ALMOND LEAF SCORCH DISEASE

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1. OBJECTIVES:

To determine the cause and host range of the disease; to determine means of spread of the causal agent in commercial orchards; to develop an efficient and rapid diagnostic method for the disease and to develop control measures that will be compatible with general practices in commercial orchards.

2. INTERPRETIVE SUMMARY:

The causal agent of almond leaf scorch (ALS) is a bacterium that invades the water and mineral conducting tissues and is indistinguishable from the causal agent of Pierce's disease (PD) in grapes, based on symptoms and vector relationships. The ALS bacterium is transmitted from diseased to healthy trees by grafting and budding, and by leafhoppers. The leafhoppers *Hordnia circellata*, *Draeculacephala minerva* and *Keonolla dolobrata* effectively transmitted the ALS organism from almond to almond and also to grape, and the PD organism from grape to grape and also to almond. The role of several other leafhopper and spittlebug species as vectors of ALS is being investigated. The population and behavior of these vectors are being studied in commercial almond orchards and on vegetation near almond orchards. In 1976, infected trees with ALS were detected for the first time in Kern and Lake Counties; presently, ALS is known to occur in 16 almond producing counties. ALS was also observed for the first time in the Thompson, Davey and Merced varieties. Up to date, ALS has been observed to infect 14 different varieties including: NonPareil, Mission, NePlus and Peerless. Surveys of commercial orchards indicate that the incidence, severity, rate of spread within an orchard and within trees depends on the susceptibility of the variety, the availability of various sources of inoculum and the abundance and efficiency of vectors. For example, the percent of infected trees in two of the surveyed orchards in Contra Costa County was: NonPareil, 37%, NePlus Ultra, 15%, and Drake, 4% in orchard #2; and Long IXL, 67%, Mission (possibly several Languedoc in the Mission rows), 16% in orchard #1. In orchard #1, the number of infected Long IXL and Mission trees increased by 31% and 22% respectively within 5 years. Trees of 16 almond varieties have been artificially inoculated and are being evaluated for their resistance to ALS. Removal of infected branches with incipient symptoms of ALS resulted in no further spread of ALS within naturally-infected orchard trees. Naturally-infected trees when injected with 5-7 gram/tree of Terramycin[R] showed dramatic remission of ALS symptoms. Injection of less than 5 grams, or more than 7 gram/tree was ineffective or phytotoxic respectively. A simple technique was developed for isolation of the ALS bacterium from diseased trees. This technique will aid in development of a rapid and reliable diagnosis of ALS and detection of other possible hosts of the organism that may serve as a source of inoculum for infection of almond trees.

3. EXPERIMENTAL PROCEDURE:

Tolerance of almond varieties and spread of ALS in orchards and trees.--Relative tolerance and resistance of various almond cultivars are being studied under commercial orchard conditions. Five selected commercial orchards were surveyed in the fall of 1976 and the incidence of diseased trees, severity of symptoms and distribution of disease within the individual trees was determined. The same orchards will be surveyed over a period of several years to assess the varietal susceptibility as well as the rate of spread of ALS in orchards and within the trees.

Twenty trees of 16 principal varieties of almond were planted in an experimental plot at Davis and 10 trees of each variety were bud-chip inoculated at single sites to determine their tolerance to ALS, the rate of spread within and between the trees and the general damage by ALS. Trees of apricot, peach, cherry and plum were also bud-chip inoculated to determine whether they are symptomless carriers and as such serve as a source of inoculum for almond trees. Ten pairs of almond orchard trees with incipient infection in a few terminal shoots were selected and infected branches were removed from one of each pair of trees. The subsequent spread of ALS within the trees was observed during one growing season.

Xylem sap extract has been used for isolation of the ALS bacterium from infected trees and over 200 different media have been tested for support of growth and isolation of the ALS organism. Mice are being used to produce specific antisera by injecting them with xylem sap extracts of healthy and diseased plants and with a suspension of several isolates of the ALS bacteria. These antisera, if produced, could be used in the rapid diagnosis of ALS-infected almond trees and other plants. Several isolates recovered from diseased almond trees have been tested for their ability to produce ALS by injecting a bacterial suspension directly into the plants and by injecting or feeding leafhoppers with the bacterial suspension and then placing these vectors to feed on healthy plants.

Alfalfa and clover varieties were tested as possible hosts of ALS and as indicator plants for a rapid diagnosis of ALS.

Chemotherapy of the disease.--Naturally-infected almond trees were pressure injected with Terramycin® in October, November, December (1975), January and February (1976). Each tree was injected with 600 ml of Terramycin® solution containing 3g, 5g, 7g or 10g of active ingredient of the chemical. The efficacy of the treatments in suppressing symptom expression plus their phytotoxicity were evaluated through August-October 1976.

Vectors and their role in the natural spread of ALS.--The efficiency of various leafhopper species in transmitting ALS from diseased almond to healthy almond and grapes, also from PD-infected grape to healthy almonds, was investigated. ALS-free leafhoppers were allowed to feed for various periods of time on diseased almonds and grapes and then transferred to feed on healthy almond and grape plants. The indicator plants exposed to infective leafhoppers were maintained in the greenhouse and observed for development of ALS symptoms.

Surveys of commercial almond orchards, weed covers and the vegetation near orchards were made to determine the type of vectors present in almond orchards, their population dynamics, migration patterns and the importance of different plants as possible sources of ALS inoculum. Vector surveys were made with traps, by sweeping, or by direct search. An aerial survey was made of almond orchards previously mapped for incidence of ALS.

4. RESULTS:

ALS was observed for the first time in Kern and Lake Counties; presently the disease is known to be present in 16 different counties. The disease was also observed for the first time to infect the Thompson, Merced and Davey varieties. Up to date, ALS is known to infect 14 different almond varieties.

Tolerance of almond varieties and spread of ALS in orchards and trees.-- Surveys of commercial orchards in Contra Costa County revealed a marked difference in the incidence, spread and disease severity in different almond cultivars naturally-infected with ALS (Table 1).

TABLE 1. The relative incidence and rate of spread of ALS in commercial almond orchards in the Brentwood area.

			Incidence of ALS			
Variety	Time of survey	Number of trees surveyed	Number infected trees	Percent infected trees	Average disease severity in infected trees[a]	California acreage as of 1975
			ORCHARD #1			
Long IXL	1971	349	56	16	2.8	179
	1976	324	223	69	4.0	
Drake	1971	180	1	<1	2.0	4,935
& Sultana	1976	162	33	20	3.0	
Mission	1971	184	22	12	4.5	51,003
(few Languedoc)	1976	174	28	16	2.7	
			ORCHARD #2			
NePlus Ultra	1976	235	35	15		23,677
NonPareil	1976	238	88	37		180,602
Drake	1976	229	9	4		4,935
			ORCHARD #3			
Jordanolo	1976	385	110	28	1.5	2,125
Long IXL	1976	435	63	14	3.5	179
NePlus Ultra	1976	161	19	12	0.9	23,677

[a] 0=no leaf symptoms; 1=light; 2=moderate; 3=heavy; 4=severe leaf symptoms in the entire top; 5=dying or dead.

However, it is not known at the present time whether these apparent differences in field resistance to ALS among almond varieties are due to a differential susceptibility of almond cultivars to ALS or to a differential preference of vectors for different varieties. These aspects are being investigated.

The incidence of ALS in bud-inoculated cultivars ranged from 0% to 60% of infected trees one year after inoculation. These preliminary results suggest that almond cultivars may differ in their susceptibility to ALS. The same varieties will again be graft-inoculated and further tested for their tolerance to ALS. Plum and prune nursery trees experimentally inoculated with bud-chips from almond trees infected with ALS developed leaf scorch symptoms identical to those in almond trees infected with ALS. The relationship of leaf scorch in plum and prune to ALS is being further investigated. Four-year-old almond trees bud-inoculated in 1973 developed symptoms in one scaffold branch in 1974 and in four scaffold branches per tree in 1976. Spread of ALS from the point of inoculation ranged 5 to 9 feet during the two year period. However, in this experiment no spread of ALS from infected to adjacent healthy trees was observed during the same period. These results indicate that ALS spreads relatively slowly through infected trees and that the relatively rapid spread of ALS observed in commercial orchards, both within infected trees and from diseased to healthy trees, is determined by the efficiency and activity of vectors occurring in commercial orchards.

The slow spread of ALS from infected to non-infected parts of individual trees prompted an experiment in which we investigated the effect of removal of branches with incipient symptoms on the subsequent spread of ALS within the trees. Removal of the infected branches resulted in no further spread of the disease in 10 orchard trees while substantial spread of ALS occurred in 10 comparable control trees within one growing season.

A simple technique was developed for obtaining pure cultures of ALS and PD causal bacteria from diseased trees and vines. This technique is essential for the following reasons: improving artificial media for isolation of the ALS organism, development of a rapid and reliable diagnostic procedure for ALS, determining possible existence of strains of the ALS organism, and studying the interrelationships between ALS and PD; it will also aid in developing control measures for ALS.

The search for an alternative host for rapid indexing of ALS has not yet been successful. Alfalfa and clover varieties were not more reliable than grape. Grape plants are being used for the bioassay of ALS.

<u>Chemotherapy of the disease</u>.--Naturally-infected trees when injected with 5 to 7 gram/tree of Terramycin[R] showed good to excellent remission of ALS symptoms. Injections less than 5 and more than 7 gram/tree were ineffective or phytotoxic respectively. Effectiveness of the chemical was determined by the uniformity of distribution of the chemical through the injected trees which in turn was inversely proportional to the severity of the disease. There was no significant difference in effectiveness of Terramycin[R] injection into trees in October through February.

<u>Vectors and their role in natural spread of ALS</u>.--Our transmission tests revealed that both adults and nymphs of the blue-green sharpshooter, <u>Hordnia circellata</u> are capable of transmitting ALS from almond to almond and grape,

and also PD from grape to almond and grape. The green sharpshooter, Draeculacephala minerva successfully vectored ALS. Survival of this species on almond was unexpectably good, despite the fact that it shows a very decided preference for grasses under natural conditions. Keonolla dolobrata, exposed for 2 days to PD-infected grapes acquired the causal agent that induced ALS disease in healthy almonds. The spittlebug, Philaenus leucophthalmus failed to transmit ALS from PD-infected grapes to almonds in two tests. These experiments are being repeated. We obtained no transmission of ALS in limited attempts using as vectors: Macrosteles faseifrons, Calladonus montanus or Ballana sp. Surveys of orchard weed cover and vegetation near four orchards with a high incidence of ALS in Contra Costa County revealed that the leafhopper population decreased greatly after annual plants dried up in the late spring. Low populations of D. minerva were found along ditches with succulent wild grasses. Barnyard grass was the most preferred host plant for D. minerva. This vector was not observed on almond trees, but it was found feeding on a grass cover crop in an almond orchard. No other species known to transmit PD was found in the surveyed areas. P. leucophthalmus-(spittlebug), was found on the margins of the surveyed orchards. This species may be important as a periodic vector. A survey of vineyards for PD in the vicinity of almond orchards with a high incidence of ALS revealed a low incidence of PD (less than 0.5%) at all but one site in Martinez, where the incidence of PD was over 30% in one portion of the vineyard adjacent to a heavily ALS-infected almond orchard.

A survey of a permanent pasture site near an almond orchard in the central valley revealed three distinct populations of D. minerva occurring from April through September with the highest population level in August. Emigration of D. minerva from pasture and alfalfa fields was highest following water stress in the pasture plants, also after cutting and drying of the alfalfa. The absence to date of ALS in Fresno and Tulare Counties where PD is present and D. minerva occurs occasionally in high populations suggests that either this species is not an important vector of ALS or that the PD causal agent in these counties is not highly virulent to almonds. This latter possibility is being investigated.

An aerial survey for ALS in commercial orchards by using color and infrared photography revealed that only trees with severe symptoms are identifiable in the photographs. However, aerial surveys seem to be useful for rapid survey of large areas and to pinpoint the relative proximity of potential vector breeding grounds, a perspective that is not so easily obtained from ground surveys only.

5. DISCUSSION:

Almond leaf scorch is an infectious disease widely distributed in California's almond producing regions. ALS is known to naturally infect 14 different almond varieties including: NonPariel, Mission, Merced, NePlus and Thompson. Incidence of the disease in affected orchards is low, with the exception of Contra Costa County where over 60% of the trees are affected in certain orchards. Capability of the disease to adversely affect the majority of commercial almond varieties and to spread naturally from affected into nonaffected regions indicates strongly that ALS has the potential to limit almond production in certain districts.

Our field surveys indicate a differential field tolerance of different varieties to ALS (Table 1). Thus the use of more tolerant almond cultivars for new plantings and for replanting almond trees in areas subject to severe ALS may be one possible control measure that will minimize losses by this disease. However, further research is needed to determine the inherent tolerance of various almond varieties.

Terramycin[R] injection (5-7 gram/tree) resulted in very good remission of ALS symptoms in treated trees. These results suggest that chemotherapy has potential as an effective control measure for ALS. However, further research is needed to determine the economic feasibility of Terramycin[R] treatments for control of ALS. In particular, the frequency of treatments needed to restore the productivity of ALS-infected trees should be determined as well as the minimum curative dosages and the amount of chemical residue in fruits at harvest. An improved application method to enable more uniform distribution of the chemical in treated trees is needed, as well as the screening of other candidate chemicals.

A new and effective technique was developed to recover the ALS organism from diseased trees. This technique will aid future research in developing a quick and reliable diagnosis for ALS, in determining a host range of ALS, and in detecting possible symptomless carriers that may serve as a source of inoculum for almond trees. Elucidation of these factors is also imperative for developing effective control measures.

The results from experimental transmission tests showed that the ALS organism is vectored by the leafhoppers *H. circellata* and *K. dolobrata* in addition to the previously-reported *D. minerva*. These results showed that that PD agent from Napa and Contra Costa Counties can cause ALS, and confirmed previous reports that the ALS agent can cause PD in grapevines.

Leafhopper surveys in and near ALS-affected orchards in Contra Costa County revealed the presence of only two potential vectors: *P. leucophthalmus* and *D. minerva*. Neither of these species was abundant in Contra Costa County but they were found in large numbers near almond orchards in Fresno County where ALS has not yet been reported.

Future research should emphasize the following studies: the possible role of other known vectors of PD in the natural spread of ALS; the breeding grounds of potential vectors; migration of vectors from vegetation near orchards into almond orchards; host plant preferences and population trends of potential vectors. These studies will provide necessary data that could aid in developing control measures for ALS through elimination or prevention of an excessive buildup of vectors at the breeding sites or in commercial almond orchards. Furthermore, a better understanding of almond variety preference by potential ALS vectors will aid in proper selection of almond varieties to minimize ALS losses where the potential of the disease may be great.

6. PUBLICATIONS:

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