Incidence and Distribution of Peach Mosaic and Its Vector, *Eriophyes insidiosus* (Acari:Eriophyidae) in Mexico

G. N. OLDFIELD and R. CREAMER, Department of Plant Pathology; C. GISPERT, Department of Entomology, University of California, Riverside 92521; F. OSORIO and R. RODRIGUEZ, Colegio de Postgraduados, Montecillo, Mexico 56230; THOMAS M. PERRING, Department of Entomology, University of California, Riverside 92521

ABSTRACT

Oldfield, G. N., Creamer, R., Gispert, C., Osorio, F., Rodriguez, R., and Perring, T. M. 1995. Incidence and distribution of peach mosaic and its vector, *Eriophyes insidiosus* (Acari:Eriophyidae) in Mexico. Plant Dis. 79:186-189.

Foliar and fruit symptoms of peach mosaic disease were observed in peach trees during a study of commercial peach orchards and dooryard peaches in 11 states in Mexico during 1991–1993. Symptomatic trees were discovered in the central highlands of Mexico extending from northern Chihuahua southward to Michoacan and Puebla. Greenhouse-grown Rio Oso Gem seedlings grafted with pieces of bark collected from symptomatic peach trees from seven states developed foliar symptoms of mosaic. The bud-inhabiting mite vector of the mosaic virus, *Eriophyes insidiosus,* was discovered for the first time in Mexico from buds of wild *Prunus munsoniana* in Chihuahua and from buds of criollo peaches in the central highland peach-growing areas. On criollo peaches, mites were commonly found in unopened buds along small branches distributed throughout the canopy of the tree, rather than in the adventitious buds typically found on infested commercial varieties of peach in the U.S. Mites from two central Mexican states were naturally inoculative based on positive transmission to greenhouse-grown peach seedlings. An isolate of the pathogen from southern California that causes severe symptoms on Rio Oso Gem peach was transmitted by naturally inoculative field-collected mites to greenhouse-grown criollo rojo, criollo blanco, and criollo naranja seedlings, causing symptoms similar to those found on criollos in the field.

Additional keyword: *Prunus serotina capuli*

Peach mosaic first was reported as a new virus disease in Texas in 1932 (5). Shown to be graft transmissible, it soon was reported in Colorado (1) and southern California (10,12). The disease has been found throughout the peach-growing states of Texas, Arizona, Utah, New Mexico, Oklahoma, and Arkansas (9), but is limited in California to areas south of the Tehachapi Mountains (11). As a result of the disease, nearly one million peach trees have been removed over the course of several decades (L. C. Cochran, *personal communication*).

In the mid-1950s, Wilson and coworkers discovered a previously undescribed species of eriophyid mite in buds of trees affected by the peach mosaic disease in southern California and demonstrated that it was a vector of the peach mosaic pathogen (13). *Prunus munsoniana* W. Wight and Hedr. and several other native plum species planted in a plot near Riverside, California, were among the first *Prunus* spp. to be identified as hosts of the eriophyid mite (13), and these native species were susceptible to infection by peach mosaic (3).

The mite, *Eriophyes insidiosus* Keifer & Wilson (8), was found subsequently on peach and on several native species of plum in other areas of the southwestern U.S. (9). In areas of the U.S. where this vector infests freestone peaches, the mite usually is limited to adventitious buds on the trunk or on the lower scaffold branches. Infested buds are swollen and reddened, and growth remains retarded; buds eventually die (G. N. Oldfield, *unpublished*).

The causal agent of peach mosaic has not been successfully purified; however, it is theorized to be viral, because it is transmitted readily by grafting. Grafting is the only method currently used to detect peach mosaic. Cherry mottle leaf virus, which is transmitted by *Eriophyes inaequalis* Wilson and Oldfield (9), a close relative of *E. insidiosus,* recently was characterized as a closterovirus (6). Cherries, the primary host of cherry mottle leaf virus, are not a host of peach mosaic (3).

Peaches constitute an important crop in Mexico with approximately 40,500 ha in production (S. Perez, *personal communication*). Over 80% of the peaches grown are criollo types, which have been grown in Mexico for hundreds of years. Criollos are clingstones having various colors of flesh, and are referred to as *blanco* (white), *amarillo* (yellow), *naranja* (orange), or *rojo* (red) and may be produced either from seed or by grafting. Criollo peach production is based in the highlands of central Mexico, with the largest acreage in the state of Zacatecas. A much smaller area, over 2,500 ha, is planted to freestone peaches, primarily in the state of Chihuahua.

In Mexico, mosaic symptoms were reported on peach trees in the states of Chihuahua, Coahuila, and Baja California by the early 1950s (2). During the past 20 yr, U.S. agricultural scientists familiar with the disease visited the commercial peach-growing area around Nuevo Casas Grandes, Chihuahua, and observed numerous trees with peach mosaic-like symptoms (L. S. Jones and H. Larsen, *personal communications*). Indigenous hosts of the peach mosaic pathogen or vector have not been identified in Mexico. During 1991 and 1992, the authors conducted a study in the peach-growing areas of Mexico to *(1)* obtain the first direct evidence for the presence of peach mosaic disease, *(2)* estimate the geographic distribution and severity of the disease in various peach-growing areas, and *(3)* document the presence and distribution of the vector mite. This paper reports the first documentation of the eriophyid mite vector of peach mosaic in Mexico, the distribution of the mite in several peach-growing states, and the first verification of peach mosaic disease in Chihuahua and the central highlands of Mexico.

MATERIALS AND METHODS

Field evaluations. Trees in commercial peach orchards and dooryard peach trees were randomly selected and inspected for symptoms of peach mosaic in 11 Mexican states during 1991, 1992, and 1993 (Fig. 1). The orchards also were surveyed for the vector mite by inspecting trees for swollen, retarded, adventitious buds on the main trunk, on the lower main scaffold branches, and on smaller branches. In a 20-yr-old orchard in Chihuahua planted to Jefferson, Rio Oso Gem, Loring, and Redskin trees, disease incidence was investigated by systematically inspecting leaves and fruit of approximately 160 trees of each variety.

Buds from peach trees at each location were inspected for vector mites in the field with a 20× hand lens, and in the laboratory with a stereoscopic microscope. Wild *P. munsoniana* located near several peach orchards in northern Chihuahua also were inspected for mosaic symptoms and for signs of the vector mite. Three twig cuttings (20–25 cm long) were collected from each of 40 plants over two locations near Nuevo

Accepted for publication 4 November 1994.

Casas Grandes and inspected for *E. insidiosus*.

Greenhouse experiments. Criollo seeds of three flesh types, blanco, naranja and rojo, were obtained from fresh fruit, vernalized at 2 C for 4–8 wk in damp peat moss, germinated, and held for 1 wk prior to inoculation. *Prunus serotina capuli* (Cavanilles) McVaugh, a wild cherry widely distributed in central Mexican highlands, was found growing near mosaic-infected criollo orchards. Therefore, this wild *Prunus* was suspected as an alternate host of peach mosaic. Seeds of *P. serotina capuli* were germinated in the greenhouse and treated similarly to the criollos.

The presence of the peach mosaic pathogen was tested by bioassay using the mite vector or grafting to a susceptible host. Grafting was done using patches of bark from symptomatic trees onto 1-yr-old Rio Oso Gem seedlings. Bud mites found at various locations were tested for inoculativity by transferring them to the growing tip of newly germinated Rio Oso Gem peach seedlings using a transfer tool consisting of a human eyelash attached to a wooden probe (13). Mites were allowed to feed until they died, approximately 2 days after transferring. Inoculated seedlings showed symptoms within a minimum of 7 days, but were observed for symptom development for one growing season (approximately 3 mo). Mites were cleared and mounted on microscope slides according to Keifer (7) and their identities were determined by phase contrast microscopy.

In order to determine the effect of a severe isolate of the mosaic pathogen on criollos and *P. serotina capuli,* seedlings were inoculated with the "Chino" isolate of the pathogen using mites (approximately 25 mites per plant) from infected flowering peach trees in San Bernardino County, CA. *Prunus serotina capuli* also was tested for susceptibility to mosaic by grafting pieces of bark from peach trees infected with the "Chino" isolate, or an isolate from symptomatic freestone peach in central Mexico.

Serological detection. Indirect enzyme-linked immunosorbent assay was done following the procedure of Creamer (4) using monoclonal antisera to cherry mottle leaf virus obtained from D. James (6). This monoclonal was utilized because of the close relationship between the mite vectors of the two pathogens. Monoclonal antisera was used at a 1:2,000 dilution and alkaline phosphatase/conjugated rabbit anti-mouse IgG (Sigma Chemical Co., St. Louis, MO) was used at a 1:2,000 dilution.

Total protein extracts were prepared as reported previously (4). Western blot analysis was done by transferring sodium dodecyl sulfate–polyacrylamide gel electrophoresis gels to Immobilon-P membrane (Millipore, Bedford, MA), except that membranes were blocked with 5% nonfat dry milk (4).

RESULTS

Symptoms of peach mosaic were observed on freestone peaches in Chihuahua and on criollo varieties in seven states in central Mexico (Fig. 1, Table 1). While disease incidence differed in various orchards, a tree by tree inspection of one orchard in Chihuahua indicated that 49% of the trees were infected. The incidence of the peach mosaic disease in four freestone varieties in this 20-yr-old orchard is shown in Table 2. Many of the standing Rio Oso Gems trees, which appeared to be the variety with the most severe disease symptoms, were stunted, with only one or two severely damaged branches remaining.

Symptoms of peach mosaic varied between freestone and criollo peach types and between orchards. Generally, symptoms of peach mosaic on freestone peaches in northern Chihuahua included tattering, mosaic, and decrease in size of leaves, shortened internodes, and small bumpy fruit, whereas, on criollo varieties in central Mexico, milder symptoms were observed on leaves and fruit, and shortened internodes were not as prevalent. The mild symptoms in criollos precluded an accurate tree by tree inspection based solely on symptoms. In an experimental block in an orchard in Aguascalientes in which criollos had been grafted with buds of freestone varieties, the freestone branches exhibited clear mosaic symptoms, while the criollos to which they were grafted were virtually free of symptoms.

To confirm that the trees surveyed were infected with peach mosaic, bark from symptomatic freestone and criollo trees from seven states was grafted onto Rio Oso Gem seedlings grown in the greenhouse. The bark grafts induced characteristic vein clearing within 1–2 wk, and typical mosaic symptoms within a month on each potted tree. Generally,

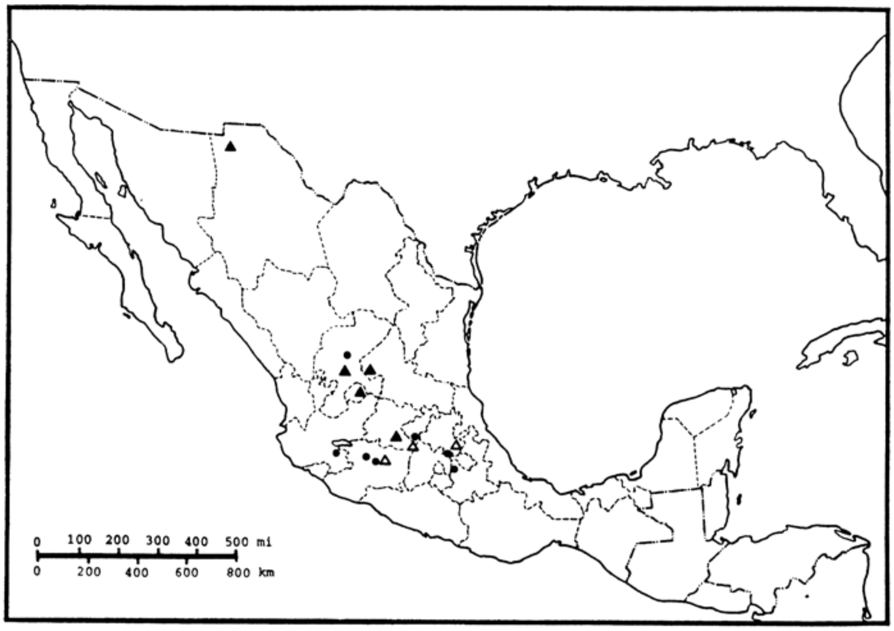

Fig. 1. Map of Mexico showing states where peach mosaic and its vector mite were found. Locations where peach mosaic was found (△). Locations where *Eriophyes insidiosus* was collected (●). Locations where peach mosaic and its vector mite were found (▲).

Table 1. Sites evaluated for peach mosiac and its mite vector in Mexico[a]

		Number of positive sites/ number sites sampled	
State sampled	**Variety evaluated**	**Peach mosaic infection**	***Eriophyes insidiosus***
Aguascalientes	Criollo	2/2	1/2
Chihuahua	Freestone peaches	3/6	0/6
Guanajuato	Criollo	1/1	1/1
Hidalgo	Criollo	2/4	0/4
Jalisco	Criollo	0/1	1/1
Mexico	Criollo	0/1	1/1
Michoacan	Criollo	2/4	3/4
Puebla	Criollo	0/1	1/1
Queretaro	Criollo	0/2	1/2
San Luis Potosi	Criollo	1/1	1/2
Zacatecas	Criollo	3/6	4/6

[a] Site indicates a distinct peach-growing property.

however, isolates from central Mexican criollos caused mild symptoms on Rio Oso Gem seedlings to which they were transmitted by grafting or by naturally inoculative mites. Further confirmation that trees were infected with peach mosaic was obtained by enzyme-linked immunosorbent assay (Table 3) and Western blot (Fig. 2) of grafted trees. In extracts from peach mosaic–infected plants inoculated by mites or grafting, the cherry mottle leaf antisera identified a protein band of approximately 22 kD, which co-migrated with a band from cherry mottle leaf virus–infected cherries. No protein bands of similar size were detected from healthy peach or cherry leaves (Fig. 2).

Although criollo peaches did not show especially strong symptoms of peach mosaic, passage through this peach type did not appear to attenuate the intensity of severe isolates. Criollo blancos, naranjas, and rojos grown from seed and inoculated using the mite vector with the severe "Chino" (California) isolate of mosaic developed mild, ephemeral symptoms of vein clearing on a few leaves, although the concentration of virus was similar to that found on Rio Oso Gem when inoculated with the same isolate (Table 3). When healthy Rio Oso Gems seedlings were back-inoculated by grafting with bark from the infected criollos, severe symptoms developed.

Table 2. Incidence of peach mosaic in a 20-yr-old freestone peach orchard in Nuevo Casas Grandes, Chihuahua[a]

Variety	No. of trees evaluated	% trees symptomatic
Loring	77	74
	82	66
Redskin	71	62
	65	18
Jefferson	77	44
	78	32
Rio Oso Gem	50	50
	58	38

[a]Two rows of each variety were inspected in June, 1991.

The mite vector was found in buds of various types of criollo peach at 14 locations within nine states of the central highlands of Mexico, including several of the sites where peach mosaic symptoms were observed (Fig. 1). However, swollen adventitious buds (symptomatic of mite infestation on freestone peaches in California) were not found on peaches in Chihuahua, and the mite was not detected in buds that were inspected. Mite-infested buds of criollos in most of central Mexico did not exhibit swelling or reddened bud scales, but buds infested with mites were distributed throughout trees on branches and twigs of various ages. All stages (adults, nymphs, and eggs) of the mite were found in criollos on all collection dates from April to December. Mites collected from criollo trees in Zacatecas and Guanajuato during June 1992 transmitted the naturally occurring mosaic isolate when allowed an inoculation access feeding upon Rio Oso Gem seedlings.

Table 3. Enzyme-linked immunosorbent assay analysis of peach mosaic isolates from Mexico and California[a]

Isolate	Isolate origin	Test host	A_{405nm}
Chino	California	Criollo	0.11
Criollo	San Luis Potosi, Mexico	Rio Oso Gem	0.07
Chino	California	Rio Oso Gem	0.08
Healthy peach		Rio Oso Gem	0.02

[a]Absorbance readings three times greater than that for healthy peach were considered positive for peach mosaic.

Wild *Prunus* spp. were surveyed for peach mosaic and its vector. Of the over 1,200 *P. munsoniana* buds collected from northern Chihuahua, only 10 buds were infested with *E. insidiosus*. These buds were collected from two branches of a single tree that exhibited shortened internodes (Fig. 3) and was growing adjacent to a freestone orchard with a high incidence of mosaic. Trees with normal internode length were not infested with mites. Although the infested buds of *P. munsoniana* did not exhibit obvious signs of mites, they were inhabited by numerous adults, nymphs, and eggs of the vector.

Inspection of over 100 buds from eight *P. serotina capuli* plants growing near mosaic-infected orchards in Hidalgo and Michoacan failed to reveal any *E. insidiosus*. The *P. serotina capuli* plants did not show symptoms of peach mosaic and the disease could not be recovered from the plants by grafting patches of bark to Rio Oso Gem seedlings.

DISCUSSION

The successful graft transmission of mosaic from trees collected from several states in southern Mexico and the transmission of naturally occurring isolates of mosaic pathogen harbored by mites from the states of Zacatecas and Guanajuato clearly demonstrate that peach mosaic is widespread in the Mexican highlands, the region where most peach production is centered. In addition, this documentation of *E. insidiosus* from *P. munsoniana* is the first evidence for the presence of the mite vector of peach mosaic in Mexico.

The discovery of *E. insidiosus* on wild *P. munsoniana* plants near mosaic-infected orchards in Chihuahua is significant. While we found mites on a low percentage of buds sampled, the large numbers of mites found on these buds

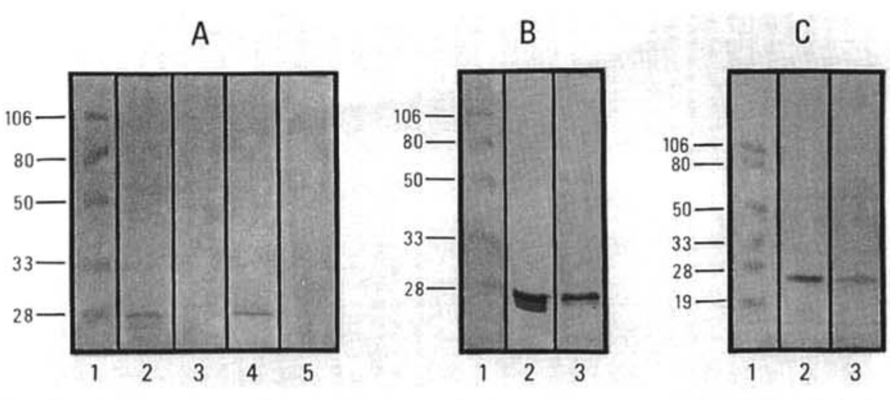

Fig. 2. Western blot analysis of proteins from peach mosaic and cherry mottle leaf infected trees. Lane 1 from all blots show molecular weight size standards indicated in kDa. **(A)** Lane 2, peach infected with "Chino" (California) isolate; Lane 3, healthy peach; Lane 4, cherry infected with cherry mottle leaf; Lane 5, healthy cherry. **(B)** Lane 2, peach infected with "Chino" (California) isolate; Lane 3, peach inoculated by *Eriophyes insidiosus* found on criollo peach in Zacatecas. **(C)** Lanes 2 and 3, peach grafted with "Redskin" var. peach from Chihuahua.

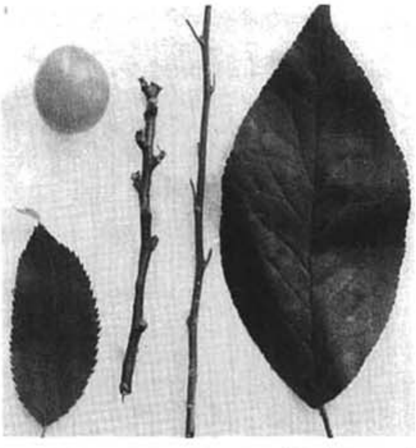

Fig. 3. *Prunus munsoniana* with shortened internodes and buds in which *Eriophyes insidiosus* was found in Chihuahua in 1991. The stunted leaf, twig, and fruit (left) are from an infested plant compared with an uninfested twig and leaf (right).

and the high efficiency with which mites can transmit peach mosaic (C. Gispert, *unpublished*) suggest that this wild host may play a role in the epidemiology of peach mosaic in this area. In the southern highlands, however, *P. serotina capuli*, the only known wild *Prunus* relative of peach, was not infected with the peach mosaic pathogen, nor was it infested with the mite vector. While it is likely that peach mosaic is transmitted naturally by *E. insidiosus* between peach trees in Mexico, as it is in California, the roles of *P. munsoniana* and *P. serotina capuli* in the epidemiology of peach mosaic may differ and should be elucidated further.

Although we failed to find the vector on mosaic-infected freestones in Chihuahua, mosaic has spread in southern California by *E. insidiosus* existing at population levels that were nearly undetectable and where no known wild hosts of mosaic or the vector exist. Considering the frequency with which we encountered the vector on criollos, any attempts to manage mite-mediated transmission of the virus would require reducing vector populations as well as inoculum sources. Eliminating mosaic and vectors from primary stock trees is of critical importance in preventing initial introduction of the pathogen and vector.

The demonstration that the peach mosaic isolates described here from central Mexico produce only mild symptoms on Rio Oso Gem seedlings, that peach mosaic–infected criollos appeared to be mildly symptomatic in the field, and that experimentally inoculated criollos of the three flesh types were only minimally symptomatic when inoculated with a severe California isolate, suggests that criollos may be less affected by peach mosaic than the freestone variety tested and might be somewhat tolerant to the disease. Previous studies showed that several other clingstone peach cultivars grown in the U.S. do not develop severe symptoms when inoculated with some U.S. isolates of the mosaic virus (3). A systematic study of peach mosaic pathogen isolates and the pathogenicity on many commercially important criollo selections is needed.

Our observations suggest that criollos may play a more important role in the epidemiology of peach mosaic than do freestone peaches. Criollos support higher populations of the vector. Criollo trees that we inspected possessed many buds that were infested with vector mites; at most, only a few mite-infested buds have ever been found on single freestone peach trees.

The fact that peach mosaic reacts specifically with a monoclonal antibody made to cherry mottle leaf virus suggests that the two pathogens are closely related. The approximately 22-kD band identified through Western blot analysis is similar in size to that identified for cherry mottle leaf virus (6). While we are in the process of characterizing the peach mosaic pathogen further, the identification of peach mosaic with cherry mottle leaf antisera gives us a useful interim tool by which to study peach mosaic, and could also be useful for development of virus-free stock plants.

After acceptance of this paper, the authors found *E. insidiosus* in buds of a mosaic-symptomatic peach tree growing in Sonora, Mexico. Mites from this tree transmitted peach mosaic to peach seedlings.

ACKNOWLEDGMENTS

Financial assistance from UCR-Mexico is gratefully acknowledged. We thank Salvador Perez, CENID-COMES, San Jose Iturbide, Guanajuato, for providing us with valuable information on the peach industry in Mexico, and for assistance in contacting members of the Mexican peach industry. Thanks are also extended to Tony Hatch, Extension Pomologist, Utah State University, and the several members of Empacadora Paquime, Nuevo Casas Grandes, Chihuahua, including Ron Walser, Fred Hatch, Merriner Jones, and Ron Belnap, for their help in conducting the studies in northern Chihuahua. We thank Delano James for monoclonal antisera to cherry mottle leaf virus.

LITERATURE CITED

1. Bodine, E. W. 1934. Occurrence of peach mosaic in Colorado. Plant Disease Rep. 18:132.
2. Cavanagh, A. H., and Rothe, C. H. 1953. Phony peach and peach mosaic. Pages 681-684 in: Plant Diseases - The Yearbook of Agriculture. USDA, Washington, DC.
3. Cochran, L. C., and Pine, T. S. 1958. Present status of information on host range and host reactions to peach mosaic virus. Plant Disease Rep. 42:1225-1228.
4. Creamer, R. 1992. Purification and protein characterization of sorghum stunt mosaic rhabdovirus. Phytopathology 82:1473-1476.
5. Hutchins, L. M. 1932. Peach mosaic - A new virus disease. Science 76:123.
6. James, D., and Mukerji, S. 1993. Mechanical transmission, identification, and characterization of a virus associated with mottle leaf in cherry. Plant Disease 77:271-275.
7. Keifer, H. H. 1952. Eriophyid Studies. Bull. Calif. Dep. Agric. 41:65-74.
8. Keifer, H. H. and Wilson, N. S. 1955. A new species of eriophyid mite responsible for the vection of peach mosaic virus. Bull. Calif. Dep. Agric. 44:145-146.
9. Oldfield, G. N. 1970. Mite transmission of plant viruses. Annu. Rev. Entomol. 15:343-380.
10. Reed, H. S. and Thornberry, H. H. 1935. A peach tree disease recently discovered in California. Phytopathology 25:897.
11. Stout, G. L. 1937. Peach Mosaic. Bull. Calif. Dep. Agric. 28:177-200.
12. Thornberry, H. H. 1935. Peach mosaic in southern California. Pages 27-30 in: Riverside County, Cal. Farm Bur. Monthly, December.
13. Wilson, N. S., Jones, L. S., and Cochran, L. C. 1955. An eriophyid mite vector of the peach mosaic virus. Plant Disease Rep. 39:889-892.