Epidemiology and Control of Bacterial Spot of Almond in California

Project No.:   **18-PATH5.Adaskaveg**

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A. Summary

Bacterial spot caused by *Xanthomonas arboricola* pv. *pruni* (*Xap*) is a relatively new disease in California that has been reported in locations from Butte to Kern Co. It is most severe on cv. Fritz but may also occur on other cultivars. Wet, warm springs are favorable for the disease, but frequent wetness from dew, rain, or high-angle sprinklers can also result in outbreaks. With high rainfall but low temperatures, the incidence of bacterial spot was low to moderate in the spring of 2020. This is in contrast to 2019 when rainfall occurred during warmer environments. In surveys, we detected no copper resistance in the pathogen populations. Based on DNA fingerprinting, *Xap* strains from different locations were very homogeneous which may indicate a recent introduction.

Pathogen isolations over several years confirmed that fruit mummies and peduncles remaining on the tree are the major primary inoculum sources, but *Xap* was also detected in healthy flower buds, emerging leaves, and spurs close to infected mummies in the tree. No tree cankers have been observed in California, and cankers did not develop in our wound inoculations of twigs at different ages in early spring 2020. In inoculations of cv. Fritz trees at petal fall or early fruit set, disease on fruit was first observed after 8 weeks, whereas inoculations of young fruit developed symptoms after 5 weeks, coinciding with increasing temperatures. In each inoculation, some disease developed without providing additional wetness. Disease incidence increased when at least 4 h of wetness was provided. It is not clear if flower inoculations directly result in an infection, or if surviving epiphytic inoculum later infects the developing fruit. Still, bactericide applications at full bloom and petal fall were shown to be most effective among in-season treatment timings suggesting early carpel infections. The temperature optimum for growth of *Xap* was determined to be 25C, there was very little growth at 10C and 15C, and moderate growth at 20C and 35C.

We evaluated the efficacy of dormant and in-season applications with bactericides on the management of bacterial spot over several seasons. We have been evaluating new copper alternatives and treatments that could be used in organic almond production. In small scale studies in the spring of 2020 on cv. Fritz, branches with flowers and developing fruit at UC Davis were treated and then inoculated. Champ/Manzate, Kasumin/Manzate, and ε-poly-L-lysine/ManniPlex Zn were the most effective treatments that reduced the incidence of bacterial spot from 46% in the control to ≤18.2% with no disease present in the Champ/Manzate treatment. Similarly, in a field trial with three applications that started at full bloom and where 31.7% disease incidence developed in the control, Champ, Champ/Manzate, Kasumin/Manzate, and ε-poly-L-lysine/ManniPlex Zn/ZnO were the most effective treatments. The chlorine doxide-generating compound TDA-NC-1, Nisin/ManniPlex Zn/ZnO, and BacStop also showed good efficacy with an incidence of less than 10%. In a second trial, disease incidence in the untreated control was only 3.8%, and all treatments evaluated significantly reduced the

disease. In previous trials with organic products, Blossom Protect/buffer and Serenade ASO that were not evaluated in 2020 were also effective.

Based on our results from several years of field studies, in wet winter/spring seasons, a delayed dormant bactericide application to reduce inoculum should be followed by bloom and petal fall treatments around rainfall events and rising temperatures to prevent new infections. Bloom applications with copper cause minimal phytotoxicitywhen applied at rates (starting a 1 lb MCE) reduced by 50% with each consecutive application. In drier spring seasons, only a dormant treatment or bloom/petal fall applications may be necessary for effective disease management. Kasugamycin is pending registration on almond in 2021. Integration of different compounds should reduce the potential of resistance to any one mode of action and overuse of copper that may cause phytotoxicity.

In trials for the management of bacterial blast, kasugamycin was effective and reduced the disease and increased fruit set. A Section 18 emergency registration was successfully submitted in 2019 for use in 2020 and was re-submitted for renewal this past fall for 2021.

B. Objectives *(Milestones are indicated in bold italics)*

I) **Disease epidemiology** –

    A. Pathogen identification and characterization of populations. ***Based on DNA fingerprinting, the pathogen population was found to have very little genetic diversity, and this may indicate a recent introduction.***

    B. Identify minimum temperature for growth. ***The optimum temperature for in vitro growth was 25°C; there was very little growth at 10C and 15C.***

    C. Field evaluation of inoculations using wetness periods and fruit stages. ***Short wetness periods can result in disease, but disease increased with 4 h of wetness.***

II) **Management of bacterial spot** -

    A. In vitro sensitivity to new food grade biobactericides (e.g., nisin, poly-L-lysine, and activators). ***Nisin and ε-poly-L-lysine reduced growth of* Xap*, but in combination with EDTA, both antimicrobial food additives completely prevented growth.***

    B. Field testing of dormant and in-season bactericides and biobactericides (EPA biopesticide classification):

        i. Kasugamycin, ZTD, and adjuvants. ***In season-treatments with kasugamycin effectively reduced bacterial spot, but it was more effective in combination with copper or mancozeb. A US registrant for ZTD could not be identified.***

        ii. Nisin, ε-poly-L-lysine, and activators (biobactericides). ***Both significantly reduced the disease. Efficacy was very good in combination with Dart or ManniPlex Zn. Evaluations are ongoing with new registrant formulations.***

        iii. Biological controls – Serenade ASO, Blossom Protect, and others. ***Both biocontrols showed good efficacy and provide treatment options for organic almond production. Other modes of action are also being identified.***

III) **Management of bacterial blast** -

Kasugamycin applied immediately prior to frost events, decreased flower blast by ca. 80% as compared to the control. A Section-18 was obtained in 2020.
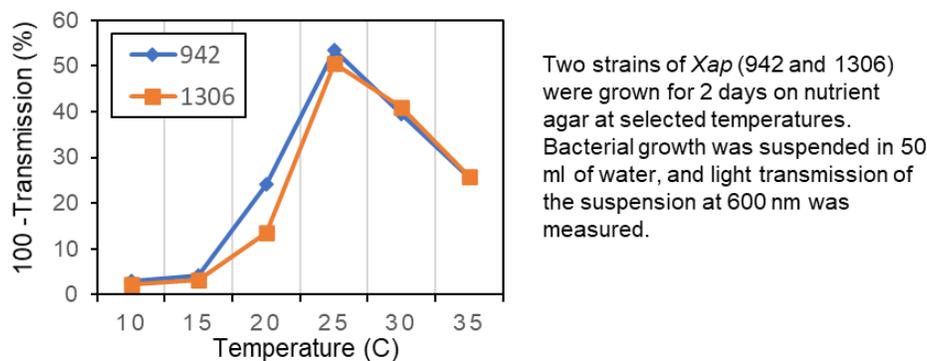
C. Results and Discussion
I) **Disease epidemiology** –

A. *Pathogen identification and characterization of populations.* In repetitive element sequence-based polymerase chain reactions (rep-PCR), a limited number of intense bands 100 to 1,500 bp in size was consistently amplified for 113 *Xap* isolates evaluated. Fingerprint patterns were identical among isolates indicating a high degree of genetic homogeneity for the California *Xap* population based on this method. This limited diversity may indicate a recent introduction. The primers employed are a standard tool to detect variability within bacterial populations including *X. arboricola* pv. *juglandis* where a high degree of genetic heterogeneity was demonstrated by us and others.

B. *Identify minimum temperature for growth.* The two strains of the pathogen used in the study responded very similar in their in vitro growth at temperatures between 10C and 35C (Fig. 1). Very little growth occurred at 10C and 15C. The optimum temperature was 25C, and growth was moderate at 20C and 35C. These results support our field observations where symptoms on fruit only develop when temperatures are increasing in the spring.

Fig. 1.Temperature optimum for in vitro growth of *Xanthomonas arboricola* pv. *pruni*

Two strains of *Xap* (942 and 1306) were grown for 2 days on nutrient agar at selected temperatures. Bacterial growth was suspended in 50 ml of water, and light transmission of the suspension at 600 nm was measured.

C. *Field evaluation of inoculations using wetness periods and fruit stages.* In inoculations of cv. Fritz trees at petal fall or early fruit set, disease on fruit was first observed 8 weeks after inoculation, and in inoculations during early fruit development, disease was first observed after 5 weeks. The onset of disease symptom development coincided with increasing temperatures (average daily temperatures above 20C; Fig. 2 bottom). At each inoculation timing, some disease developed without providing additional wetness by bagging (Fig. 2 A,B,C) and was highest (i.e., 45%) in the inoculation of young fruit (Fig. 2C) and lowest (i.e., 4.2%) in inoculations at early fruit set (Fig. 2B). Thus, sufficient wetness for development of disease was provided by applying the inoculum suspension. Disease incidence increased when at least 4 h of wetness were provided by bagging, and the increase was different between inoculation timings. In the inoculations at late petal fall, disease increased with increasing wetness periods of up to 24 h. For the other two inoculation timings, there was a steep increase in disease incidence from 0 to 4 h of wetness, and longer wetness periods did not lead to further increase. The highest levels of disease developed in the young fruit inoculations. These results indicate that short wetness periods are sufficient for high disease development in inoculation studies. Under natural conditions, however, multiple wetness periods will be required because inoculum first has to be produced and dissemined before infection can occur. It is not clear if flower inoculations directly result in an infection, or if surviving inoculum later infects the

developing fruit. Because applications at full bloom and petal fall were shown to be more effective than later in-season timings, it appears that the host is most susceptible during this period. The lower disease incidence in petal fall and early fruit set inoculations as compared to young fruit inoculations in our studies may be due to low temperatures in early spring.

Fig. 2. Effect of wetness duration after inoculation of cv. Fritz almond on bacterial spot development on fruit at UC Davis in the spring of 2020
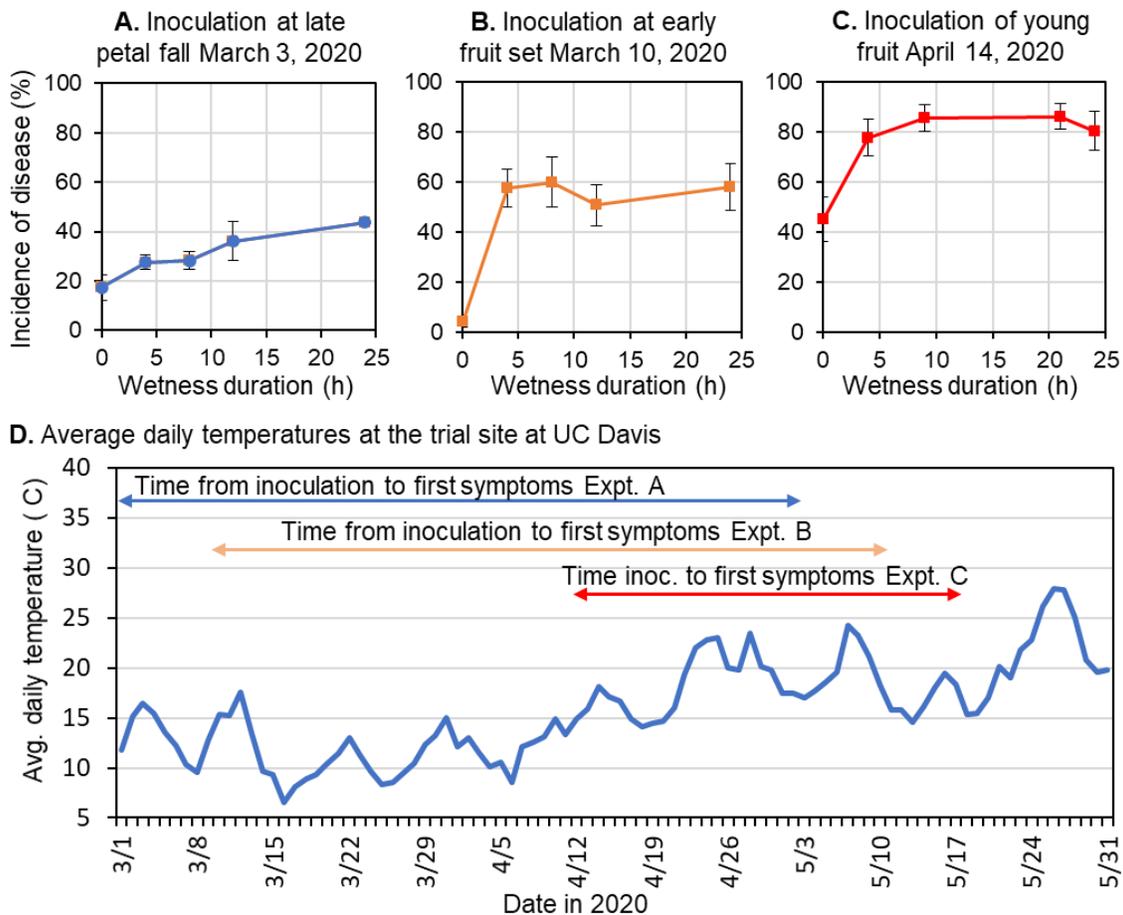
Almond twigs with flowers at petal fall, early fruit set, or with developing fruit were spray-inoculated *X. arboricola* pv. *pruni* (10^8 cfu/ml), and branches were bagged for 0, 4, 8, 12, or 24 h (petal fall and early fruit set inoculations) or for 0, 4, 9, 21, or 24 h (young fruit inoculations). The time when first symptoms were observed on fruit was recorded, and a final disease assessment was done on May 6, May 12, or May 19, 2020 for experiments A, B, or C, respectively. Temperature data were obtained from CIMIS station 6 (Davis).

II. Management of bacterial spot -

A. *In vitro sensitivity to new food grade biobactericides.* In 30-min direct exposures, nisin and ε-poly-L-lysine at 500 ppm reduced growth of *Xap* by 38 and 24%, respectively. In combination with 500 ppm EDTA, however, both antimicrobial food additives completely prevented growth.

B. *Field testing of dormant and in-season bactericides and biobactericides.* Dormant treatments with copper or copper-mancozeb applied in December or January were evaluated for their efficacy from 2014 to 2017. Both significantly reduced the incidence as compared with the untreated control, probably by reducing inoculum levels and pathogen dispersal. Additional in-season treatments with selected treatments reduced the disease to very low levels. We

established that among in-season treatments, full bloom and petal fall treatments are more effective in reducing the disease than later treatments. Data are presented for a 2019 study (Table 1).

Table 1. Effect of delayed dormant applications with copper and timing of in-season treatments with copper-mancozeb on the incidence of bacterial spot of cv. Fritz almond in San Joaquin Co. 2019

| | | In-season treatments | | | | Incidence of bacterial spot | | | |
| | | Dates and rates | | | | No dormant application | | Dormant application Champ 6 lb 2-6-18 | |
No.	Treatment	FB 2-21	PF 3-14	LPF 4-3-19	LLPF 4-25	Dis.**	LSD^	Dis.	LSD
1	Control	---	---	---	---	60.7	a A	43.3	a A
2	Champlon + Manzate	52 + 64 oz	---	---	---	32.0	ab A	17.4	bc A
3	Champlon + Manzate	52 + 64 oz	26 + 64 oz	---	---	12.0	b A	6.9	c A
4	Champlon + Manzate	---	26 + 64 oz	---	---	36.5	ab A	17.3	bc A
5	Champlon + Manzate	---	---	13 + 38 oz	13 + 38 oz	25.5	b A	30.1	ab A
6	Champlon + Manzate	---	---	---	13 + 38 oz	42.5	ab A	29.8	ab A
			Dormant - No dormant average			33.7	A	24.1	B

* - Treatments were applied using an airblast sprayer at 100 gal/A. The copper rate was reduced by half with each subsequent application from an initial rate of 52 oz/A. LPF = late petal fall, LLPF = very late petal fall.

**- Fruit were evaluated for the presence of bacterial spot on 6-13-19. Values are the incidence of diseased fruit of 100 fruit for each of four single-tree replication.

Table 2. Effect of in-season bactericide treatments on the incidence of bacterial spot of cv. Fritz almond in San Joaquin Co. 2020

| | | | | Applications | | | Incidence of bacterial spot | |
No.	Program	Treatment	Rate/A	2-19-20	3-4-20	4-1-20	%	LSD^
1	---	Control	---	---	---	---	31.7	a
2	Biologicals	BacStop	64 fl oz	@	@	@	9.6	bc
3		BacStop + EF400	38 + 38 fl oz	@	@	@	13.7	b
4		TDA-NC-1	20 oz	@	@	@	6.2	bcd
5		PolyLysine + ManniPlex Zn + ZnO	13.5 oz + 32 fl oz + 32 oz	@	@	@	2.7	cd
6		Nisin + ManniPlex Zn + ZnO	13.5 oz + 32 fl oz + 32 oz	@	@	@	8.8	bc
7		Serenade ASO + Luna Experience	10 fl oz + 64 fl oz	---	@	@	14.8	b
8	Copper	Champ	64 oz	@	32 oz	16 oz	1.7	d
9	and	Kasumin 4L + Manzate	32 fl oz + 38 oz	@	@	@	2.5	cd
10	antibiotics	Champ + Manzate	64 oz + 38 oz	@	32 oz	16 oz	0.8	d
11		New Mycoshield + Dart	8 oz + 48 fl oz	---	@	@	14.8	b

* - Treatments were applied using an airblast sprayer at 100 gal/A. The copper rate was reduced by half with each subsequent application from an initial rate of 4 lb/A. Disease on fruit was evaluated on 6-16-20. Values are the incidence of diseased fruit of 130 fruit evaluated for each of four single-tree replications.

In numerous trials, Kasumin was identified as an effective alternative to copper, and its efficacy was often increased when mixed with mancozeb. In a field trial in 2020 where the incidence of bacterial spot in the control was 31.7%, three applications of Kasumin-Manzate reduced the disease to 2.5%, and this treatment was statistically similar to Champ or Champ-Manzate (Table 1). Having several modes of action registered including copper, mancozeb, kasugamycin, and potentially others (e.g, food preservatives, natural products, biological controls) should delay the selection of resistance indefinitely. Based on our efforts, Kasumin is

pending registration on almond in the spring of 2021. The other antibiotic, oxytetracycline – Mycoshield was sometimes not as effective. Because oxytetracycline is an antibiotic used in human and animal medicine, it was not submitted to IR-4 due to potential residues on hulls that are often used for animal feed.

The antimicrobial food preservatives nisin and ε-poly-L-lysine both significantly reduced the disease when used by themselves (Table 3). We have been evaluating several additives to potentially increase their efficacy. EDTA that significantly increased the toxicity in vitro, was not effective in field applications (Table 3). The addition of zinc oxide and ManniPlex Zn to nisin improved the efficacy in 2020 (8.8% incidence), and these two additives in combination with ε-poly-L-lysine reduced the incidence of bacterial spot from 31.7% in the control to 2.7% (Table 2). The addition of Dart (capric/caprylic acids) also looked promising in another study (data not shown). Nisin and ε-poly-L-lysine were tested as pure active ingredients, and they are promising new treatments for managing bacterial plant diseases. These natural products are are US FDA-approved food preservatives and potentially can be registered as biopesticides that are exempt from residue tolerances. We will continue to improve their performance and we will work with an agrochemical company to formulate these active ingredients as agricultural products. We anticipate to have new formulations available for testing in 2021.

Table 3. Effect of in-season bactericide treatments on the incidence of bacterial spot of cv. Fritz almond in San Joaquin Co. 2019

No.	Treatment	Rate	Applications				Incidence of bacterial spot	
			7-Mar	19-Mar	3-Apr	25-Apr	%	LSD^
1	Control	---	---	---	---	---	46.2	a
2	ε-poly-L-lysine + EDTA	13.5 oz + 500 ppm	@	@	@	@	28.5	b
3	Nisin + EDTA	13.5 oz + 500 ppm	@	@	@	@	26	b
4	ε-poly-L-lysine	13.5 oz	@	@	@	@	23.8	b
5	Nisin	13.5 oz	@	@	@	@	20	b
6	Champ	32 oz - 16 oz - 8 oz	@	@	@	@	3.5	c

* - Treatments were applied using an airblast sprayer at 100 gal/A. The copper rate was reduced by half
with each subsequent application from an initial rate of 2 lb/A. Disease on fruit was evaluated on
6-13-19. Values are the incidence of diseased fruit of 100 fruit for each of four single-tree replication.

Table 4. Effect of in-season biological and copper treatments on the incidence of bacterial spot of cv. Fritz almond in San Joaquin Co. 2019

No.	Treatment	Rate	Applications				Incidence of bacterial spot	
			Full bloom 2-20	Petal fall 3-19	Late petal fall 4-3	5 wk after petal fall 4-25	%	LSD^
1	Control	---	---	---	---	---	44.7	a
2	Serenade ASO + NuFilm-P	64 fl oz + 8 fl oz	@	---	@	---	17.3	bc
3	Serenade ASO + NuFilm-P	64 fl oz + 8 fl oz	---	@	---	@	26.8	ab
4	Serenade ASO + Nufilm-P + Sugar	64 fl oz + 8 fl oz + 32 oz	@	---	@	---	21.5	bc
5	Serenade ASO + Nufilm-P + Sugar	64 fl oz + 8 fl oz + 32 oz	---	@	---	@	18.8	bc
6	Blossom Protect + buffer	20 oz + 80 oz	@	---	@	---	8.0	c
7	Blossom Protect + buffer	20 oz + 80 oz	---	@	---	@	15.3	bc
8	Champ	32 - 16 oz	@	---	@	---	20.3	bc
9	Champ	32 - 16 oz	---	@	---	@	6.0	c

* - Treatments were applied using an airblast sprayer at 100 gal/A. The copper rate was reduced by half with each
subsequent application from an initial rate of 2 lb/A. Disease on fruit was evaluated on 6-6-17. Values are the incidence
of diseased fruit of 100 fruit evaluated for each of four single-tree replication.

The biological controls products Serenade ASO and Blossom Protect showed good efficacy in two-application programs in the spring of 2018 and 2019, similar to copper (Tables 4,5). There was a trend of Blossom Protect to be the more effective one, and the efficacy of Serenade ASO was not improved when mixed with Luna Experience, a fungicide that others claim has antibacterial activity (Table 2). Zinkicide, another experimental bactericide, also showed good efficacy (Table 5), but EPA will not register this compound because it contains nanoparticles.

Table 5. Effect of in-season biological treatments on the incidence of bacterial spot of cv. Fritz almond in San Joaquin Co. 2018

| | | | PF | 3-wk | Disease** | |
No.	Treatment*	Rate(/A)	2-28	3-15	Incid. (%)	LSD^
1	Control	---	---	---	28.4	a
2	Vacciplant	64 fl oz	@	@	15.5	ab
3	Nisin + Zinkicide	13.5 oz + 64 fl oz	@	@	12.8	bc
4	Serenade Opti + Nufilm-P	64 + 8 fl oz	@	@	12.0	bc
5	Champ 50WDG	2-0.5 lb	@	@	7.3	bcd
6	Zinkicide	64 fl oz	@	@	6.8	bcd
7	Serenade Opti + Nufilm-P + Sugar	64 + 8 fl oz + 32 oz	@	@	4.0	cd
8	Blossom Protect + Buffer	20 oz + 143 oz	@	@	2.8	d

* - Treatments were applied using an air-blast sprayer at 100 gal/A. The copper rate was reduced by half with each subsequent application from an initial rate of 2 lb/A.

**- Disease on fruit was evaluated on 6-20-18. Values are the incidence of diseased fruit of 100 fruit evaluated for each of four single-tree replications.

In summary, our research provides insight into the epidemiology of this new disease in California, and this information can be used to optimize management practices. Overwintering of the pathogen in mummies emphasizes the importance of mummy removal in the winter. Short wetness periods appear to be sufficient for high disease development once viable is present. Kasumin was identified as an effective copper alternative or rotation treatment, and its registration is pending. Other alternatives are the biological control products Serenade ASO and Blossom Protect, and these can also be used in organic almond production. Additional bactericides that may also qualify for organic use (i.e., nisin and ε-poly-L-lysine) are in development.

III. Management of bacterial blast

In field studies where Kasumin was applied immediately prior to a frost event at pre-bud break or during bloom in 2019, the incidence of infected flowers was reduced from 19.7% to 5.9% or 4.1%, respectively (Table 6). An application of nisin/EDTA was moderately effective. ManKocide was not effective in this study possibly because the pathogen population was copper-resistant. Studies were also conducted in 2020, but environmental conditions were not favorable for disease development. Kasumin, however, was also very effective in previous studies we reported on. Use of Kasumin against bacterial blast will be included in the pending registration for management of bacterial spot.

Table 6. Efficacy of bactericide treatments against bacterial blast of cv. Independence almond, Merced Co. 2019

No.	Treatment*	Rate	Application timing 2-6-19 pre bud break	Application timing 2-20-19 flowering	Incid. of blast %	Incid. of blast LSD^
1	Control	---	---	---	19.7	a
2	Kasumin 4L	32 fl oz	@	---	5.9	c
3	Kasumin 4L	32 fl oz	---	@	4.1	c
4	ManKocide	5 lb	---	@	17.8	a
5	Nisin + EDTA 0.5 M	6.75 oz + 40 fl oz	---	@	11.3	b

* - Treatments were applied using an air-blast sprayer at a rate of 100 gal/A. Low/high temperatures on 2-7-19 were 31F/54F.

**- The incidence of blast was assessed based on 100 random flowers counted on each tree on 3-7-19.

^ - Values followed by the same letter are not significantly different based on ANOVA and LSD mean separation ($P > 0.05$).

B. Outreach Activities

1. Jan. 9, 2019: Managing diseases in almonds. Bayer CropScience 2019 Tree, Nut, and Vine Meeting, Universal Studies, Universal City, CA. 150 people. Mainly PCAs.
2. Jan. 17, 2019: Almond Disease Management - Colusa Co. Williams, CA. 35 people. Growers and PCAs.
3. Jan. 18-19, 2019: Diseases - Key economic pests, identification, biology, and treatments in almond. Independent PCA Symposium. Monterey, CA. 65 people. Mainly PCAs.
4. Jan. 21, 2019: IPM of Almond Diseases. Syngenta Crop Protection, Bakersfield CA Pre-recorded. 45 people. Growers and PCAs.
5. Jan. 29, 2019: Almond flower, foliar, and fruit diseases and their management- Spring time and Summer diseases. Cortez, CA. 45 people. Growers and PCAs.
6. Feb. 5, 2019: Bloom and foliar diseases. UCCE Annual Almond Production Meeting, Woodland, CA. 45 people. Growers and PCAs.
7. Nov. 5-7, 2019. Almond diseases and their management. 2019 UC Almond Short Course. Visalia Convention Center, Visalia, CA. 100 people. Growers and PCAs. State and Inernational audience.
8. Jan. 17, 2020: Key economic pests, identification, biology, and treatments in almond. Bayer Crop Science Annual Almond Disease Management Meeting, Monterey, CA. 70 people. Growers and PCAs.
9. Jan. 21, 2020: Almond foliar diseases. North Valley Nut Conference, Orland, CA. 250 people. Growers and PCAs as well as regulators and industry representatives.
10. Jan. 22, 2020: Fungal and bacterial almond disease management during bloom. UCCE Colusa Winter Almond Meeting, Williams, CA. 35 people. Growers and PCAs.
11. Jan 29, 2020: Key economic diseases, identification, biology, and treatments in almond. Bayer Crop Science Chico Tree Nut Meeting, Chico, CA. 250 people. Growers and PCAs as well as regulators and industry representatives.
12. Jan. 31, 2020: Bacterial blast - Why it was a problem in 2019 and can we protect against it? 2020 North San Joaquin Valley Almond Day. Modesto, CA. 250 people. Growers and PCAs as well as regulators and industry representatives.

13. Feb. 5, 2020: Almond flower, foliar, and fruit diseases and their management. Yolo-Solano-Sacramento Almond Meeting, 50 people. Growers and PCAs.

C. Materials and Methods:

I. Disease epidemiology.

A. *Pathogen identification and characterization of populations.* Published PCR primers were used to correctly identify the pathogen. The genetic variability (population structure) of *Xap* within and among orchards was investigated using rep-PCR primers REP1R-I and REP2-I, ERIC1R and ERIC2, or BOXA1R that have been used to demonstrate variability within several other bacterial species. Amplification products were separated in agarose gels, and banding patterns were compared.

B. *Identify temperature effects on growth of Xap.* Two strains of *Xap* (942 and 1306) were grown for 2 days on nutrient agar at 10, 15, 20, 25, 30, or 35C. Bacterial growth was suspended in 50 ml of water, and light transmission of the suspension at 600 nm was measured.

C. *Field evaluation of different wetness conditions.* At UC Davis, cv. Fritz almond twigs with flowers at petal fall, early fruit set, or with developing fruit were spray-inoculated *Xap* (10^8 cfu/ml), and branches were bagged for 0, 4, 8, 12, or 24 h (petal fall and early fruit set inoculations) or for 0, 4, 9, 21, or 24 h (young fruit inoculations). The time when first symptoms were observed on fruit was recorded, and a final disease assessment was done on May 6, May 12, or May 19, 2020 for the three experiments, respectively. Temperature data were obtained from CIMIS station 6 (Davis).

II. Management of bacterial spot.

A. *In vitro sensitivity of Xap to new food grade biobactericides.* In direct contact studies, bacterial suspensions were exposed to test substances for 30 min. Suspensions were then diluted 1:100 with sterile water and plated onto agar media. Colonies were enumerated after 2 days.

B. *Management of bacterial spot in the field.* Field studies were done in commercial cv. Fritz orchards where the disease is known to occur. Dormant treatments were applied in previous years' studies. In-season treatments were applied using an air-blast sprayer at 100 gal/A starting at bloom, and were continued prior to rain events in the spring. In treatments with standard copper, rates were decrease from 1 to 0.5 to 0.25 and to 0.2 lb MCE/A for each application timing. Bactericide rates were based on their current labels on almond or other crops. For each treatment, there were four single-tree replications. Disease was evaluated in late spring and the incidence was calculated based on the number of diseased fruit of the total number of fruit evaluated. Data were analyzed using analysis of variance and means separation procedures of SAS 9.4.

The only challenge that we encountered in our studies was that in some years, only a low incidence of disease developed at some field trial sites due to unfavorable environmental conditions. Therefore, studies were conducted at several locations.

D. Publications that emerged from this work
1. Peer review publication:

S. E. Haack, L. Wade, H. Förster, and J. E. Adaskaveg. 2020. Epidemiology and management of bacterial spot of almond caused by *Xanthomonas arboricola* pv. *pruni*, a new disease in California. Plant Dis. 104:1685-1693.

2. Abstracts:
a. Haack, S., Wade, L., Forster, H., and Adaskaveg, J. E. 2015. Epidemiology and management of bacterial spot of almond, caused by *Xanthomonas arboricola* pv. *pruni*. Phytopathology 105(Suppl. 4):S4.55.

b. Haack, S.E., Förster, H.C., and Adaskaveg, J.E. 2018. Evaluation of copper alternatives and enhancers for managing almond bacterial spot caused by *Xanthomonas arboricola* pv. *pruni* in California. Phytopathology 108: S1.147 https://doi.org/10.1094/PHYTO-108-10-S1.147.

3. Article in trade magazine:
Adaskaveg, J.E., Forster, H., and Wade, L. 2020. Bacterial blast of almond caused crop losses in 2019. CAPCA Advisor Magazine Feb. 2020:34-36.

Please provide copies of publications