Stimulation of Growth and Yield of Virus-Infected Cantaloupe with Pyrethroids

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ABSTRACT

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Application of pyrethroid insecticides to field-grown cantaloupes had a significant impact on the number of fruit produced per plant but did not influence the rate at which cantaloupes were infected with aphid-transmitted zucchini yellow mosaic potyvirus (ZYMV). Plots that received permethrin had significantly more fruit per plant than control plots, whereas treatment with fenvalerate did not affect yield. In a greenhouse experiment, noninfected cantaloupe plants and plants infected with ZYMV during early vegetative growth, perfect flower production, and fruit set were treated with the same pyrethroid insecticides. Plant response to these treatments was measured as number of perfect flowers, plant weight, fruit weight, and number of fruit. Plants treated with permethrin produced a higher average fruit weight than plants that were not treated. All growth parameters were significantly lower among control plants infected with ZYMV during the vegetative stage; however, the first three were significantly increased by treatment with fenvalerate at that growth stage. These experiments suggest that pyrethroids have a stimulatory effect on plants infected with virus, which is manifested in increased plant growth and yield.

Aphid-vectored viruses, particularly zucchini yellow mosaic potyvirus (ZYMV), have caused substantial yield losses in spring-planted cucurbits in the desert southwestern United States for many years (4,10,24–26,29). In Imperial County, California, ZYMV was responsible for most of the 40–50% reduction in average production of the nearly 7,500 ha of melons (*Cucumis melo* L.) grown in the spring of 1984 (25). In the spring of 1990, estimated loss in this area was in excess of $22 million (29).

Nonpersistently transmitted viruses typically can be acquired and inoculated in a matter of minutes (13, 27), usually less time than that required for a toxic material to kill the insect vector (6,20). However, synthetic pyrethroids have been reported to be effective in reducing the impact of several nonpersistently transmitted viruses (1,3,22,28,31,33,36) especially when mixed with oils (14,15, 22,30). There are several hypotheses proposed for virus disease reduction through the use of pyrethroids. First, the pesticide may cause rapid aphid knock down or mortality prior to virus transmission (3,5,16,21,33). Second, pyrethroids may repel insects (17,33,34), although this action can cause increased aphid activity, actually resulting in increased virus spread (21,23). Third, pyrethroids reduce the probing times of aphids (3,21,36), which may reduce transmission efficiency. An additional mode of action that has not yet been addressed is the possible direct influence of pyrethroids on the plant and its response to virus infection.

We evaluated the impact of two pyrethroids on ZYMV disease incidence in the field and their effect on infected and noninfected cantaloupe plants in the greenhouse. In this paper we describe a direct positive response in the productivity of plants infected with ZYMV when treated with pyrethroids.

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MATERIALS AND METHODS

Field study. Research was conducted at the Imperial Valley Desert Research and Extension Center located in Holtville (Imperial County), California. Cantaloupe (*Cucumis melo* L. 'Topmark') was planted on 1.52-m center-raised beds and furrow irrigated. Preplant fertilization consisted of 202 kg/ha of P_2O_5 and 180 kg/ha of N. At emergence, plants were thinned to 30-cm spacing within rows. These and other practices standard for the region were followed.

Plots (five rows by 50 m) were arranged in a randomized complete block design, and four replicates were used. At 50% emergence, insecticide treatment was initiated. Fenvalerate (Pydrin 2.4 EC, E.I. Du Pont de Nemours & Co.) and permethrin (Pounce 3.2 EC, FMC Agriculture Chemicals Group) were applied at a rate of 0.17 kg a.i./ha with a CO_2-driven backpack sprayer calibrated to deliver 61.27 L of mixture per hectare. Applications were made twice weekly, so that coverage of new plant growth could be maximized. A nonsprayed control served as a third treatment.

Each week 50 plants from the middle three rows of each plot were evaluated visually for mosaic symptoms; plants were selected randomly each week from emergence through harvest. Just before harvest, the two youngest fully expanded leaves on the growing tip of the primary branch were collected from 10 randomly chosen plants that exhibited mosaic symptoms in each plot. These samples were tested for the presence of ZYMV, watermelon mosaic potyvirus (WMV), cucumber mosaic cucumovirus (CMV), and papaya ringspot potyvirus type W (PRSV-W) with indirect enzyme-linked immunosorbent assay (ELISA) following the procedures outlined by Fereres et al (13). Positive controls were zucchini squash (*Cucurbita pepo* L. 'Chefini') leaves infected with isolates of the respective viruses.

Yields were determined by harvesting all fruit from a 12-m section of two of the middle rows of each plot. Fruit were counted and weighed, and the number of plants in each 12-m section was counted. The total number of fruit and the fruit weight per plot were divided by the number of plants in the plot to give an average production per plant. Analyses of variance were conducted on the total number of fruit per plant, the average fruit weight, and the weight of fruit per plant.

Greenhouse study. In a greenhouse at the University of California, Riverside, 16-day-old cantaloupe (cv. Topmark) seedlings with one true leaf were transplanted into 7.57-L pots and watered and fertilized daily with a modified Ward's (37) solution. As plants grew, they were trellised on a supporting rod inserted into the soil of each pot. Greenhouse conditions were maintained between 20 and 27 C and 40 and 90% RH. Plants were hand-pollinated daily by rubbing staminate flowers from noninfected, nonsprayed plants over the stigma of perfect flowers of plants in all treatments. To maintain an insect-free environment, the greenhouse was fumigated with dichlorvos (Vapona, Shell International

Chemical Co.) 30 days and 65 days after transplanting.

A completely randomized design with a 4 × 3 factorial arrangement was used with four replicates of each treatment. Treatments consisted of inoculation time, which included four levels (noninoculated control, and inoculation at 5- to 10-leaf vegetative stage, perfect flower stage, and first fruit set stage), and insecticide application, which included three levels (nonsprayed control, fenvalerate, and permethrin).

An Imperial Valley, California, strain ZYMV culture established in 1987 and maintained in squash by mechanical and aphid transmission was used as the inoculum source. ZYMV inoculum was obtained by grinding the infected leaves in deionized water with a mortar and pestle, and plants dusted with carborundum were lightly rubbed with a pestle dipped in the slurry.

Fenvalerate (Pydrin 2.4 EC) and permethrin (Pounce 3.2 EC) were applied at a rate of 0.17 kg a.i./ha with a CO_2-driven backpack sprayer after moving the plants to a separate room of the greenhouse. Applications were made approximately every other week on days 37, 51, 63, and 81 after transplanting.

Growth variables measured for each plant included the total number of perfect flowers produced, the dry weight of vegetative plant material, the total number of fruit produced, and the total fruit weight. Harvest took place 115 days after transplanting and consisted of counting and weighing each fruit on each plant (subsequently calculating the total fruit weight per plant). After harvest, the plants were excised from the roots at the soil line, placed in paper bags, and allowed to dry. Following drying, the plants were weighed to determine total vegetative biomass production.

Analyses of variance of data from the field and greenhouse experiments were conducted using the PROC GLM procedure of SAS (35).

RESULTS AND DISCUSSION

Field experiment. No CMV or PRSV-W was detected by ELISA from any of the leaf samples. Infection with ZYMV averaged 85, 56, and 64%, respectively, in permethrin, fenvalerate, and control plots, whereas WMV was detected in 43, 49, and 57% of the samples from those treatments. Samples harboring mixed infections, included in the above percentages, were found at rates of 38, 34, and 39%. There were no significant ($P > 0.05$) differences in these average percentages between treatments; therefore, visual incidence data for both viruses were pooled for treatment comparison.

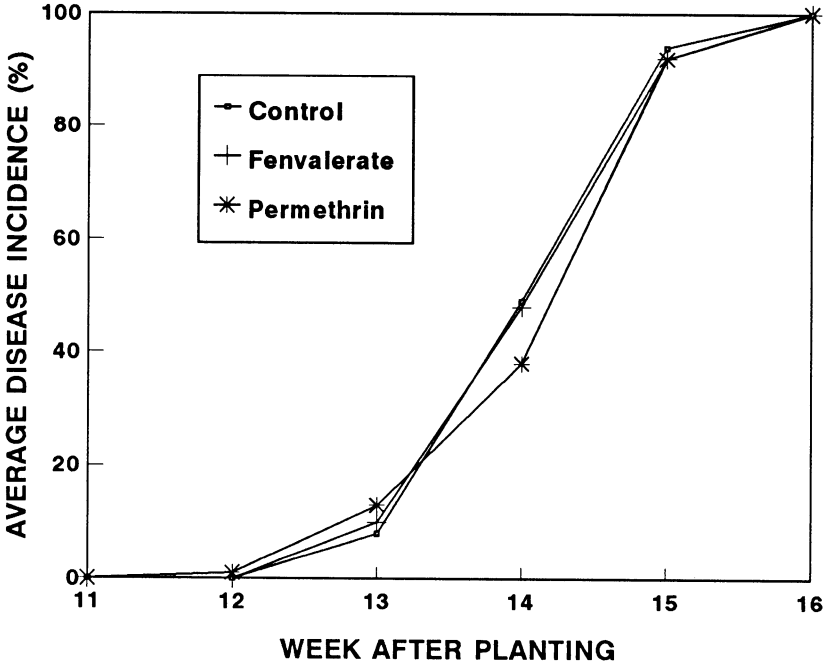

Fig. 1. Average percent disease for control plots and plots sprayed with fenvalerate and permethrin in the field study.

Table 1. Average per plant effect of pyrethroids on the growth and yield of cantaloupe plants in the greenhouse after inoculation with ZYMV[y]

Treatment	Flowers (no.)	Plant weight (g)	Fruit (no.)	Fruit weight per plant (g)
Control	7.8 a[z]	52.0 a	0.60 a	241.8 b
Fenvalerate	13.3 a	62.4 a	1.07 a	436.1 ab
Permethrin	13.1 a	52.5 a	1.07 a	463.9 a

[y] Results were averaged across inoculation times.

[z] Means within columns followed by the same letter are not significantly different ($P > 0.05$) according to Duncan's multiple range test (11).

Even though insecticides were sprayed twice weekly, there was no reduction in disease incidence compared with control plots (Fig. 1). Disease incidence increased slightly during the 12th week after planting and rose sharply from this date to 100% infection by the 16th week of the study (Fig. 1).

Although there were no differences in disease incidence between treatments, there were differences in yield. These differences were not due to differential insecticidal activity in the treatments, because weekly inspection of the plants revealed no pest infestations in any of the plots. The total number of fruit per plant was significantly higher ($P < 0.05$) in the plots sprayed with permethrin (4.6 fruit) compared with the control plots (3.6 fruit); the yield of fenvalerate-treated plants (3.8 fruit) was statistically similar to both of the other treatments. There were no differences ($P > 0.827$) in average weight per fruit between treatments (1,037.9, 1,018.0, and 1,000.6 g, respectively, in permethrin, fenvalerate, and control); however, there were indications ($P < 0.111$) of spray treatment enhancement of total fruit weight per plant (3,293.0, 2,790.2, and 2,739.4 g for permethrin, fenvalerate, and control).

Greenhouse experiment. There were no significant interactions between levels of insecticide and inoculation times for any of the variables measured ($P > 0.338$ or higher); therefore, contributions of insecticide treatment and inoculation time could be evaluated separately. There were statistical differences ($P < 0.05$) between insecticide treatments in the average fruit weight per plant across inoculation treatments (Table 1). The fruit weights of plants treated with permethrin were significantly higher than the weights from the nontreated control plants, whereas the fenvalerate-treated plant yield was not significantly different from the other treatments. There were no statistical differences in the average number of perfect flowers produced, the average plant weight, or the total number of fruit per plant between treatments when averaged across all inoculation times (Table 1).

Averaged over insecticide treatments, plants inoculated in the vegetative stage produced fewer flowers and lower plant weights than noninfected controls or plants inoculated at perfect flower or fruit set (Table 2). The average number of fruit and fruit weight per plant were higher when plants either were infected with ZYMV during fruit set or not infected.

We conducted analyses of variance

within each inoculation treatment to determine the impact of pyrethroid application on growth and productivity of plants differentially inoculated. For noninfected plants, there were no differences between treatments in plant weight, number of fruit, or fruit weight per plant (Table 3). However, the average number of flowers produced on permethrin-treated plants was significantly higher than that for the fenvalerate-treated and control plants.

For plants inoculated with virus during the vegetative stage, there were significantly fewer flowers and lower fruit weights in nonsprayed plants than in those receiving insecticide treatments (Table 3). Additionally, the average plant weight of control plants was significantly lower than than that of plants treated with fenvalerate. There were no treatment differences in the total number of fruit.

In plants infected at the perfect flower stage, there was a higher number of fruit produced by plants in the fenvalerate treatment than by those not sprayed. Additionally, although statistical separation between the fruit weight per plant in fenvalerate and control plots was not apparent at $P = 0.05$, there was a difference in these means at $P = 0.10$. There was no evidence for statistical differences between spray treatments in the number of flowers or the average plant weight.

The plants inoculated at fruit set had no differences among the measured variables (Table 3).

DISCUSSION

Disease curves from the field experiment (Fig. 1) indicated that pyrethroid insecticides did not prevent or slow virus spread, which contradicts most other research conducted on aphid-borne viruses (3,14,16,28,30,33,36). However, among other field-based studies (14, 16,28,30), virus control was accompanied by insecticidal control of the vectors that also infested the tested crops. *Myzus persicae* (Sulzer), the most abundant and efficient vector of ZYMV in the Imperial Valley (7), does not readily colonize cantaloupe. Thus, in addition to the absence of aphids observed in our studies, direct insecticidal control of the vector in this system is unlikely. Another possible reason for the discrepancy between our work and others' may lie in the environmental conditions present at the times of our respective studies. When virus was being inoculated to plants in our study, daytime temperatures and light conditions, which play a role in pyrethroid degradation (19), might have been more extreme than in the temperate regions of others' work, thus reducing the pyrethroids' effectiveness. Such degradation might have been exacerbated by cantaloupe plant architecture; the axial surface of foliage upon which aphids are likely to land is oriented more horizontally than in crops such as beets and potatoes.

More substantial than the direct impact of pyrethroids on virus incidence, our results, obtained under the different growing conditions of the field and greenhouse experiments, demonstrated that pyrethroids can have a positive impact on the yield of virus-infected plants. The field experiment showed that, despite similar disease incidence curves among treatments (Fig. 1), there were differences in plant production, measured by total number of fruit per plant. Similarly, across inoculation treatments in the greenhouse experiment, permethrin-treated plants had significantly higher fruit weights than did the controls (Table 1). Most interesting was the impact of pyrethroids as a function of the growth stage at which virus inoculation occurred. The influence of the pyrethroids was strongest on plants inoculated during the early vegetative stage, whereas later inoculation times resulted in reduced effect of the sprays (Table 3). Increases in yield were obtained under quite different circumstances. In addition to varied light, weather, and nutritional environments, plants were subjected to different levels of pyrethroids. Field plots were sprayed 24 times, twice weekly, while greenhouse plants received four sprays, biweekly; however, pyrethroids degrade more quickly outdoors, primarily because of increased ultraviolet light (19).

Direct positive impacts of insecticides on plant yield have been demonstrated previously. In an insect-free environment, Rao and Rao (32) showed a 40–53% yield increase when rice was treated with various organophosphate insecticides. Enhanced plant growth with pyrethroid use has been reported in cotton (8,9). In addition, Hill (18) showed that under conditions of low pest pressure, the yields of cotton, broccoli, cabbage, potatoes, and apples are consistently, but nonsignificantly, increased by pyrethroid sprays.

The direct effect of pyrethroids on plant disease has seldom been addressed. Several studies comparing pyrethroids with other classes of insecticides found that, in spite of equivalent vector control, pyrethroids were more effective in reducing virus disease (17). Authors attributed the disease reduction to changes in vector behavior and virus transmission, but the possibility of direct effects on the infected plants was not acknowledged. Atiri and Ligan (2) found that, in addition to reducing the percent

Table 2. Average per plant effect of time of ZYMV inoculation on the growth and yield of cantaloupe plants in the greenhouse[y]

Treatment	Flowers (no.)	Plant weight (g)	Fruit (no.)	Fruit weight per plant (g)
Noninfected	13.5 a[z]	61.1 a	1.00 a	462.9 a
Vegetative	6.0 b	37.2 b	0.55 b	253.6 b
Perfect flower	11.8 a	63.6 a	0.90 ab	321.3 ab
Fruit set	13.9 a	60.4 a	1.18 a	466.7 a

[y] Results were averaged across insecticide treatments.

[z] Means within columns followed by the same letter are not significantly different ($P > 0.05$) according to Duncan's multiple range test (11).

Table 3. Effect of pyrethroids on growth and yield of cantaloupe plants in the greenhouse infected with ZYMV at different growth stages

Growth stage Treatment	Flowers (no.)	Plant weight (g)	Fruit (no.)	Fruit weight per plant (g)
Noninfected				
Control	8.8 b[z]	55.0 a	0.75 a	356.0 a
Fenvalerate	12.0 b	63.3 a	1.00 a	405.3 a
Permethrin	19.8 a	65.0 a	1.25 a	627.5 a
Vegetative				
Control	1.0 b	18.5 b	0.25 a	41.0 b
Fenvalerate	8.3 a	56.3 a	0.67 a	338.0 a
Permethrin	9.3 a	41.5 ab	0.75 a	402.8 a
Perfect flower				
Control	9.7 a	80.3 a	0.33 b	141.7 a
Fenvalerate	15.3 a	58.5 a	1.50 a	504.3 a
Permethrin	9.3 a	53.7 a	0.67 ab	257.0 a
Fruit set				
Control	12.3 a	61.3 a	1.00 a	403.5 a
Fenvalerate	17.3 a	72.7 a	1.00 a	484.7 a
Permethrin	13.0 a	50.3 a	1.50 a	516.5 a

[z] Means within columns within each inoculation time followed by the same letter are not significantly different ($P > 0.05$) according to Duncan's multiple range test (11).

infection by cowpea aphid-borne mosaic virus, increased doses of cypermethrin resulted in fewer infected plants exhibiting severe symptoms. In another study, pyrethroid sprays caused foliar symptoms of potato virus Y on potato to decrease 49%, while assays of tubers revealed an actual reduction in infection of 14% (33). Such studies demonstrate the importance of not relying solely upon visual rating of infection when treatments might affect symptoms directly.

The mechanisms by which plants infected with virus respond favorably to pyrethroid application are unknown. Neither pyrethroids nor their metabolites are readily translocated within plants (19), but pyrethroids, which are lipophilic, are readily absorbed onto waxy layers on plant surfaces and then, as more polar metabolites, penetrate to the aqueous inner phases (12). In this phase, the metabolites are in close proximity to potyvirus replication (J. E. Polston, *personal communication*), but how pyrethroids affect virus or plant metabolism has not been investigated.

Researchers studying the impact of pyrethroids on disease progression of insect-vectored viruses have suggested that the mechanisms by which this occurs relate to the dynamics of arthropod vectors. The data presented in this paper indicate that pyrethroid application to plants infected with virus, especially at an early growth stage, may enhance productivity. This added benefit should be considered when researchers, through measurement of plant yield, seek to determine the impact of pyrethroid applications on insect control or arthropod-vectored plant viruses.

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