

Comparison of Sprayable and Film Mulches in Delaying the Onset of Aphid-Transmitted Virus Diseases in Zucchini Squash

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ABSTRACT

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Silver spray mulch and two silver polyethylene film mulches, applied to the planting beds before seeding, were effective in repelling alate aphids and delaying the onset of several virus diseases in spring- and fall-planted zucchini squash (*Cucurbita pepo*) in California's San Joaquin Valley. Disease symptoms in plants growing over these mulches appeared 7 to 10 days later than in plants growing on unmulched beds. In spring seeded squash, approximately 30% of the plants on unmulched beds were infected with one or more viruses by the first harvest while only 10 to 15% of those grown over the silver mulches showed virus symptoms. In fall-planted trials, 100% of the plants grown on unmulched beds, with and without insecticide applications, were virus-infected by the first harvest; less than 10% of plants grown over silver mulches were diseased at first harvest. Silver-pigmented mulches were generally more effective in repelling aphids and delaying virus onset than were white-pigmented mulches. Marketable fruit yields in the spring planting were approximately 70% higher in plots mulched with silver than the unmulched control. In the fall trial, yields of marketable fruit were 75 and 80% greater in plots mulched with silver polyester film and silver spray, respectively, than those from the unmulched control, either with or without an insecticide application. Although plants grown over the silver mulched plots eventually became infected, they continued to produce a significantly higher percentage of marketable fruit throughout most of the season than did the unmulched controls. Water-soluble, biodegradable silver sprays may be advantageous over polyethylene films because they can be incorporated into the soil at the end of the season, rather than requiring removal and disposal in a landfill.

Zucchini squash, *Cucurbita pepo* L., is susceptible to a number of viruses. Among the most important are cucumber mosaic cucumovirus (CMV) and watermelon mosaic (WMV) and zucchini yellow mosaic (ZYMV) potyvirus. All are transmitted in a stylet-borne, nonpersistent manner by several aphid species (17,26). Infected plants are stunted, yield fewer fruit than do healthy ones, and fruit are frequently misshapen or mottled, which renders them unmarketable. Insecticides offer little relief, and under certain circumstances may increase the rate of virus transmission (5,13). In California's San Joaquin Valley,

virus diseases in squash may shorten the spring harvest period and frequently cause total failure of the fall crop.

Reflective mulches have been used successfully to reduce the incidence of aphid-borne virus diseases in squash and other crops (6-8,23,28). These mulches reflect short-wave light (15,21), which confuses incoming alate aphids and reduces the incidence of their alighting on plants (30). Brown et al. (6) found silver plastic mulch superior to white, yellow, or black with yellow edges in repelling aphids in yellow crookneck summer squash. Plants grown on silver mulch produced significantly higher yields of marketable fruit than did those grown on bare soil (6). Other materials and colors used to successfully reduce virus incidence in various crops include aluminum foil (10,15,16,18,23,24,29), white plastic (15,29), aluminum powder sprayed on the soil (15), and aluminum strips painted on black plastic (20).

Plastic mulches, while effective, are not without their drawbacks. Disposal following crop termination presents particular problems. Disking merely chops the plastic into small pieces, and because most of these plastics are not biodegradable, the

pieces persist in the soil. These may blow onto adjacent property creating a nuisance and can interfere with routine farming practices in succeeding crops by fouling cultivators, planter shoes, or scraper blades. The best approach is to remove the entire mulch from the field. This can be labor intensive, however, and the problem of ultimate disposal remains. Biodegradable, water miscible spray mulches are a promising alternative to plastic films for conferring reflectivity to the soil surface. These mulches were originally developed as a possible replacement for polyethylene used in sealing soil following fumigation. They can be incorporated into soil after use, rather than requiring removal and disposal in a landfill. They may also be less expensive to use than are plastic mulches (27). We reasoned that with the addition of a silver or white pigment, this sprayable mulch might be useful in repelling aphids and delaying the onset of aphid-transmitted viruses. This paper reports the results of a study comparing biodegradable sprayable mulches with several plastic mulches, an unmulched control (bare soil), and insecticide-treated plants.

MATERIALS AND METHODS

Field site and land preparation. Two experiments were conducted, one each in the spring and fall, 1993, at the University of California Kearney Agricultural Center, Parlier, Fresno County, Calif., on a Hanford sandy loam (Typic Xerorthents) soil. Field preparation consisted of pre-irrigation followed by a broadcast application of N and P, 60 and 75 kg/ha, respectively, which was incorporated to a 15-cm depth by disking. Planting beds, 102 cm between centers, were formed with a mechanical bed shaper. Bensulide (Prefar 4-E) herbicide, 5.6 kg/ha, was applied to the beds in 189 liters of water per ha with an FMC hydraulic sprayer Model DP20 3PT (FMC Corp., Jonesboro, Ark.) equipped with Tee Jet 8004 nozzles (Spraying Systems Co., Bellwood, Ill.) and incorporated with a Culti-mulch bed shaper/tiller (B. W. Implement Co., Buttonwillow, Calif.) that incorporated the herbicide to a depth of approximately 5 cm and reshaped the beds in a single operation.

Experimental design and mulch treatments. Each plot was three beds wide

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(3.06 m) × 7.6 m long. The center row was used for all data collection; the outside rows were used as guard rows. Individual plots were separated from adjacent plots by 3.06 m of bare soil on all sides. Treatments (mulches) were arranged in a randomized complete block design with five replications. In the spring experiment, eight mulches were used: (i) silver embossed polyethylene (silver poly 1) (AEP Industries, Moonachie, N.J.); (ii) white embossed polyethylene (white poly) (AEP Industries, Moonachie, N.J.); (iii) silver-brown co-extruded polyethylene (silver poly 2) (Polyon Barkai Ltd., Israel); (iv) silver embossed polyethylene netting placed on the soil surface (silver net 1) (Specialty Ag, Reedley, Calif.); (v) silver embossed polyethylene netting suspended on wire hoops approximately 61 cm above the surface of the bed to form a tunnel (silver net 2) (Specialty Ag); (vi) silver-pigmented "Styrofoam" synthetic latex spray mulch (silver spray) (BASF Corp., Charlotte, N.C.); (vii) white "Styrofoam" synthetic latex spray mulch (white spray) (BASF Corp.); (viii) unmulched control (control). The plastic mulches were cut to the proper length, placed over the beds and anchored around the edges with soil. The silver and white spray mulches were cast by applying a primer coat of white spray mulch (1:3 aqueous dilution, approximately 2 liters/m²) with a custom-built, tractor-mounted, PTO-driven ground sprayer equipped with Tee Jet 8004 nozzles (Spraying Systems Co.) followed by a second coat of either white spray mulch or silver pigment (Standard Brands Paint Inc., Los Angeles, Calif.), the latter delivered in a 1:4 (approximately 1.78 liters/m²) dilution with mineral spirits.

In the fall experiment, the same mulches were used with one exception: a silver embossed polyester film (silver polyester) was substituted for the silver net 2 mulch. An insecticide control, consisting of plants on unmulched beds but receiving two insecticide applications (insecticide control), was added. In the latter treatment, diazinon at 0.56 kg a.i./ha was applied with a backpack sprayer equipped with 8002 Tee Jet nozzles (Spraying Systems Co.) in 75.6 liters of water per ha on 17 and 27 August.

Crop establishment and cultural practices. Holes, 7.6 cm in diameter, were cut in the mulches every 45 cm and three seeds per hill were planted 3.8 cm deep. The spring experiment was planted on 3 June (cv. Grey) and the fall experiment was planted on 3 August (cv. Sunre 9718). The field was sprinkle irrigated daily until seedlings emerged and then furrow irrigated every 5 to 7 days. Following seedling emergence, the stand was thinned to one plant per hill leaving 15 plants per plot row. After the first harvest, additional N (liquid 17-0-0) at 33.6 kg/ha was delivered in the irrigation water every 2 to 3 weeks. Weeds were controlled by applying a 2%

solution of glyphosate (Roundup) with a wick applicator 2 to 3 times during the season.

Aphid sampling. Plants were sampled weekly, beginning at the two-leaf stage, by selecting one newly expanded leaf per plant and visually counting the number of alate aphids present. Leaves were then placed in a Ziploc bag and transported to the laboratory, where the aphids were identified to species.

Virus determinations. All plants were visually rated for foliar symptoms of virus infection at the first (12 July) and fifth (22 July) harvest in the spring experiment and the first (7 September), fourth (15 September), and seventh (21 September) harvest in the fall experiment. Symptomatic leaves were selected from plants in the center data row, placed in plastic bags and returned to the laboratory for processing. Samples were subjected to enzyme-linked immunosorbent assay (ELISA) using commercial WMV, ZYMV, and CMV Patho-Screen kits (AgDia Inc., Elkhart, Ind.). Each sample was placed in two wells per plate. Samples were also evaluated for the presence of papaya ringspot virus type W (PRSV-W) and squash mosaic virus (SqMV) as described above.

Harvesting and grading. As fruit reached marketable size, plots were hand harvested approximately every other day; 9 times in the spring experiment beginning on 12 July, and 12 times in the fall experiments, beginning on 7 September. Fruit from the center data row was picked and collected at the end of each plot, where it was sorted as either marketable or unmarketable culls. Fruit were judged to fall into the latter category if they were misshapen or mottled. Fruit were weighed on an electronic FWC model DMW II

scale (Flex-Weight Corp., Santa Rosa, Calif.). Yields were calculated as kg of marketable fruit per plant.

Statistical analysis. The number of aphids, percent virus infected plants, and yield of marketable fruit were evaluated by analysis of variance (ANOVA) followed by mean separation using Duncan's multiple range test (1,11). Percent virus infected plants were transformed to arcsin (\sqrt{x}) before analysis. Relationships between cumulative yield and the harvest frequency of representative mulches were compared with the unmulched control using simple linear regression (1). Regression coefficients ($b \pm SE$) of the resulting relationships were compared using the procedures of Gomez and Gomez (14) to determine if there were significant differences between the slopes of the regression lines.

RESULTS

Aphid populations and virus incidence (spring experiment). Cotton-melon aphid (*Aphis gossypii* Glover) constituted 98% of the alate aphids recovered. Green peach aphid, *Myzus persicae* (Sulzer), and cowpea aphid, *Aphis craccivora* Koch, constituted the remainder of the species collected. On the first two sample dates (22 and 29 June), all mulch treatments significantly reduced the number of alate aphids per leaf over those in the unmulched control (Fig. 1). By the third and fourth sample dates, 6 and 13 July respectively, the number of aphids increased substantially in all treatments. The earlier repellency afforded by the net mulches (silver net 1 and silver net 2) failed at this point and aphid numbers were not significantly different ($P > 0.05$) from those in the unmulched control plots (Fig. 1). The net "tunnels" (silver net 2) had been

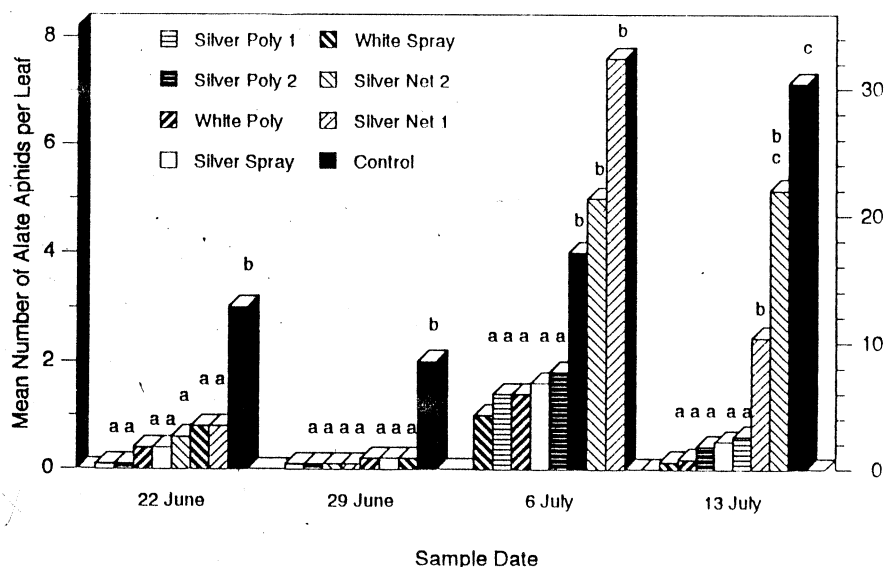


Fig. 1. The effect of reflective mulches on alighting of alate aphids on spring-planted zucchini squash plants, Parlier, Calif., 1993. Values on the right-hand vertical axis refer to the number of alate aphids per leaf in the sample taken on 13 July only. Means sharing the same letter(s) are not significantly different at $P = 0.05$ according to Duncan's multiple range test. Comparisons are valid only within individual sample dates.

removed on 3 July in preparation for the first harvest. Alate numbers in the remaining treatments remained significantly ($P < 0.05$) below those in the control and the net mulches on both sampling dates. A major aphid flight occurred between 6 and 13 July. Counts made on 13 July showed that all mulches, except silver net 1 and silver net 2, were still effective in repelling alate aphids. By 20 July, alate aphid populations in all plots had declined to near zero.

At the first harvest (12 July), all plots contained plants with symptoms of virus diseases. The mulched plots, except silver net 2, had a significantly ($P < 0.05$) lower percentage of virus infected plants, however, than did the controls (Table 1). Plots

mulched with silver poly 1, silver poly 2, and silver spray contained fewer than half the number of infected plants found in the other mulch treatments. By the fifth harvest (26 July), all plants in all plots showed virus symptoms. All of the plants gave a positive response to tests for the presence of WMV and ZYMV. None of the plants tested gave a positive response for the presence of PRSV-W, SqMV, or CMV.

Aphid populations and virus incidence (fall experiment). All alate aphids recovered in the fall trial were *A. gossypii*. The majority of the aphid activity occurred during the first two sample periods. All mulched plots had significantly ($P < 0.05$) fewer alate aphids than the insecticide

treated control or the unmulched control (Fig. 2). Silver poly 1, silver poly 2, silver spray, and silver polyester mulches were equally effective in repelling alate aphids during this period and were generally superior to the white poly, white spray and silver net 1 (Fig. 2). Aphid populations declined over the remaining two sample dates and there were no significant differences ($P > 0.05$) among treatments on either 7 or 15 September (Fig. 2).

The incidence of plants showing symptoms of virus diseases at first harvest (9 September) closely mirrored the distribution of alate aphids recovered from the respective plots during the first two sample periods. Plots mulched with silver poly 1, silver poly 2, silver spray, and silver polyester had a significantly ($P < 0.05$) lower occurrence of diseased plants (<17%) than the remaining plots (Table 1). Plots mulched with white poly, white spray, or silver net 1 had an intermediate level of virus incidence, while the insecticide-treated control plots and the unmulched controls were nearly 100% infected. By the fourth harvest (15 September), approximately 25% of the plants in the silver poly 1 and silver polyester mulched plots remained symptomless, while all other plots were at or near 100% of the plants showing symptoms. By 22 September (fifth harvest), the infection rate in all plots was near 100% and there were no significant ($P > 0.05$) differences among the treatments (Table 1). All plants gave a positive response to the tests for the presence of WMV and ZYMV and 17% gave a positive response for the presence of CMV. No PRSV-W or SqMV was detected.

Marketable fruit yield. In the spring experiment, total yields (kg marketable fruit per plant), with the exception of those from silver net 1 and 2, were significantly ($P < 0.05$) higher in all mulched plots than in the unmulched control (Fig. 3). Total seasonal yields/plant from silver poly 1 and silver poly 2 were approximately twice those of the unmulched control. Yields from the white poly and silver spray mulched plots were 40% higher than the unmulched controls. Production in the silver net 1 and 2 and the unmulched control was lower than in the remaining plots at the first harvest and remained so through the season (Fig. 3).

The cumulative yield increase over time was nearly linear. While total yields from silver poly 2 and silver spray were significantly greater than the control, the rate of yield increase in all three was approximately the same. There was no significant difference ($P > 0.05$, $F = 0.048$, $df = 2, 18$) in the slope (b) of the regression lines for silver poly 2, $b = 0.197 \pm 0.011$; silver spray, $b = 0.166 \pm 0.010$; and the unmulched control, $b = 0.116 \pm 0.007$.

In the fall experiment, plots mulched with silver spray and silver polyester produced significantly ($P < 0.05$) higher total

Table 1. Percentage of plants showing foliar virus symptoms at selected harvest dates in spring and fall seeded zucchini squash planted on reflective soil mulches. Parlier, Calif., 1993^w

Mulch treatment	Spring planting ^x		Fall planting ^x		
	1st harvest	5th harvest	1st harvest	4th harvest	7th harvest
Silver poly 1	10.2 a	100.0 a	17.2 a	74.6 a	95.2 a
Silver poly 2	9.0 a	100.0 a	6.8 a	90.4 ab	100.0 a
Silver net 2	40.4 c	100.0 a	— ^y	— ^y	— ^y
Silver spray	6.1 a	100.0 a	17.1 a	98.7 b	100.0 a
White poly	20.2 b	100.0 a	57.3 c	98.6 b	100.0 a
White spray	18.8 b	100.0 a	62.8 c	100.0 b	100.0 a
Silver net 1	23.2 b	100.0 a	36.5 b	97.3 b	100.0 a
Silver polyester	— ^y	— ^y	7.3 a	70.4 a	93.8 a
Insecticide control ^z	— ^y	— ^y	98.8 d	100.0 b	100.0 a
Control ^z	36.3 c	100.0 a	97.3 d	100.0 b	100.0 a

^w Viruses identified were cucumber mosaic cucumovirus, watermelon mosaic potyvirus, and zucchini yellow mosaic potyvirus. See text for the identity and percentage of individual viruses identified from plants in each experiment.

^x Means followed by the same letter(s) are not significantly different ($P < 0.05$) according to Duncan's multiple range test. Percentages were transformed to arcsin (\sqrt{x}) before analysis. Comparisons are valid within individual harvests only.

^y Treatment not included in this planting.

^z Unmulched (bare soil) plots.

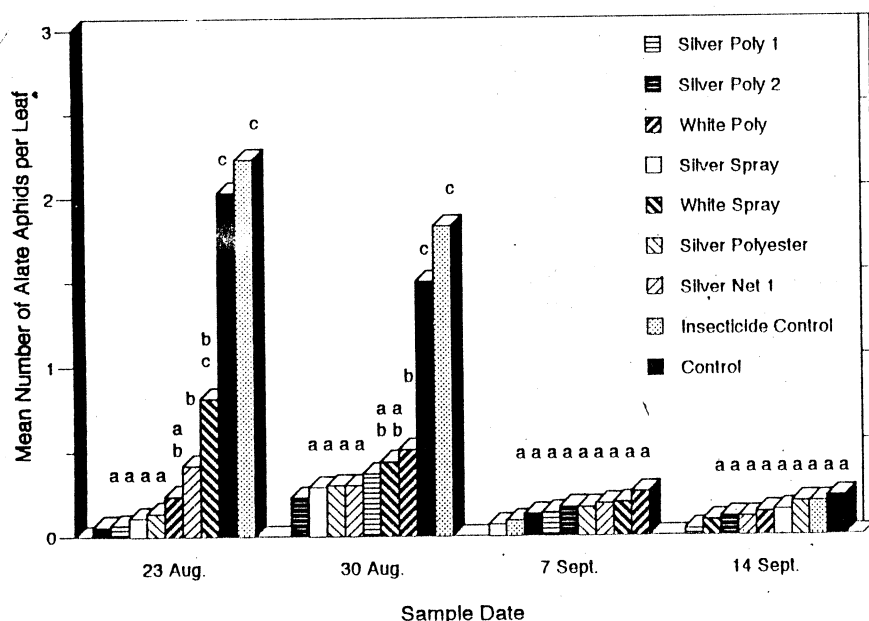


Fig. 2. The effect of reflective mulches on alighting of alate aphids on fall-planted zucchini squash plants, Parlier, Calif., 1993. Means sharing the same letter(s) are not significantly different at $P = 0.05$ according to Duncan's multiple range test. Comparisons are valid only within individual sample dates.

yields (kg/plant) than all other treatments with 80 ± 2.7 (SE) and $75 \pm 3.2\%$ more marketable fruit (kg/plant), respectively, than the unmulched control (Fig. 4). All other treatments, while less productive than the silver spray and polyester mulches, produced significantly ($P < 0.05$) greater quantities of marketable fruit than did the unmulched controls (Fig. 4). The silver netting 1 was considerably more effective in the fall experiment than it was in the spring experiment and cumulative yield was significantly ($P < 0.05$) higher than the control (Fig. 4). Although plants grown on the silver mulches eventually became infected (Table 1), they continued to produce a higher percentage of marketable fruit throughout the season than did plants grown without mulch (Fig. 5). Over the season, the percentage of marketable fruit averaged 67 ± 4.0 and $71 \pm 4.2\%$ for silver poly 2 and silver spray, respectively, while the percentage of marketable fruit for the unmulched control averaged only $24.8 \pm 10.7\%$. Two applications of insecticide to unmulched plots failed to significantly reduce alate aphid populations (Fig. 2) or delay the onset of virus infection (Table 1) and yields were equivalent to those of the unmulched, unsprayed control (Fig. 4). While cumulative yields in all mulched plots showed a steady increase, cumulative yield in the control and insecticide treated plots remained virtually flat (Fig. 4). The slope (b) of the regression lines for silver poly 2, $b = 0.099 \pm 0.009$, and silver spray, $b = 0.121 \pm 0.017$, were significantly different ($P < 0.05$, $F = 19.41$, $df = 2, 30$) from the unmulched control, $b = 0.029 \pm 0.005$.

DISCUSSION

The silver spray and silver polyethylene mulches afforded better overall repellency of incoming alates than did the silver netting or white mulches. The silver netting 2 used as a row cover (tunnels) in the spring experiment was initially as effective as the other silver mulches in repelling alate aphids but, upon its removal prior to the third sampling date, number of aphids increased rapidly to the level of the unmulched control (Fig. 1). The plants became virus infected almost immediately (Table 1) and yields in the previously net-covered plots were identical to those in the unmulched control plots (Fig. 3). Our results were similar to those of Perring et al. (25), who found that aphid populations on melons protected by row covers increased to the level of those on the uncovered control plots within 1 week following removal of the covers. Our results differ from those of Webb and Linda (28), however, who found that populations of aphids increased slowly on squash plants previously grown under row covers. In their studies, the effect of the row covers lasted for up to 14 days following removal. In plots where silver netting was used as a

soil mulch over the planting beds (spring experiment), aphid populations also increased significantly by the third sample date. In the fall experiment, aphid numbers were similar to those in the other mulch treatments throughout the sampling period. In the latter experiment, however, aphid populations in all treatments declined significantly after the second sample date. The level of virus infection at first harvest is a better indicator of mulch effectiveness than the aphid counts them-

selves. The chain of events that must occur for a host plant to become infected in nature is complex. Irwin and Ruesink (12) listed eight events that must occur before a plant becomes infected with an aphid-borne virus. Each event has its own probability of success. While we measured vector abundance, we did not determine vector propensity or vector activity. Generally, vector density only indirectly influences virus spread (12). Therefore, statistical differences in vector abundance may

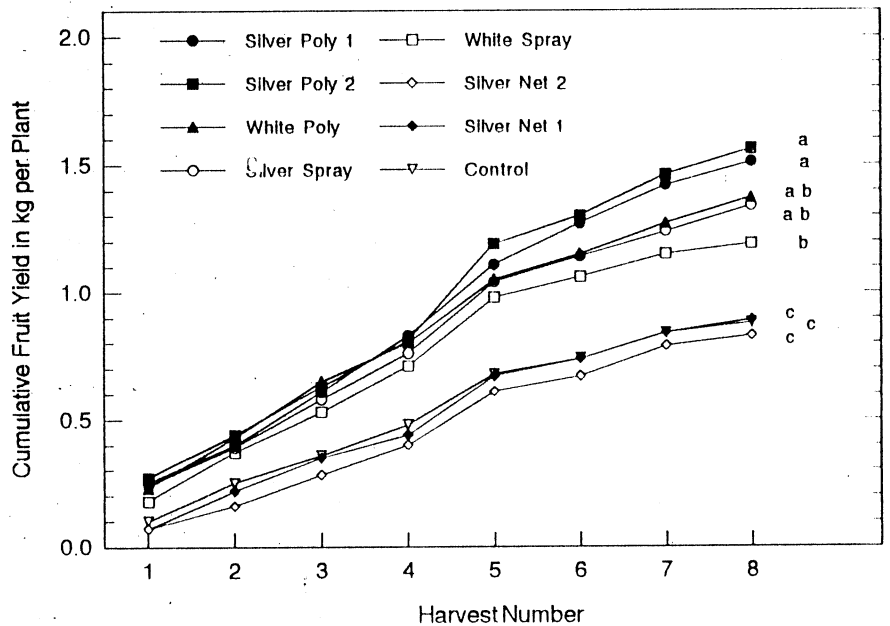


Fig. 3. Cumulative yields (kg/plant) of spring-planted zucchini squash over eight harvest dates, Parlier, Calif., 1993. Yields on harvest number eight represent total yields for the season and were evaluated by analysis of variance. Means followed by the same letter(s) are not significantly different at $P = 0.05$ according to Duncan's multiple range test.

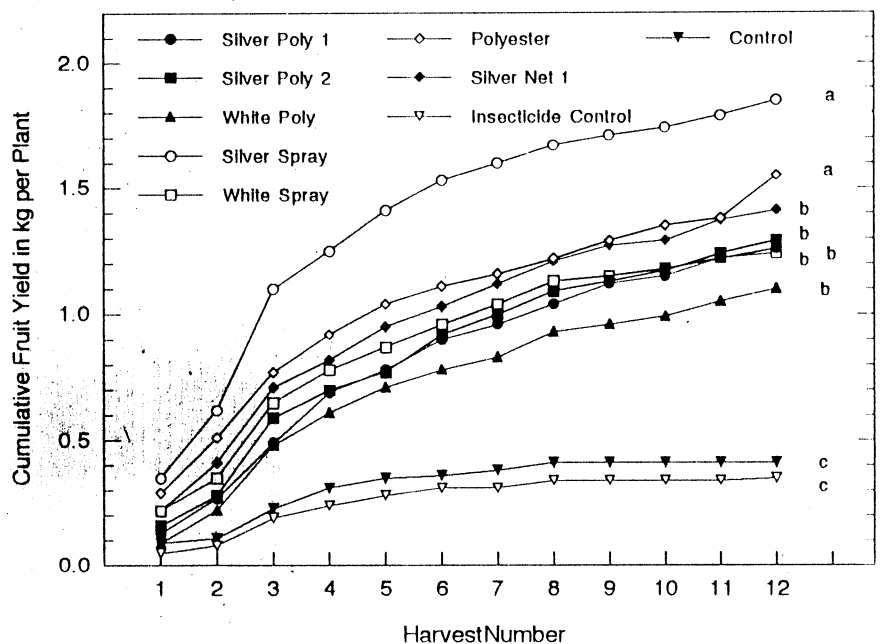


Fig. 4. Cumulative yields (kg/plant) of fall-planted zucchini squash over 12 harvest dates, Parlier, Calif., 1993. Yields on harvest number 12 represent total yields for the season and were evaluated by analysis of variance. Means followed by the same letter(s) are not significantly different at $P = 0.05$ according to Duncan's multiple range test.

not always translate into differences in the number of virus-infected plants.

The percentage of virus-infected plants in the silver net mulched plots at first harvest was significantly higher in both experiments than in the other silver mulched plots, but was generally less than in plots with white mulches. The silver netting covered <50% of the soil surface, which, while providing some protection, was not as effective as the total coverage provided by the other silver mulches. Maelzer (22) noted that, to be most effective, mulches must cover approximately 60% of the soil surface.

We obtained mixed results with the white mulches. Plants on sprayable and polyethylene white mulches had fewer aphids than those planted on unmulched beds. The white spray mulch was significantly less effective than silver in repelling aphids in both experiments. Wyman et al. (29) and Brown et al. (6) collected more aphids in yellow pan traps positioned over white mulches than in pan traps positioned over silver mulches. White may not always serve to repel aphids; the same aphid, depending on its physiological state, may at one time be repelled by white and later be attracted to it because white both transmits and reflects all colors (19). When first applied, the white mulches were extremely bright. A combination of weathering and dust and soil accumulation soon resulted in a dulling or slight yellowing of the white color. This may have led to the white becoming less repellent and perhaps even slightly attractive. Kring (18,19) found that tints of hue colors (mixtures with white) were accepted or rejected by aphids based on the amount of white in the mixture; the more white, the greater the repellency.

The early protection from incoming viruliferous aphids (Figs. 1 and 2) afforded by the silver mulches and the concomitant delay in the onset of virus diseases (Table 1) were key components in maximizing yields of marketable squash. Susceptibility to infection often decreases with increasing plant age (5, 31) and older plants are generally better able to tolerate infection than young ones (5). Although plants grown over the silver spray and silver poly mulches eventually became infected, they continued to produce a higher percentage of marketable fruit than did plants infected prior to the initiation of fruit set. Blua and Perring (4) and Perring et al. (25) showed that delaying ZYMV infection until early cantaloupe fruiting stage prevented significant yield losses. Similar results were shown for WMV infections in squash and watermelon (3,9).

The silver spray mulches produced yields equivalent to those of the silver polyethylene in the spring experiment and significantly higher yields of marketable fruit in the fall experiment. The rapid increase in percent infected plants in plots mulched with silver that occurred between the samples taken at first harvest and the subsequent sample (Table 1) was due to several factors. The plants had reached sufficient size that canopy cover significantly decreased the effectiveness of the mulches in repelling aphids (2,29). Furthermore, the high incidence of virus in the control plots provided ready inoculum for the infection of the remaining plants once the effectiveness of the mulches had been compromised by increasing canopy cover. Perring et al. (25) noted a similar increase in the incidence of melon viruses following the removal of row covers. Un-

der conditions in which the entire field was mulched and virus incidence was low throughout, virus spread would likely occur much more slowly. Much of the previous work on the repellency of aphids by various colored mulches has centered largely on green peach aphid, *M. persicae*, because of its cosmopolitan distribution, extensive host range, and efficiency in vectoring a great number of viruses to a wide range of crops (17). Authors frequently lump all aphids collected into a single category without specific species identification (6,23,28) or list the percentage of various aphid species collected (24), oftentimes from yellow pan traps and not from the plants themselves (6,24,25, 28,29). Our data clearly showed that silver spray mulch is effective in repelling cotton-melon aphid, *A. gossypii*. This species constituted the bulk of the aphid population encountered on fall-planted squash in the San Joaquin Valley and is likely the principal vector of viruses affecting the crop at that time.

Use of reflective spray mulch rather than plastic films in strategies for integrated management of aphid-borne virus diseases in susceptible row crops is likely to be considerably more cost effective. In addition to lower cost of material, problems and expenses relating to removal and disposal of plastic films are avoided. Additional work is underway to develop compatible methods for providing season-long protection against virus diseases in conjunction with the use of reflective sprays.

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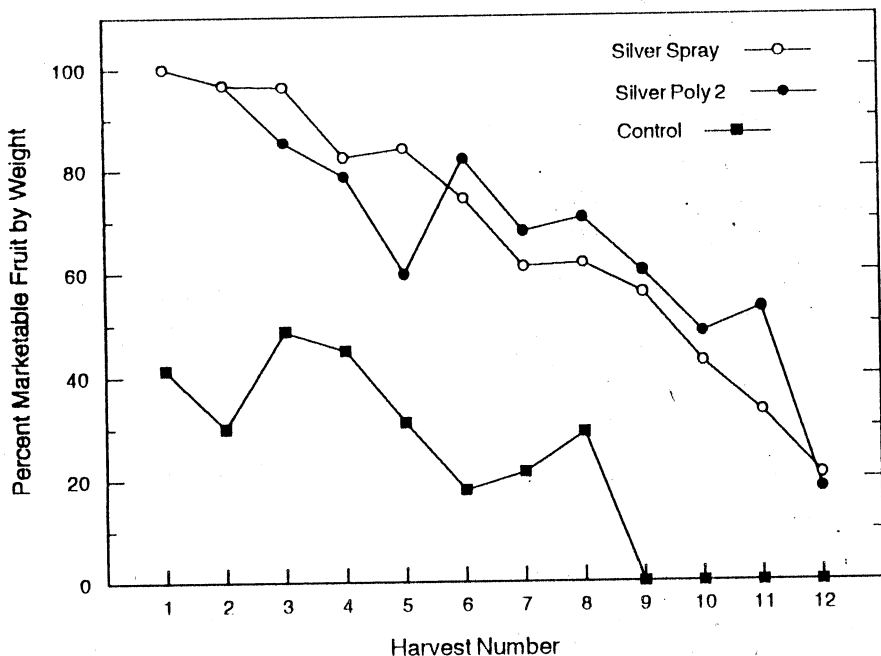


Fig. 5. Percent marketable fruit by weight of fall-planted zucchini squash from plots mulched with silver spray and silver poly 2, and from the unmulched control, over 12 harvests, Parlier, Calif., 1993.

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