BIOLOGY AND MANAGEMENT OF REPLANT DISORDER AND LETHAL PHYTOPHTHORA CANKER

Project No.: 02-GB-00

Project Leader: G.T. Browne USDA-ARS, Department of Plant Pathology UC Davis

Cooperating Personnel: J.H. Connell (UCCE, Butte Co.), M.Viveros, (UCCE, Kern Co.), H.E. Becherer (USDA-ARS, Davis), , S.T. McLaughlin (USDA-ARS, Davis), P.L. Schrader (UCCE, Kern Co.), S.M. Schneider (USDA-ARS, Parlier), E. Hosoda (Cardinal Professional Products, Woodland), Paramount Farming (Kern Co.), Paiva Farms (Chico).

Background

This research is concerned with two important problems that affect almond production—"replant disorder" (RD) (also known as "replant problem") and "lethal Phytophthora canker" disease (LPC). Replant disorder can occur when stone fruits or nuts are planted without precautions at sites previously devoted to a closely related crop. Symptoms of RD include poor growth, delayed crop production, and, in severe cases, tree death. It is known that parasitic nematodes, oak root fungus, and *Phytophthora* can all interfere with establishment and maintenance of productive almond orchards, but additional poorly understood biological factors contribute strongly to RD, because it occurs in apparent absence of the known pests, and pre-plant fumigation can eliminate or reduce severity of RD. Due to the phase out of methyl bromide, new knowledge and management strategies are needed for RD and are a central focus of our work.

Lethal Phytophthora canker, caused by either *Phytophthora cactorum* or *P. citricola*, affects full-size almond trees and can kill them within 1 to 3 years after infection. It is recognized by profusely gumming cankers that expand rapidly, especially in vertical directions. Vertical growth of LPC cankers is usually much more rapid than that of "mallet-wound" cankers caused by *Ceratocystis*. LPC cankers can persist from year to year until the scion is girdled, unlike pruning wound cankers caused by *P. syringae*, which die out in hot weather. We previously determined that experimental sprays with phosphonate-containing products (such as Nutriphite, Phostrol, and Aliette, for example) afforded preventive and curative management of LPC. In 2001/02, we compared efficacy of foliar phosphonate sprays with that of micro sprinkler applications of the material.

Objectives

1. Improve management strategies for replant disorder (RD) (also known as replant problem) on California almonds.
2. Determine unknown causes of RD on almond.
3. Improve management strategies for lethal Phytophthora canker (LPC).

I. Improving management strategies for RD and determining its unknown causes.

Chico trials. Almond replanting trials were established at Paiva Farms, Chico, in collaboration with Cardinal Professional Products (Woodland, CA) and Tri-Cal, Inc. (Hollister, CA). The experimental site had been devoted to almond for about 20 years before the grower cleared and replanted the block to almond on Marianna 2624 rootstock in spring 2000 without pre-plant fumigation. More than 80% of the grower replants failed to produce commercially acceptable growth in spring and summer 2000. Sampling did not detect significant populations of plant parasitic nematodes at the site. In a 2000/01 replant experiment, a fall pre-plant "broadcast" shank injection with chloropicrin (374 lb/A) prevented the RD, but similar broadcast treatments with Telone II or methyl bromide were not effective (2001 Almond Board Report). Tree site treatments with methyl bromide or Telone II (1 lb/tree site) were more effective than the broadcast treatments.

New experiments were established at Paiva Farms in fall 2001 and monitored in 2002. They were designed to determine the following: relative susceptibility of Lovell peach and Marianna 2624 rootstocks to the RD in the Chico area and relative effectiveness of tree site treatments with methyl bromide-chloropicrin mixture or chloropicrin for RD prevention (Table 1); the possibility that limited graft compatibility with almond is responsible for the high severity RD on Marianna 2624 rootstock, and the potential for avoiding RD by planting "green" potted almond trees with an established fine root system rather than "bare root" trees (Table 2); relative efficacy of pre-plant tree-site treatments with methyl bromide, Telone II, and chloropicrin for preventing RD (Table 3); the effect of cropping history on severity of the RD (Table 4). The required fumigation treatments all were applied on 10/26/01 to tree sites without plastic mulch (one injection per tree site at 45 cm depth; soil moisture 0.18 to 0.25 g of water per gram of dry soil; ambient soil temperature 17 to 20° C). The experimental plots were planted on 2/08/02, and all trials had 6 to 9 replicate multiple- or single-tree plots per treatment, arranged in randomized complete blocks. Vigor and survival of the trees was used to assess treatment effects. Post-plant samples of roots and soil were subjected to fungal, bacterial, and nematode isolations to determine associations between certain soil microbes and plant growth responses to fumigation.

Growth measurements and disease ratings in Experiment 1 indicated that Lovell rootstock is less susceptible than Marianna 2624 to the RD, but performance of trees on both rootstocks suffered without pre-plant fumigation (Table 1). Either fumigation treatment, methyl bromide:chloropicrin mixture (75:25) or chloropicrin (each at 1 lb per tree site), resulted in acceptable growth of most trees (Table 1).

Results of Experiment 2 indicated that susceptibility of Marianna 2624 to the RD does not result from its marginal graft compatibility with almond scions. Marianna 2624 with French prune as a scion (a known compatible combination) also was subject to PRD (Table 2). The trees started from the "green" potted plants were as susceptible as those started from bare root plants (Table 2), indicating that RD can not be avoided just by using the "potted-plant" propagation system.

In Experiment 3, pre-plant fumigation with chloropicrin at 0.5 or 1 lb per tree site or Telone at 1.8 lb per site was as effective as methyl bromide at 1 lb per tree site, but use of chloropicrin at 2 lb per tree site may have been phytotoxic (Table 3).

In Experiment 4, replacement of the tree planting hole soil with soil from an adjacent alfalfa field significantly reduced PRD severity (Table 4), suggesting that cropping history is indeed an important factor in development of RD and that there may be potential for reducing RD severity by growing a rotation cover crop during a fallow year.

Parlier trials. To facilitate additional RD research under San Joaquin Valley conditions and investigate cultural as well as chemical approaches to its management, we established micro plots in spring 2002 at the USDA-ARS farm near Parlier, CA. The micro plots (18 or 24" diameter, depth 4 ft.) were filled in early April 2002 with soil from an adjacent peach orchard expressing symptoms of RD. One set of micro plots holds a crop rotation-fallow test and will be used to determine effects pre-plant fallow, non-fallow (planted to almond on Nemaguard), and cover crop treatments (corn, sudan, wheat, or sudan followed by wheat); the plots will be replanted and evaluated in 2003. Another set of the micro plots involves pre-plant soil fumigation treatments with methyl bromide, chloropicrin, and a non-fumigated control. The fumigants were applied at 400 or 2700 lb per acre of micro plot area to simulate typical field "broadcast" and tree-site rates, respectively. A 4 ft x 4 ft area was assumed for tree sites, which typically receive 1 lb of fumigant during a commercial fumigation involving methyl bromide. The fumigation was completed on 4/30/02 (soil moisture content was 0.07 to 0.12 g per gram of dry soil; ambient soil temperature was approx. 28° C), and the plots were replanted to Nemaguard peach seedlings on 6/3/02. Growth of the plants was used to assess treatment effects, and isolations from soil and root samples are being used to determine microbes associated with the growth responses. Pre- and post-plant sampling of the soil used for the micro plots did not detect significant numbers of nematodes that are recognized as important pests on peach rootstock.

As of August 2002 in the Parlier micro plot trials, pre-plant fumigation with chloropicrin at either the 400 or 2700 lb/ treated acre rate (representing "broadcast" and tree-site rates, respectively) produced strong and equivalent growth responses relative to the control, but methyl bromide was only highly effective at the higher rate (Table 5).

Determining the unknown cause(s) of RD. To help determine unknown cause(s) for RD, roots of diseased trees (in non-fumigated plots) and those of healthy trees (in chloropicrin and/ or methyl bromide fumigated plots) in Chico and Parlier trials were sampled systematically on multiple occasions during the growing season. Both at Chico and Parlier trials, damage of the fine roots as well as a lesser abundance of fine roots are associated with the RD in the non-fumigated plots. The healthy and diseased roots were subjected to isolations on multiple fungal and bacterial isolation media. All isolated fungi are being identified, and representative bacteria were randomly selected from healthy tree and diseased tree samples and preserved for subsequent characterization. Representative fungal isolates are being tested for pathogenicity on peach seedlings in controlled greenhouse experiments, and similar screening is planned for bacterial isolates. To date, *Fusarium* and *Cylindrocarpon* fungi have shown some association with occurrence of RD symptoms on roots, and pathogenicity tests are underway to clarify their role in the problem.

Practical implications of Chico and Parlier trials. The commercial orchard (Chico) and micro plot trials (Parlier) both indicate that chloropicrin is a promising alternative to methyl bromide for control of almond RD, at least in absence of recognized nematode pests and virulent fungal pathogens. There may be a risk of phytotoxicity due to chloropicrin if the fumigant rate exceeds 0.5 to 1 lb per tree site or if sufficient dissipation time under appropriate conditions is not allowed. Results of the rootstock comparison and soil amendment tests at Chico suggest potential for RD management through rootstock selection and pre-plant cropping practices.

2002/03 RD trials. Trials have been initiated in the Chico area with growers Anthony Martinez and Jim Mead to further evaluate methyl bromide alternatives of chloropicrin, Telone II, Telone C35, iodomethane, and iodomethane:chloropicrin and confirm relative RD susceptibility of almond rootstocks Lovell peach, Nemaguard, and Marianna 2624. The Parlier micro plot studies are continuing and will be repeated to provide data on effects of cover crop rotations and alternative fumigants for managing RD. Both the Chico and Parlier trials are designed to facilitate lab and greenhouse determinations of underlying causes of RD.

II. Improving control strategies for lethal Phytophthora canker (LPC).

Relative effectiveness phosphonate applied by chemigation vs. by foliar spraying. In October 2001 a trial was established with Paramount Farming in Kern County to compare effectiveness of phosphonate treatment by foliar spraying vs. by chemigation for management of LPC (Fig. 1 A). On 10/19/01, the experimental treatments were applied to full-size almond trees and included 1) a foliar spray with Phostrol (Nufarm Americas, Inc., not yet registered for almond) (3.3 pints of formulation per acre in 150 gallons of water per acre), 2) chemigation with Phostrol through micro sprinklers (3.3 pints of formulation per acre injected 3 to 4 hr before completion of an 18-hr irrigation set that applied 1.2" water), and 3) a water control (150 gallons per acre of foliar water spray and 1.2" of irrigation water, both without Phostrol). To avoid confounding Phostrol treatments with water application, the trees that received the foliar Phostrol spray also received 1.2" of irrigation water, and trees given the Phostrol by chemigation received a foliar water spray (150 gal/A). There were four replicate 15-tree main plots per Phostrol and control treatment, arranged in randomized complete blocks. Single trees within each main plot were randomly allocated to each of nine subplot treatments, which included the possible combinations of three inoculants (*P. cactorum*, *P. citricola*, or a control) and three inoculation dates (11/16/01, 2/15/02, or 4/1/02). Phostrol treatment efficacy was assessed according to the length and width of bark cankers resulting after inoculation with the pathogens; incubation periods of 90, 45, and 45 days were allowed after the inoculations on 11/16/01, 2/15/02, and 4/1/02, respectively, before the cankers were measured.

The foliar spray/chemigation experiment was repeated on previously untreated trees in spring 2002 (Fig. 1 B). The treatments were applied on 4/24/02, and the experimental design and procedures were generally the same as for the fall trial, except that in the spring test the Phostrol application rate was 3.8 pints per acre for both foliar and micro sprinklers, the foliar spray was applied in 175 gallons of water per acre, the micro sprinkler chemigation occurred with 1.6" water, and inoculations with the pathogens occurred on 15 May, 18 June, and 22 July.

In the fall test, the pre-inoculation foliar Phostrol spray treatment significantly suppressed cankers caused by *P. cactorum* and *P. citricola*, but the Phostrol chemigation treatment had no significant effect (Fig 1 A). Depending on the inoculation/ incubation interval, cankers on trees inoculated with *P. cactorum* averaged 57 to 43% smaller (based on canker lengths) in Phostrol-sprayed plots than in the water control plots. Similarly, cankers on trees inoculated with *P. citricola* averaged 86 to 59% smaller in Phostrol-sprayed plots than in control plots.

Longer cankers developed in the spring test than in the fall test, even though shorter incubation periods were used in the fall. This was probably primarily the result of warmer temperatures in the latter test. The spring foliar spray with Phostrol suppressed subsequent canker development, but only by 13 to 35 % for *P. cactorum* and 7 to 51% for *P. citricola* (depending on incubation period, compared to canker lengths without Phostrol) (Fig. 1 B). In contrast to results in the fall test, chemigation with Phostrol resulted in some canker suppression in the spring test (17 to 31%, Fig. 1 A,B).

The degree of canker suppression following the fall and spring Phostrol sprays was not as dramatic in 2002 as we reported following previous years' tests with Nutriphite (2000-01 Almond Board reports). Although many factors could be responsible for this the effect, it is considered unlikely that either product has much greater or lesser chemical effectiveness per amount of phosphonate active ingredient; less phosphonate active ingredient was applied in the Phostrol tests than in the previous experiments involving Nutriphite formulations. It should be considered that our wound inoculations present a severe test to the preventive treatments, and that repetitive phosphonate sprays or chemigations, when allowed by product labels, would likely be more effective than the single applications that we used for experimental reasons.

It is encouraging that the spring Phostrol chemigation caused some canker suppression, because the method saves application cost compared to foliar spraying. Chemigation may work better in the spring than in the fall chemigation because of less water uptake and translocation of the phosphonate during tree dormancy compared to that during the growing season. Over all of our tests, the strongest canker suppression has occurred following a fall foliar spray with a full rate of phosphonate, but our 2002 results suggest that spring chemigation with a phosphonate fungicide may have merit as a follow-up to the fall foliar spray in a fall/spring LPC prevention program.

Table 1. Effects of rootstock and pre-plant fumigation treatments on severity of almond replant disorder, Chico

Rootstock	Fumigation treatment (rate per tree site)[a]	Increase in trunk diameter (mm)[b]	Final tree height (m)[c]	Final disease severity rating[d]	Acceptable trees (%)[e]
Lovell peach	Control	7	1.7	1.7	67
	MB:Pic 75:25 (1 lb)	15	2.2	0.3	94
	Pic (1 lb)	17	2.3	0.0	100
Marianna 2624	Control	4	1.2	2.9	6
	MB:Pic 75:25 (1 lb)	15	2.0	0.4	94
	Pic (1 lb)	14	1.9	0.6	83
LSD from 95% confidence intervals:		4	0.3	1.1	--

[a]MB:Pic= methyl bromide / chloropicrin mixture; Pic= chloropicrin; one probe per tree site at 30 to 45 cm depth
[b]From 2/27/02 to 10/11/02
[c]On 10/11/02
[d]On 10/11/02; on a scale where 0 = healthy tree with good growth, 5= dead tree and 1, 2, 3, and 4 were intermediate gradations
[e]Only trees taller than 1.5 m (5.5 ft) on 10/11/02 were considered acceptable.

Table 2. Effects of scion/ rootstock combination and pre-plant fumigation treatments on severity of almond replant disorder, Chico

Scion/ rootstock combination	Fumigation treatment (rate per tree site)[a]	Increase in trunk diameter (mm)[b]	Final tree height (m)[c]	Final disease severity rating[d]	Acceptable trees (%)[e]
Carmel almond / Mar. 2624 (bare root)	Control	0	1.0	4.3	0
	MB:Pic 75:25 (1 lb)	14	1.9	0.7	100
Carmel almond / Mar. 2624 (potted)	Control	3	1.0	3.8	0
	MB:Pic 75:25 (1 lb)	13	1.8	1.0	67
French prune / Mar. 2624 (bare root)	Control	3	1.2	2.8	17
	MB:Pic 75:25 (1 lb)	13	2.1	0.2	100
LSD from 95% confidence intervals:		6	0.4	1.6	--

[a]MB:Pic= methyl bromide / chloropicrin mixture; one probe per tree site at 30 to 45 cm depth
[b]From 2/27/02 to 10/11/02
[c]On 10/11/02
[d]On 10/11/02; on a scale where 0 = healthy tree with good growth, 5= dead tree and 1, 2, 3, and 4 were intermediate gradations
[e]Only trees taller than 1.5 m (5.5 ft) on 10/11/02 were considered acceptable.

Table 3. Effect of tree-site fumigation treatments on severity of replant disorder, Chico

Fumigant (and rate per tree site)	Increase in trunk diameter (mm)[b]	Final tree height (m)[c]	Final disease severity rating[d]	Acceptable trees (%)[e]
Control	8	1.4	2.0	37
Methyl bromide (1 lb)	12	1.8	1.0	81
Telone (1.8 lb)	12	1.8	1.0	81
Chloropicrin (0.5 lb)	17	2.0	0.4	89
Chloropicrin (1 lb)	17	2.0	0.5	93
Chloropicrin (2 lb)	12	1.7	2.0	63
LSD from 95% confidence intervals:	6	0.4	1.4	--

[b]From 2/27/02 to 10/11/02
[c]On 10/11/02
[d]On 10/11/02; on a scale where 0 = healthy tree with good growth, 5= dead tree and 1, 2, 3, and 4 were intermediate gradations
[e]Only trees taller than 1.5 m (5.5 ft) on 10/11/02 were considered acceptable.

Table 4. Effect of history of planting hole soil on severity of replant disorder, Chico

Soil used to refill planting hole	Increase in trunk diameter (mm)[b]	Final tree height (m)[c]	Final disease severity rating[d]	Acceptable trees (%)[e]
Resident soil from hole	2.3	1.0	3.9	0
Soil from adjacent alfalfa field	7.9	1.4	2.2	40
LSD from 95% confidence intervals:	8.9	0.5	1.1	--

[b]From 2/27/02 to 10/11/02
[c]On 10/11/02
[d]On 10/11/02; on a scale where 0 = healthy tree with good growth, 5= dead tree and 1, 2, 3, and 4 were intermediate gradations
[e]Only trees taller than 1.5 m (5.5 ft) on 10/11/02 were considered acceptable.

Table 5. Nemaguard peach performance in the Parlier micro plots as of 8/14/02[a]

Date of plant sampling	Pre-plant fumigant	Fumigant rate[b]	Shoot height (cm)	Shoot weight (g)	Root weight (g)[c]	Root rot (%)[c]
8/14/02	None (control)	None	32	8	3	46
	Methyl bromide	Low	41	13	5	42
		High	66	60	13	13
	Chloropicrin	Low	64	53	14	18
		High	64	56	13	11
9/24/02	None (control)	None	31	11	--	--
	Methyl bromide	Low	49	62	--	--
		High	92	218	--	--
	Chloropicrin	Low	75	166	--	--
		High	92	225	--	--
LSD from 95% confidence intervals:			26	87	10	30

[a]Data for each date of sampling from four replicate micro plots per treatment. Each micro plot was 18" in diameter and 4 ft deep.
[b]For both methyl bromide and chloropicrin, the low rate was 400 lb per acre (micro plot area basis) and represented a "broadcast" treatment, and the high rate high rate was 2700 lb per acre (micro plot area basis) and represented a typical concentrated dose delivered by a "tree site treatment" when 1 lb of fumigant is delivered to a 4-ft. x 4-ft. tree site area.
[c]"--" indicates data points that were not taken

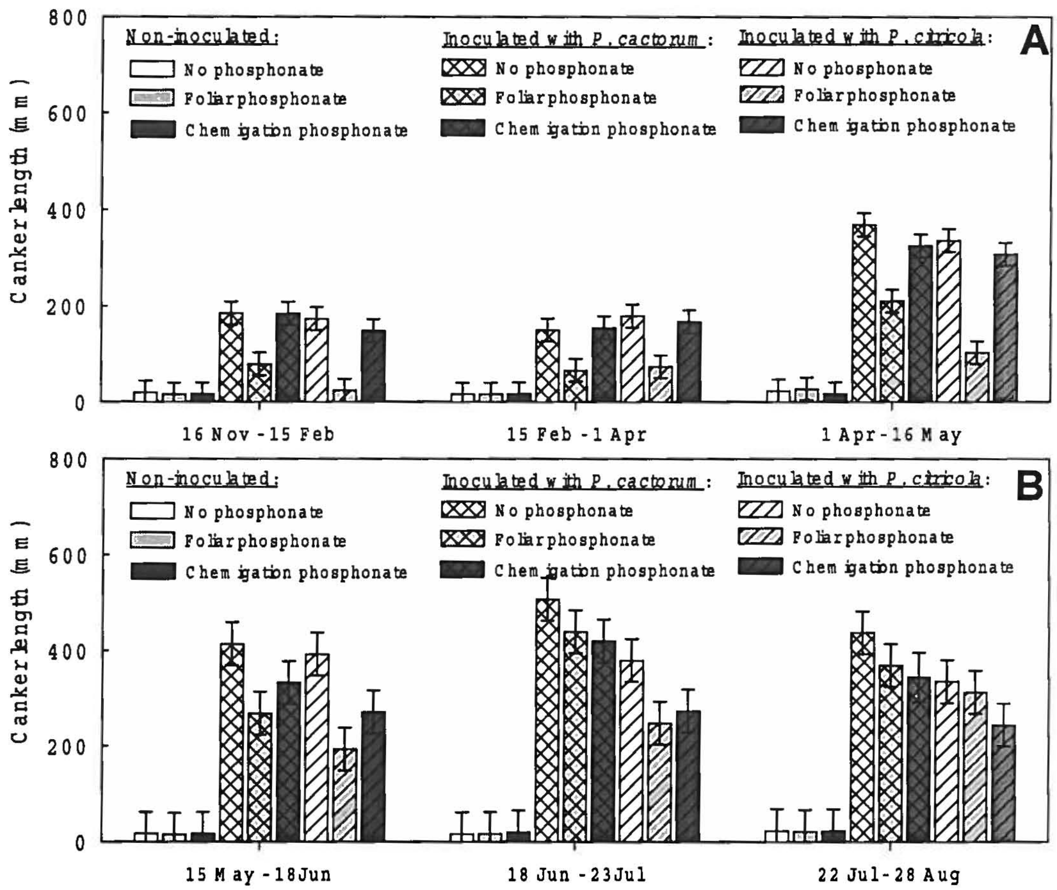

Figure 1. Relative efficacy of foliar and micro sprinkler applications of phosphonate for control of lethal Phytophthora cankers on commercial almond trees **A**, in a fall 2001-spring 2002 experiment and **B**, in a spring/summer 2002 experiment. Phostrol (an experimental phosphonate, Nufarm Americas Inc.) was applied **A**, at 3.3 pints/acre on 10/19/01, or **B**, at 3.8 pints/acre on 4/24/02 either by a complete foliar spray (in 150-175 gal water per acre depending on date, conventional air blast) or by chemigation through micro sprinklers (in 1.2" to 1.6"water, depending on date). Treatment effectiveness was determined by inoculating groups of trees with *Phytophthora cactorum* or *P. citricola* at intervals of time after the phosphonate treatments. Control trees received the same amounts of water by spray and irrigation, but without Phostrol. Date ranges on the X-axis start with the date of inoculation and end with the date of canker measurement for the test intervals. Note that the Phostrol foliar spray suppressed canker development whereas the Phostrol chemigation did not. The experiment is being repeated in spring/summer 2002.