Title: Grape trunk disease survey in North Carolina and Georgia

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Abstract:
Grape Trunk Disease (GTD) is a wood borne, deadly disease in grapes, associated with a host of different fungal pathogens. Good management of GTD often is the decisive factor for the longevity of a vineyard. GTD related disease symptoms were found in virtually all vineyards in North Carolina, both muscadine and bunch grape, mostly related to *Botryopshoaeria* species. However, management methods of GTD are not investigated for the humid and wet climate in the Southeast. Therefore we asked the SRSFC to funding initial investigations of a new pruning wound protectant and a survey of trunk disease in North Carolina and Georgia. However, in 2020, COVID-19 restriction as well as very late frost events had a devastating effect on grape research in North Carolina. Although efforts were made to collect plant material for fungal isolations, harvest the field trial and assess berry chemistry, results are not complete and inconclusive at the best. However, our group has managed to acquire funding not just to repeat the experiments, but to conduct a more comprehensive investigation into the distribution and management of grape trunk diseases in 2021 and 2022. In this regard, this project still was a success for the Southeast and North Carolina.

Objectives:
1. Assessment and documentation of the abundance of trunk disease damage in selected vineyards in North Carolina and Georgia
2. Identification of the main trunk disease pathogens present in North Carolina and Georgia
3. Development of a field trial to assess trunk disease management methods

Introduction:
Over the last two years, several die-back and sudden death incidences in vineyards in North Carolina were linked to grape trunk disease (GTD), caused by wood borne fungal pathogens. In the year 2018 alone, the Plant Disease and Insect Clinic in North Carolina processed approx. 20 samples linked to GTD. Moreover, die-back symptoms were consistently observed in both vinifera/hybrid style wine grape vineyards and well as muscadine vineyards by extension personnel (Figure 1). GTDs occur in all grape growing regions in the world, and if unattended, lead to a reduced lifespan of vineyards, increased costs of production and to less favorable berry parameters. A variety of fungal pathogens are related to GTD, with physical damage to the trunk or cordon facilitate infection. We assume that cold damage and open pruning wounds are the number one entry points. Infection usually occurs by spores intruding into the wood vessels, transported by rain and water (Gramaje et al. 2018, Appel and Brown, 2017). GTD infections occur many years before a vine becomes symptomatic. However, dying plants can have a detrimental effect on vineyard productivity and therefore on the economic value of a vineyard (Gramaje et al. 2018, Fontaine et al. 2016). If unattended, vineyard productivity can decrease up...
to 80% over a span of 3-5 years (Baumgartner et al. 2019). Typical symptoms can reach from dead spurs, stunted shoots, crispy leaves to shriveled fruit and even sudden collapse of a vine (Figure 1).

While management methods are well established in other grape growing regions and part of a vineyard management routine, in North Carolina (and in the Southeast for that matter), GTDs are not addressed in most vineyard management routines. That is particularly a problem due to the high amounts of rainfall and the frequency of cold damage in Southeastern vineyards. Initial isolations from a few symptomatic vineyards have shown that mostly organisms of the large group of *Botryosphaeria* are present in North Carolina and Georgia (Cline, Brennan, Hoffmann, Villani pers. comm.). However, multiple questions are still unanswered, regarding the unique growing conditions in the Southeast. First, it still is not clear which other organisms are involved in the observed symptoms. Secondly, the epidemiology of those organisms in the Southeast is not very well understood. And third, trunk disease management methods established in dryer climates cannot be transferred 1:1 to the Southeast and will need exploration.
Figure 1. *Upper left:* Muscadine vines affected by trunk disease often lose productivity and show dead spurs or dead parts of the cordon. *Upper right:* Typical indication for trunk diseases are pruning wounds with dark areas. *Lower left:* Trunk diseases can lead to sudden death of vines. *Lower right:* Leaf symptoms sometimes can reassemble Pierces’ Disease symptoms (all pictures by Mark Hoffmann).

COVID-19 and spring frost damage in 2020:

We originally proposed to survey selected muscadine and vinifera/hybrid wine grape vineyards in North Carolina and Georgia for the presence and severity of trunk disease organisms, to identify those organisms via molecular methods, and to establish a field trial for the initial investigation of cultural and chemical methods to control trunk diseases under our regional high precipitation viticulture conditions. However, due to COVID-19 we were not able to conduct a trunk-disease survey and we needed to limit the amount of fungal isolations from the field trial. Moreover, the field trial was compromised by several late spring frosts in 2020, leading to more than 70% of average yield loss in the vineyard. Here we report berry chemistry, yield data and isolation results of this trial. However, the funding provided through the SRSFC has served as seed for PIs Hoffmann and Villani to secure a fully funded project through the USDA Specialty Crops Block Grant Program (SCBGP) to investigate grape trunk disease distribution, identification and management in the years 2021 and 2022 in North Carolina. In summary, while the 2020 grape season in North Carolina was compromised by limited research capacity due to COVID-19 and cluster loss due to several late spring frost events, we were still able to generate preliminary data and funding for a comprehensive two-year study on identification and management of grape trunk diseases in the Southeast.

Methods:

To investigate the impact of pruning and chemical practices, a field trial will be established in December 2019 at Burnshirt vineyards in a block of ‘Merlot’, affected by trunk disease symptoms. Following treatments will be established in four replicates in a randomized complete block design (Figure 2).

1. Single pruning
2. Double (delayed) pruning
3. Single pruning + VitiSeal (1 application directly after pruning)
4. Single pruning + VitiSeal (2 applications)
5. Double (delayed) pruning + VitiSeal (late application)

All vines were pruned to two buds per spur, and shoot thinned to 24 fruitful spurs per vine. Prior to the initiation of pruning treatments, samples were arbitrarily collected from each vine, with no greater than five samples per vine. At harvest, cane tissue exhibiting dieback symptoms were
collected from each treatment replicate with no greater than five sample collections per vine. The majority of samples have been stored at 4 deg. C until labor can be hired (postponed due to COVID19) to isolate fungal pathogens from woody tissue. However, an initial subset of pre-treatment isolations were conducted prior to the COVID shutdown at MHREC. To measure the impact of pruning and chemical techniques on yield and berry composition, harvest data and berry composition parameters were be taken at the end of the trial.

Results and Discussion:

**Figure 2:** Application of VitiSeal in one of the treatments in March 2020.

The late spring frost events end of April 2020 and mid May 2020 have led to the loss of primary clusters and the growth of secondary shoots and the breaking of latent buds. The number of clusters per vine, while usually standardized in research trials, ranged after the frost events from 2 clusters per vine to 18 clusters per vine, depending on the location of the vine, but not in the treatment. Results are shown in Table 1 and 2.

**Table 1:** Average harvest parameters (lbs/vine; number of clusters; average cluster weight) ± Standard Deviation. No significant difference were detected.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yield (lbs/vine)</th>
<th>Number of Clusters</th>
<th>Cluster Weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Delayed Pruning</td>
<td>2.5 ± 1.2</td>
<td>8.5 ± 3.8</td>
<td>135.3 ± 36</td>
</tr>
<tr>
<td>Delayed Pruning + VitiSeal</td>
<td>2.1 ± 1.2</td>
<td>7.5 ± 3.1</td>
<td>114.1 ± 42.7</td>
</tr>
<tr>
<td>Single Pruning</td>
<td>2.6 ± 1.1</td>
<td>8.9 ± 3.4</td>
<td>133.6 ± 26.8</td>
</tr>
<tr>
<td>Single Pruning + 1x VitiSeal</td>
<td>3.1 ± 1.5</td>
<td>10.7 ± 5</td>
<td>136.6 ± 31.9</td>
</tr>
<tr>
<td>Single Pruning + 2x VitiSeal</td>
<td>2.8 ± 1.2</td>
<td>10.3 ± 3.7</td>
<td>125.8 ± 18.4</td>
</tr>
</tbody>
</table>
Several species were isolated from wood and identified through sequencing. Due to COVID-19, only qualitative isolations could be performed. No quantitative assessments related to the treatments could be conducted, due to COVID-19 related University restrictions at NC State University. Following species or genera could be identified over the course of 2020 in infected plant material: *Fusarium* sp.; *Fusarium graminearum*; *Colletotrichum fioriniae*; *Pestalotiopsis*; *Neopestalotiopsis clavispora*; *Epicoccum nigrum*; *Guignardia* sp.; *Botryoshpaeria* sp. The remaining isolations will be conducted and reported as labor and increases in lab capacity are permitted at MHCREC, NCSU.

**Conclusion:**

In 2020, COVID-19 and the related labor issues as well as late frost events had a devastating effect on grape research in North Carolina. Although efforts were made to collect plant material for fungal isolations, harvest the field trial and assess berry chemistry, results are not complete and inconclusive at the best. However, our group has managed to acquire funding not just to repeat the experiments, but to conduct a more comprehensive investigation into the distribution and management of grape trunk diseases in 2021 and 2022. In this regard, this project still was a success for the Southeast and North Carolina.

**Literature:**

