



Although originally found associated with red blotch disease, GRBaV has also been detected in white grape varieties such as chardonnay.

KNOW YOUR VIRUSES

The most important virus is the one in your vineyard

BY JUDIT MONIS, PH.D.

Throughout my career I have been asked many times: Which is the most important virus affecting the vineyard? The answer is easy: the one in your vineyard (or neighbor's vineyard). But seriously, many factors must be taken into consideration to answer this question.

This article describes known

viruses that affect grapevines and their effect in the vineyard, as well as new viruses to watch out for.

GRAPEVINE ASSOCIATED VIRUS

More than 60 viruses have been described to infect grapevines. Some have serious effects on vine health and subsequent wine quality

to warrant vigilant field testing and pre-planting screening.

When a virus is discovered, it is important to follow certain steps known as "Koch's postulates" to determine if it causes disease. The steps involve the isolation of the virus from grapevines and reproducing specific disease symptoms in a healthy vine. There are a few

roadblocks that make it challenging or even impossible to prove a virus found in grapevines causes a particular disease. First, most grapevine viruses can only infect grapevines. Virus transmission in woody plants, such as grapevines, is much more difficult to accomplish in the lab as compared to in herbaceous plants, which are much easier to manipulate and infect. Second, grapevine viruses are found as mixed infections and it is difficult to isolate one from another. Third, virus transmission in vines occurs only by grafting or mediated by specific insects (mealybugs, soft scale insects) or nematodes known as virus vectors.

Consequently, many of the grapevine-infecting viruses are known as “grapevine associated” with different diseases (e.g. leafroll, rugose wood, red blotch, etc.). When mixed virus infections occur, it is difficult to determine which one of the many present might be causing the problem. Regardless, the recurrent association of viruses can be correlated over the years with symptoms in the vineyard.

DECLINE-CAUSING VIRUSES

The most important group of viruses relative to vineyard dis-

ease management is the Nepoviruses. Nepoviruses are spread by nematodes and cause severe vine decline, leaf deformations and uneven fruit set and maturation in the vineyard. The most common decline-causing viruses are Arabis mosaic (ArMV), Grapevine fanleaf (GFLV), tobacco (TRSV) and tomato ringspot (ToRSV). Infected vineyards rapidly decline and die within three or four years after detection. There is a high degree of specificity between the viruses and the species of nematode able to transmit them: ArMV is transmitted by *Xiphinema diversicaudatum*, GFLV is transmitted by the dagger nematode *Xiphinema index*, and ToRSV

1 to -9) grouped in one family (Closteroviridae) and placed into three taxonomic sub-groups or genera, Ampelovirus, Closterovirus and Velarivirus. The Ampeloviruses – GLRaV-1 and GLRaV-3 being the most common in the vineyard – are transmitted non-specifically by mealybugs and soft-scale insects. GLRaV-2 is a Closterovirus transmitted primarily by grafting. Finally, GLRaV-7 has been proposed to be a member of the Velarivirus because it induces mild leafroll symptoms in grapevines. No insect transmission vector has been described for GLRaV-2 and GLRaV-7.

Viruses associated with rugose wood complex diseases in grape-



This vineyard's leafroll spread was assisted by mealybugs. The virus moves from vine to vine within the row, then to the adjacent row.

and TRSV are transmitted by *Xiphinema americanum*. In infected vineyards (even after removal) remaining nematodes are able to transmit the viruses for many years.

GRAPEVINE LEAFROLL AND RUGOSE WOOD ASSOCIATED VIRUSES

Another important group of grapevine-associated viruses cause leafroll and rugose wood diseases. The virus species that cause leafroll disease are named *Grapevine leafroll associated virus* (GLRaV) followed by a number (e.g., GLRaV-

vines (Grapevine virus A, B, D, E, and F) are members of the Vitiviruses. These viruses are transmitted by the same mealybug and soft scales that spread GLRaV-1 and GLRaV-3 and other Ampeloviruses. Furthermore, GVA and GVB naturally occur as mixed infection with GLRaVs. Reports suggest that Ampelovirus species play a role in aiding the transmission of Vitivirus species that otherwise would not be transmitted by insect vectors.

Grapevine virus A (GVA) is associated with Kober stem grooving and syrah disease. Grapevine virus B (GVB) and D (GVD) are associ-

AT A GLANCE

- + Whether long-known or newly discovered, the most important virus is the one affecting your own vineyard.
- + Certain viruses can have serious effects on vine health and resulting wine quality.
- + Two different DNA viruses were recently discovered: Grapevine vein clearing and Grapevine red blotch-associated viruses.
- + Effective disease control requires clean planting stock, plus frequent disease and insect monitoring.

ated with grapevine corky bark disease. Typical symptoms are related to grafted vines and include cracks and swelling above the graft union, and foliar discoloration.

NEWLY DISCOVERED VIRUSES

Historically, new viruses are typically found by isolating the virus from a diseased host (e.g. grapevine), observed in the electron microscope, and eventually sequenced. Once discovered, testing laboratories can develop testing methods for routine detection of new viruses.

With increased technology in automation and computing, it is now possible to sequence several viruses at once without isolating them from the vine. This method is called Next Generation Sequencing (NGS). While NGS is useful for finding viruses, it does not distinguish between disease-causing and innocuous viruses. It still requires demonstrating Koch's postulates or, at the very least, demonstrating some association with a disease before significant attention should be given to a newly found virus.

Recently NGS was applied to grapevines, resulting in the discov-

ery of two different DNA viruses: Grapevine vein clearing (GVCV) and Grapevine red blotch-associated viruses (GRBaV). Both of these DNA viruses had not been previously detected in grapevines. Neither has satisfied Koch's postulates nor has there been enough time to determine the biological characteristics of the viruses or the diseases with which they have been associated. What is known is that GVCV is a *Badnavirus* found infecting cabernet sauvignon, chardonnay, chardonnay, cabernet franc, riesling, vidal blanc and corot noir in the Midwest. Because GVCV was found in mixed infections with GFLV – a severe pathogen in grapes – it is yet to be isolated and tested separately to determine if this virus has any damaging effect in the vineyard.

The second virus found, GRBaV, is a circular DNA virus isolated from symptomatic cabernet franc vines. Although originally found associated with red blotch disease, GRBaV has also been detected in other grapevine cultivars including white fruited varieties (e.g., chardonnay, pinot gris, etc.) not exhibiting red blotch symptoms. When found in red grape varieties, GRBaV has been detected in vines exhib-

iting red blotchy leaves, red veins and lower Brix levels. The initial genomic characterization of GRBaV found a genetic sequence identity and structural relationship to *Geminiviridae* (50% similarity to chickpea chlorotic dwarf Syria virus). Members of *Geminiviridae* include several viruses that cause serious diseases in crops such as tomato, cassava, beet, cotton, etc. It is too early to know whether GRBaV causes a devastating disease. The good news is that according to a recent report from the UC Davis Foundation Plant Services, no GRBaV-infected vines were found in the newly planted Russell Ranch foundation block and only three hits in the "classic" block.

In the RNA-containing virus world, UC Davis, using deep sequencing, was able to detect a new *Vitivirus* highly related to GVA and GVD in a black table grape variety displaying graft incompatibility. This virus, named Grapevine virus F (GVF), was subsequently detected by Eurofins STA in samples originating in a California chardonnay vineyard displaying decline symptoms. The virus was detected using standard methods because of the multiple complementary diagnostic methodologies applied in our laboratory. It is interesting to note that, in the latter case, the declined vines were co-infected with GLRaV-3, supporting the notion that *Ampeloviruses* (such as GLRaV-1 or -3) may be assisting the transmission of *Vitiviruses* in the vineyard.

While the discovery of new viruses will allow rapid detection and elimination of harmful viruses in planting stock, it seems prudent to first complete research and vineyard surveys to prove that novel viruses play an active role in disease development before focusing attention and investment in their eradication.

For example, research (or cause and effect data) have not been able to correlate that Grapevine Rupes-tris associated virus-syrah strain (GRSPaV-Sy) or grapevine syrah virus -1 (GSyV-1) causes syrah decline (a disease for which its cause still remains unknown). Both



In red grape varieties, GRBaV has been found in vines exhibiting red blotchy leaves, red veins and lower Brix levels.

GRSaV-Sy and GSyV-1 appear to be widespread in vineyards throughout the world and so far have not been linked to a harmful disease.

EFFECTS ON VINE PERFORMANCE AND WINE QUALITY

Research on the effects and advantages of virus elimination on vine performance and wine quality supports the concept that healthy vineyards produce both greater yield and higher quality of grapes, translating into better wine quality. Studies that compare infected and uninfected vines consistently show that the presence of certain viruses have drastic negative effect on both fruit set and quality.

In addition to the decline of vine health, GFLV was shown to interfere with the physical features of the berries, alcohol, tartaric acid and anthocyanins (color) in various grapevine varieties. The elimination of leafroll associated (e.g., GLRaV-1, -2, -3) and Vitiviruses (e.g., GVA, GVB) showed increased vine vigor and fruit production in the vineyard. In the winery, the sugar content was increased while the juice acidity was reduced. Another study noted the beneficial effects on the aroma of produced wine; fruit from a muscat blanc variety subjected to disease elimination produced higher soluble solids and higher amount of free and bound terpene, favorably affecting aroma character.

In the vineyard, viruses and even strains of the same virus may be more or less aggressive than others causing disease. For example, certain strains of GLRaV-2 (red globe in the U.S., BD in Italy, and/or Alfie in Australia) are associated with graft incompatibility, vine decline and death. Other reports suggest that GLRaV-1 and -3 induce more severe symptoms than GLRaV-4 or -5. Certain strains of GVA can cause shiraz disease, a syndrome that induces delayed bud break, rubbery canes and premature death in syrah, merlot and gamay cultivars. So far, the disease has been observed in Australia and South Africa, in spite of Eurofins STA Laboratories detecting the same strains of GVA

in grapevines from California vineyards without displaying characteristic disease symptoms.

Finally, mixed infections of leafroll associated viruses and Viti-viruses can be lethal in some rootstock/scion combinations. Current studies are in progress at UC Davis with different rootstocks grafted to different leafroll species, which will enlighten our understanding on the effect viruses have in the vineyard and how it applies to winemaking.

FREQUENT MONITORING

While there is no clear answer to the question of which is the most important virus in the vineyard, we can definitely conclude that if a decline-causing virus such as GFLV is present, we can expect vine mortality within a few years. On the other hand, if leafroll and/or rugose wood viruses are present (depending on species or combination), we may expect vine decline over time accompanied by a decrease in the quality of fruit and subsequent reduction of wine quality.

It may seem simple to discover and characterize new viruses infecting grapevines; however, it is much harder to determine their effect on cultivated wine grapevine varieties.

Representative studies are required under many different climatic conditions using many rootstock and scion combinations to describe virus effects. In the meantime, effective disease control will require clean planting stock, along with frequent disease and insect monitoring to avoid potential infection.

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Judit Monis is manager of the Plant Health Services Division at Eurofins STA Labs in Gilroy, Calif. The lab provides disease testing services and specializes in grapevine clean planting stock and vineyard disease management. Monis may be reached at juditmonis@eurofinsus.com or (408) 846-9964.

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