ANNUAL REPORT 1992 Almond Board of California

Project No. 92-T18 -	Control of Almond Diseases
Project Leader:	Dr. James E. Adaskaveg University of California Department of Plant Pathology Davis, CA 95616
Cooperating Personnel:	Dr. J.M. Ogawa, K. Conn, L. Wade, and P. Rener, Dept. of Plant Pathology, Univ. of CaliforniaJoe Connell, Farm Advisor, Coop. Extension, Univ. of California

SUMMARY

Studies were conducted to investigate brown rot blossom blight, shot hole, scab, and anthracnose of almond. In brown rot blossom blight studies using our mist generating system (MGEMS) or bagging techniques, wetness periods of 4, 8, 12, or 16 hrs duration increased brown rot blossom blight from 6% to 16.5% (MGEMS) and from 19% to 36% (bagging method). Natural wetness periods from rainfall (0.86 in) occurred during the field incubation period. These additional, non-continuous wetness periods totaled 8 hr (day 6, 7, and 8) and affected all treatments. During incubation, ambient high temperatures ranged from 63-70 F, while low temperatures ranged from 41-48 F. The almond bloom coincided with the conducive environment for disease and this resulted in a high natural incidence of brown rot. Almond varieties were evaluated and are listed from highly susceptible to somewhat resistant: Drake, Carmel, Mission, and Nonpareil. Furthermore, we identified new genotypes (bud sports) that were highly resistant to brown rot. This information is being utilized towards developing new strategies for brown rot blossom blight control using fewer applications of fungicides. Additionally, in laboratory and field tests, Rovral and Rovral-oil mixtures provided excellent control of brown rot blossom blight. Other fungicides evaluated in the field included Funginex 1.6EC (triforine), Ronilan 50 DF (vinclozolin), and a new WG formulation of iprodione. The incidence of brown rot strikes was significantly reduced with the use of these fungicides compared to nontreated trees and provided control similar to Rovral 50WP treatments.

In the 1992 season, we verified our shot hole forecasting system-reduced spray program at two sites: one commercial orchard in Merced and one experimental orchard at UC Davis. In the fall of 1991, early natural leaf fall occurred by the first week of November and a zinc sulfate application to defoliate trees was unnecessary. Thus, sporodochia of <u>Wilsonomyces</u> <u>carpophilus</u> in late November and December were infrequent and the potential for disease was considered low (low risk) for the spring of 1992. In the Merced orchard, only trace amounts of rainfall were recorded in March when leaf emergence occurred. No differences were observed between recommended (petal fall-leaf emergence and 5-wk after petal fall) and timing (incidence of sporodochia) treatments when leaves were evaluated through mid-April. Since no sporodochia were produced, only one spray application of ziram was needed (5-wk after petal fall). In the Solano Co. orchard, however, rainfall occurred during leaf emergence and by March 15, sporodochia developed. A recommended ziram spray at petal fall (March 8) was compared to a timing spray of ziram after sporodochia formation. No differences were observed between spray treatments; while both treatments provided control when compared to the nontreated trees (check). Thus, shot hole was controlled with the timing spray but, in this plot, the total number of ziram sprays was the same. Benefits from this program include spray applications only when they are needed and reduction of spray applications would occur over a several year period.

In studies on scab control, the efficacy of dormant applications of liquid lime sulfur (LLS) and summer applications of captan and wettable sulfur were evaluated. Delayed dormant (early February), air-blast spray applications of LLS were made on Carmel and NePlus Ultra almond in Merced County and on Butte and Carmel almond in Butte County. In all test plots, LLS treatments suppressed lesion expansion and sporulation of the fungus on shoots. In summer evaluations of leaves and fruit in all plots, disease incidence was consistently lower, while disease severity was significantly lower in the LLS treatments than in check (no dormant spray) treatments. In the spring of 1992, ICI Chemical Corporation made a label change allowing the use of captan up to 30 days before harvest while still allowing treated hulls to be fed to livestock. Applications may be made up to 12 days before harvest provided hulls are not fed Our project confirmed this label change with California to dairy animals or livestock. Department of Pesticide Regulation and Pesticide Enforcement. In the first Merced plot, percent infection of fruit was reduced and disease severity of fruit was significantly reduced by the application of captan and sulfur when compared to the nontreated trees; in the second Merced plot, only disease severity of fruit was significantly reduced. In both Merced plots, no significant differences between treatments were observed on leaves but disease incidence and severity were consistently lower in the spray treatments. In the Butte plot, disease incidence and severity on fruit were consistently reduced when compared to the check. On Carmel leaves, no differences were observed between spray treatments; while on Butte leaves, disease incidence and severity were significantly reduced with sulfur or captan treatments.

During our studies of shot hole and scab in Merced county, we observed a new occurrence of anthracnose of almond caused by a species of <u>Colletotrichum</u>. Anthracnose was last reported in the early 1970's, but has not been documented in California since the 1930's. The disease was first observed 2-3 wk after petal fall and occurs primarily on the fruit. Infected fruit shrivel, become light rusty orange, and appear similar to almond blanks. Leaves attached to fruit spurs often wilt and remain attached (similar to leaf blight). Laboratory and field inoculation studies demonstrated pathogenicity of the <u>Colletotrichum</u> species. Monitoring the incidence of anthracnose and development of control strategies should be initiated in 1993.

OBJECTIVES

 Brown Rot: To determine wetness periods and temperature effects on the development of brown rot infections using the mist generator and environmental monitoring system (MGEMS) and bagging techniques. Additionally, fungicide applications were made after infection periods to determine their efficacy in disease suppression.

- 2) Shot hole: To continue to verify the shot hole forecasting system-reduced spray program in two test plots and to evaluate new formulations of Rovral for shot hole control.
- 3) Scab: To evaluate the effect of liquid lime sulfur, ziram, and sulfur treatments for the control of scab.
- 4) **Rust and Scab:** To determine the effect of early and mid-season defoliation on yield and quality of an almond crop (simulating early season scab or rust defoliation).
- 5) Supplemental: Identification of anthracnose of almond, a new occurrence in California.

MATERIALS AND METHODS

BROWN ROT BLOSSOM BLIGHT STUDIES

Influence of Wetness Periods on Incidence of Brown Rot Blossom Blight. To determine the effects of wetness period and temperature on the incidence of brown rot blossom blight, newly opened blossoms (anthers exposed) of Drake almonds in a field test plot at UC Davis were inoculated (25,000 conidia/ml) and exposed to wetness periods of 4, 8, 12, or 16 hrs duration using the mist generating system (MGEMS) or bagging techniques. In studies using MGEMS, a datalogger (Campbell 21X, Logan Utah) was programmed to mist blossoms with water for 3 sec when leaf wetness sensors were below an arbitrary critical value of 3 on a scale of 0 (dry) to 10 (wet). Misting lines and water application were as described by Adaskaveg et al. (1990). In studies using bagging methods, brown paper bags lined with plastic bags were placed over inoculated blossoms. A wet paper towel was wrapped around the branch (approximately 30 cm from the branch end) and the bags were tied with wire to enclose the paper towel and blossoms. For both misting and bagging methods, each wetness period was replicated on four single branches of four different trees and the experiment was repeated once. Blossom blight with sporulation was recorded for 2 weeks following wetness periods treatments. Data were analyzed using analysis of variance, linear regression, and general linear model procedures of the statistical package SAS 6.04.

Comparison of Blossom Blight Susceptibility among Almond Varieties. The almond bloom coincided with a conducive environment for disease and resulted in a high natural incidence of brown rot. The following varieties were evaluated: Drake, Carmel, Mission, and Nonpareil, as well as new genetically distinct genotypes. For this, percent shoot infection (infected shoots/200 shoots) was determined for twelve trees of each variety three to four weeks after bloom. Data were analyzed using analysis of variance and least significant difference (LSD) mean separation procedures of SAS.

Evaluation of Fungicides for Brown Rot Control. Fungicides were hand-gun sprayed (400 gals/A; 400 psi) on almonds at pink bud and full bloom and included: Funginex 1.6EC (triforine; 12 oz/A), Ronilan 50 DF (vinclozolin; 1 lb/A), Rovral 50WP (iprodione; 1 lb/A), Rovral 4F, and a new WG formulation of Rovral (RP10259A). Ziram 76WDG (6 lbs/A) was applied at full bloom and at petal fall and all treatments had six single tree replications. In laboratory studies, Rovral 50WP (1 lb/A) or Rovral (1 lb/A)-Omni Oil (1 gal/A) mixtures were

evaluated for control of brown rot anther infection of almond. For this, Drake blossoms were removed from branches, placed in sand (water-saturated) in a plastic container, treated with fungicides, air-dried, spray-inoculated with conidia of <u>M</u>. <u>laxa</u> (40,000 conidia/ml), and incubated for 5 days at 25C. Additionally, apricots (1 gal Omni oil/A) and almonds (4 gal Omni oil/A) were air-blast sprayed (100 gal/A) at full bloom with Rovral or Rovral-oil as described previously. Treatments consisted of four single-tree replications of apricots or six single-tree replications of almonds. Blossom blight was evaluated after 3-4 wk and data was statistically analyzed using SAS as described previously.

SHOT HOLE CONTROL

Evaluation of Fungicides. As described in the previous section, fungicides bloom sprays were evaluated for shot hole control of almond. Disease incidence (number of infected/total leaves counted), disease severity (average number of lesions/leaf), and mean number of sporodochia were evaluated for each of the treatments. Data were analyzed using analysis of variance and LSD mean separation treatments of SAS.

Confirmation of Shot Hole Forecasting. In the 1992 season, we verified our shot hole forecasting system-reduced spray program in one commercial orchard in Merced and one experimental orchard at UC Davis. In the fall of 1991, early natural leaf fall occurred in the commercial and experimental almond orchards by the first week of November and a zinc sulfate application to defoliate trees was unnecessary. Numbers of sporodochia on leaves in November of 1991 and in March of 1992 were recorded for the two orchards. In the spring of 1992, treatments of ziram (8 lbs/A) were applied with an air-blast sprayer (100 gal/A) in the commercial orchard or applied with a hand sprayer (400 gal/A; 400 psi) in the experimental orchard. Timing of application was based on petal fall-leaf emergence (current recommended practice) or on detection of sporodochia on leaves (proposed timing practice) collected 7-10 days apart through 4-wks after petal fall from 12 non-sprayed trees in each orchard. For both orchards, incidence and severity of shot hole was determined for nontreated, recommended, and timing treatments in the last week of March and April. Data were analyzed using analysis of variance and LSD mean separation procedures of SAS.

SCAB CONTROL

Evaluation of Fungicide Spray Programs for Control of Scab. In studies to evaluate the efficacy of dormant applications of liquid lime sulfur (LLS) and summer applications of captan and wettable sulfur, three test plots were established in commercial orchards in two counties (one in Butte and two in Merced). These plots were selected because trees had severe perennial, stem infections of scab. Delayed dormant (early February), air-blast spray applications of LLS (8 lbs/100 gal) were made on Carmel and NePlus Ultra almond in plots in Merced County, and on Butte and Carmel almond in the Butte County plot. In addition to the dormant spray, summer applications of captan and sulfur were compared to nontreated trees in all three plots (one in Butte, two in Merced). Spray applications of captan and sulfur were made once in mid-June and once in mid-July. Evaluations were also made in mid-July and mid-August. Air-blast spray programs were: 1) Merced plot No. 1: Benlate 50WG (benomyl) - 1 lb/A; 2) Merced plot No. 2: Rovral 50WP (iprodione) - 1 lb/A; and 3) Butte plot: a 1/2 spray application of Topsin 50WP (benomyl) - 1 lb/A, 1/2 spray application of Rovral 50WP - 1 lb/A. Additionally, in only the latter plot, ziram was applied at petal fall (1/2 spray), 2-wk after petal fall (1/2 spray), and 5-wk after petal fall (full spray). Evaluations were made of sporulation of the fungus on perennial infections (May), disease incidence, and disease severity on fruits and leaves (July and August). Experimental design was a split plot with the main effect the dormant liquid lime sulfur treatment and the sub-plots the wettable sulfur and captan treatments. Data were analyzed using analysis of variance and LSD mean separation procedures of SAS.

SIMULATION OF RUST AND SCAB DEFOLIATION

Effect of Early Season Defoliation of Almond. Studies to determine the effect of early and mid-season defoliation on yield and quality of an almond crop. These investigations were made to simulate early season scab or rust defoliation on NePlus Ultra almond in Merced County. Two and three applications of zinc sulfate (30 lb/A) were made using a handgun sprayer (400 gal/A, 400 psi) in the last two months before harvest. The concentrations used were based on tests conducted in November of two previous years for early defoliation of almond trees to control shot hole (see 1990, 1991 Almond Reports).

ANTHRACNOSE OF ALMOND

Disease Etiology. During our studies of shot hole and scab in Merced County, we observed a new occurrence of anthracnose of almond caused by a species of Colletotrichum. Samples were collected from diseased trees throughout the orchard and almond varieties were recorded. Isolation of the pathogen was done using standard procedures. For this, diseased tissue was excised from the margins of lesions occurring on fruit. The tissue was surface sterilized in 400 µg/ml Cl (dilution of 5% NaOCl) for 1 to 2 min, washed in autoclaved distilled water, blotted dry, and placed on potato dextrose agar (PDA). The fungus isolated was transferred to PDA and maintained in axenic culture at 0-2 C (32-33 F). Optimal temperature for growth was determined by measuring daily mycelial growth on PDA at 10, 15, 20, 25, 30, 35, and 40 C. To determine pathogenicity of the fungus isolated, Drake almond fruit were either wounded and inoculated with 25 μ l of a conidial suspension (13 X 10⁵ conidia/ml) or were non-wounded and spray inoculated with the same conidial suspension of isolate CG92-10. Fruit were then incubated for 7 days at 20 C, >95% RH. In field studies conducted in April 1992, fruit were either wounded or non-wounded and inoculated as described previously with either a 2 X 10⁵ or a 13 X 10⁵ conidia/ml suspension of isolate CG92-10. Fruit and shoots were covered with a plastic and brown paper bag for 16 hr as described in the brown rot section. After 14 days under ambient conditions, symptoms and disease incidence were recorded.

RESULTS AND DISCUSSION

BROWN ROT BLOSSOM BLIGHT AND SHOOT INFECTION

Susceptibility of Almond Varieties to Brown Rot Blossom Blight. Over two inches of rain occurred in the Davis area during the last week of February and the first week of March (Fig. 1). Previously, varietal differences in susceptibility to brown rot have been noted, however, comparisons of cultivars and disease incidence have not been well documented. The

following varieties were evaluated and are listed from highly susceptible to somewhat resistant: Drake, Carmel, Mission, and Nonpareil (Fig. 2). Additionally in this survey, we identified new genetic material (bud sports) that were highly resistant to brown rot. Ideally, the use of resistant cultivars would be the best strategy for brown rot control. Studies to evaluate potential brown rot-resistant genotypes of almond need to be continued in 1993.

Effect of Wetness Periods on Incidence and Severity of Brown Rot Blossom Blight. During incubation, ambient high temperatures ranged from 63-70 F, while low temperatures ranged from 41-48 F. Natural wetness periods from rainfall (0.86 in) occurred during the field incubation period (Fig. 1). These additional, non-continuous rains totaled 8 hr (day 6, 7, and 8) and affected all treatments. With increasing wetness periods, brown rot blossom blight increased from 6% to 16.5% (MGEMS) and from 19% to 36% (bagging method). The regressions of incidence of disease on wetness period were significant and linear for bagging (R^2 = 0.78) and MGEMS (R^2 = 0.93) methods (Fig. 3). Higher incidence was observed in the bagging study due to the static wetness period as opposed to the cyclic wetness periods generated by MGEMS. This study indicated the potential incidence of brown rot blossom blight for inoculated blossoms and selected wetness periods under field conditions.

Efficacy of Fungicide Treatments. The incidence of brown rot strikes was significantly reduced with the use of Funginex, Rovral 50WP, Rovral 50WG, and Rovral 4SC when compared to nontreated trees or trees with ziram (Fig. 4). In the laboratory, Rovral and Rovral-oil mixtures provided excellent control of brown rot anther infection of almond blossoms and were significantly better than the control (Fig. 5). In field trials on apricots, brown rot blossom blight (Fig. 6) and infected spurs (Fig. 7) were significantly reduced with Rovral-oil mixtures than with just Rovral alone, whereas both of these treatments were significantly better than the nontreated check trees. No injury was observed from the oil treatment (1 gal/A) on apricot. On almonds, brown rot blossom blight was low and no differences were detected between treatments. The higher rate of oil (4 gal/A), however, caused some leaf injury (marginal necrosis).

SHOT HOLE LEAF INFECTION AND CONTROL

Efficacy of Fungicides for Control of Shot Hole. Incidence and severity of shot hole was significantly decreased with petal fall applications of iprodione (Rovral) or ziram, whereas triforine (Funginex 1991 formulation) did not significantly reduce the disease (Fig. 8). The three formulations of iprodione (WP, SC, or WG) used were equivalent in their performance. Sporodochial formation was not effected by any of the fungicide treatments.

Effectiveness of Shot Hole Forecasting. In both the commercial and experimental almond orchards, early leaf fall resulted in low to no production of sporodochia of <u>Wilsonomyces carpophilus</u> in late November and December. Thus, the potential for disease was considered low (low risk) for the spring of 1992. In the experimental orchard (Solano Co.), defoliation with zinc sulfate in the fall did not affect disease incidence (P > 0.31) or severity (P > 0.15) in the spring since sporodochia were not observed on leaves in the fall of 1991. In the spring of 1992, rainfall occurred during leaf emergence and sporodochia developed by March 15 (Fig. 1). A recommended ziram spray at petal fall (March 8) was compared to a timing

spray of ziram (March 17) after sporodochia formation. No differences were observed between spray treatments; while both treatments (P < 0.01) were significantly lower than the nontreated trees (Fig. 9). Thus, shot hole was controlled with the timing spray but, in this plot, the total number of ziram sprays was the same as the recommended treatment.

In the commercial orchard, one ziram spray was applied at petal fall (March 9) as a standard recommended treatment. The timing treatment based on the production of sporodochia of the shot hole fungus was not applied since sporodochia were not found during any of the three collection periods in March. Only trace amounts of rainfall were recorded for the first three weeks of March when leaf emergence occurred (Fig. 10). For the dates evaluated, no differences were observed between recommended and timing treatments when leaves were evaluated (Fig. 11). Disease was detectable for all treatments but the incidence and severity did not cause leaf drop. Thus, one less spray application (ziram) was needed. In 1992 and in previous years, a five-wk-after petal fall spray of ziram was applied to both the timing and recommended treatments; whereas the check was not sprayed. The higher incidence of disease in the nontreated trees (checks) indicates an increase in the resident population of the fungus over a several year period when the disease was not controlled. Regardless of the differences in disease occurred between the timing and recommended treatments when sporodochia were not observed in March and April.

Benefits from this program include spray applications only when they are needed. The total number of fungicide sprays needed for control of shot hole were decreased in all study orchards over a several year period. For additional details of our shot hole forecasting system, please see our 1990 and 1991 Annual Reports to the Almond Board of California.

CONTROL OF SCAB - SHOOT AND LEAF INFECTION

Efficacy of Fungicides for Scab Control. In all test plots, liquid lime sulfur treatments suppressed lesion expansion and sporulation of the fungus on shoots. Sporulation on perennial twig lesions was decreased in LLS treated trees but not in non-treated trees. In summer evaluations of leaves and fruit in all plots, disease incidence was consistently lower, while disease severity was significantly lower ($\underline{P} < 0.05$) in the LLS treatments than in no-dormant-spray (check) treatments (Figs. 12-15).

In the spring of 1992, ICI Chemical Corporation made a label change allowing the use of captan up to 30 days before harvest while still allowing treated hulls to be fed to livestock. Applications may be made up to 12 days before harvest provided hulls are not fed to dairy animals or livestock. Our project confirmed this label change with California Department of Pesticide Regulation and Pesticide Enforcement. In the first Merced plot, percent infection of leaves and fruit was reduced, whereas disease severity of fruit was significantly reduced by the application of captan and sulfur when compared to nontreated trees (Figs. 14-17). In the second Merced plot, only disease severity of fruit was significantly reduced (Figs. 18, 19). In both Merced plots, no significant differences between treatments were observed on leaves but disease incidence and severity were consistently lower in the spray treatments. In the Butte plot, disease incidence and severity on fruit were consistently reduced when compared to the check. On Carmel leaves, no significant differences were observed between spray treatments (Fig. 12, 13); on Butte leaves, disease incidence and severity were significantly reduced with sulfur or captan.

Additional studies are needed to determine the effectiveness of LLS and captan or sulfur treatments for scab control. The slow growth of the fungus (<u>Cladosporium carpophilum</u>) and the long incubation time for disease development would indicate that early spring sprays (before 5-wk after petal fall) followed by sprays of captan and sulfur in the following months (May-July) should provide control of the disease. Studies will continue to evaluate these spray programs and experiments will be repeated in the 1993 season.

RUST AND SCAB STUDIES

Effect of Early Season Defoliation. In the 1992 study, the applications of zinc sulfate caused only limited defoliation, little to no regrowth of shoots, and no sporadic blooming before dormancy in the fall. Thus, early to mid-season application of zinc sulfate (30 lbs/A) did not defoliate trees. This prevented simulation of early season defoliation and the study was not continued.

ANTHRACNOSE OF ALMOND

Identification and Pathogenicity Studies. The disease, observed in one commercial orchard in Merced in the spring of 1992, was confirmed to be anthracnose of almond caused by a species of Colletotrichum. Anthracnose was first reported in the 1930's and since, has not been documented in California, although there were unconfirmed reports of its occurrence in the early 1970's. In 1992, the disease was first observed 2-3 wk after petal fall and occurs primarily on the fruit. NePlus was the cultivar mostly infected, whereas Nonpareil were apparently free of the disease. Infected fruit shrivel, become light rusty orange, and appear similar to almond blanks. Leaves attached to fruit spurs often wilt and remain attached (similar to leaf blight). The fungus had an optimum temperature of growth at 25 C and did not grow at 40 C (Fig. 20). Laboratory and field inoculation studies demonstrated pathogenicity of the Colletotrichum species on wounded and non-wounded fruit (Fig. 21). For fruit treated with a low conidial concentration, 3.6% of the spray-inoculated fruit and 28% of the wound inoculated fruit were infected; whereas for fruit treated with a high conidial concentration, 5% of the sprayinoculated fruit and 52.4% of the wound-inoculated fruit were infected. In these inoculation studies, symptoms including sunken lesions, collapsed fruit, and blighted leaves on infected spurs were similar to those observed in the field and to reports in the literature (Ogawa and English 1991). Monitoring the incidence of anthracnose and development of control strategies should be initiated in 1993.

REFERENCES

- Adaskaveg, J. E., D. A. Shaw, and J. M. Ogawa. 1990. A mist generator and environmental monitoring system for field studies on shot hole disease of almond. Plant Dis. 74:558-562.
- Ogawa, J. M. and H. English. 1991. Diseases of Temperate Zone Tree Fruit and Nut Crops. University of California, Division of Agriculture and Natural Resources, Oakland, CA. Publication No. 3345. 461 pp.

Figure 1.



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Figure 2.

Percent Natural Incidence of Brown Rot Shoot Infection of Almond Cultivars at the UCDavis Field Station-1992 Season



Natural incidence of shoot infection was determined based on 200 (50/quadrant) randomly selected shoots for 10 trees.

Figure 3.

Regression of Incidence of Brown Rot Blossom Blight caused *Monilinia laxa* on Wetness Period of Drake Almonds



Blossoms were inoculated with conidia of *M. laxa* (25,000/ml) exposed to wetness periods using bagging or misting techniques, and evaluated after 2 wk.

Figure 4.











Blossoms were treated with iprodione (Rovral 50WP-1 lb ai/A) or iprodione-oil emulsions (4 gals/A), dried, and then inoculated with conidia of *Monilinia laxa* (25,000/ml), and incubated for 5 days at 25 C (72 F).

Figure 6.

Incidence of Brown Rot Blossom Blight of Apricot - Evaluation of Bloom and Petal Fall Fungicide Sprays -





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Incidence of Brown Rot Infected Spurs of Apricot - Evaluation of Bloom and Petal Fall Fungicide Sprays -



Figure 8.

Incidence and Severity of Shot Hole of Almond - Evaluation of Bloom and Petal Fall Fungicide Sprays -



Figure 9.

Incidence and Severity of Shot Hole of Almond

- Evaluation Recommended and Timing Sprays of Ziram -



See text for explanation of treatments.

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Figure 10.

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Incidence and Severity of Shot Hole of Almond - Evaluation Recommended and Timing Sprays of Ziram -



Figure 12.

Control of Almond Scab with Dormant Applications of Liquid Lime Sulfur and Summer Applications of Sulfur or Captan



Figure 13.

Control of Almond Scab with Dormant Applications of Liquid Lime Sulfur and Summer Applications of Sulfur or Captan



Figure 14.

Control of Almond Scab with Dormant Liquid Lime Sulfur and Summer Sprays of Captan or Sulfur - Merced 1



See text for explanation of treatments.

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Figure 15.

Control of Almond Scab with Dormant Liquid Lime Sulfur and Summer Sprays of Captan or Sulfur - Merced 1



Figure 16.

Control of Almond Scab with Only Summer Sprays of Captan or Sulfur Merced Orchard - 1 B



Incidence = No.of infected fruit or leaves per total sample. See text for explanation of treatments.

Figure 17.

Control of Almond Scab with Only Summer Sprays of Captan or Sulfur Merced Orchard - 1 B



Severity = No. of lesions/fruit or leaf.

Figure 18.

Control of Almond Scab with Only Summer Sprays of Captan or Sulfur Merced Orchard - 2



Incidence = No. of infected fruit or leaves/total sample.

See text for explanation of treatments.

Figure 19.

Control of Almond Scab with Only Summer Sprays of Captan or Sulfur Merced Orchard - 2



Severity = No. of lesions/fruit or leaf.

Figure 20.

GROWTH OF SEVERAL ISOLATES OF COLLETOTRICHUM SP. FROM ALMOND AT 10-40 C





Fruit were inoculated with conidial suspensions, covered with plastic and paper bags for a 16-hr wetness period, air-dried, and incubated for 10-14 days under ambient conditions.

POTENTIAL USE OF ENZONE FOR CONTROL OF ARMILLARIA ROOT ROT OF ALMOND

- INTERIM REPORT -December 1991 to January 1993

Investigators: Dr. J. E. Adaskaveg and Dr. J. M. Ogawa

- Project: Armillaria Root Rot Control in Almonds: A Comparative Study of the Soil Fumigants: Tetrathiocarbonate and Methyl Bromide or Metam-Sodium
- Location: Department of Plant Pathology University of California Davis, CA 95616
- Cooperators: Kevin Conn, Layne Wade, and Pam Rener, Dept. of Plant Pathology, UCDavis Joe Connell, Farm Advisor, Cooperative Extension, University of California Les Herringer and Keith Emerson, M & T Ranch, Chico, CA

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SUMMARY

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Studies are ongoing to determine the effectiveness of tetrathiocarbonate (TTC) formulated as Enzone (liquid or gel formulation) compared to methyl bromide or metam-sodium for control of Armillaria root rot of almond (Prunus dulcis (Mill) W.A. Webb caused by the putative species Armillaria mellea (Vahl.: Fr.) Kummer. A test plot was established in Butte County where 10% of a 50 acre block of trees had Armillaria root rot. Experiments were done in Armillaria infection centers directly affecting approximately 5 acres. Soil treatments were: preplant TTC liquid formulation (TTC-L) at 3850 µg a.i./ml (189 L/site); post-plant TTC-L at 500 μ g/ml (189 L/site); pre- and post-plant TTC-L; pre- and post-plant TTC-L with root and soil treatments with Trichoderma harzianum; pre-plant methyl bromide (454 g/site); and nontreated, water drenches as check treatments. Recovery of the fungus from buried root segments of different sizes was variable at both 30 and 121 cm depths after 5 months for the fumigants evaluated. After 1 year, a significant reduction of recovery of A. mellea was shown for both the methyl bromide and the pre- and post-plant treatment of TTC-L when compared to the check. Tree survival after 10 months, however, decreased slightly in pre-plant or pre- and postplant TTC-L treated soil when compared to trees grown in soil fumigated with pre-plant methyl bromide or water treatments. No difference was observed in severity rating of these treatments. In trees treated only with post-plant TTC-L, a 50% reduction in tree survival and a significant increase in tree decline was observed. Soil populations of Trichoderma species did not significantly increase with root dips and soil injections of T. harzianum applied at a rate of 5.83

X 10^5 cfu, however, populations increased numerically. After 4 months, no difference was observed in tree survival or tree decline rating when mature trees were treated with one application of TTC-L (500 μ g/ml) or water during the growing season. In the 1993 season, studies will include: continued evaluations of replants; attempts to keep mature trees infected with Armillaria root rot alive with TTC treatments; improved field-application methods of TTC and other fumigants; and in vitro studies of dosage response of A. mellea to TTC.

INTRODUCTION

Armillaria root rot occurs on a wide range of hosts including <u>Prunus</u> and other species of tree fruit and nut crops (Raabe 1962, 1979a). Symptoms of the disease are poor shoot growth, premature yellowing and dropping of leaves, dieback of branches, and death of trees. Three signs of the fungus include: 1) white, fan-shaped, mycelial plaques that are formed between the bark and wood; 2) dark brown, root-like mycelial strands called rhizomorphs that are associated with plant roots; and 3) mushrooms or basidiocarps of the fungus that commonly occur in clusters around the base of tree trunks usually produced between October and February. In California, the disease is caused by the putative species <u>Armillaria mellea</u> (Vahl.: Fr.) Kummer. Other names of the disease include "Shoestring root rot" and "Oak root fungus" (Ogawa and English 1991).

The fungus attacks roots of plants, killing the cambium and degrading woody tissue. The fungus spreads root to root by rhizomorphs or by mycelium through root contact. Rhizomorphs are the principal agents of infection and can penetrate directly through the bark of roots (Thomas 1934). Under California conditions, basidiospores produced from basidiocarps of the fungus are not considered to function in the epidemiology of the disease. The fungus causes a white rot of wood, degrading all major components of wood cell walls including cellulose, hemicellulose and lignin and survives on dead roots remaining in the soil after tree removal (Ogawa and English 1991).

Soil fumigation and resistant rootstocks of tree crops are the most effective control practices for Armillaria root rot. Resistant rootstocks offer a practical method of control (Raabe 1979b), but trees are generally slower growing and dwarfed. Fumigants reduce inoculum of the fungus but eradication is rarely achieved. Fumigants currently available include metam-sodium (vapam), chloropicrin, methyl bromide, and carbon disulfide (LaRue et al. 1962; Munnecke et al. 1970, 1973, 1981; Filip and Roth 1977). Application methods previously used include treatment of soil (Munnecke et al. 1973) or treatment of stumps (Filip and Roth 1977). Control of Armillaria root rot by soil fumigation is also related to weakening of the fungus followed by antagonism by the fungus <u>Trichoderma viride</u> or other <u>Trichoderma</u> species (Munnecke et al. 1973; Ohr et al. 1973).

Due to environmental concerns about some fumigants such as methyl bromide, alternative environmentally safe materials need to be evaluated for the control of Armillaria root rot in California. Enzone by the Unocal Corp. Chemical Division, is a non-explosive liquid formulation of tetrathiocarbonate (TTC) that releases the fumigant CS_2 . Based on information provided by Unocal, Enzone can be used at high concentrations as a pre-plant fumigant or at low concentrations as a post-plant treatment possibly without causing phytotoxicity to almond trees.

OBJECTIVES

The objectives of this study were:

- 1) To determine the efficacy of TTC or other fumigants for control of Armillaria root rot of almond under commercial conditions applied by the following methods:
 - a) Pre- or post-plant soil treatment.
 - b) Stump application treatments.
- 2) To determine if TTC or other fumigants can eradicate <u>Armillaria mellea</u> from infested roots buried at one and four foot depths under field conditions.
- 3) To determine if soil populations of <u>Trichoderma</u> species can be increased by the addition of preparations of a <u>Trichoderma</u> species above population levels found in nontreated soils or in soils treated with fumigants (TTC, Methyl Bromide).

MATERIALS AND METHODS

Site selection and isolation of <u>Armillaria mellea</u>. A test plot was established in a commercial orchard in Butte County where 10% of a 50 acre block of trees had <u>Armillaria</u> root rot. Experiments were done in <u>Armillaria</u> infection centers directly affecting approximately 5 acres. One-hundred and eight trees were removed in the fall of 1991. Trees that died in the current or previous growing season were identified. Roots of removed trees were sampled and isolations for <u>A. mellea</u> were done in the laboratory. For this, root segments were broken open and wood was placed on a selective medium containing 10 μ g/ml benomyl (DuPont de Nemoeurs Co., Wilmington, DE), 100 μ g/ml streptomycin, and 2 ml/L of 25% lactic acid. The fungus was identified following cultural characteristics described by Nobles (1965).

Soil fumigation treatments and application techniques. The experimental design is in accordance with SOP GEN-004-01. A liquid formulation of Enzone (31.4% TTC; TTC-L) was used for soil treatments. After dead infested trees were removed, soil treatments were: preplant TTC-L at 3850 μ g a.i./ml (189 L/site); post-plant TTC-L at 500 μ g/ml (189 L/site); preand post-plant TTC-L; pre- and post-plant TTC-L and root and soil treatments with <u>Trichoderma harzianum</u> (see below for details); pre-plant methyl bromide (454 g/site); and nontreated, water drenches as check treatments. Pre-plant treatments were made in December 1991, whereas post-plant treatments (3 months after planting) were made in May 1992.

Methyl bromide (Dowfume 98%, methyl bromide) was applied by a commercial applicator at each site using a 45 cm soil injector. No coverings (e.g. plastic tarp) were used over the treated soil surface. For applications of TTC-L, a 2 X 3 X 0.5 m basin was prepared at each replant site following SOP CHM-001-01. Over the next two days, 2 in. (5 cm) of water were applied using sprinkler irrigation or from rain to settle the soil in each basin. At the time of treatment (2 days after irrigation), Unocal representatives (Neil Phillips, Brad Bell, and Sahag Garabedian) observed that each site needed a supplemental application of water. Therefore, another 378 L (100 gal) of water was applied to each site before treatment with TTC-L. An

aqueous solution of the fumigant (3850 μ g a.i./ml) was prepared and 189 L (50 gal) was applied to each site. Water (189 L) was applied to non-treated control sites. Each treatment had 18 replications (17 replications for the check).

Five days after treatment, soil probes were taken by Brad Bell, Joe Connell (Farm Advisor), Keith Emerson, and Dr. Adaskaveg to determine soil moisture at different depths and distances from the center of the prepared basins. Soil probes were obtained from two orchard sites, while 4-8 soil probes were obtained from each of eight treatment sites.

Evaluation of A. mellea survival in buried, infested root segments. Approximately, 200 root segments (30 cm long), infested with A. mellea, were obtained from removed trees. The segments were stored at 1-3 C for approximately 3 weeks after they were obtained. Isolations were made as previously described to verify the presence of the fungus. Root segments were cut approximately 15-18 cm long and were placed into large (1.25-2.5 cm diam.; 2.0 cm mean) and small diameter (0.5-1.25 cm diam.; 0.9 cm mean) groups. Using steel wire, two root segments (one large and one small diameter segment) were tied together. One pair of segments was tied at one end of a 2.75 m (9 ft) length of steel wire. Another pair of root segments were tied onto the steel wire 91 cm from the first pair. Two holes at treatment sites were dug using a tractor-mounted soil auger. Root segments, attached to the wire, were hung down into each hole at 30 cm (1 ft) and 120 cm (4 ft) depths from the soil line, and the holes were re-filled with soil. The end of each wire was tied to an iron stake. Roots segments were buried in 6 of 17 replications of each treatment prior to the pre-plant treatments. One set of buried root segments from each treatment was removed 5 months, while the second set was removed 12 months after the pre-plant treatments. Data were analyzed using analysis of variance and LSD mean separation procedures of SAS 6.04.

Replanting of almond trees and <u>Trichoderma</u> treatments. In February 1992, one week before planting, basins used for the Enzone drench treatment, were leveled and allowed to settle. Almond trees (Peerless, Carmel, and Butte) on Lovell peach rootstock were replanted into the soil treated sites. Treated soil from the edges of the basin was used to refill the treated area.

During isolation of <u>A</u>. <u>mellea</u> from infected root segments, isolates of <u>Trichoderma</u> <u>harzianum</u> were obtained and identified by Dr. J.E. Adaskaveg. Two isolates of the fungus were sent to Dr. J. Vartanian at Unocal for culturing of the fungus and later use in selected treatments at planting or for post-plant treatments. Dilutions of the batch culture will be applied as a root dip or directly to soil with a soil fumigation injector. For this, the isolate JEA S1-16 was grown on a molasses and yeast extract liquid media (500 ml) on a shaking incubator (25 C) for 14 days, filtered through cheese cloth, and re-suspended in an aqueous suspension of 2% carboxy-methyl cellulose gel. The final concentration was 5.85 X 10⁵ cfu of <u>T</u>. <u>harzianum</u> (mycelium and chlamydospores). Roots of 18 trees were dipped into the suspension (0.55 L/tree) and then trees were planted into sites pre-treated with TTC as previously described. Additionally, an equal volume was injected into the soil within a 30 cm radius of the crown of the replanted tree at the time of planting and 3 months later after the post-plant fumigation treatment with TTC (500 μ g/ml).

At four and nine months after planting, soil samples were collected to a depth of 30 cm using a soil probe (2 cm diam.) from: pre-plant TTC; pre- and post-plant TTC with <u>T</u>. <u>harzianum</u> added; methyl bromide; and check treatments. Eight equally spaced soil cores taken from approximately a 30 cm radius of the tree crown were combined in polyethylene bags for a composite sample from each of 10 replications of the four treatments. Soil texture and pH were recorded for the replications of each treatment following methods in Chapman and Pratt (1961). Ten grams, fresh weight of soil for each replication was weighed out and mixed with 100 ml of a 0.1% solution of water agar. A 1/2000 and a 1/10000 dilution were made and 1 ml per plate of each dilution was placed in six petri dishes. A <u>Trichoderma</u> selective media (10 ml/plate) was used in this experiment (Elad at al. 1981). The media consisted of MgSO₄ (0.2 g/L), K₂HPO₄ (0.9 g/L), KCL (0.15 g/L), NH₄NO₃ (1 g/L), glucose (3 g/L), chloramphenicol (0.25 g/L), Lesan 75WP (0.257 g/L), PCNB (0.2 g/L), rose bengal (0.15 g/L), and agar (20 g/L). Plates were incubated at 25 C and numbers of colonies of <u>Trichoderma</u> spp. and other fungal species were recorded after 7-10 days. Data were analyzed using analysis of variance and LSD mean separation procedures of SAS 6.04.

Determination of tree survival and decline of replanted trees. Ten months after the pre-plant and five months after post-plant treatments with fumigants, trees were evaluated for survival, trunk diameter, and decline (0 = healthy; 1 = chlorotic leaves; 2 = chlorotic leaves and thin canopy (no new growth); 3 = chlorosis, thin canopy, and dieback; or 4 = tree death) were evaluated. Data were analyzed using general linear model and LSD mean separation procedures of SAS 6.04.

Suppression/eradication treatments using Enzone. Living, mature trees with Armillaria root rot that bordered infection centers were used in this experiment. The fungus was isolated from wood samples obtained from infected trees as previous described. In May 1992, a soil drench was made using TTC-L (500 μ g a.i./ml; 189 l per site) applied in basins (2 X 3 X 0.5 m) made around ten infected trees. Water was applied to basins around 10 check trees. Trees were rated using a disease severity scale: 0 = healthy; 1 = chlorotic leaves; 2 = chlorotic leaves and thin canopy (no regrowth); 3 = chlorosis, thin canopy, and dieback; and 4 = tree death. Data were analyzed using analysis of variance and LSD mean separation procedures of SAS 6.04.

Direct treatment of infested tree stumps with fumigants. In January 1993, trees that died in the 1992 growing season were cut down leaving a stump approximately 15 cm in height from the ground. Stump diameter was recorded. Seven to eight equally spaced holes (1.9 cm in diam. X 10 cm deep) were made 2.5-3 cm from the perimeter of the trunk using an electric drill. The holes were filled with stock material of either: TTC-L; an Enzone gel formulation (31.4% TTC; TTC-G); vapam (32.7% sodium N-methyldithiocarbamate or Metam-Sodium); or water. The fumigants were poured into each of the stumps (300 ml/stump), bark-corks were placed in each hole, and exposed wood of the stumps was sealed with a silicone tile sealer. After the silicone sealer dried, polyethylene plastic bags were placed over the stumps, the bag edges were buried, and strapping tape was wrapped around the bags and trunks. After 3 months, plans include removal of the stumps, sampling of roots, and isolation of <u>A</u>. mellea as previously described to determine if the fungus was eradicated from the infested root system.

RESULTS AND DISCUSSION

Soil characterization and movement of TTC-L in soil. Two main soil types, a heavy clay loam and a sandy loam, characterized the upper one meter of the soil profile of fumigation treatments sampled. Below one meter, soils were generally sandy to gravelly. Soil pH ranged from 6.32 to 6.58 for all the soil fumigation sites. Movement of TTC from soil basins through the soil profile was estimated by taking soil probes at various distances from basin centers and comparing them to probes taken where no treatments were applied. Lateral movement in heavy soils was less than 2 m, while movement in sandy soils was estimated to be between 3-4 m.

Survival of A. mellea in buried root segments. Percent recovery of A. mellea after 5 months from infested buried root segments was variable for both depth (30 and 121 cm) roots were buried and for the treatments evaluated (Fig. 1). The two root sizes (0.9 and 2.0 cm) used in the experiment did not significantly influence efficacy of the treatments evaluated. A significant interaction, however, was observed between root segment depth and treatment (P < P0.02). Thus, the two depths were evaluated separately. For infested roots buried at a depth of 30 cm, percent recovery of the fungus from pre-plant TTC-L and pre-plant TTC-L + \underline{T} . harzianum was significantly reduced from the check; whereas the methyl bromide treatment was intermediate between the TTC-L and check treatments. Percent recovery was significantly reduced for methyl bromide and pre-plant TTC-L treatments when compared to the check and the pre-plant TTC-L + T. harzianum at the 121 cm depth (Fig. 1). No difference was found between the check and the pre-plant TTC-L + \underline{T} . <u>harzianum</u>. The inconsistency of methyl bromide at the 30 cm depth was possibly due to the application technique (soil injection and no covering over treated soil), while the variability of the pre-plant TTC-L at 121 cm may indicate the difficulties inherent in attempting to eradicate A. mellea from deeply buried roots.

After 1 year, at both soil depths, percent recovery of <u>A</u>. mellea was significantly reduced for the pre- and post-plant TTC-L treatment when compared to the check (Fig. 2). At the 30 cm depth, recovery of the fungus from root segments in the other treatments was not significantly different from the check, however, a general reduction was observed. At the 121 cm depth, a 100% recovery of the fungus was obtained from the check and post-plant TTC-L. Recoveries of <u>A</u>. mellea from roots in the methyl bromide, pre-plant TTC-L, or the pre- and post-plant TTC-L treatments were significantly less than the check and post-plant TTC-L treatments. Recovery of <u>Armillaria</u> from the pre-plant TTC-L + <u>T</u>. <u>harzianum</u> treatment was lower than the check, forming an intermediate group between the check and the other treatments (Fig. 2). The post-plant TTC-L treatment when used alone had no effect on recovery of the fungus. This was probably due to the low concentration (500 μ g/ml) of TTC-L used. The preand post-plant TTC-L treatment, however, was consistently effective at both soil depths evaluated.

Percent survival and decline rating of replant trees after soil fumigation treatments. After 10 months, tree survival slightly decreased in pre-plant or pre- and post-plant TTC-L treated soil when compared to trees grown in soil fumigated with methyl bromide (pre-plant only) or water treatments (Table 1). No difference was observed in decline rating of these treatments. In trees treated only with post-plant TTC-L, a significant reduction (50%) in tree survival and in tree decline was observed. <u>Armillaria</u> was not recovered during isolations from tree roots that died. Thus, the percent survival data suggests that the TTC treatments maybe slightly phytotoxic to young replanted trees. Interestingly, the post-plant treatment of TTC was the most injurious (Table 1). Perhaps soil conditions such as temperature and moisture levels may predispose almond trees to injury when post-plant applications of fumigants are used.

Percent survival and decline rating of mature infected trees treated with TTC. Percent survival and decline rating of mature trees treated with TTC or water were not significantly different six months after treatment; although, a trend was observed with a higher percent survival and a lower decline rating for trees treated with TTC (Table 2). The therapeutic treatments did not appear to be phytotoxic to the mature, infected trees based on the decline rating. Perhaps the additional planned applications of TTC-L in 1993 will maintain tree survival. Eradication of the pathogen or prevention of death from trees extensively colonized by the fungus is not promising.

Recovery of <u>Trichoderma</u> Species from Soil. Six and 12 months after soil fumigation treatments, no statistical difference was observed in the mean number of colonies of <u>Trichoderma</u> or other fungal species between check and soils fumigated with methyl bromide or TTC-L (Tables 3, 4). Colonies that grew on the selective media from the two dilutions were proportional. Numerically, however, a higher average number of <u>Trichoderma</u> colonies was found in the treatment where <u>T</u>. <u>harzianum</u> was applied to roots and injected into soil surrounding (30 cm radius form tree crown) the replant trees. Variation in the replications prevented separation of treatments, although the highest number of <u>Trichoderma</u> colonies were in the pre-plant + <u>T</u>. <u>harzianum</u>. Additional studies to determine populations of trichoderma on roots of replant trees are planned.

REFERENCES

- 1) Chapman H.D. and P.F. Pratt. 1961. Methods of Analysis for soils, Plants, and Waters. University of California, Div. of Agric. Sciences, Publ. No. 4034. Berkeley. 309 pp.
- 2) Elad, Y., I. Chet, and Y. Henis. 1981. A selective medium for improving quantitative isolation of <u>Trichoderma</u> spp. from soil. Phytoparasitica 9: 59-67.
- Filip, G.M. and L.F. Roth. 1977. Stump injections with soil fumigants to eradicate <u>Armillariella mellea</u> from young-growth ponderosa pine killed by root rot. Canadian Journal of Forest Research 7: 226-231.
- LaRue, J.H., A.O. Paulus, W.D. Wilbur, J.H. O'Reilly, and E.F. Darley. 1962. Armillaria root rot fungus controlled with methyl bromide soil fumigation. Calif. Agric. 16: 8-9.
- 5) Munnecke, D.E., M.J. Kolbezen, and W.D. Wilbur. 1970. Dosage response of <u>Armillaria mellea</u> to methyl bromide. Phytopathology 60: 992-993.

- 6) Munnecke, D.E., M.J. Kolbezen, and W.D. Wilbur. 1973. Effect of methyl bromide or carbon disulfide on <u>Armillaria</u> and <u>Trichoderma</u> growing on agar medium and relation to survival of <u>Armillaria</u> in soil following fumigation. Phytopathology 63: 1352-1357.
- 7) Munnecke, D.E., M.J. Kolbezen, W.D. Wilbur, and H. D. Ohr. 1981. Interactions involved in controlling <u>Armillaria mellea</u>. Plant Disease 65: 384-389.
- Nobles, M.K. 1965. Identification of cultures of wood-inhabiting Hymenomycetes. Can. J. Bot. 43: 1097-1139.
- Ogawa, J.M. and H.E. English. 1991. Diseases of Temperate Zone Tree Fruit and Nut Crops. University of California, Div. of Agric. and Nat. Res. Oakland, CA. Publication 3345. 461 pp.
- 10) Ohr H.D., D.E. Munneke, J.L. Bricker. 1973. The interaction of <u>Armillaria mellea</u> and Trichoderma spp. as modified by methyl bromide. Phytopathology 63: 965-973.
- Raabe, R.D. 1962. Host list of the root rot fungus, <u>Armillaria mellea</u>. Hilgardia 33: 25-88.
- Raabe, R.D. 1979a. Some previously unreported hosts of <u>Armillaria mellea</u> in California, III. Plant Dis. Rep. 63: 494-495.
- 13) Raabe, R.D. 1979b. Resistance or susceptibility of certain plants to Armillaria root rot. Univ. of Calif. Div. Agric. Sci. Leaf. 2591. 11 pp.
- 14) Thomas, H. E. 1934. Studies on <u>Armillaria mellea</u> (Vahl.) Quel., infection, parasitism, and host resistance. J. Agric. Res. 48: 187-218.

Figure 1.

Recovery of *A. mellea* from Root Segments Five Months after Soil Fumigation with Tetrathiocarbonate or Methyl Bromide

Root Size+Treatment P > 0.93



Root segments infested with Armillaria mellea were buried at 30 or 121 cm depths. Soil was treated with fumigants prior to planting of trees. Roots of some replants were treated at planting with a cellulose gel suspension of Trichoderma harzianum.



Recovery of *A. mellea* from Root Segments One Year After Soil Fumigation with Tetrathiocarbonate or Methyl Bromide



Root segments infested with *Armillaria mellea* were buried at 30 or 121 cm depths. Soil was treated with fumigants prior to planting of trees. Roots of some replants were treated at planting with a cellulose gel suspension of *Trichoderma harzianum*. Table 1.Percent survival and disease severity of almond replants (Lovell peach
rootstock) one year after fumigation treatments with tetrathiocarbonate
(TTC) or methyl bromide. 1

	Number of		
Treatment	Trees	% Survival ²	Severity Rating 2,3
Preplant TTC	18	83.33 a	0.84 a
Pre- + Post-plant TTC	18	88.88 a	0.50 a
Post-plant TTC	18	50.00 b	2.61 b
Pre-plant TTC +	18	94.44 a	0.22 a
T. harzianum 4			
Preplant Methyl Bromide	18	100.0 c	0.16 a
Check	17	100.0 c	0.11 a

 Fumigation treatments were: Preplant Tetrathiocarbonate (TTC;3850 ug/ml); Postplant TTC (500 ug/ml); Pre- and Post-plant TTC; Methyl Bromide (454 g/site); and Check (nontreated).

- 2 Treatments were evaluated using general linear model and LSD mean separation procedures of SAS 6.04. Means followed by the same letter are not significantly different (P > 0.05).
- 3 Severity rating : 0 = Healthy, 1 = Chlorotic leaves, 2 = Chlorotic leaves and thin canopy; 3 = Thin canopy and dieback; and 4 = Tree death.
- 4 Cultures of Trichoderma harzianum were grown in vitro in a molasses, yeast extract media, applied in a cellulose gel as a root dip to replant trees, and as a soil injection within 30 cm of replanted trees.

	Number of			2.2
Treatment	Trees	% Survival ²	Severity Rating	2,3
Post TTC	10	80 a	1.75 a	
Check	10	50 a	2.60 a	

Table 2.Percent survival and disease severity of mature almond trees
(Lovell peach rootstock) infected with Armillaria mellea six months after
fumigation treatments with tetrathiocarbonate (TTC) or methyl bromide. 1

- Fumigation treatments were: Preplant Tetrathiocarbonate (TTC;3850 ug/ml); and Check (nontreated).
- 2 Treatments were evaluated using general linear model and LSD mean separation procedures of SAS 6.04. Means followed by the same letter are not significantly different (P > 0.05).
- 3 Severity rating : 0 = Healthy, 1 = Chlorotic leaves, 2 = Chlorotic leaves and thin canopy; 3 = Thin canopy and dieback; and 4 = Tree death.

Table 4. Mean number of colonies of Trichoderma species and other fungi from soil six months after soil fumigation and 5 months after almond trees (Lovell peach rootstock) were treated (at planting and three months after planting) with or without *T. harzianum.*¹

Treatment	Mean No. of Colonies (1:2000 Dilution) ²		Mean No. of Colonies (1:10000 Dilution) ²	
	Trichoderma	Other Colonies	Trichoderma	Other Colonies
Pre- + Post-Plant TTC Pre-Plant TTC + <i>T. harzianum</i> ³	23.00 a 36.02 a	172.49 a 114.00 a	6.01 a 9.00 a	43.34 a 35.16 a
Preplant Methyl Bromide	25.01 a	108.33 a	5.34 a	21.99 a
Check	22.17 a	120.51 a	5.27 a	32.33 a

 Fumigation treatments were: Preplant Tetrathiocarbonate or TTC (3850 ug/ml); Postplant TTC (500 ug/ml); Pre- and Post-plant TTC; Methyl Bromide (454 g/site); and Check (nontreated).

- 2 Values are the means of colonies of Trichoderma spp. or other fungi on 6 plates of TSM/rep and 10 reps/treatment. Treatments were evaluated using general linear model and LSD mean separation procedures of SAS 6.04. Means followed by the same letter are not significantly different (P > 0.05).
- 4 Cultures of Trichoderma harzianum were grown in vitro in a molasses, yeast extract media, applied in a cellulose gel as a root dip to replant trees, and as a soil injection within 30 cm of replanted trees.
- Table 4. Mean number of colonies of Trichoderma species and other fungi from soil 12 months after soil fumigation and 10 months after almond trees (Lovell peach rootstock) were treated (at planting and three months after planting) with or without *T. harzianum.*¹

	Mean No. of Colonies (1:2000 Dilution) ²		Mean No. of Colonies (1:10000 Dilution) ²	
Treatment	Trichoderma Other Colonies		Trichoderma	Other Colonies
Pre- + Post-Plant TTC Pre-Plant TTC + <i>T. harzianum</i> ³	14.30 a 24.50 a	88.10 a 61.00 a	2.80 a 5.40 a	17.80 a 21.70 a
Preplant Methyl Bromide	15.50 a	59.10 a	2.90 a	26.50 a
Check	16.00 a	103.90 a	3.20 a	24.10 a

- Fumigation treatments were: Preplant Tetrathiocarbonate (TTC; 3850 ug/ml); Postplant TTC (500 ug/ml); Pre- and Post-plant TTC; Methyl Bromide (454 g/site); and Check (nontreated).
- 2 Values are the means of colonies of Trichoderma spp. or other fungi on 6 plates of TSM/rep and 10 reps/treatment. Treatments were evaluated using general linear model and LSD mean separation procedures of SAS 6.04. Means followed by the same letter are not significantly different (P > 0.05).
- 4 Cultures of Trichoderma harzianum were grown in vitro in a molasses, yeast extract media, applied in a cellulose gel as a root dip to replant trees, and as a soil injection within 30 cm of replanted trees.

Scout In Fall For Shothole Control

Fall scouting for almond shothole can help growers plan their disease control program in the spring

By Parry Klassen

A NEW fall scouting technique for shothole fungus in almonds is helping refine what could be called the current "shotgun" approach to controlling the disease in spring. Tests conducted in experimental and commercial orchards during the past three years showed that California almond growers could potentially expect huge savings in disease control expenses.

DISEASE CONTROL

A shothole control program typically begins in the spring, with the first fungicide treatment intended to protect newly developing leaves. Applied at bloom, the spray is used in combination with fungicides for brown rot control and other bloom diseases. Should it rain, additional shothole treatments are often made. Treatments are sometimes applied on a weekly basis for five weeks after petal fall. Rain can spread shothole fungus spores within and between trees.

Researchers agree with this control strategy — to a point. "Growers usually apply this first shothole treatment at bloom because other materials must be applied. If a shothole fungicide is included with the spray, they can wait — and watch," says Jim Adaskaveg, plant pathologist at University of California-Davis, who along with plant pathologist Joe Ogawa, have been studying shothole prevention.

Scout For Spores In Fall, Spring

Growers should look for evidence of shothole spores on newly developing leaves each spring. In the scouting program suggested by UC scientists and other researchers, this leaf inspection is the second time growers should look for evidence of shothole fungus. Scout first in the fall.

"If leaves do not show evidence of shothole in the fall, you should expect to see less

disease in the spring and you shouldn't need a shothole material in the first spray," says Adaskaveg. "If your leaves are covered with shothole spore pustules in the fall, that means you should be prepared to take care of it in the spring with your first spray."



Ogawa and Adaskaveg have been studying how shothole spores overwinter and spread in almond orchards. They found that when spore pustules are visible on leaves in early November just before leaf drop, rain or sprinklers can spread the spores into twigs and branches where they overwinter. If no rain falls before leaf drop or the trees are defoliated with zinc sulfate, disease flareups are less likely next spring.

"The key in the spring is to watch the new leaves as they emerge," says Ogawa. "If it rains during and after bloom and you don't see any spore pustules developing, you can skip some or all of the shothole sprays. If you see the spores beginning to form, then apply fungicide treatments."

Treatments Skipped When No Spores Found

In orchards monitored over the last three years with farm advisors Joe Connell (Butte County), Lonnie Hendricks (Merced County), and Mario Viveros (Kern County), Ogawa and Adaskaveg went so far as to skip treatments within plots with no detectable spores or where low levels of spores were visible on young leaves in spring. No outbreaks of shothole occurred in those orchards.

"Shothole forms a small hole on the leaf because the infected area falls out," says Adaskaveg. "We have no clear evidence that once the spore falls to the ground it makes its way

back to the foliage."

Some growers and farm advisors are concerned if the scouting program will work on large acreage orchards. Ogawa suggests dividing the fields into suitable blocks. sampling from each block, then spray each block based on its disease history and a fall evaluation.

Adaskaveg and Ogawa are opti-

mistic that the scouting strategy for shothole could save almond growers in California millions of dollars. "If we can reduce these \$20 an acre applications by one treatment, and apply that reduction to the huge almond acreage in California, that can save the industry \$10 to \$30 million each year."



Fall scouting for almond shothole can help

growers determine their spray control program

in the spring. Researchers say if there is no

evidence of shothole in the fall, there is proba-

bly no need to spray in the spring.

Klassen is a contributing editor based in Selma, Calif.