

Reducing Sour Rot Spray Applications Initiated after Symptom Development Does Not Impact Disease Control

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Summary

Goals: Previous sour rot research indicates the highest efficacy for sour rot treatments beginning prior to the onset of symptoms, yet many grapegrowers delay applications until after symptoms develop and continue weekly until harvest, which can exceed four sprays. This number of broad-spectrum insecticide applications is costly both financially and environmentally, and risks developing resistant populations of *Drosophila* fruit flies. The objective of this study was to determine the efficacy of a reduced number of spray applications beginning after symptom development, comparing weekly sprays totaling four applications to a reduced number of two to three applications based on total soluble solids.

Key Findings:

- In both years of the trial, when chemical sprays were applied post-sour rot symptom development, more sprays did not equal more control. Reducing the number of post-symptom applications is a substantial cost-saving measure and is beneficial for environmental sustainability.
- In 2018, there were no significant differences between sour rot severity in both vineyard sites used in the trial. Incidence was significantly lower for the industry standard of weekly applications initiated at 15 Brix until harvest only in one vineyard site compared to applications at 16 and 20 Brix and applications at 16, 19, and 22 Brix.
- In 2019, there were no significant differences observed in disease severity and incidence between two sprays and weekly applications beginning at 15 Brix in both vineyard sites.

Impact and Significance: Many growers choose to initiate sour rot control applications only after symptoms develop. Weekly applications are financially and environmentally costly, and the efficacy of timing applications initiated post-symptom development has yet to be researched. In this study, we show that a reduced number of chemical applications to control sour rot post-symptom development offers the same control as weekly sprays, consistent across both years of the study. Relying on weekly applications initiated only after sour rot symptoms develop does not significantly decrease incidence and severity at harvest. If sour rot symptoms are present, more spray applications do not offer better control.

Key words: bunch rot, chemical control, disease control, insecticide, spray reduction

Overview

Sour rot is a late-season bunch rot that is defined by browning of berry skin and liquefaction of berry pulp, accompanied by a strong smell of acetic acid, coinciding with the presence of *Drosophila* spp.^{1,2,3} Affected grapes include white and red cultivated *Vitis* spp. and *Vitis* interspecific hybrids, particularly ones that have high cluster compactness and are susceptible to berry splitting.⁴ Sour rot causes significant yield losses as the disease progresses rapidly in the final weeks before harvest, leading to fruit that cannot be harvested or

may be rejected in the winery due to reduced quality.^{5,6} Sour rot significantly modifies the crushed fruit juice, with higher total and volatile acidity along with a lower pH compared to healthy berries that are not affected by sour rot.^{2,5}

For sour rot symptoms to develop, grape berries require a wound site, yeast, acetic acid bacteria (AAB), and fruit flies (*Drosophila* spp.).³ Wounds are necessary to initiate sour rot development within the field, and *Drosophila* spp. play a critical role in the establishment and spread of sour rot among clusters.^{1,2,3,7} Yeast and bacteria are present within and on the surface of healthy grape berries,⁸ but wound sites provide opportunities for *Drosophilids* to lay eggs. Damage to berries occurs throughout the season by a multitude of factors. Birds can damage berries, as well as insects, including wasps, grape berry moths, and Japanese beetles.^{2,9,10,11} Additionally, fungal diseases, such as powdery mildew and botrytis bunch rot, and abiotic factors, such as berry splitting due to increased rainfall or hail damage, all increase the likelihood of sour rot development.^{11,12} Berry microbiota diversity and population size are heavily influenced by damage, allowing an opportunity for new microbial species to become established in the immediate area around these wound sites.²

Additionally, *Drosophila* spp. are known to carry microorganisms on their bodies and in their guts, particularly yeast and AAB.^{13,14,15} *Drosophila* spp. use grapes for feeding and reproduction^{16,17,18} and are attracted to volatiles in overripe grapes that contain fermenting yeast and acetic acid.^{19,20} Fruit flies are causal organisms of sour rot and are implicated in the loss of berry integrity, which does not occur without the presence of fruit flies.³ Preventing an infestation of fruit flies in the vineyard is the best control measure against the spread of sour rot.

An important observation by Bisiach et al. (1986) was that insecticidal sprays effectively targeted *Drosophila* spp. and lowered sour rot infection compared to untreated vines.¹ However, they noted that disease was best controlled when wounds were reduced and fruit flies were repressed, with the most significant reduction of sour rot from the use of plastic nets or cheesecloth that prevented fruit fly infestation. This was further demonstrated by Barata et al. (2012), who showed sour rot did not spread in wounded clusters that were excluded from fruit fly access using nylon nets around individual clusters.²

Additionally, more recent research has shown that insecticide-treated vines, when sprayed concurrently with antimicrobials, had significantly less sour rot compared to vines treated only with antimicrobials and that insecticide treatments initiated before the onset of symptoms

could successfully control sour rot.⁶ Delaying chemical treatments until symptoms were visible resulted in less control of sour rot when compared to preventive sprays initiated before symptom development. In those same trials, insecticide sprays targeting fruit flies showed up to a 99% reduction in the number of *D. melanogaster* adults reared from Mustang Maxx (zeta-cypermethrin) treated vines compared to control vines not treated with insecticide.⁶

In a separate study, preharvest timing of potassium metabisulfite (KMS) and Milstop (potassium bicarbonate) treatments were evaluated for control against sour rot and lowering of volatile acidity of must.²¹ In both years of the study, Milstop and KMS were able to reduce sour rot compared to untreated vines.²¹ However, delaying KMS applications and reducing the number of applications during the season did not affect sour rot severity or volatile acidity.²¹

Previous research shows that applying chemical applications before sour rot symptoms develop is the best measure for reducing sour rot incidence and severity,⁶ however, many growers choose to delay chemical treatments until symptoms develop because of the cost of materials, time required for spraying, environmental cost of applications, or unknowns of whether the disease will develop in certain blocks or vineyards. The goal of this research was to determine the efficacy of various spray timings initiated after the onset of sour rot symptoms, comparing a typical commercial schedule of weekly applications to a reduced number applied at specific total soluble solids (TSS) measurements. Treatments were assessed visually by sour rot incidence (percent of diseased clusters per vine) and severity (percent of affected berries per cluster).

Major Observations and Interpretations

In both Vineyards 1 and 2, weekly treatments were initiated when berries reached 16 Brix. Vineyard 1 did not keep specific dates for when applications were applied, but treatments were applied when TSS reached the selected levels. Vineyard 2 application dates in 2018 and 2019 are recorded in Tables 1 and 2. In Vineyard 2, the 20 Brix treatment was reapplied after five days due to precipitation above 25 mm on the day after treatment in 2018. For weekly treatments, a total of four sprays were applied in both 2018 and 2019.

In Vineyard 1, precipitation between the start of applications and harvest was slightly lower in 2018 than 2019, a difference of 22.36 mm (Figure 1), and average daily temperatures were very similar in both years from

the start of applications to harvest, with an average of 22.8°C in 2018 and 22.6°C in 2019. In Vineyard 2, precipitation between start of applications and harvest was higher in 2018 than 2019, a difference of 72.49 mm (Figure 2), and average daily temperatures were similar in both years, 23.9°C in 2018 and 23.3°C in 2019, between the start of applications and harvest.

All of the treatments were compared to the industry standard of weekly applications initiated once symptoms developed, instead of an untreated control, because the trials were conducted in commercial vineyards and vineyard managers would not agree to a no-spray treatment due to past prevalence of sour rot and potential crop losses that would result from a no-spray control. Results of this trial show that more post-symptom spray applications did not lead to better control of sour rot. In 2018, Vineyard 1 did not have significant differences in severity between treatments ($p = 0.0612$); however,

Table 1 Spray application dates for sour rot control in 2018 and total soluble solids (TSS) for *Vitis* interspecific hybrid Vignoles in Vineyard 2, located in Ste. Genevieve, MO.

Date	TSS (Brix)	Applications
26 July	16.20	
28 July		Weekly and 16 Brix
3 Aug	18.80	
4 Aug		Weekly and 19 Brix
6 Aug		20 Brix
10 Aug	20.30	
11 Aug		Weekly and 20 Brix ^a
16 Aug	21.80	
18 Aug		Weekly and 22 Brix
22 Aug	22.30	Harvest

^a20 Brix treatment was reapplied due to rain accumulation >25 mm.

Table 2 Spray application dates for sour rot control in 2019 and total soluble solids (TSS) for *Vitis* interspecific hybrid Vignoles in Vineyard 2, located in Ste. Genevieve, MO.

Date	TSS (Brix)	Applications
29 July	12.00	
31 July		Weekly and 13 Brix
1 Aug	13.80	
5 Aug		Weekly and 16 Brix
8 Aug	16.60	
12 Aug		Weekly and 18 Brix
14 Aug	19.40	
18 Aug		Weekly and 20 Brix
19 Aug	21.20	
21 Aug	20.90	
28 Aug	22.60	
2 Sept		Harvest

weekly applications starting at 15 Brix resulted in significantly lower disease incidence than applications at 16 and 20 Brix and applications at 16, 19, and 22 Brix ($p = 0.001$) (Figure 3). Vineyard 2 did not have a significant treatment effect on incidence ($p = 0.584$) or severity ($p = 0.987$) (Figure 4). In 2018, average sour rot severity was significantly higher ($p = 0.0001$) in Vineyard 2, in which canopies had a broad horizontal spread with significantly higher average severity ($p = 0.0087$) and incidence ($p = 0.0136$) compared to Vineyard 1.

In 2019, two treatments were compared to weekly sprays initiated after symptom development: the first consisting of applications at 16 and 20 Brix and the other with applications at 13 Brix and 18 Brix. This earlier timing was included to account for development of sour rot symptoms prior to 15 Brix. In Vineyard 1 we saw no significant effect of treatment on incidence ($p = 0.4062$) or severity ($p = 0.2680$) (Figure 5). In Vineyard 2, there were no significant differences for incidence ($p = 0.2086$)

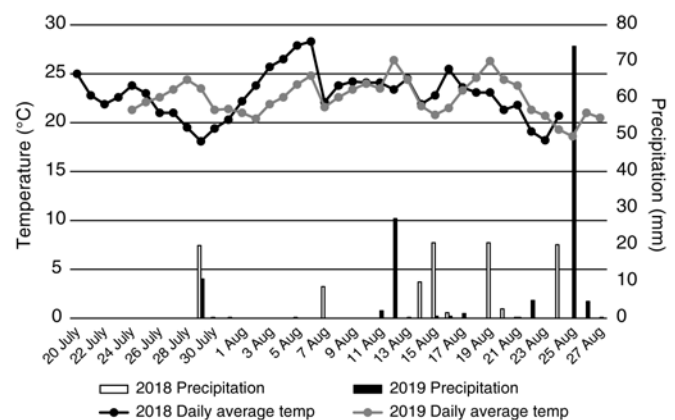


Figure 1 Average daily temperature (°C) and daily precipitation (mm) for Vineyard 1 located in Hermann, MO, from the start of sour rot applications to harvest in 2018 and 2019.

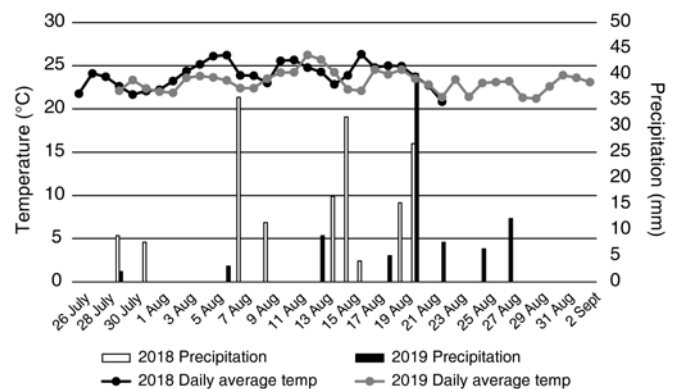


Figure 2 Average daily temperature (°C) and daily precipitation (mm) for Vineyard 2 located in Ste. Genevieve, MO, from the start of sour rot applications to harvest in 2018 and 2019.

and severity ($p = 0.1999$) between any of the treatments (Figure 6). In both vineyards, there was a slight reduction in severity and incidence in the treatment that included an application before 15 Brix, yet sour rot symptoms had already begun at the time of the earlier spray at 13 Brix, which would account for severity and incidence not being affected by harvest. During this time, fruit flies were trapped in Vineyard 2 using monitoring cups that captured *Drosophila* spp. in the week leading up to the 13 Brix application. An earlier spray timing that could have been applied before symptom development began would have been useful for comparison in this trial. Both years of spray trials suggest that when vineyards wait to apply

insecticides and antimicrobials until symptoms develop, applying more sprays does not offer more control, and two sprays can be just as effective as four.

Our experiment was conducted in two commercial vineyards in which entire rows were sprayed with the corresponding treatment, and those rows not included in the study were sprayed using the industry standard treatment of weekly applications beginning at 15 Brix. This comparison of treatments across vineyard rows provided the opportunity to examine disease control within a block, limiting the potential impact of other

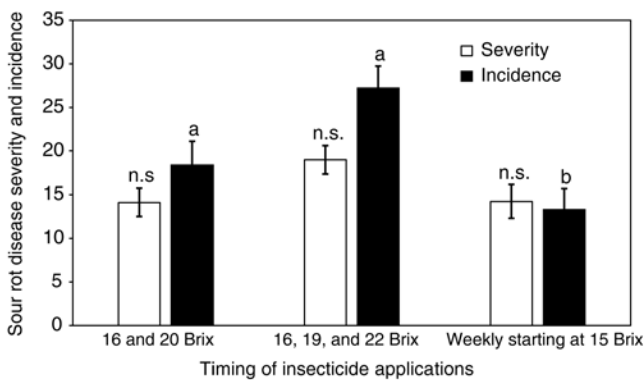


Figure 3 Average severity and incidence for sour rot treatment chemical application timings in 2018 for Vineyard 1. Error bars indicate standard error of the mean for both severity and incidence. Using one-way analysis of variance, application timings had a significant treatment effect for incidence ($p = 0.001$) but not severity ($p = 0.0612$). Using Welch’s test, incidence showed unequal variances ($p = 0.001$). Means not sharing the same letter were significantly different and means with “n.s.” were not significantly different ($\alpha \leq 0.05$, Tukey-Kramer honest significant difference test). Sample size was 75 vines.

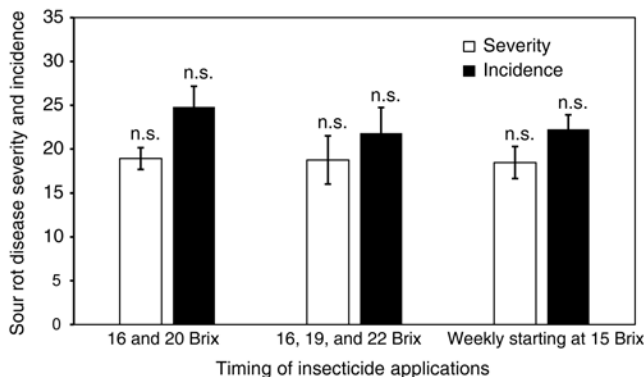


Figure 4 Average severity and incidence for sour rot treatment chemical application timings in 2018 for Vineyard 2. Error bars indicate standard error of the mean for both severity and incidence. Using one-way analysis of variance, application timings had no significant treatment effect for incidence ($p = 0.5844$) or severity ($p = 0.987$). Means with “n.s.” were not significantly different ($\alpha \leq 0.05$, Tukey-Kramer honest significant difference test). Sample size was 30 vines.

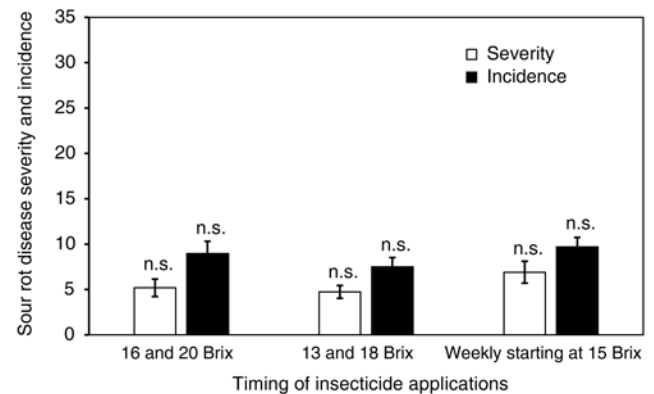


Figure 5 Average severity and incidence for sour rot treatment chemical application timings in 2019 for Vineyard 1. Error bars indicate standard error of the mean for both severity and incidence. Using one-way analysis of variance, application timings did not have a significant treatment effect for incidence ($p = 0.4062$) or severity ($p = 0.2680$). Means not sharing the same letter were significantly different and means with “n.s.” were not significantly different ($\alpha \leq 0.05$, Tukey-Kramer honest significant difference test). Sample size was 45 vines.

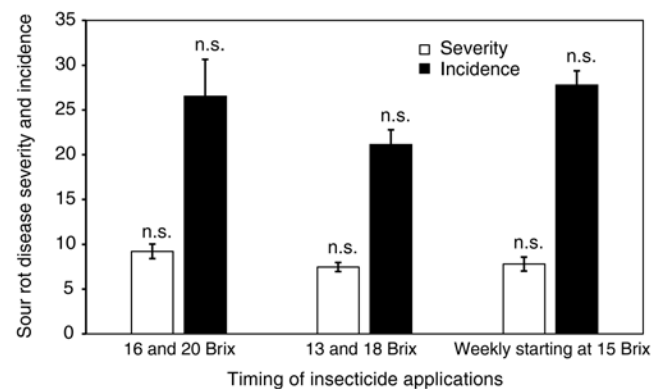


Figure 6 Average severity and incidence for sour rot treatment chemical application timings in 2019 for Vineyard 2. Error bars indicate standard error of the mean for both severity and incidence. Using one-way analysis of variance, application timings did not have a significant treatment effect for incidence ($p = 0.2086$) or severity ($p = 0.1999$). Using Welch’s test, incidence showed unequal variances ($p = 0.0360$). Means with “n.s.” were not significantly different ($\alpha \leq 0.05$, Tukey-Kramer honest significant difference test). Sample size was 30 vines.

rows to serve as a source of fruit flies, which would be a concern if rows were unsprayed. This experiment design allowed us to determine the efficacy of reducing sprays once symptoms develop in a commercial setting compared to the industry standard, clearly demonstrating the need for better prediction of symptom development, so that more precise chemical management strategies can be used.

Broader Impact

Sour rot chemical management strategies are most effective when initiated prior to symptom development, but vineyards commonly wait to treat vines until the disease is present. The reasons for this decision may vary from high costs associated with starting applications earlier in the season to a lack of scouting for fruit flies and disease symptoms. This report addresses growers who initiate sour rot control strategies after symptom development, demonstrating that weekly sprays initiated after symptom development do not significantly reduce sour rot by harvest and, by extension, shows that a reduced number of sprays is just as effective.

When sprays are initiated before the onset of sour rot symptoms, fruit fly populations can be controlled effectively using weekly sprays,⁶ but waiting until disease symptoms develop, and therefore fly populations are established, will lead to difficulty in controlling emerging new fruit flies, regardless of how many sprays are applied. Therefore, the frequency of the chemical applications can be reduced, as weekly sprays are not providing adequate control. Lowering the frequency of applications after symptom development is less costly to growers (Table 3), and because frequent applications of insecticides can select for resistant fruit fly populations, maintaining weekly sprays even with high populations of fruit flies could result in resistance of those flies to the applied insecticide.²² Moreover, more sprays do not provide better control of sour rot and do not impact sour rot incidence or severity at harvest when initiated after symptoms develop. Therefore, as demonstrated in previous studies, if growers begin weekly sprays after symptom development, there is inadequate disease control partnered with a greater cost and increased potential for fruit fly resistance. Reducing sprays initiated after symptom de-

velopment to two sprays at 16 and 20 Brix would be less costly, both financially and environmentally, with less pressure for the development of insecticide resistance.

This study demonstrates that initiating weekly chemical applications at symptom development should not be considered a management strategy that will reduce the severity and incidence of sour rot, but instead, is a strategy that will maintain sour rot at the same level as when spray applications are initiated. To determine whether sour rot symptoms may soon develop, scouting weekly for *Drosophila* fruit flies, monitoring climate characteristics such as rainfall and humidity, along with individual vineyard risk factors such as training system and cultivar susceptibility, are all important elements to consider. Limiting wound sites through the use of bird netting and bird deterrents also reduces the likelihood of symptom development.

Experimental Design

Vineyard site. To evaluate the efficacy of various spray regimens beginning after sour rot symptom development, chemical spray trials were conducted in 2018 and 2019 in two commercial vineyards of *Vitis* interspecific hybrid Vignoles in central and southeastern Missouri. This cultivar is highly susceptible to sour rot due to its very compact clusters, which are susceptible to berry splitting. Mustang Maxx (zeta-cypermethrin) and OxiDate 2.0 (hydrogen dioxide) were used for sour rot chemical applications, based on previous research.⁶ Vineyard sites are located in Hermann, Missouri (Vineyard 1; 38°37'N; 91°17'W), and in Ste. Genevieve, Missouri (Vineyard 2; 37°46'N; 90°11'W). Vineyard 1 is a 3-ha block with a 15° east-facing slope of Menfro soil. The vineyard was planted in 2000 with some replanting in 2015 and is own-rooted at a vine spacing of 2.4 m and 3 m between rows positioned north to south. Vineyard 2 is a 1-ha block with a 3% southeast-facing slope of Fourche silt loam. The vineyard was planted in 2011 on 3309C rootstock at a vine spacing of 2.1 m and 3 m between rows positioned southwest to northeast. Both vineyards are high wire-trained to two cordons from single trunks.

Experiment design. Applications of Mustang Maxx at 1.75 L/ha were tank-mixed with 1.0% OxiDate 2.0, applied at 6.43 L/ha. Vineyard 1 used a CIMA model 55 Blitz sprayer (CIMA SpA) with a double fan spray head. Vineyard 2 used a CIMA model 55 Blitz Extra with a modified 2Q2Q distribution head. In 2018, the spray timings were as follows: i) 16 and 20 Brix for a total of two applications, ii) 16 Brix, 19 Brix, and 22 Brix for a total of three applications, and iii) weekly sprays beginning at 15 Brix for a total of four or five applications, depending

Table 3 Total application cost of Mustang Maxx (1.75 L/ha) and OxiDate 2.0 (6.43 L/ha) applications for 1 ha.

Number of applications	Cost in \$USD
4	593
3	445
2	297

on the vineyard site and harvest timing. In 2019, spray trials were repeated at the two vineyard sites with slight modification to spray timings to determine the effectiveness of two applications, in which one was initiated before 15 Brix. The three-application treatment at 16, 19, and 22 Brix was replaced by applications at 13 and 18 Brix, with the other two spray timings remaining the same as 2018.

In both vineyards, treatments were assigned using a split-plot design, and treatment rows differed in 2018 and 2019. In Vineyard 1, 15 rows of the block were used for the chemical application trial. Each of the three treatments was applied to entire rows and replicated in five continuous rows. Those vineyard rows not included in the trial received weekly sprays after berries surpassed 15 Brix. In Vineyard 2, treatments were applied to a total of eight rows using two replicated entire rows for each application timing. The first and last row were used as buffer rows and received weekly spray applications after berries measured 15 Brix.

Grape maturity was determined by vineyard managers who took TSS measurements at the beginning of each week using 200 randomly selected berries from a minimum of 20 individual sampling locations that fairly represented average exposure conditions within the vineyard block. When grapes matured to predetermined TSS levels, spray applications were conducted in entire corresponding treatment rows on that day or the following day.

Disease rating. For Vineyard 1 in 2018, all five rows in each treatment were used in analysis. Data were collected on the middle three rows in each of the spray treatments in 2019, minimizing the potential influence of spray drift. Every cluster on five randomly selected vines was rated for severity and incidence in each of the three replicated treatment rows for a total of 15 vines per treatment. For Vineyard 2, sour rot disease severity and incidence ratings were taken on every cluster on five randomly selected vines per row for a total of 10 vines per treatment. Vines from which data were collected differed between 2018 and 2019 for both vineyards. To determine effectiveness of spray timings, sour rot was rated at harvest on the basis of severity and incidence. For each randomly selected data vine, the total number of clusters was counted, as well as the number of sour rot-affected clusters (incidence is the percent diseased clusters). Severity was assessed visually for each cluster affected by sour rot and recorded as a percentage of affected berries per cluster.

Weather data. Precipitation and average air temperatures for Vineyard 1 were retrieved from the Mis-

souri Historical Agricultural Weather Database (agebb.missouri.edu) using the Williamsburg weather station (Callaway County) located ~67 km southeast of the vineyard site. Precipitation data were recorded on site for Vineyard 2 and average air temperatures were retrieved using the Delta weather station (Cape Girardeau County) located ~120 km southeast of the vineyard site.

Statistics. The data were analyzed by one-way analysis of variance for both incidence and severity to compare group means in JMP Student Edition (Version 14, SAS Institute, Inc.). A significant treatment effect was found in 2018 ($p = 0.0158$) and mean separation was performed by Tukey-Kramer honest significant difference. To determine main effects, a mixed model with severity and incidence as response variables was created using full factorial for vineyard and treatment with the interaction between vineyard \times treatment.

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