

# Incidence of Nonpersistently Transmitted Viruses in Pepper Sprayed with Whitewash, Oil, and Insecticide, Alone or Combined

S. MARCO, Department of Virology, ARO, The Volcani Center, Bet Dagan 50250, Israel

## ABSTRACT

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Several methods of protecting pepper (*Capsicum annuum*) against aphid-borne virus diseases were evaluated in field experiments for 6 yr. Results of weekly sprays of the insecticide pirimicarb (Pirimor) or bifenthrin (Talstar) at 0.1% or of a mixture of pirimicarb at 0.1% and whitewash (Yalbin) at 10% did not differ from those of unsprayed controls. Applications of mineral oil (Virol) at 1% or of whitewash (Yalbin or Loven) at 10% reduced the incidence of virus infection by about 40%. The best control was achieved with Yalbin combined with bifenthrin or mineral oil, which reduced virus infection by about 60%. Whitewash treatments caused slight damage to pepper seedlings at the first-true-leaf stage but did not damage older plants. In three of four experiments, whitewash-treated plots had significantly higher yields than control plots. In the fourth, however, the yields in treated plots were lower, and the possible direct effect of whitewash treatment on the pepper plant is discussed. Yields with whitewash plus bifenthrin did not differ from yields with whitewash alone. Treatments with whitewash plus pirimicarb or whitewash plus mineral oil were more harmful, and the latter decreased yields significantly. Potato virus Y, the most prevalent virus, was found in 86% of infected plants, cucumber mosaic virus in 23%, and alfalfa mosaic virus in only one sample. Several plants had mixed infections.

Pepper (*Capsicum annuum* L.) is an important crop worldwide. Virus diseases cause serious losses and can be the most important limiting factor for production (10). In Israel, the crop is severely affected by the nonpersistently aphid-transmitted viruses potato virus Y (PVY) and cucumber mosaic virus (CMV) (16). Marketable yields are dramatically decreased because of reductions in fruit set, fruit quality, and fruit size. In the early spring and autumn, the potential for virus diseases makes cropping in certain areas of Israel unfeasible.

Bradley et al (1) showed that coating the plant surface with oil interferes with aphid transmission of nonpersistent viruses. Since then, vegetable and mineral oils have been used on many crops, including peppers, with variable levels of control (7,9,15). Variability in the effectiveness of oil sprays is related to many factors (21), such as climate and spraying equipment. Quite often, oils may damage the plants, especially when applied during hot weather (21).

Nonpersistently transmitted viruses are not adequately controlled with most insecticides (7,22). Pyrethroids were found to reduce the incidence of PVY infection in the laboratory (5), but they seldom give adequate control in the field,

and their combination with oil is preferred (4,17).

Mulching with reflective foils (aluminum or plastic) deters aphids from alighting on the crop, thereby reducing virus spread (7,20,23). However, the growing crop shades and covers the mulch, limiting repellency to a rather short, early growth period (23).

Applications of reflective whitewash to plants were shown to reduce the incidence of the nonpersistently transmitted viruses PVY in potato (13) and turnip mosaic virus (TuMV) in rutabaga (9), as well as the persistently transmitted potato leaf roll virus (PLRV) in potato (13).

Oils, insecticides, and whitewashes presumably reduce viral incidence by different mechanisms, and combinations of these materials may increase control. The present work compares the effectiveness of these materials alone and in different combinations on virus incidence and marketable yields in pepper.

## MATERIALS AND METHODS

**Experiments.** Seeds of the determinant pepper cultivar Maor were sown in beds. At the first- or second-true-leaf stage, the seedlings were transferred to small pots (7 cm in diameter) or speedling trays and kept in an insect-proof greenhouse. At the eight- to 10-leaf stage, after hardening for 2-4 days outdoors under a white, coarse net to prevent virus infection (2, 3,12), the seedlings were transplanted to the experimental field previously fumigated with methyl bromide.

Six independent experiments were carried out, with plots arrayed in a random block design and replicated four to

six times. The experimental plots were grown in spring, summer, or autumn using commercial practices. Sprinkler irrigation was provided once a week, adjusted to the evapotranspiration values. The plots were 5 × 2 m, 1.5 m apart, and consisting of three rows, for a total of 50-60 plants.

**Spray treatments.** The following materials were used: commercial whitewash (Loven [v/v] or Yalbin [w/v]) at 10% (Tapazol Co., Rishon Le-Zion, Israel) with 40% polyvinyl (Dabak [v/v], Tapazol Co.) added at approximately 0.1% to increase adherence; the insecticides pirimicarb (Pirimor 50WP [w/v]) and bifenthrin (Talstar 10%) at 0.1% concentration; and light mineral oil 80% (Virol [v/v]) at 1%. Sprays were applied once a week, 1 day after irrigation, with a Solo type 425 knapsack hand sprayer equipped with a Tee Jet 8004 nozzle. The materials were applied in the same concentrations when used alone or combined. The last spray treatments were given when the first fruits were 2-5 cm in diameter. Treatments were not continued beyond this stage because whitewash adheres to the fruit, and virus infection after fruit set in the cultivar used has little effect on fruit yield and quality (*unpublished*).

In preliminary tests, whitewashes were applied at 15%, as used in a previous work on potatoes (13), but this concentration damaged young pepper plants. Therefore, we conducted two experiments to determine the maximum non-damaging whitewash concentrations. Four pepper plants at first-true-leaf, four-leaf, 10- to 12-leaf, and branching stages of growth were dipped in increasing concentrations (5-25%) of Loven or Yalbin alone or combined with pirimicarb 0.1%, bifenthrin 0.1%, mineral oil 1%, and Dabak 0.1%. Appearance of treated plants was compared with that of nontreated controls for up to 6 wk. Test plants were grown in the greenhouse in the first trial and outdoors in the second trial.

**Incidence of virus.** Incidence of virus-diseased plants was visually assessed according to symptoms without specifying the viruses involved. This evaluation was repeated three times during the growing season, the final time about 1 wk after the last treatment spray. In plots treated with whitewash alone or combined, virus symptoms on plants were less obvious because of the whitewash cover, thereby raising the possibility of incorrect visual

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evaluations. To reduce this problem, virus evaluations were always done on young leaves the day after irrigation before a new application of whitewash. In addition, in experiments 1 and 3, about 100 plants with and 100 without symptoms, as visually assessed, were sampled from the control and the whitewash-treated plots and assayed individually by double antibody sandwich enzyme-linked immunosorbent assay (ELISA) (14) for PVY, CMV, and alfalfa mosaic virus (AMV). The visual evaluations of symptoms were related to ELISA results for the specific plants, and the correlation coefficients were highly significant in both experiments ( $r = 0.91$  and  $0.88$ , respectively), indicating the validity of the visual assessments. These ELISA results were also used for estimating the incidence of the above-mentioned viruses in the field.

#### Effect of treatments on fruit yields.

The middle rows of each plot (consisting of three rows) in experiments 3, 4, 5, and

6 were harvested each week (six to 10 harvests). The fresh weight of the marketable fruits was determined the same day in the field. Marketable fruits were defined as symptomless and larger than 100 g.

**Statistics.** Virus incidence data, after transformation to multiple infections (6), and yield data were analyzed by the multiple analysis of variance for repeated measurements, MANOVA (PROC GLM, SAS) for  $\alpha \leq 0.05$ . Means of virus incidence and of yields were separated by Duncan's multiple range test, and data of visual evaluations of infections were related to ELISA results for the same plants by calculating the coefficient of correlation. Efficiency of reducing infection was expressed as: percentage protection =  $100 (1 - \% \text{ infected in treatment} / \% \text{ infected in control})$ .

## RESULTS

### Effect of treatments on pepper plants.

No direct damage was perceptible on

pepper at different growth stages when treated with pirimicarb 0.1%, bifenthrin 0.1%, mineral oil 1%, Dabak 0.1%, Yalbin 5%, or Loven 5%. Loven or Yalbin at 10% concentrations caused slight damage in the youngest plants (first-true-leaf stage) but not in those at the 10- to 12-leaf stage; plant damage increased as whitewash concentration increased above 10%. Young plants were more sensitive than mature plants, and greenhouse-grown plants were more sensitive than plants grown outdoors. Combining whitewashes with mineral oil or pirimicarb increased damage, whereas addition of Dabak or Talstar did not. Direct damage appeared as leaf rugosity and discoloration, especially on margins, followed by some plant stunting, but symptoms did not resemble those of typical mosaic virus infection. In view of these results and because plants were transplanted at about the eight- to 10-leaf stage, 10% whitewash plus 0.1% Dabak was chosen as the standard treatment to achieve maximum effect with minimum damage. In the field, where six to eight sprays were applied, the plants in the plots treated with the mixtures of whitewash and pirimicarb or whitewash and oil were stunted and had small foliage. The other treatments did not visibly damage the plants.

**Virus incidence.** The effect of the eight treatments tested fell into three distinct categories (Fig. 1, Table 1): 1) Neither insecticide spray (pirimicarb nor bifenthrin) differed significantly from the control, nor did the combination of Yalbin whitewash and pirimicarb. 2) Both Yalbin and Loven whitewashes provided significant protection (means of 39 and 38% protection, respectively) at a level similar to that of mineral oil sprays (mean of 37% protection). 3) The best protection was obtained with the combination of Yalbin whitewash and bifenthrin (mean of 56% protection) and the combination of Yalbin whitewash and oil (mean of 65% protection). These combinations provided significantly higher levels of protection than the test materials applied separately (Table 1).

As determined by ELISA on approximately 200 symptomatic samples from the control and whitewash-treated plots in experiments 1 and 3, PVY was the

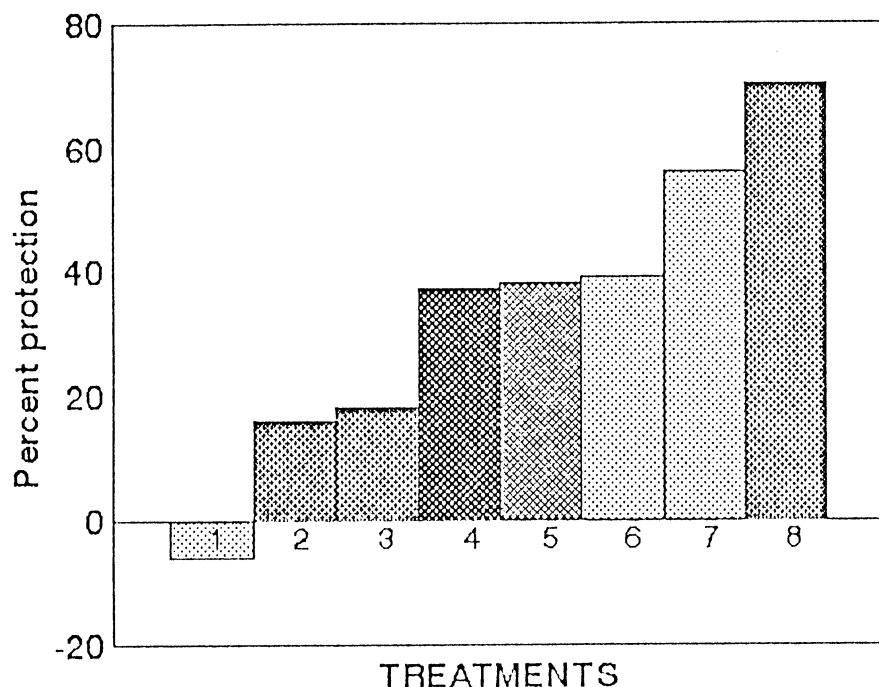


Fig. 1. Protection efficiency against viral infection in pepper plots sprayed weekly with 1 = pirimicarb, 2 = bifenthrin, 3 = whitewash (Yalbin) + pirimicarb, 4 = mineral oil, 5 = whitewash (Loven), 6 = whitewash (Yalbin), 7 = whitewash (Yalbin) + bifenthrin, and 8 = whitewash (Yalbin) + mineral oil; fruit were 2-5 cm in diameter. Data are means of the percent protection in three to five experiments.

Table 1. Incidence of virus infection in peppers (*Capsicum annuum*) sprayed with whitewash, mineral oil, and insecticide, alone or combined, and in unsprayed controls

Expt.	Date	Percent infection (transformed for multiple infections)									
		Control	Whitewash		Mineral oil	Insecticide		Whitewash plus			
			Yalbin	Loven		Pirimicarb	Bifenthrin	Pirimicarb	Bifenthrin	Mineral oil	
1	10 Aug. 1983	15 a <sup>2</sup>	9 ab	9 ab	11 a	...	...	14 a	...	...	5 b
2	24 Apr. 1984	19 a	9 bc	...	11 b	21 a	...	17 a	...	...	7 c
3	3 Sept. 1985	58 a	40 b	42 b	43 b	...	...	...	33 bc	...	25 c
4	15 Mar. 1987	73 ab	53 b	...	...	87 a	60 ab	51 b	31 c	...	...
5	15 Apr. 1988	33 a	...	21 b	...	30 a	26 ab	29 ab	15 c	...	...
6	5 May 1990	39 a	22 b	...	19 b	...	36 a	...	16 bc	...	11 c

<sup>2</sup> Data with different letters are significantly different according to Duncan's multiple range test ( $P < 0.05$ ).

**Table 2.** Fruit yield of peppers (*Capsicum annuum*) sprayed with whitewash, mineral oil, and insecticide, alone or combined, and of unsprayed controls

Expt.	Date	Yield/plant (g)								
		Control	Whitewash		Mineral oil	Insecticide		Whitewash plus		
			Yalbin	Loven		Pirimicarb	Bifenthrin	Pirimicarb	Bifenthrin	Mineral oil
3	3 Sept. 1985	320 b <sup>z</sup>	260 a	240 a	390 c	...	290 ab	...	...	220 a
4	15 Mar. 1987	380 a	610 b	...	...	380 a	450 a	570 b	710 c	...
5	10 Apr. 1988	420 a	...	560 ab	...	440 a	480 a	450 a	610 b	...
6	5 May 1990	510 a	620 b	...	580 ab	...	560 ab	...	650 b	420 a

<sup>z</sup> Data with different letters are significantly different according to Duncan's multiple range test ( $P < 0.05$ ).

most prevalent virus, occurring in 88% of infected control samples and 85% of infected whitewash-treated samples. CMV was found in 19 and 26% of these samples, respectively, sometimes occurring with PVY. One sample was found infected with AMV.

**Fruit yields.** Yields were not measured in experiments 1 and 2 because of the relative low incidences of viral infection (Table 1) and because the late appearance of virus diseases suggested a low impact on yields. In experiment 3, all the whitewash treatments (Yalbin, Loven, Yalbin + mineral oil) except Yalbin + bifenthrin significantly decreased pepper yields (81, 75, and 69%, respectively, of controls)—this despite a significant reduction in virus incidences with all of these treatments (Table 1) and despite the lack of meaningful visible damage in Loven- or Yalbin-treated plots. In experiments 4, 5, and 6, however, treatment with Loven or Yalbin significantly increased yields (133, 122, and 161%, respectively, of control) (Table 2) as well as decreasing virus incidence (Table 1). Whitewash combined with pirimicarb was not more effective than whitewash alone, whereas the effect of Yalbin + bifenthrin was significantly better, at least in experiment 4 (Table 2). The worst treatment was Yalbin + mineral oil (experiments 3 and 6), which clearly damaged the plants and significantly decreased yields. Treatment with mineral oil alone significantly increased yield in experiment 3 (122% vs. control) but had no effect in experiment 6 (Table 2).

## DISCUSSION

Whitewash sprays reduced the incidence of PVY and CMV in pepper (Fig. 1). These results confirm the reports of similar effects on the incidence of PLRV and PVY in potato (13) and TuMV in rutabaga (9). Whitewash is relatively inexpensive and safe and is easier to apply than oil, which requires sophisticated spraying machinery (19). The levels of protection achieved by whitewashes or oil are far from satisfactory but are still important, since very few alternatives are available. Our results (Table 1) confirm that insecticides provide no protection against nonpersistently transmitted viruses in pepper (7,22) even though they are most effective aphicides (8,11). Even pyrethroids that reduced PVY infection

in potatoes (5,18) and zucchini yellow mosaic virus in melons (15) did not protect pepper in the field to a significant extent (Table 1).

Oil sprays reduced virus incidence (Fig. 1), confirming many other reports (1,7,15,19). Results are not consistent (9,15,21), however, and are dependent on many factors, such as pressure and volume of the spray, which is expensive to achieve, and climate (21), which is impossible to control. The efficacy of oil in our work was also low (37% protection) (Fig. 1). This might be explained by the relatively low concentration used (1%), but in the hot climate of Israel, higher concentrations often cause damage. Also, volume and adequate pressure could not be achieved with our simple knapsack sprayer (19). Under the same conditions, however, addition of whitewash increased the protective efficiency of oil to 65%, which was significantly higher than the efficiency of either oil or whitewash alone. An additive effect apparently is obtained (Fig. 1), suggesting that whitewash and oil do interfere with virus transmission by different mechanisms. Thus, whitewash alone or mixed with oil could be a promising alternative to insecticides and could also reduce the percentage of oil needed (8,9).

Whitewash effects on yield are not yet understood, and the data obtained are rather confusing. The reduced yields in whitewash-treated plots in experiment 3 despite the decrease in virus incidence, plus the yield increase in experiments 4, 5, and 6 that was sometimes higher than could be explained by the decrease in virus incidence (Table 2), indicate a direct effect of the whitewashes on the physiology of the plant, even when symptoms were not perceptible. This effect interacts, in as yet an unknown way, with many other parameters (e.g., virus infection) in yield production. Several studies report an increase in yield of whitewash-treated sorghum, cotton, melon, artichoke, and rutabaga (8,13), while others report decreased yield in potato (13). In the present work, the same crop showed both increased and decreased yield effects, suggesting that such effects may be related to growth conditions rather than to the specific crop. Pepper yield decreased when the crop was grown in rather low temperatures (experiment 3) and increased when the crop was grown

in relatively high temperatures (experiments 4, 5, and 6). Whitewash increases leaf reflectance (3,13,15), thereby reducing its temperature. This may be the reason whitewash increases yields under supraoptimal temperature conditions and decreases yields in relatively low temperatures. Whatever the reason, the possible physiological effects of whitewashes on plants must be investigated further before recommendations for virus control can be made.

## LITERATURE CITED

- Bradley, R. H. E., Wade, C. F., and Wood, F. A. 1962. Aphid transmission of potato virus Y inhibited by oils. *Virology* 18:327-328.
- Cohen, S. 1981. Reducing the spread of aphid-transmitted viruses in peppers by coarse-net cover. *Phytoparasitica* 9:69-76.
- Cohen, S., and Marco, S. 1979. Reducing virus spread in vegetables and potatoes by net cover. (Abstr.) *Phytoparasitica* 7:40-41.
- Gibson, R. W., and Cayley, G. R. 1984. Improved control of potato virus Y by mineral oil plus the pyrethroid cypermethrin applied electrostatically. *Crop Prot.* 3:469-478.
- Gibson, R. W., and Rice, A. D. 1986. The combined use of mineral oils and pyrethroids to control plant viruses transmitted non- and semi-persistently by *Myzus persicae*. *Ann. Appl. Biol.* 109:465-472.
- Gregory, P. H. 1948. The multiple infection transformation. *Ann. Appl. Biol.* 35:412-417.
- Loebenstein, G., and Raccach, B. 1980. Control of non-persistently transmitted aphid-borne viruses. *Phytoparasitica* 8:221-235.
- Lowery, D. T., and Boiteau, G. 1988. Effects of five insecticides on the probing, walking, and settling behavior of the green peach aphid and the buckthorn aphid (Homoptera: Aphididae) on potato. *J. Econ. Entomol.* 81:208-214.
- Lowery, D. T., Sears, M. K., and Harmer, C. S. 1990. Control of turnip mosaic virus of rutabaga with applications of oil, whitewash, and insecticides. *J. Econ. Entomol.* 83:2352-2356.
- Makkouk, K. M., and Gumpf, D. J. 1974. Further identification of naturally occurring virus diseases of pepper in California. *Plant Dis. Rep.* 58:1002-1006.
- Marco, S. 1980. The use of insecticides to control potato leafroll virus in seed-potato crops in the Golan Heights. *Phytoparasitica* 8:61-71.
- Marco, S. 1981. Reducing potato leaf roll virus (PLRV) in potato by means of baiting aphids to yellow surfaces and protecting crops by coarse nets. *Potato Res.* 24:21-31.
- Marco, S. 1986. Incidence of aphid-transmitted virus infections reduced by whitewash sprays on plants. *Phytopathology* 76:1344-1348.
- Marco, S., and Cohen, S. 1979. Rapid detection and titer evaluation of viruses in pepper by enzyme-linked immunosorbent assay. *Phytopathology* 69:1259-1262.
- Mor, N. 1989. Epidemiology and control of non-persistent viruses in cucurbits. M.Sc. dissertation. University of Jerusalem, Israel.
- Nitzany, F. E., and Tanne, E. 1962. Virus diseases of pepper in Israel. *Phytopathol. Mediterr.* 1:180-182.

17. Raccach, B. 1986. Nonpersistent viruses: Epidemiology and control. Pages 387-429 in: *Advances in Virus Research*. Vol. 31. K. Maramorosch, F. A. Murphy, and A. J. Shatkin, eds. Academic Press, Toronto.
18. Rice, A. D., Gibson, R. W., and Stribley, M. F. 1983. Effects of deltamethrin on walking, flight and potato virus Y transmission by pyrethroid-resistant *Myzus persicae*. *Ann. Appl. Biol.* 102:229-236.
19. Simons, J. N., and Zitter, T. A. 1980. Use of oils to control aphid-borne viruses. *Plant Dis.* 64:542-546.
20. Smith, F. F., and Webb, R. E. 1969. Repelling aphids by reflective surface, a new approach to the control of insect-transmitted viruses. Pages 631-639 in: *Viruses, Vectors and Vegetation*. K. Maramorosch, ed. Wiley-Interscience, New York.
21. Vanderveken, J. J. 1977. Oils and other inhibitors of nonpersistent virus transmission. Pages 435-454 in: *Aphids as Virus Vectors*. K. F. Harris and K. Maramorosch, eds. Academic Press, New York.
22. Walkey, D. G. A., and Dance, M. C. 1979. The effect of oil sprays on aphid transmission of turnip mosaic, beet yellows, bean common mosaic, and bean yellow mosaic viruses. *Plant Dis. Rep.* 63:877-881.
23. Zitter, T. A. 1977. Epidemiology of aphid-borne viruses. Pages 385-412 in: *Aphids as Virus Vectors*. K. F. Harris and K. Maramorosch, eds. Academic Press, New York.