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Vine Failure Can Be Caused by Latent Viruses

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ABSTRACT

Determining the cause of failure of young vines in newly planted vineyards can be a difficult task because numerous causal agents can contribute to problems with vineyard establishment. Previously, we have documented sites where stunting and death of young vines appeared to be associated with latent virus infection of scion wood sources. We have taken virus samples from those sites and performed extensive testing involving biological tests, serological tests (ELISA), and DNA tests (PCR) to characterize the virus infections in these samples; good correlations were seen between testing techniques. A new disease, Kober Stem Grooving (Grapevine Virus A) was reported in California for the first time from a latent virus site. Vines were made with Cabernet Sauvignon 05 chip budded on Freedom, Kober 5BB, Teleki 5C and Rupestris St. George and inoculated with three virus combinations. In 3 years of trials, significant differences were seen between healthy vines and virus treatments; the severity of disease was strongly influenced by rootstock identity. Severe virus treatment resulted in the death of some young vines. The severity of the effect of virus on average pruning weights varied by rootstock. This confirms our hypothesis that latent virus severity is determined by rootstock as well as virus type. Freedom rootstock was selected for a large experiment with 22 distinct latent virus isolates varying from mild to severe in type. After 2 years, effects vary with virus from mild (Rupestris stem pitting) to severe (GLRV 2 and Grapevine Virus B). We found that many severe latent virus sources represent multiple infections which include Grapevine Leafroll Virus (GLRV) 2 and Grapevine Virus B.

INTRODUCTION

California vineyards are being planted with increasingly diverse rootstock and scion materials. A large part of the scion wood is uncertified and, therefore, more likely to be virus infected. Sick, dead or dying grapevines were observed in young vineyards throughout the state. Symptoms, vineyard case histories, and our field trials strongly suggest that in many cases, young vine decline can be caused by grapevine latent viruses (Martelli, 1993). Research in our laboratory in this area has three goals: 1) To determine if virus disease can be responsible for young vineyard failures and which virus or combination of viruses may be the causal agent(s); 2) To test selected rootstocks for response to latent viruses; and 3) To apply new molecular tests to our

characterized latent virus selections in hope of developing reliable, fast lab procedures to screen field selections. We believe that accomplishing these goals will improve the ability of growers to avoid replanting problems as well as increasing our basic knowledge of grapevine viruses in ways which will ultimately produce better disease control strategies. In this poster, we report on the results of two sets of experiments which have provided us with clear evidence that vine failure can be caused by latent viruses. First, we have documented the role rootstock genotype plays in the severity of virus response. Second, we have discovered that many latent virus sites are infected with a combination of Grapevine Leafroll Virus 2 and Grapevine Virus B.

MATERIALS AND METHODS

In earlier work, we selected samples from sites in vineyards in Napa, Sonoma, San Joaquin, Merced, and King counties where replant failure may have been caused by latent viruses. Wood was collected, propagated, and planted in a permanent site on the Davis campus. To this set we added some virus isolates from the Davis virus collection (Golino, 1992) that also demonstrated potential in field situations to be latent virus candidates.

This collection of vines, which we call the latent virus collection, has been extensively observed, tested and re-tested for viruses by methods including: ELISA , RT-PCR, herbaceous host indicators, and woody indexing (Rowhani et al., 1995; Rowhani et al., 1997; Rowhani et al., 1998). Testing was done to screen for viruses including: grapevine leafroll viruses (GLRV) -1, -2, -3, -4, and -5, grapevine virus A (GVA), grapevine virus B (GVB), grapevine virus C (GVC), grapevine fleck virus (GFkV), grapevine fanleaf virus (GFLV), Rupestris stem pitting (RSP), tomato ringspot virus (TmRSV), and arabis mosaic virus (ArMV). As new virus types are characterized and new tests become available, the collection will continue to be tested and evaluated to confirm and improve our diagnosis of the virus status of these vines. The reported virus profile of isolates described in this work reflects a diagnosis based on our best interpretation of all test results. Good correlations are seen between testing techniques; at this time, each type of test has a contribution to make to accurate diagnosis of virus status. We have not yet reached a point where a single molecular test will provide information on virus disease profile of a selection although PCR is promising.

Now that PCR techniques are available for RSP (Zhang et al., 1998), it has been demonstrated that RSP is far more wide spread than previously believed in both Registered and non-Registered selections of grapes. There is also evidence RSP may spread readily in field conditions (Rowhani, unpublished). In this work, we are currently disregarding RSP status until more information is available.

Virus effects on growth and survival of grafted plants - Three field trials are in various stages of progress to quantify the effects of latent viruses on the growth and survival of grafted grapevines. This poster reports on the results from the first and oldest trial which was budded in 1996 and planted in 1997. Healthy and virus - infected budwood of Cabernet Sauvignon-5 was chip budded on four rootstocks: Rupestris St. George, Freedom, Kober 5BB, and Teleki 5C. Virus

sources included were: grapevine leafroll virus 101 (LR101), a selection infected with GLRV-3 in which no other virus types have been detected; grapevine leafroll virus 102 (LR102), a multiple infection of GVB, GLRV-1, and -2; and grapevine leafroll virus 109 (LR109), a multiple infection of GVC, GLRV-2 and -3.

Parameters measured include: survival, symptoms, diameter above and below the grafted bud, length of the longest shoot, and pruning weight. Grafts were observed for necrosis and abnormalities.

Virus effects on Freedom rootstock -Trials are ongoing to survey the effects of a broad range of latent viruses on the rootstock Freedom. Freedom was selected as a test rootstock because it is a popular rootstock known to be especially sensitive to latent virus problems. In 1997, approximately 900 Freedom rootstocks were chip budded with 22 selected latent virus isolates from the latent virus isolate collection and planted in a completely randomized plot field design. Disease symptom observations were made throughout the following two years and vine vigor assessed by measuring the length of the longest shoot and pruning weight. Data was only recorded for those vines where the virus inoculation was judged successful because the bud from the disease isolate source survived; the bud was then removed to prevent the virus scion source from overgrowing the Freedom rootstock.

RESULTS

Virus effects on growth and survival of grafted plants- During the first two years following planting, virus treatment had a profound effect on the survival of the young vines grafted to Freedom and Kober 5BB (Figure 1). The effect of LR 101, infected with only a single virus GLRV-3, was relatively insignificant. It is interesting that survival of young vines grafted onto the rootstock Rupestris St. George seems to be relatively unaffected by these viruses, suggesting a greater tolerance of infection.

Vine vigor varied dramatically between treatments (Figure 2). Pruning weights were collected in early winter for the vines in each treatment group which were still alive in the second fall. After two years, there was a range of responses to the severe virus treatments (Figure 3). Plants grafted on Freedom and Kober 5BB were extremely stunted compared to single virus infection and the controls. The response of Teleki 5C was intermediate although there were significant differences between the plants infected with isolates with multiple viruses (LR 102, LR 109) and those with no virus or the single infection of leafroll (LR 101). Rupestris St. George appears to be relatively insensitive to virus since it exhibited only non-significant response to virus infection.

Virus effects on Freedom rootstock - Freedom appears to be particularly sensitive to multiple virus infection. Virus symptoms observed on leaves during spring of a Freedom rootstock trial exemplified the fact that virus effects on Freedom are highly variable, ranging from no observable effect to severe stunting (Figure 4), leaf deformation, yellowing, chlorotic line patterns and blotches. Necrosis of the vascular system is also associate with some virus isolates

(Figure 5). Multiple leaf symptoms and low pruning weight are correlated with infection by more than one virus.

Growth in the spring of 1999 was assessed by measuring the length of the longest shoot on plants in the trial. As can be seen in Figure 6, there is a sharp difference between the success of those plants which are infected with no virus or single infections of a virus and those with multiple infections. We do not yet know if the mild effects seen when stock is infected with LR 101, a single grapevine leafroll infection, are due to a mild strain of the virus or whether single infections are always relatively mild. Field collected latent virus isolates (LV91-1, LV91-2, LV92-1, LV92-2, LV92-6) and select Davis virus collection isolates all had profound, statistically significant effects on the growth of Freedom rootstock, supporting our hypothesis that the virus status of the scion can be a major factor in the decline of young vineyards (Golino, 1993).

Diagnosis of latent virus candidates - In Figure 6, a virus profile is provided for each latent virus isolate, virus collection isolate, and controls. In studying these profiles, an important observation can be made. In all but one case, if severe latent virus effects on Freedom rootstock are observed, both GLRV-2 and GVB are present. In the one exception, LV-91-2, GVA and GVC were both present and may be contributing the same disease factor as GVB. In our work to date, GVB is the most common grape vitivirus in California. The vitiviruses are the rugose wood causing grapevine viruses that include GVA, GVB, GVC, and GVD.

CONCLUSIONS

It is apparent from our field trials that it is possible for multiple virus infection to cause young vine failure. Infection with some virus combinations can cause stunting and death of both grafted vines and rootstock alone. This phenomena has been widely referred to as the latent virus problem. We offer the first experimental results which confirm a cause and effect relationship between the presence of certain viruses in propagating stock and the decline of young vines.

Although this research is not yet complete, the correlation between the presence of simultaneous infections with GLRV-2 and GVB with latent virus symptoms is striking. In our continuing studies, we will be making artificial mixes of these viruses to determine whether all strains of LR-2 and GVB can cause this profound disease reaction. It seems likely that the combination of some grapevine leafroll viruses with one or more of the vitiviruses can cause the phenomena known as latent virus decline of grapevines.

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Fig. 1. Percent survival 2 years after planting of healthy and virus-infected Cabernet Sauvignon scions chipbudded to 4 rootstocks.

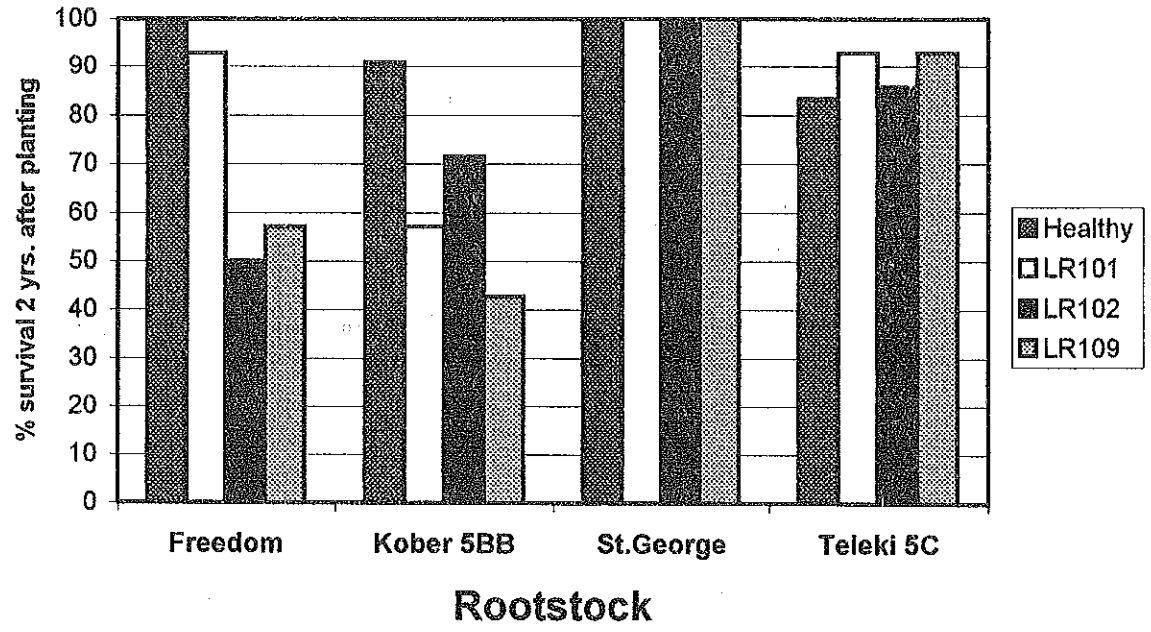
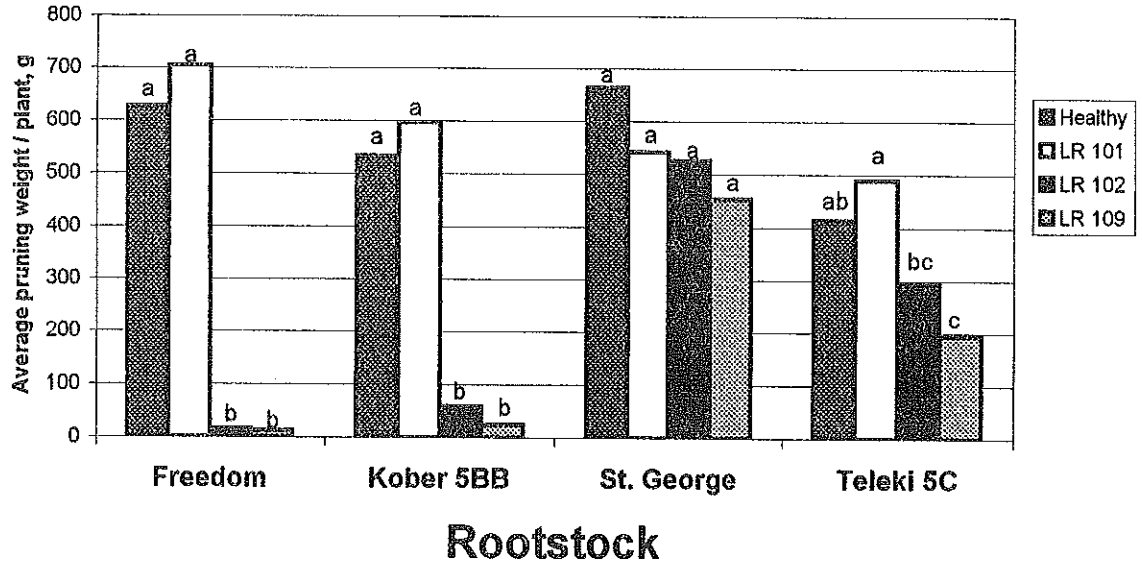


Fig. 3. Pruning weight of healthy and virus-infected Cabernet Sauvignon scions chipbudded to 4 rootstocks.



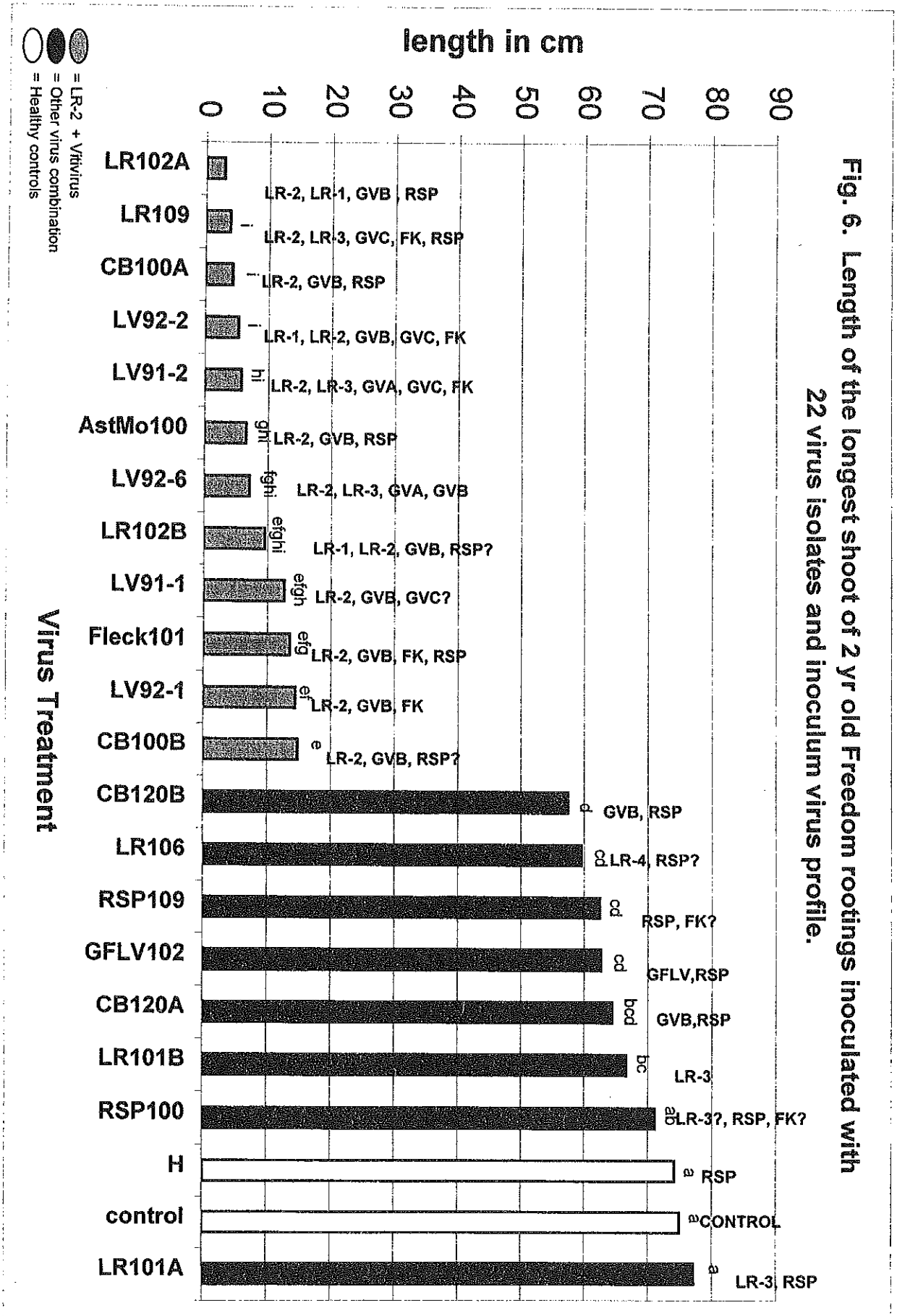


Fig. 6. Length of the longest shoot of 2 yr old Freedom rootings inoculated with 22 virus isolates and inoculum virus profile.