Project Title: Fire Blight Product Testing for Effective Recommendations
Report Type: Final Project Report

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Item	(Type year of project start date here)	(Type year start date of year 2 here if relevant)	(Type year start date of year 3 here if relevant)
Salaries	$7,800.00	$8,112.00	
Benefits	$2,955.00	$3,074.00	
Wages			
Benefits			
RCA Room Rental			
Shipping			
Supplies	$500.00	$500.00	
Travel			
Plot Fees	$3,000.00	$3,000.00	
Miscellaneous			
Total	$14,255.00	$14,686.00	$0.00

Footnotes:

Salaries for a scientific assistant 2 month/ yr.

Benefits at 38% for scientific assistant.

Fire Blight Product Testing for Effective Recommendations

OBJECTIVES
1. Test new fire blight prevention products.
2. Provide research-based information to growers and consultants.

SIGNIFICANT FINDINGS
- Alum (potassium aluminum sulfate) provided good control similar to antibiotic checks as well as biological Blossom Protect (*A. pullulans*) and several copper products (Previsto, Mastercop, Instill).
- Several essential oil, and peracetic acid-peroxide products (Oxidate 5.0, Jet Ag, Thyme Guard, Thymox, Cinnerate) provided moderate disease suppression similar to some other biological and copper products (Serenade Opti, Cueva) and may be best incorporated as rotational products as part of an integrated program during lower risk periods.
- In order to minimize the risk of fruit marking managers should consider drying times for essential oils and peracetic acid-peroxide products as well as soluble coppers.

RESULTS and DISCUSSION

Alum
Potassium aluminum sulfate compounds are aseptic, astringent compounds known to inhibit the growth of bacteria, fungi and oomycetes potentially due to pH, cellular ionic imbalance or disruption of membranes (Kolaei et al. 2013; Mecteau et al. 2002).

Alum (Potassium aluminum sulfate) has been tested for six years in Washington. This compound is experimental (non-labeled). It has had generally consistent positive results with an average of 72% control relative to the untreated check in 2016 to 2022 trials when the product was applied at an 8 to 10 lb per 100 gal rate. This control is comparable to 74% in the oxytetracycline standard and 85% in the streptomycin standard (2013 to 2022 median). Marking from chemical russet was negligible in all WA trials (< 1 on a 0 to 15 scale). In 2022 relative control from alum was 88%, not significantly different than the streptomycin standard and significantly better than the water treated check (Table 1). In 2021 relative control was approximately 50%, but still significantly different from the water treated check and comparable to the relative control obtained using oxytetracycline check (56% relative control) and streptomycin check (58% relative control) (Table 2). However, in 2020 relative control from alum was 28% compared to the water treated check (Table 3).

Suppression of fire blight by alum was similar to trials in Germany where potassium-aluminum sulfate averaged 72% efficiency in eight trials (Kunz and Donat 2013). Alum also had high relative disease suppression in recent Oregon trials averaging 74% (*n*=8) (Johnson et al. 2022).

Table 1. Effect of mineral based biopesticides to pear, cv. Anjou on infection of *E. amylovora* in pear blossoms in Wenatchee, WA in 2022 [u]

Treatment	Rate per 100 gallons water	Application timings [z]	Infections per 100 clusters [y]				Fruit russet [v]
Streptomycin standard (Firewall 50WP) [x]	8 oz	3	4.4	±	1.2	c [w]	0.2
Oxytetracycline standard (Fireline 45WP) [x]	9 oz	3,6	15.7	±	4.8	b	0.2
Alum [t]	8 lb	3,4,6	3.9	±	1.4	c	0.5
Alum [t]	8 lb	3,4,6,8,9,10	4.1	±	0.4	c	1.8
Blossom Protect + Buffer Protect Alum [t]	1.25 lb + 5 lb 8 lb	1,2 3,6	6.8	±	1.6	bc	1.5

Blossom Protect + Buffer Protect	1.25 lb + 5 lb	1,3	15.5	±	4.4	b		0.3
Water treated check	NA	3,4,6	35.5	±	5.4	a		0.3

[z] Timings 1: 70% bloom, 2: 90% bloom, 3: morning before evening inoculation (full bloom), 4: morning after inoculation, 5: 2 days after inoculation, 6: 3 days after inoculation (petal fall), 7: 4 days after inoculation, 8: 6 days after inoculation, 9: 2 weeks after inoculation, 10: 3 weeks after inoculation

[u] Inoculation was conducted on the evening of 22 Apr 2021 at full bloom (of king blooms) using a suspension of freeze-dried cells of *Erwinia amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 17x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[x] Amended with Regulaid: 16 fl. oz. per 100 gallons. Buffered to 5.6 pH.

[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).

[t] Amended with Regulaid: 16 fl. oz. per 100 gallons. pH verified at 4.0.

[v] Fruit marking is rated from an average of 25 fruit per tree. In 2022 less than 25 fruit were often present. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades. No statistical differences were observed between treatments.

Table 2. Effect of mineral based biopesticides on *E. amylovora* infection of apple blossoms cv. Red Delicious in Wenatchee, WA, in 2021[z]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters[y]				Fruit russet[t]
Streptomycin (Firewall 17)[x]	8 oz	100% bloom	16.1	±	2.3	ab[w]	0.06
Oxytetracycline (Fireline 17)[x]	16 oz	100% bloom, petal fall	17.0	±	5.7	a	0.00
Organic standard apple Blossom Protect + Buffer Protect Previsto	1.24 lb+ 8.75 lb 3 qt	70% bloom, 100% bloom, 100% bloom + 1 day, petal fall	17.8	±	4.5	ab	0.69
Organic standard pear Blossom Protect + Buffer Protect Serenade Opti[u]	1.24 lb + 8.75 lb 20 oz	70% bloom, 100% bloom, 100% bloom + 1 day, petal fall	14.0	±	2.6	a	0.73
Alum[v]	8 lb	100% bloom, 100 bloom + 1 day, petal fall	19.3	±	2.4	ab	0.19
TDA-NC-1[u]	571 g	pink, 50% bloom, 100% bloom, petal fall	26.7	±	3.9	bc	0.05
Water-treated check	NA	100% bloom, petal fall, petal fall + 3 days	38.6	±	5.1	c	0.00

[z] Application dates were:18 Apr (70% bloom), 19 Apr (full bloom), 20 Apr (full bloom + 1 day), 23 Apr (petal fall), 26 April (petal fall + 3 days). Inoculation was conducted on the evening of 19 Apr 2021 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells and 50% live cells of *E. amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 40-94 x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[x] Amended with Regulaid: 16 fl. oz. per 100 gallons. Buffered to 5.6 pH.

[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).

[v] Amended with Regulaid: 16 fl. oz. per 100 gallons.

[u] Amended with Swilet spreader sticker 23 fl. oz per 100 gallons.

[t] Fruit marking, average of 25 fruit per tree. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades.

Table 3. Effect of Mineral Product Treatments on *E. amylovora* infection of apple blossoms in Wenatchee, WA, in 2020[‡]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters				Fruit russet[t]
Streptomycin (Firewall 17)[yz]	28.8 oz	50% bloom, 100% bloom, petal fall	2.8	±	1.2	a	0
Oxytetracycline (Fireline 17)[yz]	28.8 oz	50% bloom, 100% bloom, petal fall	8.2	±	2	b	0
Organic Standard Blossom Protect + Buffer Protect + Soluble Copper (Previsto)	1.24 lb + 8.75 lb 3 qt	50% bloom, 80% bloom, 100% bloom, petal fall	9.5	±	1.3	bc	0.02
Alum[y]	8 lb	100% bloom, petal fall	22	±	4.2	d	0.02
TDA-NC-1[x]	17.1 g	Tight cluster, 50% bloom, 100% bloom + 1 day, petal fall	13	±	2.3	bc	0
Water-treated check	NA	100% bloom, +1 day, petal fall	31	±	7.1	d	0

[‡]Application dates were: April 14 (20% bloom), April 16 (50% bloom), April 17 (80% bloom) and April 18 (full bloom), April 19 (full bloom plus 1 day), April 22 (petal fall). Inoculation was conducted on the evening of April 18, 2020 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells of *E. amylovora* strain 153N (streptomycin and oxytetracycline sensitive pathogen strain) and 50% live cells, which was prepared at 24 x 10^6 CFU per ml.
[y] Amended with Regulaid: 30 fl. oz. per 100 gallons.
[Z]Buffered to 5.6 pH.
[x] Amended with Silwet oil at 0.0125%. Copper sulfate product.
[t]Fruit marking, average of 25 fruit per tree. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades.

Oxidizers

Several new peroxide products with higher levels of peracetic acid have recently been released (e.g. Jet Ag, Oxidate 5.0). Peracetic acid denatures proteins, disrupts cell wall permeability, and oxidizes sulfhydryl and sulfur bonds in proteins, enzymes, and other metabolites. Peracetic acid and peroxide oxidizers generally have little residual activity.

Oxidizing agents (Jet Ag and Oxidate 5.0) produced median relative disease suppression of 53% and 62% with 2 to 3 applications post inoculation (4 trials: 2019-2022). In 2022, with applications the day after inoculation, petal fall and 6 days after inoculation (petal fall plus 3 days), oxidizers provided 53 and 62% relative control, but when applied at full bloom, day after inoculation and petal fall the relative control was 42.4% (Table 4). In 2021 control relative to the water treated check for peroxide + peracetic acid treatments was 63-67% with three applications (100% bloom + 1 day, petal fall and petal fall + 3 days), not significantly different than the organic standard (Table 5). In 2020 with two applications relative control for peroxide + peracetic acid treatments was not significantly different than the water treated check (Table 6). At these application timings no significant fruit marking was observed (less than 1 on a 0 to 15 scale). In comparison 2013 to 2022 Washington long term averages are 85% relative control for the streptomycin standard (N=35), 72% relative control Blossom Protect + Buffer Protect (N=25), 74% relative control oxytetracycline standard and 73% relative control Previsto. Enumeration of bacterial populations in the flower suggest that the 3-day post petal fall application in 2021 and 2022 was important to keep populations lower compared to in 2020 when 1 week post petal fall *Erwinia* numbers reached high levels in peroxide + peracetic acid treated trees (Fig 1-3).

In a previous study, peroxide + peracetic acid products were applied after antibiotics during the post petal fall period (Fireline at: 50% bloom, 100% bloom, PF peroxide/peracetic acid product at: 5, 7, 10, 14 days after full bloom). Multiple post petal fall applications resulted in significant fruit marking which would have resulted in culled fruit (average 8.2 on 0 to 15 scale). In order to limit fruit marking potential peroxide + peracetic acid products should be applied only in fast drying conditions and up until the early post-petal fall period.

Table 4. Effect of hydrogen peroxide, peracetic acid treatments applied to pear, cv. Anjou on infection from *E. amylovora* in pear blossoms in Wenatchee, WA in 2022[u]

Treatment	Rate per 100 gallons water	Application timings [z]	Infections per 100 clusters[y]			Fruit russet [v]
Streptomycin standard (Firewall 50WP) [x]	8 oz	3	4.4	± 1.2	c [w]	0.2
Oxytetracycline standard (Fireline 45WP) [x]	9 oz	3,6	15.7	± 4.8	b	0.2
Organic standard apple Blossom Protect + Buffer Protect, Previsto	1.25 lb + 5 lb 3 qt	1,2 3,6	11.1	± 4.0	bc	1.1
Organic standard pear Blossom Protect + Buffer Protect Serenade Aso	1.25 lb+ 5 lb 96 fl oz	1,3 4,6	16.9	± 2.6	ab	0.6
Blossom Protect + Buffer Protect	1.25 lb + 5 lb	1,3	15.5	± 4.4	b	0.3
hydrogen peroxide (26.5%), peracetic acid (4.9%) (Jet Ag)	128 fl oz	4,6,8	13.5	± 3.3	b	0.4
hydrogen peroxide (27%), peracetic acid (5%) (Oxidate 5.0)	128 fl oz	4,6,8	16.7	± 3.0	ab	0.6

hydrogen peroxide (27%), peracetic acid (5%) Oxidate 5.0	128 fl oz	3,4,6	20.4	±	5.7	ab	0.8
Blossom Protect + Buffer Protect hydrogen peroxide (26.5%), peracetic acid (4.9%) Jet Ag Stargus	1.25 lb + 5 lb 128 fl oz 2 qt	1,3 5 7	16.1	±	2.8	ab	1.3
Blossom Protect + Buffer Protect hydrogen peroxide (27%), peracetic acid (5%) (Oxidate 5.0)	1.25 lb + 5 lb 128 fl oz	1,3 5,7	17.2	±	2.2	ab	0.8
Water treated check	NA	3,4,6	35.5	±	5.4	a	0.3

[z] Timings 1: 70% bloom, 2: 90% bloom, 3: morning before evening inoculation (full bloom), 4: morning after inoculation, 5: 2 days after inoculation, 6: 3 days after inoculation (petal fall), 7: 4 days after inoculation, 8: 6 days after inoculation, 9: 2 weeks after inoculation, 10: 3 weeks after inoculation.

[u] Inoculation was conducted on the evening of 22 Apr 2021 at full bloom (of king blooms) using a suspension of freeze-dried cells of *Erwinia amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 17x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[x] Amended with Regulaid: 16 fl. Oz. per 100 gallons. Buffered to 5.6 pH.

[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).

[v] Fruit marking is rated from an average of 25 fruit per tree. In 2022 less than 25 fruit were often present. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades. No statistical differences were observed between treatments.

Table 5. Effect of hydrogen peroxide, peracetic acid treatments applied to apple, cv. Red Delicious on infection from *E. amylovora* in apple blossoms in Wenatchee, WA, in 2021[z]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters[y]				Fruit russet[v]
Streptomycin (Firewall 17) [x]	8 oz	100% bloom	16.1	±	2.3	a[w]	0.06
Oxytetracycline (Fireline 17) [x]	16 oz	100% bloom, petal fall	17.0	±	5.7	a	0.00
Organic standard apple Blossom Protect + Buffer Protect Previsto	1.24 lb + 8.75 lb 3 qt	70% bloom, 100% bloom, 100% bloom + 1 d, petal fall	17.8	±	4.5	a	0.69
Organic standard pear Blossom Protect + Buffer Protect Serenade Opti	1.24 lb + 8.75 lb 20 oz	70% bloom, 100% bloom, 100% bloom + 1 d, petal fall	13.9	±	2.6	a	0.73
hydrogen peroxide (26.5%), peracetic acid (4.9%) (Jet Ag)	128 oz	100% bloom + 1 day, petal fall, petal fall + 3 days	12.8	±	1.6	a	0.75
hydrogen peroxide (27%), peracetic acid (5%) (Oxidate 5.0)	128 oz	100% bloom + 1 day, petal fall, petal fall + 3 days	14.2	±	1.2	a	0.51
Blossom Protect + Buffer Protect hydrogen peroxide (26.5%), peracetic acid (4.9%) (Jet Ag) *Bacillus amyloliquefaciens* (Stargus)	1.24 lb + 8.75 lb 128 oz 2 qt	70% bloom, 100% bloom petal fall petal fall + 3 days	11.4	±	0.7	a	0.99
Water-treated check	NA	100% bloom, petal fall, petal fall + 3 days	38.6	±	5.1	b	0.00

[z] Application dates were: 18 Apr (70% bloom), 19 Apr (full bloom), 20 Apr (full bloom + 1 day), 23 Apr (petal fall), 26 April (petal fall + 3 days). Inoculation was conducted on the evening of 19 Apr 2021 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells and 50% live cells of *E. amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 40-94 x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[x] Amended with Regulaid: 16 fl. Oz. per 100 gallons. Buffered to 5.6 pH.

[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).

[v] Fruit marking, average of 25 fruit per tree. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades.

Table 6. Effect of hydrogen peroxide and peracetic acid treatments applied to Red delicious apple trees on infection from *E. amylovora* in apple blossoms in Orondo, WA, in 2020[‡]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters [u]				Fruit russet[t]
Streptomycin standard (Firewall 17)[zy]	28.8 oz	50% bloom, 100% bloom, petal fall	2.8	±	1.2	a	0
Oxytetracycline standard (Fireline 17) [zy]	28.8 oz	50% bloom, 100% bloom, petal fall	8.2	±	2	b	0

Organic standard (Blossom Protect/ Buffer Protect + Previsto)	1.24 lb 8.75 lb 3 qt	50% bloom, 80% bloom, 100% bloom, petal fall	9.5	±	1.3	b	0.02
hydrogen peroxide (26.5%), peracetic acid (4.9%) (Jet Ag)	128 fl oz	Day after inoc and 3 days after inoc[v]	27.8	±	3.9	c	0
hydrogen peroxide (27%), peracetic acid (5%) (Oxidate 5.0)	128 fl oz	Day after inoc and 3 days after inoc	24.1	±	3.8	c	0.02
hydrogen peroxide (27%), peracetic acid (5%) (Oxidate 5.0)	50 fl oz	Day after inoc and 3 days after inoc	28	±	4.1	c	0.07
Untreated water check	----	100% bloom, +1 day, petal fall	30.7	±	7.1	c	0

[‡]Application dates were: April 15, pink, April 19 (20% bloom), April 21 (50% bloom), April 23 (full bloom), April 24 (full bloom plus 1 day), April 28 (petal fall). Inoculation was conducted on the evening of April 23, 2020 at full bloom (of king blooms) using a suspension of freeze-dried cells of *E. amylovora* strain 153N (streptomycin and oxytetracycline sensitive pathogen strain), which was prepared at 1.3 x10^6 CFU per ml.

[y] Amended with Regulaid: 30 fl. Oz. per 100 gallons.

[z] Buffered to 5.6 pH.

[u] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[v] Note inoculation was done at dusk. Day after spray is done early morning next day. 3 days after inoculation coincided with petal fall sprays.

[t] Fruit marking, average of 25 fruit per tree. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades.

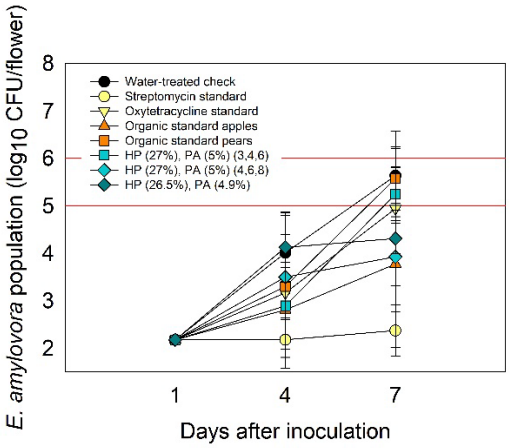

Figure 1. Effect of hydrogen peroxide (HP), peracetic acid (PA) treatments applied to pear cv. Anjou trees to suppress fire blight on the population size of *E. amylovora* strain 153N on flowers 1, 4 and 7 days post-inoculation of the pathogen in Wenatchee, WA, in 2022.

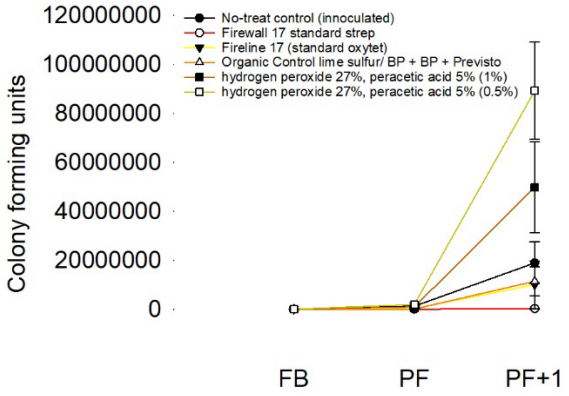

Figure 2. Effect of treatments applied to Red delicious apple trees to suppress fire blight on the

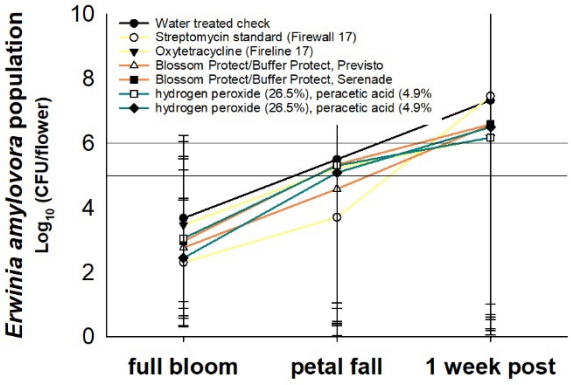

Figure 3. Effect of hydrogen peroxide and peracetic acid treatments applied to Red delicious

population size of *E. amylovora* strain 153N on flowers at Full Bloom (FB), Petal Fall (PF) and Petal Fall + 1 week (PF+1) in WA in 2020.

apple trees to suppress fire blight on the population size of *E. amylovora* strain 153N on flowers at full bloom, petal fall and 1 week post petal fall in Wenatchee, WA, in 2021.

Essential Oils

Essential oils (e.g. from thyme, mint, cinnamon, oregano) have known antimicrobial activity. In one laboratory study, active compounds from *Origanum compactum* (oregano family) and *Thymus vulgaris (Thyme)* were most effective (Kokoskova *et al.*, 2011). In another study, Apium graveolens (celery seed) and Curcuma longa (turmeric) essential oils showed a reduction in *E. amylovora* virulence (Akhlaghi *et al.* 2017). These oils are rich in antioxidative phenolic compounds, which are believed to be responsible for their antimicrobial activity (Chizzola *et al.*, 2008). Several essential oil products are available commercially, which may be of interest including Thyme Guard, Thymox, and Cinnerate.

Essential oil plant extracts from thyme and cinnamon (Thyme Guard, Thymox, Cinnerate) resulted in median relative disease suppression of 49% (thyme oils 3 to 6 applications) and 45% (cinnamon oils 3 to 4 applications) between 2019 and 2022. Thyme oils (Thyme Guard and Thymox) had infection incidence significantly lower than water treated controls in 2021 and 2022 but not in 2019 and 2020 (Tables 7-10). Cinnamon oil compounds (Cinnerate) significantly reduced infection incidence compared to water treated controls in 2021 with four applications, but not in 2020 with three applications. Essential oil products with 3 to 4 applications resulted in low fruit marking but with 7 applications in 2019 the thyme oil product resulted in significant fruit marking (average of 4 on a 1 to 15 scale). In 2021 and 2022 the alternative organic program Blossom Protect + Buffer Protect at 50% and 100% bloom followed by Previsto at 100% bloom + 1 day and by thyme oil product at petal fall was not significantly different than organic apple and pear standard programs where Blossom Protect + Buffer Protect were followed by Previsto or Serenade Opti at 100% bloom and petal fall. Enumeration of bacterial populations in the flower showed no significant reduction of E. amylovora after the application of essential oils in any of the years (Fig. 4 - 8).

Table 7. Effect of essential oil/plant extract treatments applied to pear, cv. Anjou on infection of *E. amylovora* in pear blossoms in Wenatchee, WA in 2022 [u]

Treatment	Rate per 100 gallons water	Application timings [z]	Infections per 100 clusters [y]				Fruit russet [v]
Streptomycin standard (Firewall 50WP) [x]	8 oz	3	4.4	±	1.2	c [w]	0.2
Oxytetracycline standard (Fireline 45WP) [x]	9 oz	3,6	15.7	±	4.8	ab	0.2
Organic standard apple Blossom Protect + Buffer Protect Previsto	 1.25 lb + 5 lb 3 qt	 1,2 3,6	11.1	±	4.0	bc	1.1
Organic standard pear Blossom Protect + Buffer Protect Serenade Aso	 1.25 lb+ 5 lb 96 fl oz	 1,3 4,6	16.9	±	2.6	ab	0.6
Blossom Protect + Buffer Protect	1.25 lb + 5 lb	1,3	15.5	±	4.4	b	0.3
Thyme Guard [t]	2 qt	3,4,6	11.2	±	2.3	bc	0.9
Cinnerate	32 fl oz	3,4,6,8,9,10	16.1	±	4.0	ab	0.9
Cinnerate	32 fl oz	3,4,6	18.5	±	3.3	ab	0.5
Blossom Protect + Buffer Protect Previsto Thyme Guard [t]	1.25 lb + 5 lb 3 qt 2 qt	1,2 3 6	11.2	±	7.5	c	0.4
Blossom Protect + Buffer Protect Cinnerate	1.25 lb + 5 lb 32 fl oz	1,2 4,6	21.3	±	4.3	ab	0.4
Problad Verde [s] Cinnerate	40 fl oz 32 fl oz	1,3 2,6	15.3	±	3.1	ab	0.8

Water treated check	NA	3,4,6	35.5	±	5.4	a	0.3

[z] Timings 1: 70% bloom, 2: 90% bloom, 3: morning before evening inoculation (full bloom), 4: morning after inoculation, 5: 2 days after inoculation, 6: 3 days after inoculation (petal fall), 7: 4 days after inoculation, 8: 6 days after inoculation, 9: 2 weeks after inoculation, 10: 3 weeks after inoculation

[u] Inoculation was conducted on the evening of 22 Apr 2021 at full bloom (of king blooms) using a suspension of freeze-dried cells of *Erwinia amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 17x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[x] Amended with Regulaid: 16 fl. oz. per 100 gallons. Buffered to 5.6 pH.

[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).

[t] Acidified to pH 4.

[s] Amended with NuFilm: 16 fl. oz. per 100 gallons.

[v] Fruit marking is rated from an average of 25 fruit per tree. In 2022 less than 25 fruit were often present. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades. No statistical differences were observed between treatments.

Table 8. Effect of essential oil/ plant extract treatments applied to apple, cv. Red Delicious on infection of *E. amylovora* in apple blossoms in Wenatchee, WA, in 2021[z]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters[y]				Fruit russet[u]
Streptomycin standard (Firewall 17)[x]	8 oz	100% bloom	16.1	±	2.3	a[w]	0.06
Oxytetracycline standard[y] (Fireline 17)[x]	16 oz	100% bloom, petal fall	17.0	±	5.7	a	0.00
Organic standard apple Blossom Protect + Buffer Protect Previsto	1.24 lb + 8.75 lb 3 qt	70% bloom, 100% bloom, 100% bloom + 1 day, petal fall	17.8	±	4.5	a	0.69
Organic standard pear Blossom Protect + Buffer Protect Serenade Opti	1.24 lb + 8.75 lb 20 oz	70% bloom, 100% bloom, 100% bloom + 1 day, petal fall	13.9	±	2.6	a	0.73
Blossom Protect + Buffer Protect Previsto Thyme oil (23%) (Thyme Guard)[v]	1.24 lb + 8.75 lb 3 qt 2 qt	50% bloom, 100% bloom, 100% bloom + 1 day, petal fall	16.0	±	1.9	a	0.34
Thyme oil (23%) (Thyme Guard)[v]	2 qt	100% bloom, 100% bloom + 1 day, petal fall	21.4	±	3.9	ab	0.24
Thymol (23%) (Thymox)	2 qt	100% bloom, 100% bloom + 1 day, petal fall	22.9	±	5.7	ab	0.35
ET91[v]	640 oz	100% bloom, 100% bloom + 1 day, petal fall	21.7	±	5.3	ab	0.06
ET91[v]	320 oz	100% bloom, 100% bloom + 1 day, petal fall	21.9	±	3.7	ab	0.06
Cinnamon oil (60%) (Cinnerate) + Lupine[h]	32 oz + 40 oz	100% bloom, 100% bloom + 1 day, petal fall, petal fall + 3 days	17.6	±	3.2	ab	0.02
Cinnamon oil (60%) (Cinnerate)	32 oz	100% bloom, 100% bloom + 1 day, petal fall, petal fall + 3 days	20.8	±	3.7	ab	0.01
Thyme oil (3%) (G)	256 oz	100% bloom, 100% bloom + 1 day, petal fall	35.9	±	8.4	bc	0.00
Water-treated check	NA	100% bloom, petal fall, petal fall + 3 days	38.6	±	5.1	c	0.00

[z] Application dates were: 18 Apr (70% bloom), 19 Apr (full bloom), 20 Apr (full bloom + 1 day), 23 Apr (petal fall), 26 April (petal fall + 3 days). Inoculation was conducted on the evening of 19 Apr 2021 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells and 50% live cells of *E. amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 40-94 x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[x] Amended with Regulaid: 16 fl. oz. per 100 gallons. Buffered to 5.6 pH.

[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).

[v] Acidified to pH 4.

[u] Fruit marking, average of 25 fruit per tree. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades.

[h] Banda de Lupinus albus doce (20%).

Table 9. Effect of Essential Oil/ Plant Extract Treatments on infection of *E. amylovora* in apple blossoms in Orondo, WA, in 2020 [‡]

Treatment	Application timings	Infections per	Fruit

	Rate per 100 gallon water		100 clusters				russet
Streptomycin (Firewall 17)[yz]	28.8 oz	50% bloom, 100% bloom, petal fall	2.8	± 1.2	a		0
Oxytetracycline [y] (Fireline 17)[yz]	28.8 oz	50% bloom, 100% bloom, petal fall	8.2	± 2	b		0
Organic Standard (Blossom Protect/Buffer) + Soluble Copper (Previsto)	1.24 lb 8.75 lb 3 qt	50% bloom, 80% bloom, 100% bloom, petal fall	9.5	± 1.3	bc		0.2
Thyme oil (23%) (Thyme Guard)	2 qrt	80% bloom, 100% bloom +1, petal fall	17	± 2.3	cd		0
Thymol (23%) (Thymox)	2 qrt	80% bloom, 100% bloom, petal fall	22	3.5	d		0
Cinnamon oil (60%) (Cinnerate)	1 qt	50% bloom, morning after inoc, petal fall	19	± 3.5	d		0
TS28	21.9 ml	100% bloom, +1 day, petal fall	23	± 5.5	cd		0
TS108	25 ml	100% bloom, +1 day, petal fall	31	± 5.8	d		0
ET91	38.4 oz	100% bloom, +1 day, petal fall	10	± 6.6	b		1.9
Lupine[u]	40 oz	50% bloom, morning after inoc, petal fall	22.6	± 4.1	cd		0
Water-treated check	NA	100% bloom, +1 day, petal fall	31	± 7.1	d		0

[y]Amended with Regulaid: 30 fl. oz. per 100 gallons.
[z]Buffered to 5.6 pH.
[‡]Application dates were: April 14 (20% bloom), April 16 (50% bloom), April 17 (80% bloom) and April 18 (full bloom), April 19 (full bloom plus 1 day), April 22 (petal fall). Inoculation was conducted on the evening of April 18, 2020 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells of *E. amylovora* strain 153N (streptomycin and oxytetracycline sensitive pathogen strain) and 50% live cells, which was prepared at 24 x 10^6 CFU per ml.
[u]Banda de Lupinus albus doce (20%).

Table 10. Effect of Essential Oil/Plant Extract Treatments on infection of *E. amylovora* in apple blossoms in Wenatchee, WA, in 2019[‡]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters**			Fruit russet
Streptomycin (Firewall 17)[yz]	28.8 oz	50% bloom, 100% bloom, petal fall	4.6	± 2.7	a	0
Oxytetracycline (Fireline 17) [yz]	24 oz	50% bloom, 100% bloom, petal fall	5.8	± 3.2	a	0
Organic standard (lime sulfur, Blossom Protect+ Buffer Protect, Previsto)	6 gal 1.24+8.75 lb 3 qt	LS: 70% bloom BP: 20% bloom, 80% bloom PR: 100% bloom, petal fall	6.1	± 1.2	a	0
Cueva/ Previsto	4qt/3qt	day before and day after 100% bloom, petal pall	9.7	± 2.7	a	0
Thyme oil (23%) (Thyme Guard)	2 qrt	50%, 100% bloom, petal fall, + 4 post petal fall apps	9.2	± 5.3	a	4.1 ± 0.9
Untreated, Inoculated check	NA	100% bloom	20.9	± 11.1	b	0

[z]Buffered to 5.6 pH.
[y] Amended with Regulaid: 32 fl. oz. per 100 gallons.
[‡]Application dates were: April 21 (pink), April 23 (20% bloom), April 24 and 25 (50% bloom), April 26 (full bloom minus 1 day), April 27 (full bloom), April 28 (full bloom plus 1 day), May 1, 2019 (petal fall), May 2, May 4 and May 6, and May 10, 2019. Inoculation was conducted on the evening of April 27, 2019 at full bloom (of king blooms) using a suspension of freeze-dried cells of *E. amylovora* strain 153N (streptomycin and oxytetracycline sensitive pathogen strain), which was prepared at 1.3 x10^6 CFU per ml and on May 1, 2019 using live culture prepared at 1x10^6 CFU ml^{-1}.

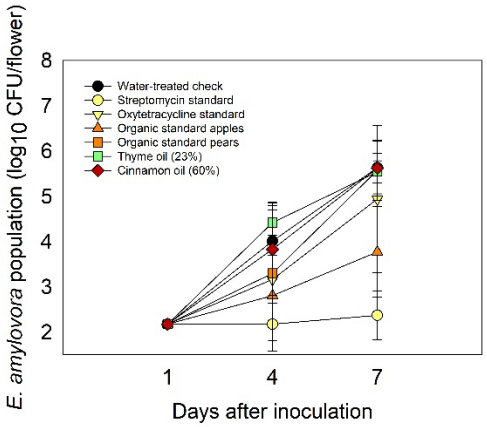

Figure 4. Effect of essential oil/plant extract treatments applied to pear cv. Anjou trees to suppress fire blight on the population size of *E. amylovora* strain 153N on flowers 1, 4 and 7 days post-inoculation of the pathogen in Wenatchee, WA, in 2022.

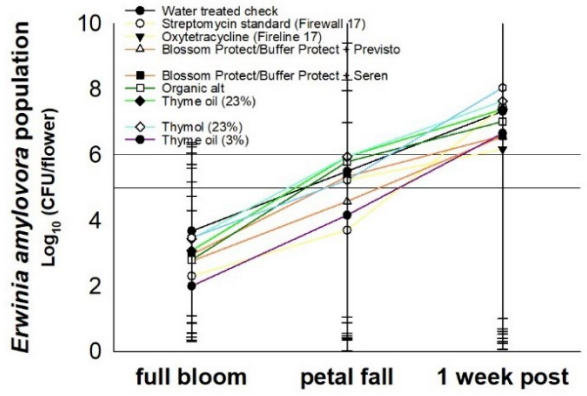

Figure 5. Effect of thyme treatments on the population size of *E. amylovora* strain 153N on flowers at full bloom, petal fall and 1 week post petal fall in Wenatchee, WA, in 2021.

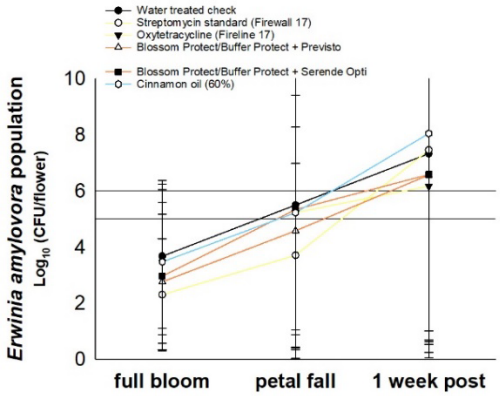

Figure 6. Effect of cinnamon oil products on the population size of *E. amylovora* strain 153N on flowers at full bloom, petal fall and 1 week post petal fall in Wenatchee, WA, in 2021.

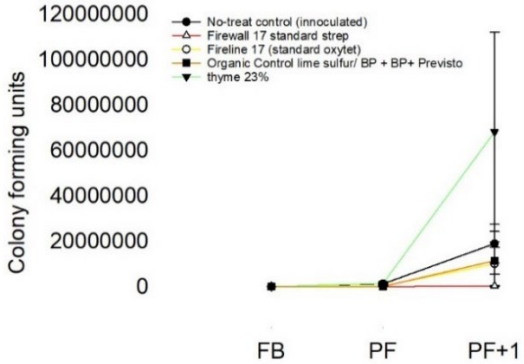

Figure 7. Effect of thyme oil treatments on the population size of *E. amylovora* strain 153N on flowers at full bloom (FB), petal fall (PF) and petal fall + 1 week (PF+1) in Orondo, WA, in 2020.

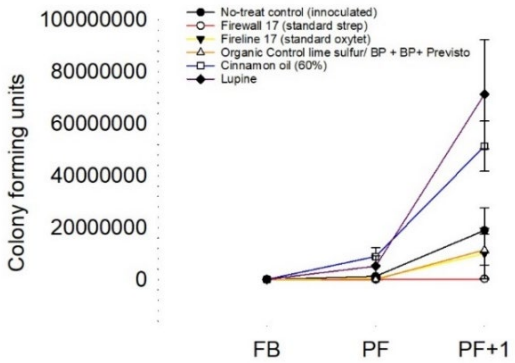

Figure 8. Effect of cinnamon oil treatments on blight on the population size of *E. amylovora* strain 153N on flowers at full bloom (FB), petal fall (PF) and petal fall + 1 week (PF+1) in Orondo, WA, in 2020.

Biological Control Products

There is interest in bacteriophage products for control of fire blight. A *bacteriophage* is a type of virus that infects bacteria. "*Bacteriophage*" literally means "bacteria eater," because *bacteriophage* destroy their host cells. *Bacteriophage* infect bacteria and multiply inside the host (lytic cycle), killing the host and releasing the progeny. *Bacteriophage* are composed of a nucleic acid molecule that is surrounded by a protein structure. *Bacteriophage* are very specific to a type of bacteria which make them an attractive option for IPM management. However, *bacteriophage* have some challenging features. *Bacteriophage* can only replicate in bacterial cells and are sensitive to environmental conditions. pH, UV, and precipitation can all reduce their ability to live on the leaf surface (Gill and Abedon 2003). Interestingly, there is some evidence that *bacteriophage* can be effective when they penetrate and translocate through the plant (Nagy et al. 2015). For example, *bacteriophage* have been effective for bacterial wilt of tomato in greenhouse trials (Fujiwara et al. Vol. 77, No. 12; Iriarte et al. 2012).

In 2020 and preliminary trials in 2019 *bacteriophage* products performed no better than the water treated check (Tables 13,14). In 2022 the *bacteriophage* product provided 58% relative control not significantly lower than oxytetracycline standards. Based on work by Sundin (Michigan State University) it was hypothesized that the addition of a particle film sun protectant would reduce *bacteriophage* die-off due to UV and enhance control potential. In 2020 addition of kaolin clay (Surround) did not improve control (Table 12).

Bacteriophage active against *E. amylovora* have had variable results. For example in Michigan OmniLytics phage tested in 2018 with 74% relative disease suppression and Fire Quencher phage with 42% relative disease suppression while in 2019 AgriPhage had 35 to 39% relative disease suppression (Outwater et al. 2019; Sundin et al. 2018). This variation has also been observed in Washington, where a relative disease suppresison of *bacteriophage* products below 20% was observed in 2019 and 2020 (Tables 13,14), while in 2022 the application of Agriphage at full bloom, one day after full bloom and petal fall provided 58% relative disease suppression (Table 11). One factor explaining this variation could be UV light from the sun, as it has been reported that *bacteriophage* of *E. amylovora* are sensitive to it (Buttimer et al. 2017). In fact, the total solar radiation in 2019 and 2020 was above 20 MJ/m^2 more days than in 2022.

Table 11. Effect of biological treatments applied to pear, cv. Anjou on infection of *E. amylovora* in pear blossoms in Wenatchee, WA in 2022 [u]

Treatment	Rate per 100 gallons water	Application timings [z]	Infections per 100 clusters [y]				Fruit russet [v]
Streptomycin standard (Firewall 50WP) [x]	8 oz	3	4.4	±	1.2	d [w]	0.2
Oxytetracycline standard (Fireline 45WP) [x]	9 oz	3,6	15.7	±	4.8	bc	0.2
Blossom Protect + Buffer Protect	1.25 lb + 5 lb	1,3	15.5	±	4.4	c	0.3
Bacillus Subtilis (Serenade Aso)	96 fl oz	3,4,6	16.7	±	2.8	bc	0.6
Phage 7 (Agriphage)	2 qt	3,4,6	14.9	±	1.2	bc	0.2
Citric acid (F)	1.4 gal	3,4,6	15.9	±	3.2	bc	0.4
PSU1	200 g	2,4,6	25.5	±	3.2	ab	0.2
PSU2	1.7 kg	2,4,6	18.3	±	6.5	bc	0.3
PSU3	500 g	2,4,6	15.0	±	3.5	bc	0.2
Water treated check	NA	3,4,6	35.5	±	5.4	a	0.3

[z] Timings 1: 70% bloom, 2: 90% bloom, 3: morning before evening inoculation (full bloom), 4: morning after inoculation, 5: 2 days after inoculation, 6: 3 days after inoculation (petal fall), 7: 4 days after inoculation, 8: 6 days after inoculation, 9: 2 weeks after inoculation, 10: 3 weeks after inoculation

[u] Inoculation was conducted on the evening of 22 Apr 2021 at full bloom (of king blooms) using a suspension of freeze-dried cells of *Erwinia amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 17x10^6 CFU ml^{-1}).

[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.
[x] Amended with Regulaid: 16 fl. oz. per 100 gallons. Buffered to 5.6 pH.
[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).
Fruit marking is rated from an average of 25 fruit per tree. In 2022 less than 25 fruit were often present. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades. No statistical differences were observed between treatments.

Table 12. Effect of biological treatments applied to apple, cv. Red Delicious on infection of *E. amylovora* in apple blossoms in Wenatchee, WA, in 2021[z]

Treatment	Rate per 100 gallons water	Timing	Infections per 100 clusters[y]				Fruit russet[s]
Streptomycin standard (Firewall 17)[x]	8 oz	100% bloom	16.1	±	2.3	ab[w]	0.06
Oxytetracycline standard (Fireline 17)[x]	16 oz	100% bloom, petal fall	17.0	±	5.7	a	0.00
Organic standard apple Blossom Protect + Buffer Protect Previsto	1.24 lb+ 8.75 lb 3 qt	70% bloom, 100% bloom, 100% bloom + 1 day, petal fall	17.8	±	4.5	a	0.69
Organic standard pear Blossom Protect + Buffer Protect Serenade Opti	1.24 lb + 8.75 lb 20 oz	70% bloom, 100% bloom, 100% bloom + 1 day, petal fall	13.9	±	2.6	a	0.73
RejuGro[u]	15.1 g	100% bloom, 100 bloom + 1 day,	19.1	±	1.8	ab	0.00
UW37_4RLE	400 ml	100% bloom, 100% bloom + 1 day, petal fall	30.4	±	4.5	bc	0.00
UW58_4DLA	400 ml	100% bloom, 100% bloom + 1 day, petal fall	17.0	±	4.4	a	0.05
UW29_2ALA1	400 ml	100% bloom, 100% bloom + 1 day, petal fall	23.4	±	3.5	abc	0.00
PSU1[t]	1x10^9 CFU ml^{-1}	100% bloom, 100% bloom + 1 day	14.5	±	4.3	a	0.05
Water-treated check	NA	100% bloom, petal fall, petal fall + 3 days	38.6	±	5.1	c	0.00

[z] Application dates were:18 Apr (70% bloom), 19 Apr (full bloom), 20 Apr (full bloom + 1 day), 23 Apr (petal fall), 26 April (petal fall + 3 days). Inoculation was conducted on the evening of 19 Apr 2021 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells and 50% live cells of *E. amylovora* strain Ea153 (streptomycin and oxytetracycline sensitive strain) prepared at 1 x10^6 CFU ml^{-1} (verified at 40-94 x10^6 CFU ml^{-1}).
[y] Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.
[x] Amended with Regulaid: 16 fl. oz. per 100 gallons. Buffered to 5.6 pH.
[w] Treatments followed by the same letter are not significantly different at P=0.05 Fisher's T test (LSD).
[u] Amended with PEG4000 and Regulaid: 16 fl. oz. per 100 gallons.
[s] Fruit marking, average of 25 fruit per tree. Rated on a 0 to 15 scale where ratings below 3 indicate no commercial downgrades.

Table 13. Effect of Biological Control Product Treatments on *E. amylovora* infection of apple blossoms in Wenatchee, WA, in 2020.[‡]

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters**				Fruit Russet
Untreated, Inoculated Check	water	100% bloom, +1 day, petal fall	31	±	7.1	c	0
Streptomycin standard (Firewall 17)[zy]	28.8 oz	50% bloom, 100% bloom, petal fall	2.8	±	1.2	a	0
Oxytetracycline standard (Fireline 17)[zy]	28.8 oz	50% bloom, 100% bloom, petal fall	8.2	±	2.0	b	0
Organic standard (Blossom Protect/Buffer Protect +Previsto)	1.24 lb 8.75 lb 3 qt	50% bloom, 80% bloom, 100% bloom, petal fall	9.5	±	1.3	b	0.02
Phage7 (Agriphage)	2 qt	100% bloom 12hr before ap, +1 day, +3 days	24	±	4.8	c	0

Phage7 + Surround (Agriphage)	2 qt + 0.1 lb	100% bloom 12hr before ap, +1 day, +3 days	31	±	3.7	c	0

** Transformed log(x + 1) prior to analysis of variance; non-transformed means are shown.

[y] Amended with Regulaid: 30 fl. oz. per 100 gallons.

[z] Buffered to 5.6 pH.

‡Application dates were: April 14 (20% bloom), April 16 (50% bloom), April 17 (80% bloom) and April 18 (full bloom), April 19 (full bloom plus 1 day), April 22 (petal fall). Inoculation was conducted on the evening of April 18, 2020 at full bloom (of king blooms) using a suspension of 50% freeze-dried cells of *Erwinia amylovora* strain 153N (streptomycin and oxytetracycline sensitive pathogen strain) and 50% live cells, which was prepared at 24 x 10^6 CFU per ml.

Table 14. Effect of Biological Control Product Treatments on *E. amylovora* infection of apple blossoms in Wenatchee, WA, in 2019‡

Treatment	Rate per 100 gallons water	Application timings	Infections per 100 clusters			
Streptomycin standard (Firewall 17)[zy]	28.8 oz	50% bloom, 100% bloom, petal fall	4.6	±	2.7	a
Oxytetracycline standard (Fireline 17) [zy]	24 oz	50% bloom, 100% bloom, petal fall	5.8	±	3.2	ab
Organic standard (lime sulfur, Blossom Protect+ Buffer Protect/ Previsto)	6 gal 1.24 lb/8.75 lb 3 qt	LS: 70% bloom BP: 20% bloom, 80% bloom PR: 100% bloom, petal fall	6.1	±	1.1	ab
Cueva/ Previsto	4qt/3qt	day before and day after 100% bloom, petal pall	9.7	±	2.7	abc
Phage7[y]	1 qt	50% bloom, 100% bloom, petal fall	17.3	±	3.6	bc
Phage7 + oxytet (Fireline)[y]	1 qt + 0.1 lb	50% bloom, 100% bloom, petal fall	12.4	±	3.4	abc
Bacillus Subtilis (A)	30 oz	50% bloom, 100% bloom, petal fall	22.5	±	7.1	c
Bacillus Subtilis QST 713 strain (Serenade Opti)	20 oz	day before and day after 100% bloom, petal fall	16.0	±	3.2	abc
Untreated, Inoculated Check	water	100% bloom	20.9	±	11.1	c

[y]Amended with Regulaid: 32 fl. oz. per 100 gallons.

[Z]Buffered to 5.6 pH.

‡Application dates were: April 21 (pink), April 23 (20% bloom), April 24 and 25 (50% bloom), April 26 (full bloom minus 1 day), April 27 (full bloom), April 28 (full bloom plus 1 day), May 1, 2019 (petal fall), May 2, May 4 and May 6, and May 10, 2019. Inoculation was conducted on the evening of April 27, 2019 at full bloom (of king blooms) using a suspension of freeze-dried cells of *E. amylovora* strain 153N (streptomycin and oxytetracycline sensitive pathogen strain), which was prepared at 1.3 x10^6 CFU per ml and on May 1, 2019 using live culture prepared at 1x10^6 CFU ml^{-1}.

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Executive Summary

Fire Blight Product Testing for Effective Recommendations

Keywords: fire blight, apple, pear, biopesticides, essential oils, biologicals

Abstract:

The efficacy of several alternatives to antibiotics for the control of fire blight were tested in four Washington trials. Alum (potassium aluminum sulfate) provided good control similar to antibiotic checks as well as biological Blossom Protect (*A. pullulans*) and several copper products (Previsto, Mastercop, Instill). Alum provided a median of 72% control relative to the water treated check in 2016 to 2022 trials when the product was applied at an 8 to 10 lb per 100 gal rate. This control is comparable to comparable to 74% in the oxytetracycline standard and 85% in the streptomycin standard (2013 to 2022 median). Marking from chemical russet was negligible in all WA trials (< 1 on a 0 to 15 scale). Several essential oil, and peracetic acid-peroxide products (Oxidate 5.0, Jet Ag, Thyme Guard, Thymox, Cinnerate) provided moderate disease suppression similar to some other biological and copper products (Serenade Opti, Cueva) and may be best incorporated as rotational products as part of an integrated program during lower risk periods. Oxidizing agents (Jet Ag and Oxidate 5.0) produced median relative disease suppression of 53% to 62% with 2 to 3 applications post inoculation (4 trials: 2019-2022). Essential oil plant extracts from thyme and cinnamon (Thyme Guard, Thymox, Cinnerate) resulted in median relative disease suppression of 49% (thyme oils 3 to 6 applications) and 45% (cinnamon oils 3 to 4 applications) between 2019 and 2022. In 2020 and preliminary trials in 2019 bacteriophage products performed no better than the water treated check. In 2022 the bacteriophage product provided 58% relative control not significantly lower than oxytetracycline standards. In order to minimize the risk of fruit marking managers should consider drying times for essential oils and peracetic acid-peroxide products as well as soluble coppers.