

## PROTECTION OF GRAPEVINES FROM RED BLOTCH BY UNDERSTANDING MECHANISTIC BASIS OF ITS INFECTION

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### Abstract:

**Context and purpose of the study** - Currently, grapevine is host to a large number of pathogenic agents, including 65 viruses, five viroids and eight phytoplasmas. Needless to say, these pathogens, especially viruses responsible for several 'infectious degeneration' or 'decline' cause great distress to wine makers and grape growers, let alone the large economic losses incurred by the wine industry. A recent addition to this wide repertoire of grapevine viruses is a new viral disease known as Red Blotch in viticulture parlance. Its causal organism, *Grapevine red blotch associated virus* (GRBaV), discovered in 2008 is a newly identified virus of grapevines and a putative member of a new genus within the family Geminiviridae. Typical symptoms in red cultivars include red blotches in leaves of afflicted vines with pinkish red-colored veins without any rolling of the margins observed at the onset of ripening. The objective of this study was to determine as to how the virus enters grapevine and once it enters the vine, how it is distributed throughout the vine.

**Material and methods** - During the growing season, vine samples were collected from vineyards with a history of Red Blotch (both red and white cultivars) located in Napa/Sonoma counties and the state of Washington. Starting at flowering, shoots (leaf and stem tissues) were sampled for microscopy analysis. These samples were used to determine the structure and functionality of the vascular strands (xylem and phloem) using callose specific dye, aniline blue and various techniques of microscopy.

**Results** - The afflicted fruits were high in acid but low in sugars causing delay in harvest. Canematuration was poor as evident from uneven browning characterized by areas of green and brown color. Typically, green stems fail to turn brown when the periderm is not formed indicating that the GRBaV interferes with cane maturation process. Callose (a carbohydrate substance) accumulated in the phloem cells of afflicted vines. Such deposition indicated that the GRBaV is primarily confined to phloem sieve tubes. Since callose deposits limit pathogen dispersal, this phenomenon could be used as a diagnostic indication of Red Blotch. Similarly, in the context of vascular blockage, the xylem vessels of afflicted canes showed tylosis, which is ballooning of neighboring xylem parenchyma cells into the lumen of the vessels. These observations indicated that even though the GRBaV enters grapevine via the phloem, both xylem and phloem responded to the virus invasion. This is an interesting observation and needs further investigation as tylosis typically occurs only when the xylem is injured either by pruning or occluded with bacteria. Vessels of healthy canes remained free of tylose. These results added a new dimension to viral diseases, especially Red Blotch and might provide a framework for developing management strategies to minimize the incidence of Red Blotch in grape growing regions worldwide.

**Keywords:** Callose, Grapevine, Phloem, Red blotch, Tylosis, Xylem.

### 1. Introduction.

Protection of grapevines from Red blotch by understanding mechanistic basis of its infection

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Introduction and Objectives

Fed Blotch caused by Grapevine red blotch associated virus (GRBaV) cause great distress to wine makers and grape growers, let alone the large economic losses incurred by the wine industry (Sudarshna et al., 2015). Typical symptoms include red blotches in leaves with pinkish red-colored veins without any rolling of the margins observed at the onset of ripening. The objective of this study was to determine as to how the virus enters grapevine and once it enters the vine, how it is distributed throughout the vine.

Materials and Methods

During the growing season, vine samples were collected from vineyards with a history of Red Blotch (both red and white cultivars) located in Napa/Sonoma counties and the state of Washington. Starting at flowering, shoots (leaf and stem tissues) were sampled for microscopy analysis. These samples were used to determine the structure and functionality of the vascular strands (xylem and phloem) using callose specific dye, aniline blue and various techniques of microscopy.

Results and Discussion

The xylem and phloem pathways in the infected vines was altered, which explains the frequently observed diminished sugar accumulation in the berries of infected vines. For instance, the infected vines showed callose accumulation as typically occurs in distressed phloem (Bondada 2014) or tylosis in xylem vessels as found in vines afflicted with physiological disorders (Bondada and Keller, 2012). Other details are shown in figures 1 through 3.

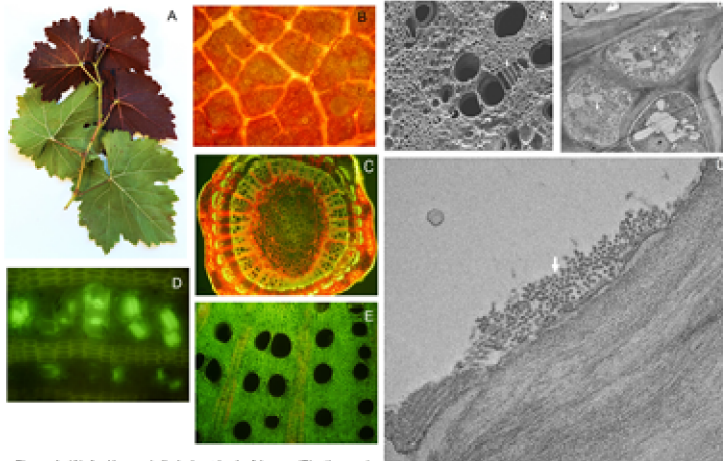


Figure 1. (A) A phloem girdled shoot by leaf hoper, (B) anthocyanin accumulation in leaves, (C) cross section of infected (Red Blotch) shoot, (D) phloem tubes showing callose, and (E) healthy xylem vessels.

Figure 2. (A) Xylem with radial multiples, (B) phloem with dense cytoplasm, and (C) phloem particles in the infected vines.

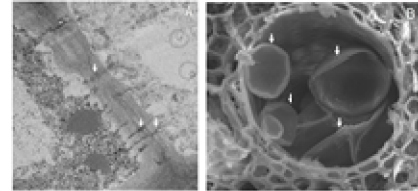


Figure 3. (A) numerous plasmodesmata and tylosis in xylem vessels of infected vines.

Conclusions

Such a wide range of knowledge will ensure that the causal factors behind the spread of Red Blotch over time can be accurately determined, and will undoubtedly be necessary to deliver effective management solutions in the near future.

References

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