

GRAPE (*Vitis vinifera* ‘Pinot noir’, ‘Pinot gris’)
Powdery Mildew; *Erysiphe necator*

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Efficacy of tank mixes of biological fungicides and micronized sulfur with phenological timing while using an Intelligent and airblast sprayer for management of grape powdery mildew, 2024.

Micronized sulfur and biological fungicides were applied during critical periods for powdery mildew management on Pinot noir and Pinot gris vines at the Botany and Plant Pathology Field Laboratory in Corvallis, Oregon. In the past, use of biological products alone all season long resulted in poor powdery mildew control. In 2024 trials, treatments were focused on evaluating biological fungicides tank mixed with micronized sulfur until 4 weeks after bloom when sulfur is typically not used in commercial vineyards (Figure 1). Sulfur is an effective material for management of powdery mildew alone but was used at below label-rate quantities to reduce GPM disease pressure (Figure 1). Tank mixing with below label-rate sulfur was also evaluated in previous years, and was found to be a more realistic way to evaluate the activity of biological fungicides. Additionally, each of the biological fungicide treatments were applied using the Intelligent Spray System (ISS) to compare the efficacy of using the system with the same program.

The sprayer used (50 gallon Pak-blast, Rears Mfg., Coburg, OR) was a standard air blast sprayer retrofitted with a Lidar laser sensor, Doppler speed sensor, embedded computer, and individual pulse width modulation (PWM) solenoid valves at each sprayer nozzle. These components make up the ISS that adjusts pesticide application volume in real time to match plant canopy characteristics. In doing so, the sprayer can minimize pesticide use and off target drift while keeping pest management similar to standard sprayers. A spray console wired to the system allowed use of either the ISS components or standard constant-rate operation mode. When the ISS was used it was referred to as “intelligent mode,” and when the system was off and standard operation occurred it was referred to as “standard mode.” The sprayer was operated using a Kubota M5N-111 tractor and the nozzles in the sprayer were TeeJet ceramic D3 discs and DC25 cores.

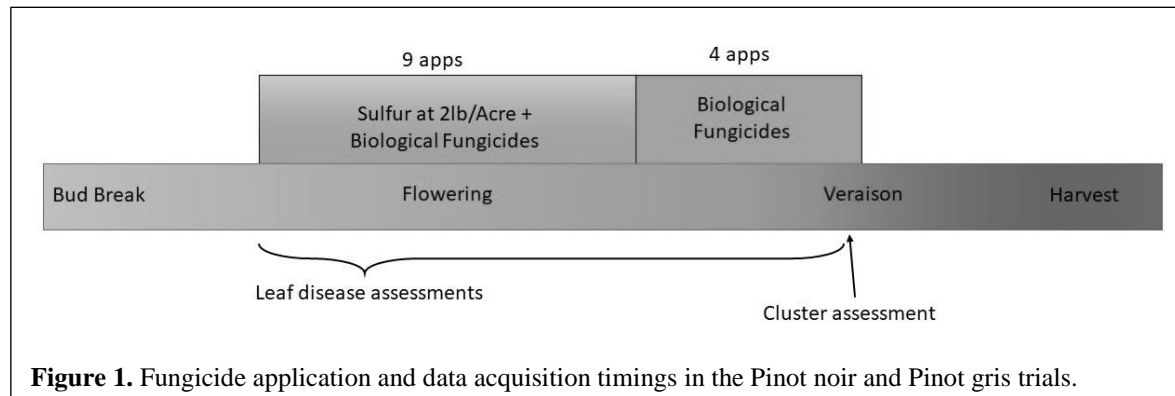


Figure 1. Fungicide application and data acquisition timings in the Pinot noir and Pinot gris trials.

Treatments (Table 1) were arranged in a randomized complete block design in each cultivar. For both the Pinot noir and Pinot gris trials, fungicide programs were initiated on 14 May just after powdery mildew was detected through intensive scouting. Microthiol Disperss was tank mixed with biological fungicides at 2 lb/Acre for each application until approximately 4 weeks post bloom. After which, all succeeding applications were with biological fungicides alone until veraison. The sulfur alone treatment was not continued beyond 4 weeks post flowering.

The fungicide schedule used in the study was based on the different stages of a grape powdery mildew (GPM) epidemic. Initially, before grapevines flower, fungicide programs are focused on preventing GPM infections primarily on leaves. During flowering and for approximately 3 weeks thereafter, fungicide programs are focused on

preventing infection of the grape clusters. Flowering and fruit set are the most critical stages for management of GPM, as this is when the inflorescences and subsequently the developing berries, are most susceptible to infection. After three weeks post flowering fungicide programs focus on keeping GPM levels low on both leaves and fruit to keep overall inoculum to a minimum. Fungicide program intensity is usually not as aggressive after bloom and early fruit set as the earlier two sections of the season as both leaves and fruit develop ontogenic resistance to infection as they age. Additionally, many viticulturists cease sulfur applications two to four weeks before véraison, so as to avoid residual sulfur on grapes that could contribute to wine off flavors. The trial regime allowed testing of whether using biological products would augment the control of a below-label rate of sulfur, while not overwhelming the biological fungicide products with the high disease pressure of the research vineyard.

The blocks used consisted of ‘Pinot noir’ or ‘Pinot gris’ planted in 1998 on *V. rupestris* x *V. riparia* 101-14 rootstock with 7x8 ft spacing. A single buffer rootstock vine was trained between each set of treatment vines and a buffer row of rootstock vines separated each varietal row, which helped minimize plot-plot interference. Vines were trained to a Guyot (vertical shoot position) system and pruned from 10 January to 27 Mar. Shoot thinning by hand occurred from 9 to 25 May and sucker removal by hand was continuous throughout the season. Shoots were cut above the top wire on 15 Jun and maintained at this height throughout the growing season. Fungicide treatments were applied every 7-10 days. Each treatment was replicated on 4 sets of 5 vines.

Rainfall during the dormant season (Oct 2022 to March 2023) was 4.1 inches above normal while spring weather conditions were close to long term norms, however, precipitation dropped off rapidly at the end of April. Symptoms of powdery mildew were first found on 9 May as a flag shoot, with additional infections found on 13 May. Bloom took place from approximately 7 to 16 Jun with caps rapidly detaching due to warm, dry weather.

Leaf and cluster data were taken on the middle three vines of each experimental plot by randomly examining either 25 clusters or leaves on both the east and west side of the row for a total of 50 units examined per plot. The incidence of powdery mildew on leaves was recorded weekly from 10 Jun through 5 Aug. The severity of powdery mildew on clusters was taken on 7 Aug. Leaf incidence data was analyzed by calculating the area under disease progress curve (AUDPC) which was calculated by multiplying the mean incidence from two observation dates by the number of days between observations ($\sum [Y_{i+1} + Y_i]/2][X_{i+1}-X_i]$ where Y_i is incidence of mildew at i th observation and X_i is the day of the i th observations) and adding together the values. AUDPCs were calculated using the agricolae package and modeled with linear models. Cluster severity percentages were modeled using a generalized linear mixed model with block fitted as a random effect. Cluster severity treatment contrasts were conducted using the emmeans package and model fit was checked with the DHARMA package. Uncertainty was estimated using asymptotic 95% confidence intervals. The Tukey Honest Significant Difference (HSD) and Bonferroni P value adjustments were used. All data was analyzed in R version 4.0.3.

Table 1. Biological fungicide treatments applied to Pinot noir or Pinot gris vines.

Pinot noir		Pinot gris	
Treatment ^x	Sprayer Mode ^y	Treatment ^x	Sprayer Mode ^y
Non-treated	N/A	Non-treated	N/A
2 lb/A Sulfur	Standard	2 lb/A Sulfur	Standard
Theia 3 lb/A ^z + 2lb/A Sulfur (then Theia alone)	Standard Intelligent high Intelligent low	Serenade ASO 4 Qt/A ^z + 2lb/A Sulfur (then Serenade ASO alone)	Standard Intelligent high Intelligent low

^xTreatments applied at 80psi at approximately 430 PTO rpm and 3 mph.

^yIntelligent high and low treatments applied at 0.12 fl oz/ft³ and 0.06 fl oz/ft³ of grape canopy, respectively.

^zTreatment mixed at the rate indicated if the sprayer was used in standard mode. Actual rates applied by the ISS were lower due to the lower volume applied in intelligent mode.

Table 2. Area under disease progress curve (AUDPC, leaf disease) and percent infected berries on Pinot noir in 2024.

Treatment ^{wx}	AUDPC ^{yz}	Percent Infected Berries ^{yz}
Non-treated	2757 (2605-2909) A	97.7 (96.6-98.4) A
2 lb/A Sulfur	1880 (1728-2032) BC	27.7 (20.8-35.9) C
Theia 3 lb/A + 2lb/A Sulfur	1341 (1189-1493) D	14.2 (10.2-19.5) D
Theia 3 lb/A + 2lb/A Sulfur Intelligent High	1686 (1534-1838) C	20.3 (14.8-27.1) CD
Theia 3 lb/A + 2lb/A Sulfur Intelligent Low	2082 (1930-2233) B	48.5 (39.2-57.9) B

^wAll sulfur treatments were discontinued after 10 July, approximately 4 weeks post flowering.

^xAll treatments were applied at 80psi at approximately 430rpm PTO; intelligent high and low sprayer treatments applied at 0.12 fl oz/ft³ and 0.06 fl oz/ft³ of grape canopy, respectively.

^yEstimates are followed by asymptotic 95% confidence intervals in parentheses. Treatments followed by different letters are significantly different from each other, marginal means contrast ($p < 0.05$) with p values adjusted using Tukey (HSD) method.

^zEstimates are followed by asymptotic 95% confidence intervals in parentheses. Treatments followed by different letters are significantly different from each other, marginal means contrast ($p < 0.05$) with p values adjusted using Bonferroni method.

In the Pinot noir and Pinot gris trials the non-treated controls had significantly higher AUDPC values and percent infected berries than all fungicide treated plots (Table 2, 3). In the Pinot noir trial, the Theia intelligent low treatment resulted in a significantly higher AUDPC than all other fungicide treatments, while the Theia standard treatment resulted in a lower AUDPC value than the sulfur control (Table 2). In the Pinot gris trial, all fungicide treatments resulted in AUDPC values that were not significantly different from each other when using HSD (Table 3). However, when using the Bonferroni adjustment, the Serenade ASO standard mode treatment resulted in a significantly lower AUDPC value than the sulfur control and intelligent mode treatments.

In the Pinot noir trial, the Theia intelligent low treatment resulted in significantly higher cluster severity than the sulfur control (Table 2). The Theia standard treatment resulted in significantly lower cluster severity than the sulfur control. In the Pinot gris trial, the Serenade ASO intelligent low treatment resulted in significantly higher cluster severity than the sulfur control (Table 3). However, the sulfur control resulted in cluster severity that was not significantly different from both the Serenade ASO intelligent high and Serenade ASO standard mode when using HSD (Table 3). When the Bonferroni correction was used, the Serenade ASO standard mode treatment resulted in significantly lower cluster severity than the sulfur control and Serenade ASO intelligent high treatment.

Table 3. Area under disease progress curve (AUDPC, leaf disease) and percent infected berries on Pinot gris in 2024.

Treatment ^{wx}	AUDPC		Percent Infected Berries (HSD)	
	(HSD) ^y	(Bon) ^z	(HSD) ^y	(Bon) ^z
Non-treated	2746 (2468-3023) A	2746 (2355-3136) A	93.7 (90.0-96.2) A	93.7 (87.9-96.8) A
2 lb/A Sulfur	1952 (1674-2229) B	1952 (1561-2342) B	22.4 (14.4-33.0) CD	22.4 (12.5-36.8) C
Serenade ASO 4 Qt/A + 2lb/A Sulfur	1466 (1188-1743) B	1466 (1075-1857) C	14.6 (9.1-22.7) D	14.6 (7.8-25.8) D
Serenade ASO 4 Qt/A + 2 lb/A Sulfur Intelligent High	1906 (1628-2183) B	1906 (1515-2297) B	24.2 (15.8-35.4) C	24.2 (13.6-39.3) C
Serenade ASO 4 Qt/A + 2 lb/A Sulfur Intelligent Low	2007 (1730-2285) B	2007 (1616-2398) B	43.2 (30.8-56.5) B	43.2 (27.3-60.6) B

^wAll sulfur treatments were discontinued after on 10 July approximately 4 weeks post flowering.

^xAll treatments were applied at 80psi at approx. 430 rpm PTO; intelligent high and low sprayer treatments applied at 0.12 fl oz/ft³ and 0.06 fl oz/ft³ of grape canopy, respectively.

^yEstimates are followed by asymptotic 95% confidence intervals in parentheses. Treatments followed by different letters are significantly different from each other, marginal means contrast ($p < 0.05$) with p values adjusted using Tukey (HSD) method.

^zEstimates are followed by asymptotic 95% confidence intervals in parentheses. Treatments followed by different letters are significantly different from each other, marginal means contrast ($p < 0.05$) with p values adjusted using Bonferroni method.

Spray volume applied in the Pinot noir and Pinot gris trials were very similar due to using the same settings in both trials. For standard mode treatments, the volume applied remained close to 60 gallons per acre (GPA) for the entire season (Figure 2A and 2B). Intelligent mode treatments in both trials started the season at 9.3 GPA and 13 GPA for intelligent low and high, respectively, with both quickly trending up to 35 GPA and 43 GPA at the beginning of bloom, respectively. After bloom, intelligent high mode remained just under 45 GPA while intelligent low mode remained between 35 GPA and 40 GPA (Figure 2A and 2B).

In the Pinot noir trial, Theia standard mode resulted in approximately 3.4 lb/A at each application throughout the trial, while in intelligent mode, a lower than recommended rate of 0.64 lb/A to 0.94 lb/A was applied for the first application in intelligent low and high modes, respectively (Figure 3A). The intelligent low and high mode treatments reached the minimum suggested rate of 1.5 lb/A at the fourth and third application, respectively (Figure 3A). For the sulfur applied in the Pinot noir study, in standard mode the quantity was consistent for the season and ranged from a low of 2.2 lb/A to a max of 2.6 lb/A (Figure 4A). In intelligent mode, the quantity of sulfur applied ranged from 0.43 lb/A and 0.62 lb/A, to 1.4 lb/A and 1.6 lb/A for intelligent low and intelligent high, respectively.

In the Pinot gris trial, 4.3 to 5.4 Qt/A of Serenade ASO applied on a per-acre basis in standard mode. In intelligent mode the amount ranged from 0.84 Qt/A to 3.0 Qt/A and 1.3 Qt/A to 3.4 Qt/A in intelligent low and intelligent high modes, respectively (Figure 3B). The suggested product application rate range for Serenade ASO when tank mixed is from 1 to 4 Qt/A, so most applications were above the lowest rate except for the first application using intelligent low mode (Figure 3B). For the quantity of sulfur applied in the Pinot gris study, in standard mode the quantity was consistent for the season and ranged from a low of 2.2 lb/A to a max of 2.7 lb/A (Figure 4B). In intelligent mode, the quantity of sulfur applied ranged from 0.43 lb/A and 0.68 lb/A, to 1.4 lb/A and 1.7 lb/A for intelligent low and intelligent high, respectively (Figure 4B).

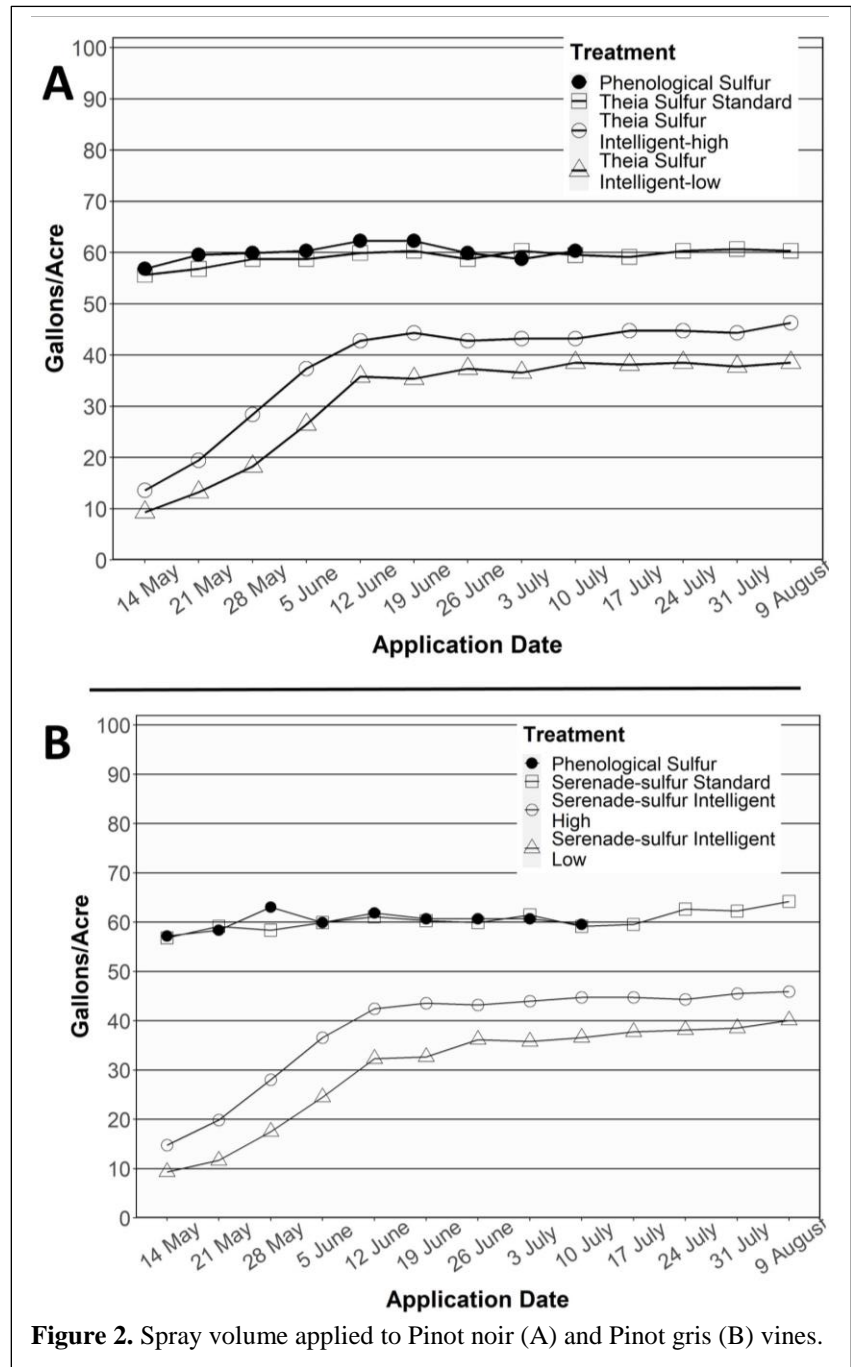


Figure 2. Spray volume applied to Pinot noir (A) and Pinot gris (B) vines.

Both Theia and Serenade ASO applied in standard mode resulted in significantly better disease control on clusters than when sulfur alone was applied. However, this was not necessarily the case when the mixture was applied in

other modes. In standard mode, the highest volume of fungicide mixture was applied (Figures 2A, 2B), which should have resulted in the largest amount of fungicide deposited on the vines compared to the treatments applied in intelligent mode. Higher spray volume typically also means higher coverage, as the larger amount of spray droplets can physically cover a larger area. In previous experiments with contact action fungicides such as Microthiol, we have observed that there is a threshold amount of the product applied per area that is needed for efficacy, even if coverage is not significantly different than that of other application settings.

In plots treated with Theia, the amount of product applied did not reach the lowest recommended rate on the label until the 3rd or 4th application for plots treated with Intelligent high, or low, respectively (Figure 3A). For plots treated with Serenade ASO, the lowest label rate for standalone treatments is 2 Qt/A, while the lowest label rate for tank mix treatments is 1 Qt/A. The standalone rate is likely more relevant as the sulfur applied was lower than that required for efficacy. Plots treated in intelligent high or low did not reach those thresholds until the third application or 5th application for the standalone rate, while for the tank mix rate the intelligent low treatment reached the 1 Qt/A threshold at the second application (Figure 3B). Theia and Serenade ASO may be similar to Microthiol in that there needs to be a threshold amount applied per area for efficacy.

The AUDPC and cluster severity data between both the Pinot noir and Pinot gris trials were remarkably similar, while the biological fungicides used themselves are different in formulation. They both have similar modes of action, each containing the same species of bacteria with the exact same CFU count. However, it is known that the live organisms in Serenade do not need to be viable to get efficacy in the field, while the mode of action of Theia is likely benefitted by active organisms on the leaf surface. Theia is a dry flowable (powder) that contains 100% of the active ingredient organism, whereas Serenade ASO is a liquid formulation that contains 1.34% of its active ingredient

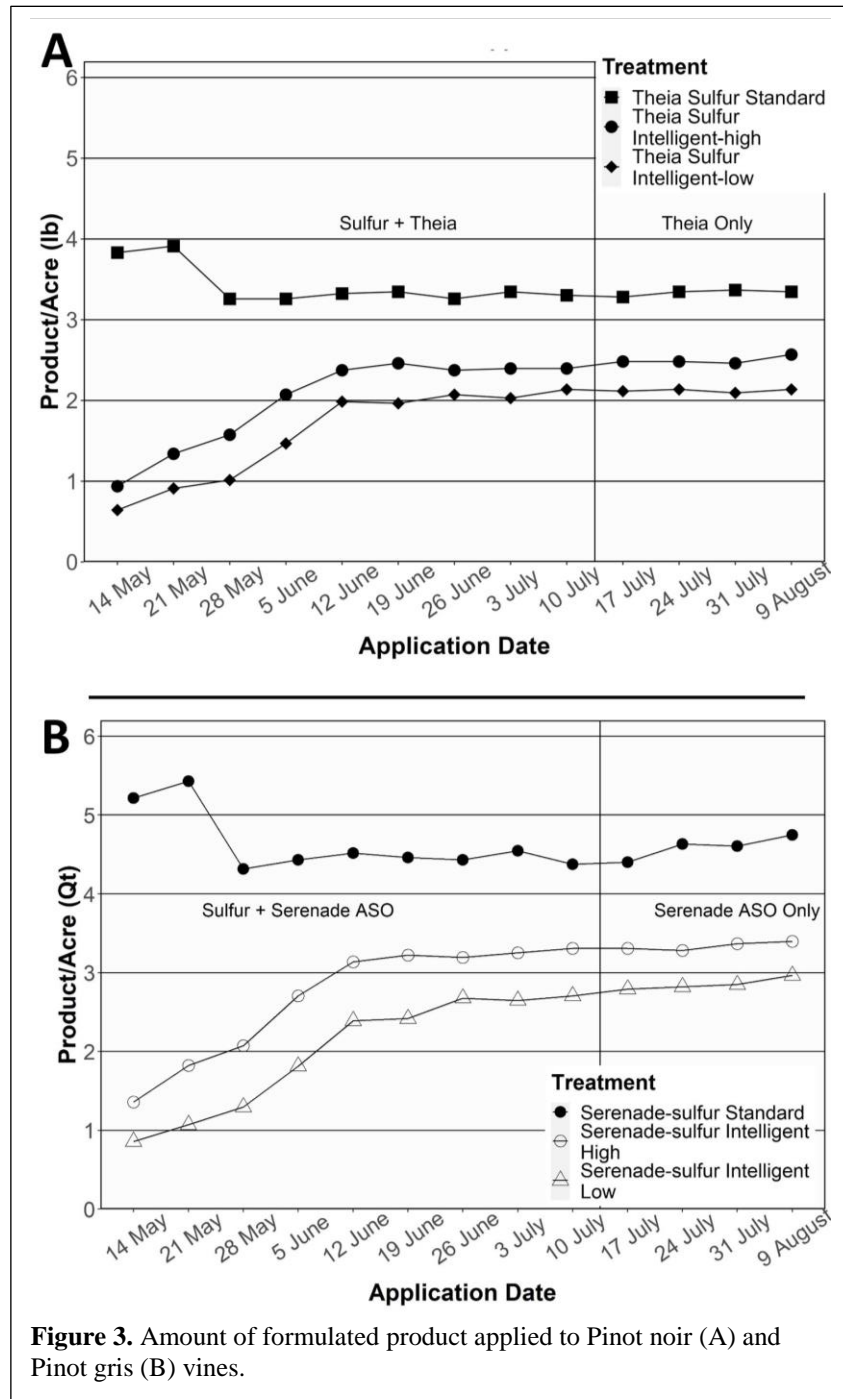


Figure 3. Amount of formulated product applied to Pinot noir (A) and Pinot gris (B) vines.

organism. These differing formulations may be due to the delivery strategy of the active ingredient, but in the end, led to similar efficacy in the trials.

Several experiments were conducted in the lab to determine the effect of mixing Microthiol Disperss with Theia and Serenade ASO on the Colony Forming Units (CFU) of each product on plates of nutrient agar. At rates of 5 lb/A and 7 lb/A Microthiol was observed to increase the number of CFU counts on plates relative to when each product was mixed with water alone. When Microthiol was mixed at 3 lb/A there was no significant effect on CFU count, however 2 lb/A was not tested. Additional testing on Microthiol mixes in the lab and field may more fully elucidate the impact of mixing Theia and Serenade ASO with Microthiol. Additional testing of biological fungicide product viability during and after application with airblast sprayers may more fully elucidate any effect of application practices on the viability of bacteria in the tank mixes.

