Trunk and Scaffold Canker Diseases of Almond in California

Project No.: **PATH12.Trouillas**

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A. Summary

Trunk and scaffold canker diseases (TSCD) caused by fungal pathogens are destructive diseases of almond trees. Recent surveys conducted by our laboratory confirmed the occurrence of four major canker diseases of almond in California: band canker, Ceratocystis canker and the newly identified Cytospora and Eutypa cankers. Nonetheless, up to 21 fungal pathogens were found associated with almond cankers diseases, suggesting that a comprehensive approach is required to provide effective management strategies against this wide range of fungal pathogens. Pruning wounds made for primary and secondary scaffold selections as well as maintenance pruning are common entry sites for canker pathogens. The goal of this project was to develop pruning wound protection strategies to prevent infection of almond trees and promote the establishment of orchards as well as tree longevity. During the winter 2018-2019, we completed pruning wound protection and susceptibility trials in experimental orchards to determine best fungicidal products as well as best pruning timing to reduce risks of infection with canker pathogens. Experiments confirmed the superior efficacy of the fungicide Topsin M (Thiophanate-methyl) for which a 2(ee) recommendation was granted on almond for the protection of pruning wound against several canker pathogens. Additionally, Vintec, a biocontrol product formulated with the fungus *Trichoderma atroviride* SC1 provided excellent pruning wound protection thus allowing sustainable control solutions against canker diseases. Vintec is currently in the registration phase in California for use against canker diseases of almond. Additional studies were conducted in orchards to investigate the seasonal susceptibility of pruning wound and duration of pruning wound susceptibility according to the time (month) of pruning. Results of 2018-2019 experiments confirmed that the duration of pruning wounds susceptibility is lowest when pruning is done in January. Overall pruning wound susceptibility declined substantially after 1 to 2 weeks following pruning and continue to decrease overtime. In conclusion, this work indicates that one application of a pruning wound protectant such as Topsin M (conventional fungicide) or Vintec (biological control agent) following late pruning in January can significantly reduce risks of infection of pruning wounds by canker pathogens. Research is continuing in our laboratory to determine best application methods for these products in commercial orchards.

B. Objectives

1. Determine the seasonal susceptibility of pruning wound and duration of pruning wound susceptibility according to the time (month) of pruning. This will help identify low risk periods for pruning of almond trees.
2. Identify best fungicidal products including biocontrol agents for the protection of pruning wounds. This will permit the registration of new fungicides and biocontrol agents that are effective for the protection of pruning wounds in almond.
3. Provide management guidelines and IPM strategies for the control of almond canker diseases.

C. Annual Results and Discussion

Pruning wound susceptibility

Pruning wounds inoculated immediately after pruning in September through January were most susceptible to infection by the various canker pathogens, in comparison to wounds inoculated 1 to 8 weeks after pruning (Figures 1 and 2). Overall, a steady decrease in pruning wound susceptibility was observed over time, with wounds susceptibility declined substantially after 2 weeks for all pruning months (September, October, November, December and January). The duration of pruning wounds susceptibility was lowest (up to 2 weeks) overall in pruning wounds made in January, with the exception of pruning wounds inoculated with *B. dothidea* in Trial 1 (Fresno Co). In several instances, some pathogens were able to infect pruning wounds up to 8 weeks after pruning at the different pruning months. However, the percent of pruning wounds infected after 2 weeks following pruning remained low overall across the experiments. The high susceptibility of pruning wounds at the time of pruning and up to 2 weeks after pruning stresses the need for protecting pruning wounds after pruning. Interestingly, the 2-week susceptibility window of pruning wound overlaps with the 2-week residual activity of most chemical fungicides, suggesting that one application of a pruning wound protectant right after pruning should significantly reduce risks of infection. Factors influencing the natural healing processes of pruning wounds are less known, however it is likely that variations in the duration of pruning wound susceptibility may occur from year to year, depending on environmental conditions such as temperatures and wetness.

Pruning wound protection

Following three years of experiments for the protection of pruning wound using fungicidal products, Topsin M and the *Trichoderma* biocontrol product Vintec continued to provide excellent disease control (80-100% disease control) when applied to fresh pruning immediately after pruning (Figures 3 and 4). Fungicide products such Quilt Xcel, Merivon and Quash provided moderate disease control with efficacy ranging from 40 to 70% disease control. Acrylic paint was not effective as a pruning wound protectant against the various canker pathogens tested (20-40% disease control).

A final trial at KARE investigated the efficacy of Topsin M and Vintec using air blast sprayer applications. Results indicated that Topsin M remained effective at protecting pruning wounds when applied with an air blast system, reducing disease incidence by nearly 70%. However, air blast applications of the *Trichoderma* biocontrol product

Vintec at both high and low rates did not significantly reduce disease incidence (20-25% disease reduction). On the other hand, application of Vintec using a hand-held spray bottle provided up to 80 to 100% disease control. These results suggest that air blast system may not be well adapted for the application of *Trichoderma* products. This may be due to the difference in droplet size produced by air blast system (mist) in comparison to larger droplets produced with hand-sprayed bottles. Ongoing trials in our laboratory are investigating the use of a backpack manual pressure sprayer to protect pruning wounds with a biological control agent.

In conclusion, combined results of the pruning wound protection and susceptibility trials provided strong evidence that successful protection of almond trees from canker diseases can be achieved. Overall the duration of pruning wounds susceptibility was lowest when pruning was done in January and pruning wound susceptibility decreased significantly after two weeks following pruning. This suggests that a single application of a pruning wound protectant such as Topsin M or Vintec right after pruning in January should significantly reduce risks of infection of pruning wounds by canker pathogens. In addition, Topsin M and Vintec provided great protection of almond pruning wounds against a broad spectrum of canker pathogens. These finding are particularly existing as almond canker diseases are caused by multiple, taxonomically unrelated pathogens. Following this research, Topsin M received a 2(ee) label recommendation for use as a pruning wound protection in almond against several canker pathogens. The biological control product Vintec is currently in the registration phase in California for use against canker diseases of almond in California. Field trials are ongoing in our laboratory to optimize application methods and effective delivery of Vintec to pruning wounds. The availability of a biological control product to protect almond trees will help to enhance the sustainability of almond production systems, which align with the current objectives of the Almond Board of California.

D. Outreach Activities

1. Diagnosis and Management of Canker Diseases of Almond, Invited Speaker, Butte/Glenn/Tehama Almond and Walnut Day 2018. Chico, CA, 01/31/2018, 400 Attendees.
2. Diseases of fruit and nut crops, Invited Speaker, 2018 California Plant and Soil Conference, Fresno, CA, 02/07/2018. 80 Attendees.
3. Management of Trunk and Scaffold Canker Diseases of Almond in California, Invited Speaker, 2018 South Valley Nut Show, Tulare, CA, 10/26/2018, 200 Attendees.
4. The Latest on Almond Canker Disease Management. Mid- Valley Nut Conference Modesto, CA, Nov 2018. 150 Attendees.
5. Canker diseases of almond, Invited Speaker, Almond Conference, Sacramento, CA, 12/06/2018, 500 Attendees.
6. Prevention and Management of Canker Diseases in Tree Crops, Invited Speaker, Growers and Pest Control Advisers, Madera, Dec 12, 2018, 60 Attendees.
7. Update on almond canker disease research, Invited Speaker, 2018 Almond Workgroup Meeting, Davis, CA, 12/07/2018, 40 Attendees.
8. Diseases of fruit and nut crops, spring update, Invited Speaker, PCA/Growers Breakfast Meeting, Modesto, CA, 04/03/2019, 60 Attendees.

9. Diagnosis and Management of Almond Canker Diseases, Invited Speaker, 2019 Kern County Almond Day - UCCE Kern County, CA, 04/11/2019, 80 Attendees.
10. Plant Pathology Research Update in the Fruit and Nut crops in California, Invited Speaker, ANR Pest Mgmt. Program Team meeting, Davis, CA, 06/05/2019, 30 Attendees.
11. Limb, Trunk and Root Diseases. 2019 Almond Short Course, Visalia, CA, 11/07/2019, 150 Attendees.
12. Almond Disease Diagnostic. Almond Short Course Field Tour, KARE, Parlier, CA, 11/08/2019, 58 Attendees.

E. Materials and Methods:

Pruning wound susceptibility

Two independent field trials were conducted to determine the month (Sept, Oct, Nov, Dec, Jan) and duration (+0, 1, 2, 3, 5, 8 weeks following pruning) for which pruning wounds are most/less susceptible to infection by canker pathogens. Nonpareil trees in two orchards (Fresno and Colusa Counties) were pruned either in September 2018, October 2018, November 2018, December 2018, or January 2019, respectively. For each pruning time, subsets of branches were inoculated immediately after pruning (0), or after 1, 2, 3, 5 and 8 weeks following pruning. After wetting wounds with a spray bottle application of sterile water to mimic rain, spore suspensions (5×10^3 spores/wound) of canker pathogens *Cytospora sorbicola, Eutypa lata, Neofusicoccum parvum, Botryosphaeria dothidea*, and *Neoscytalidium dimidiatum* were applied separately to individual pruning wounds in all the various treatments. After 4 months, branches were processed in the laboratory to assess percent fungal recovery for each treatment and determine pruning wounds susceptibility according to the time (month) of pruning and timing of infection following pruning. High or low recovery of fungal pathogens corresponded to high or low susceptibility of pruning wound in each treatment, respectively.

Pruning wound protection

Two independent field trials were conducted in experimental almond orchards in Colusa and Kern counties to evaluate the efficacy of Topsin M (FRAC 1), Quash (FRAC 3), Quilt Xcel (FRAC 3/11), Rhyme (FRAC 3), Merivon (FRAC 7/11), acrylic paint and Vintec (*Trichoderma atroviride* SC1). All products were selected based on high performance results in previous year trials as well as to represent different FRAC groups. Vintec is a biological control product. For each trial, 2 to 3 year-old branches were pruned to roughly 6-to-12 inches in length in December 2018. Following pruning, the various fungicidal products were applied to fresh wounds with a hand-held spray bottle until runoff. Twenty-four hours after treatment, wounds were sprayed with sterilized water to imitate rain and inoculated with the respective fungal pathogens *Eutypa lata, Ceratocystis destructans, Cytospora sorbicola, Neofusicoccum parvum* and *Botryosphaeria dothidea* (1,000 spores/wound). Each trial was set up in a randomized complete block design with 10 tree replicates per treatment (treatment = product + fungal pathogen). Treatment branches were collected three months after inoculation and brought back to the laboratory to assess the percent of fungal recovery. Presence (1) or absence (0) of the inoculated fungal species was recorded and averaged for each treatment. Low rates of fungal recovery correlated with high product efficacy. Infection rate was calculated as the percentage of pruning wounds from which the pathogen was recovered, out of the total number of inoculated pruning wounds.

Finally, a last trial in Fresno County was conducted using Topsin M, Vintec and Clonostachys (biocontrol) to test the efficacy of an air blast system (backpack sprayer) to deliver sufficient coverage for effective pruning wound protection. The experiment was conducted at KARE in a young almond orchard following pruning in Dec 2018. Following pruning and product applications, pruning wounds were inoculated with *Eutypa lata, Neofusicoccum parvum* and *Botryosphaeria dothidea*. Data were processed after 3 months.

F. Publications that emerged from this work

1. Peer review publications

   Holland L.A., et al. (2020) A Re-examination of the Etiology of Almond Trunk and Scaffold Canker Diseases in California. *Plant Disease*. (*Accepted*).

   Holland L.A., et al. (2019) Taxonomic revision and multi-locus phylogeny of the North American clade of *Ceratocystis*. *Fun Syst and Evol* 3: 135-156. View this Article

   Lawrence D.P., et al. (2019) Molecular phylogeny of *Cytospora* species associated with canker diseases of fruit and nut crops in California, with the descriptions of ten new species and one new combination. IMA Fungus, 9(2):333-370. View this Article

2. Conference abstracts

   Holland L.A. et al. 2019. Etiology, biology and management of almond trunk and scaffold canker diseases in California. American Phytopathological Society Annual Meeting (Cleveland, OH)

   Trouillas F.T. 2018. The rise of fungal canker and vascular diseases in cultivated and native woody plants: a California case study. International Congress of Plant Pathology, Boston, Massachusetts, USA, July 2018.

   Holland L.A., Nouri M.T., Lawrence DP, Travadon R, and Trouillas F.P. Biological control of canker diseases in the fruit and nut crops. 2019 APS Pacific Division Meeting, Fort Collins, CO, (Jun 24- 27).

3. Broadcast, Print or Electronic

   Early Detection Key to Managing Ceratocystis Canker in Almonds, Other, 06/22/2018, Growing Produce. View this Media

   Pruning wounds can lead to cankers, UC specialist warns, Other, 07/03/2018, Western Farm Press.View this Media

   Canker diseases with Florent Trouillas, Radio Interview, 08/19/2019, UC ANR. View this Media

   Pruning wound protection: How to reduce the impact of canker diseases, 12/04/2019, Sacramento Valley Orchard Source. View this Media

Figures

Figure 1: Trial 1, Fresno County - Percent fungal recovery over an 8-week period in September, October, November, December and January, respectively.

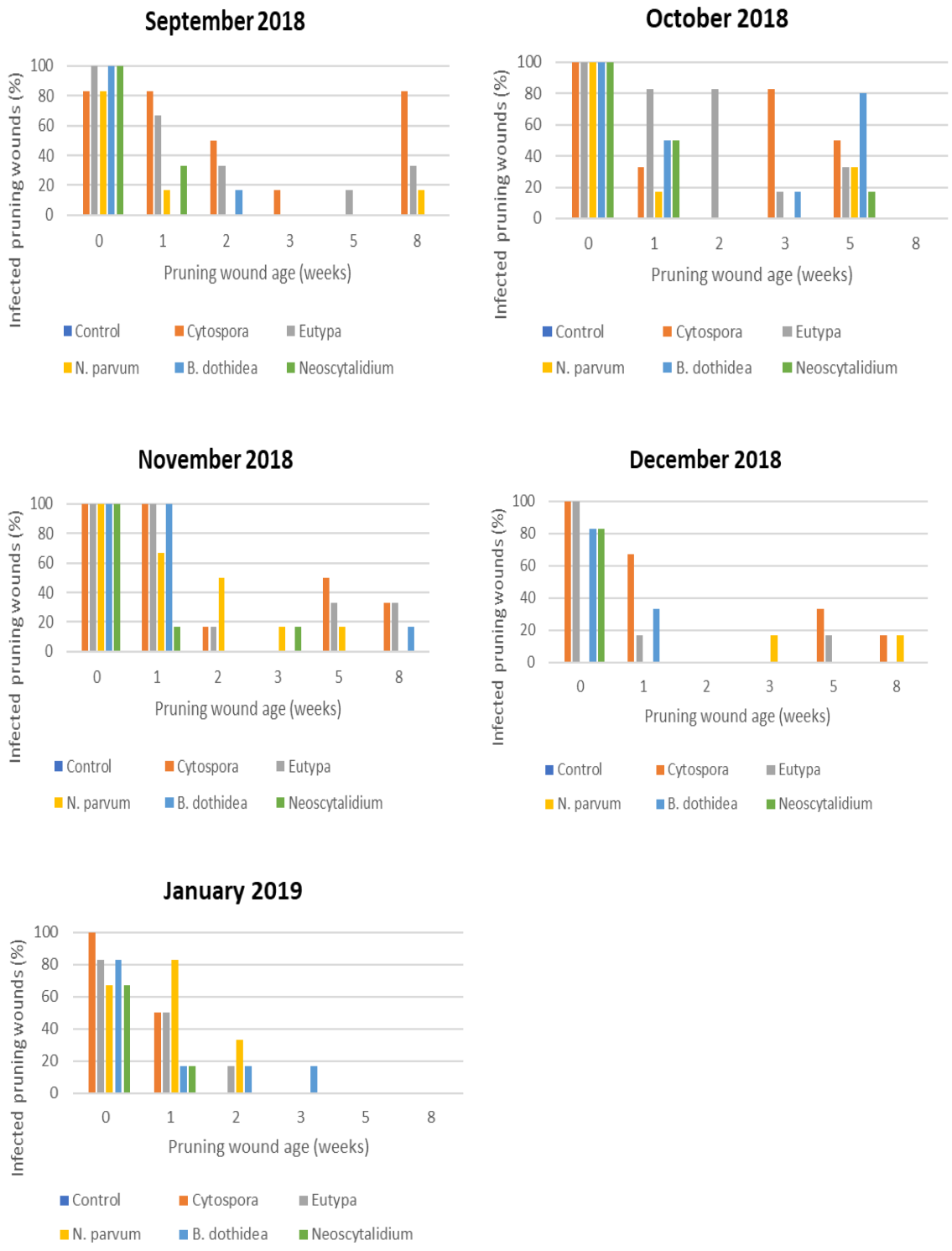

Figure 2: Trial 2, Colusa County - Percent fungal recovery over an 8-week period in September, October, November, December and January, respectively.

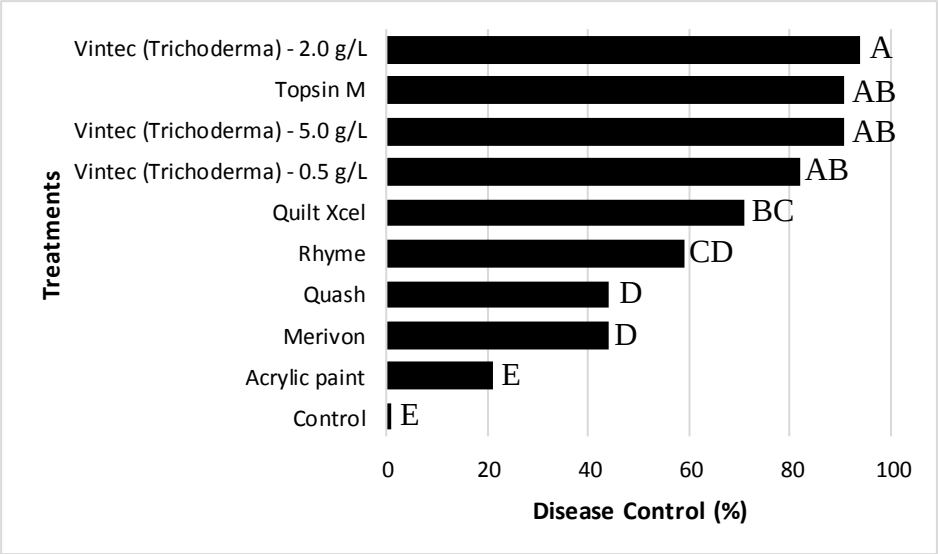

Figure 3: Year 3, trial 1, Colusa County – Percent disease control in pruning wound protection field trials for combined fungal canker pathogens (*Eutypa lata, Ceratocystis destructans, Cytospora sorbicola, Neofusicoccum parvum,* and *Botryosphaeria dothidea*) tested against 10 products.

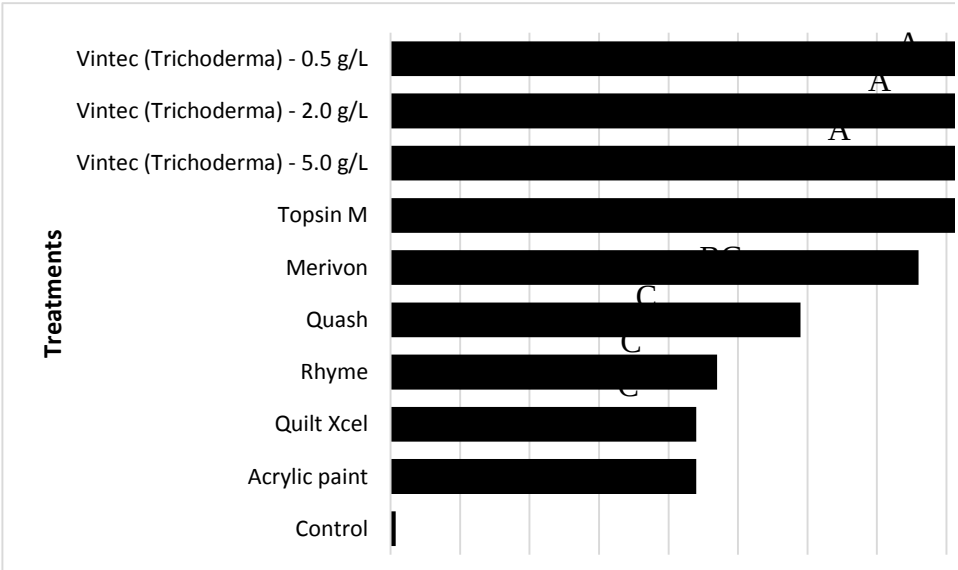

Figure 4: Year 3, trial 2, Kern County - Percent disease control in pruning wound protection field trials for combined fungal canker pathogens (*Eutypa lata, Ceratocystis destructans, Cytospora sorbicola, Neofusicoccum parvum,* and *Botryosphaeria dothidea*) tested against 10 products.