignon Blanc. It is heavily infected with GLRaV-3. Examination of the distribution pattern of the virus in this source vineyard, as seen in Figure1, shows that the right side of the vineyard is much more heavily infected than the left side. The right side is infected at a rate of 22.5%. The bud-wood used to plant this side of the vineyard was from a different source to the left part of the vineyard. The left side of the vineyard is only 1.8% infected.

There are numerous clusters or "Hot spots" of infection scattered apparently randomly throughout the right side of the vineyard. Such a distribution pattern of infection is consistent with what would be expected with the use of infected propagation material. The "Hot spots" may later increase in size and also spread to other parts of the vineyard as insect vectors spread the introduced infection further.

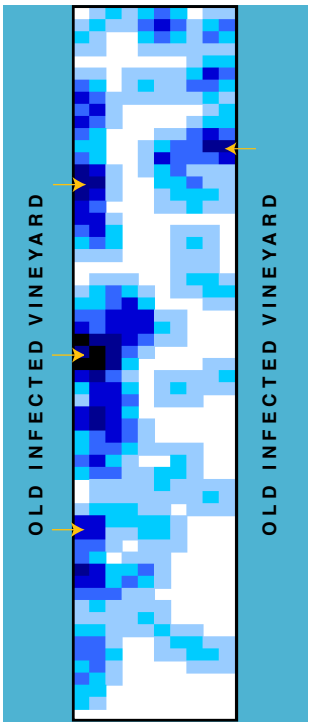


left side *right side*

VINEYARD 2

Infection spreading by insect vectors into a new vineyard from a virus infected neighbouring vineyard

This is a source vineyard of four year old Merlot vines in Hawkes Bay. The vineyard is planted between two much older Cabernet Sauvignon vineyards which are both heavily infected with GLRaV-3. The Merlot Material was supplied as 10% virus tested through the NZGVIG system, but is now showing a high rate of virus infection. Examination of the virus infection density map clearly shows that the infection is associated with the edges of both sides of the vineyard, it is moving (see arrows) into the new Merlot vineyard from the older Cabernet Sauvignon vineyards on each side. Mealybugs have been very common in this vineyard and they will most certainly be the insect vectors which are spreading the virus into the new vineyard.



Some important "Do's" and "Do nots"

to minimise the risk of Leafroll virus infections when developing new vineyards

DO

test all mother vines or source vines by ELISA methods before taking cuttings.

Using infected propagation material will certainly result in infected new vines.

DO

make sure the root stock vines used are ELISA tested as well,

Many viticulturists and vineyard managers are unaware that root stock vines can carry Grapevine Leafroll infections. Root stock vines generally show no visual symptoms of Leafroll when infected and they will certainly transmit the infection when grafted. Root stock vines can be infected with any of the Grapevine Leafroll viruses.

DO

use certified high-health vines for planting new vineyards,

Certified material is fully tested for Grapevine Leafroll viruses, it is of a known high-health standard, it is of a known and traceable clonal identity and source and it has been prepared using best industry practices.

If the material you use is not certified, then what is it?

DO NOT

graft "Nursery tops"

When using this method of propagation, it is extremely difficult to properly monitor and test for Grapevine Leafroll Virus infections. If some infection is introduced into the system (by Mealybugs for instance), or there is some small amount of infection already present, the infection can be rapidly multiplied with very unfortunate results,

DO NOT

top-work onto old untested vines,

Such older and no-longer wanted vines are very likely to be already infected with Grapevine Leafroll viruses, and this may be why they are poor performers and in poor health. Grafting onto old infected vines will guarantee that the new vines become infected. This can be a very expensive mistake for a vineyard development.

DO NOT

under-plant beneath infected vines,

Mealybugs will simply drop out of the older infected vines and land on the new young vines and infect them. Our observations show that under-planting is a very efficient way to spread the virus. It may be possible to save one crop in the change-over process by under-planting, but subsequent crops may then be badly compromised in quality by the presence of the disease in the new vines, the life expectancy of the new vines may also be shortened.

How to get your vineyard tested

If you are concerned that you may have virus infected vines in your vineyard and you wish to get some vines tested, here is how you can do it.

During the growing season mark the vines you think are showing symptoms. Pictures of the typical symptoms for red and white varieties are shown in Fig 1 and 2.

The best time to look for the symptoms is late in the season. Symptoms will start to appear after veraison and will remain until leaf-fall.

Leaf testing

You can have leaf material tested between January and April. For this you should take 3 leaves from each vine. Take a leaf from between the 1st and 5th nodes from 3 different canes and select the canes from mid-cordon positions.

Dormant wood testing

You can get dormant wood tested from April onwards, and anytime during winter.

You should take one 15 cm long section with at least three buds from the mid section of each of two different canes. Select the canes from the mid cordon part of the vine. Note: Root stock vines can only be tested using dormant wood, leaves do not give reliable results for root stocks.

For further information on Leafroll virus testing and sampling procedures, call the Linnaeus Laboratory on (06) 867 5279.

The infection developed from an initial 5% or less to around 95% in a period of only five years.

In new or relatively young viticultural regions such **Central Otago** or **Marlborough** it is highly likely that vines infected with Grapevine Leafroll viruses will have been introduced with the original propagation material unless due care was exercised. How long it takes for this introduction to firmly establish itself and then begin to spread will depend upon several factors as discussed previously. In Marlborough the common belief among the local vignerons seems to be that they do not have any spread of Grapevine Leafroll viruses and that they do not have Mealybugs. Both of these beliefs are incorrect. Mealybugs were reported as being very common in Marlborough this year, and it is known that the species of Mealybugs in the Marlborough region are quite capable of transmitting the Grapevine Leafroll viruses. Our mapping studies have also clearly shown that there is actually spread of Grapevine Leafroll Virus Type 3 in Marlborough vineyards. Some vignerons would say that this is just the virus expressing itself late, what we would call a latent infection. However the vineyards we have been surveying at are too old for latency of disease expression to be remotely probable, we are certainly observing real spread of virus in these vineyards.

While this spread of GLRaV-3 that we are studying in the Marlborough region is not yet as dramatic as the spread seen in the North Island, it is only early days yet, who knows what will happen in the next few years? It is quite likely that the scale of the problem is currently hidden in that nearly all the GLRaV-3 infected vines recently found in Marlborough are Sauvignon Blanc which does not show much in the way of visual symptoms when infected, so most people do not even realise their vines are infected. If the infection starts to spread into the Marlborough or Central Otago Pinot noir vineyards, the problem will become much more apparent as Pinot Noir is very susceptible to Grapevine Leafroll viruses. Unfortunately there is not any really good reason to think the problem will remain hidden or quiescent in the Marlborough region, despite the local optimism. There are regions of Europe quite equivalent in climate to Marlborough, and these regions have very significant problems with Grapevine Leafroll viruses.

What can be done about Grapevine Leafroll disease in a vineyard?

Once a vine has acquired infection with a grapevine Leafroll virus, there is nothing that can be done. The best course of action is to make sure it does not happen in the first place. **Hot water treatment of vine cuttings does not kill the grapevine leafroll viruses.**

To prevent infection with Grapevine Leafroll viruses, you should:

- Only plant your vineyard with vines which are certified as virus tested to a specific level and are produced in a certified production process.
- Monitor for Mealybugs, especially well into the later part of the season and after harvest.
- If Mealybugs are present, they should be controlled. For control of Mealybugs, advice should be sought from appropriate insect pest specialists.
- Monitor your vines for Vinescale, and control them if found.
- If you think you may have some virus infected vines in your vineyard, test some vines to find out,
- If you do have some virus infected vines in your vineyard, you should monitor them for performance, check for:
  - Fruit set, how successful?
  - Berry size, shot?
  - Bunch structure, changed?
  - Brix, Total acids and pH, normal or different?
  - Time of ripeness, late?
  - Yield, reduced or variable?
  - Flavours, reduced?

You may find that the infection is having an effect upon your vines performance, and this may convince you to remove the infected ones and replace them with new vines.

Cooperative Action by Growers

In areas where there are many vineyards all close together, Leafroll viruses will spread very easily if the Mealybugs are not controlled. In regions such as the Gimlett Road area of Hawkes Bay, or the Opou Road area in Gisborne where there are many vineyards which have Leafroll virus infection and there are also high numbers of Mealybugs and perhaps also vinescales, a cooperative approach to control measures will be much more successful than any individual action. If you are applying control measures at a different time to your neighbours, you may be reducing the effectiveness of everyones actions. Control of the mealybugs in such a situation is best done on a cooperative basis. Organise a group of the local growers to discuss and act in unison on the problem. It can be controlled, but most successfully by cooperative action based on a common understanding of the problem.

VINEYARD 3

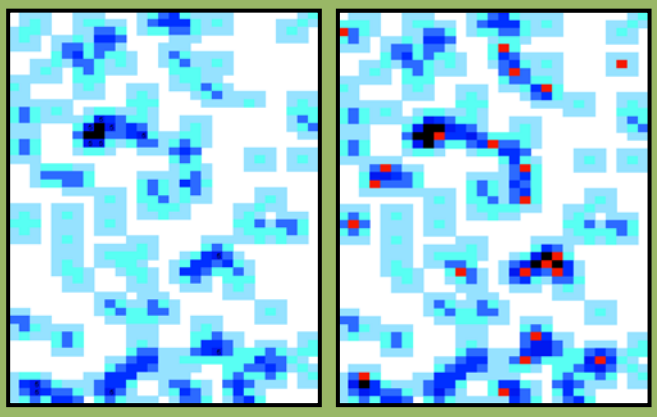
Introduction of the virus into a vineyard by infected propagation material and subsequent further spread within the vineyard by insect vectors

This is a six year old source vineyard in Marlborough planted to Sauvignon Blanc. Examination of the virus infection-density map for 2000 shows multiple small clusters or “Hot spots” of infection that are apparently scattered randomly around the vineyard. Statistical analysis of the distribution of the clusters suggests that it is random and this indicates that the vineyard was planted with infected propagation material.

Figure One: *Red variety (Merlot) infected with GLRaV-3. Note typical red leave symptoms with green veins.*

The virus infection-density map for 2001 shows that there is significant spread of the infection since 2000 and that the new infections are mostly, but not exclusively concentrated around the previous years “Hot spots”. The infection may even be even more advanced than this now, because the vineyard was only cluster tested in 2001, rather than 100% tested as we did in 2000. This vineyard was not re-tested at 100%, despite the newly infected vines because it was rejected by us for use as a source block. In 2000 the vineyard was 10.5% infected, in 2001 it is 12% infected, maybe even more. The interpretation of this situation is that while the initial infection was established by using infected propagation material of an unknown source, the infection is now being spread further by other means, probably Mealybugs or vinescale insects.

■ ***red squares denote newly infected vines***

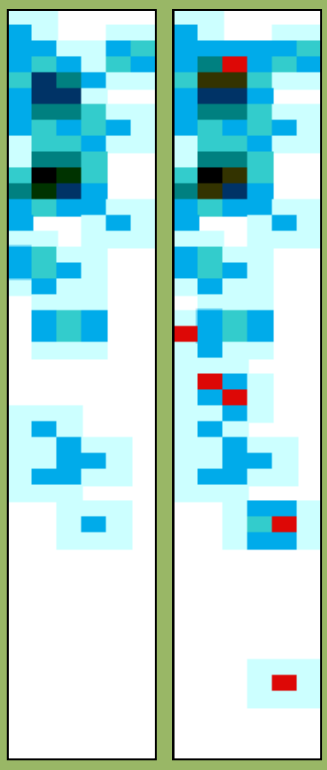


year 1, 2000 ***year 2, 2001***

VINEYARD 4

Grapevine Leafroll virus type 1 infection spreading in a root stock block

This root stock source block is in the Hamilton area on the North Island. It has a significant level of infection with GLRaV-1, and this infection is spreading within the vineyard. Examination of the virus infection density map shows that the infection is concentrated towards the top end of the vineyard in two main clusters. Statistical analysis of the clustering shows it is strongly clustered, it is not random. This indicates it is most likely a naturally (insect) introduced infection which is now being actively spread in the vineyard by insect vectors. The second years data shows that there is further spread of the infection around the areas of the original clusters and also further down into the vineyard.



year 1, 2000 ***year 2, 2001***

Riversun Te Kaha Nursery

Photo: Scott Cowan

RIVERSUN

257 Awapuni Rd

PO Box 1199

Gisborne

New Zealand

Tel +64-6-867 1120

Fax +64-6-867 8800

riversun@riversun.co.nz