Effect of Carbofuran on Transmission of Maize Dwarf Mosaic Virus in Sorghum Mechanically and by the Aphid Schizaphis graminum

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Seifers, D. L., and Harvey, T. L. 1989. Effect of carbofuran on transmission of maize dwarf mosaic virus in sorghum mechanically and by the aphid Schizaphis graminum. Plant Disease 73:61-63.

Significant (P < 0.05) increases in levels of natural infection by maize dwarf mosaic virus (MDMV) occurred in carbofuran-treated sorghum in the field, compared with untreated sorghum. In greenhouse studies, carbofuran-treated sorghum increased infection in aphid (Schizaphis graminum) transmission tests with the A strain of MDMV. In mechanical inoculation tests, virus infection was reduced in carbofuran-treated sorghum by strain A and no difference was observed for strain B.

During field insecticide trials for control of greenbugs (Schizaphis graminum (Rondani)) in sorghum (Sorghum bicolor (L.) Moench), we observed that

Contribution 88-489-J from the Kansas Agricultural Experiment Station, Kansas State University, Manhattan 66506. Research was supported in part by a grant from the Kansas Sorghum Commission.

Accepted for publication 11 September 1988 (submitted for electronic processing).

carbofuran-treated plots had greater numbers of virus-infected plants than untreated plots. These observations were confirmed, and studies were initiated in the laboratory to determine if this response could be duplicated using the greenbug and mechanical transmission trials with different isolates of maize dwarf mosaic virus (MDMV). We report that the response of insecticide-treated sorghum to infection by MDMV, by natural or mechanical means, is altered and varies with virus isolate. Portions of this study were previously published (10).

MATERIALS AND METHODS

Virus maintenance. Three isolates of MDMV were used: 1) strain A (MDMV-A), originally obtained from R. E. Ford (University of Illinois); 2) the virus isolate observed in field studies in Kansas (MDMV-A Hays); and 3) strain B (MDMV-B), obtained from S. G. Jensen (USDA-ARS, Lincoln, NE). The identity of virus isolates was confirmed by host range and serological tests. Preparation of antisera to strains A and B and ELISA conditions were as previously described

Field studies. DeKalb sorghum hybrids DK 28 and DK 18 were planted on 8 and 10 July 1985, respectively, in four-row plots 20 m long with a row spacing of 0.9 m and a plant spacing of approximately 10 cm within each row. Carbofuran (15%

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granular formulation) was applied infurrow at 1.12 kg a.i./ha for DK 18 and 2.24 kg a.i./ha for DK 28.

Four sticky traps, each 9 cm in diameter and facing the cardinal directions, were used to monitor flight activity of alate greenbugs. Traps were present continuously near the plots during the field tests. The number of greenbugs trapped was recorded at weekly intervals, and the number of greenbugs per plant was determined by counting greenbugs on 10 plants selected randomly from the center rows of each plot. Greenbug numbers were recorded at 3, 4, and 5 wk after planting.

Infection notes (plants having mosaic symptoms) were taken on 23 August 1985 at both planting sites. Leaf samples were taken from both sites for verification of virus identity by means of host range and serological tests.

Vector transmission studies. Greenbugs (biotype E) maintained on wheat in a greenhouse were increased on virusinfected sorghum cv. Colt that had been mechanically inoculated 10 days previously (8). After 7 days, the greenbugs were removed from the infected plants, and about 10 greenbugs per plant were immediately placed, in the greenhouse, onto DK 18 and DK 28 sorghum planted in 50×30 cm soil-filled flats with 11 rows of 20 plants each. Carbofuran was added (at planting) to each row of four flats of each cultivar at 0.15 g a.i. per row. This concentration of carbofuran approximated that used in the field studies (1.12 kg a.i./ha). Four flats of each cultivar without carbofuran served as controls. Greenbugs were eliminated 8 hr after infestation by spray treatment with

Experiments were repeated twice, with four replications per experiment for the MDMV-A Hays isolate and three replications for the MDMV-A and MDMV-B isolates.

Mechanical inoculation studies. Inoculum preparation and the method of inoculation used throughout this investigation were reported previously (8). In all experiments, Colt was used as the source plant for inoculum production and DK

18 for the assay host, and plants were incubated in a greenhouse at 30 ± 4 C.

Preliminary tests were conducted to determine the linear portion of infectivity curves for each virus isolated. In these tests, 200 sorghum seedlings were inoculated with a series of twofold dilutions of inoculum (1:10-1:2,560, prepared with a mortar and pestle from MDMV-infected leaf tissue by grinding in 0.02 M potassium phosphate buffer, pH 7.0, with all dilutions made in the same buffer) 6 days after planting. The number of systemically infected plants was assessed 2 wk later.

Greenbug transmission and mechanical inoculation tests were done sequentially for each virus isolate, to avoid contamination. In these trials, sorghum was planted in 50×30 cm soil-filled flats with 11 rows per flat and 20 seedlings per row. Four flats had carbofuran applied in each of the 11 rows, as previously described. Four flats at the same planting rate but without carbofuran were used as controls. The plants were inoculated at 6 or 12 days after planting with a 1:800 dilution of inoculum (prepared as described above) for MDMV-A and MDMV-B and a 1:1,600 dilution for MDMV-A Hays. The number of systemically infected plants was recorded after 2 wk.

Experimental replication and statistical analysis. Data from each replicate were pooled for comparison of treatments. Values obtained were subjected to analysis of variance. All percentage data were arc sine transformed before statistical analysis. Data in the tables are actual percentage values.

Field tests were a randomized complete block with treatments replicated four times for each cultivar for each test. All comparisons were subjected to analysis of variance. Significant treatment effects were compared by the Student-Newman-Keuls multiple range test.

RESULTS

Significant increases (P < 0.05) in MDMV infection occurred in carbofuran-treated sorghum cultivars (Table 1). Numbers of greenbugs trapped on four 9-cm-diameter traps in the vicinity of plots after planting were: 1 wk = 16, 2 wk = 91, 3 wk = 1,314, 4 wk = 1,192, 5 wk = 96, and 6 wk = 2. The peak flights occurred during the third and fourth weeks after planting and ended about 1 wk before MDMV infection was seen. After 3 wk, carbofuran gave almost complete greenbug control (98%) at the 2.24 kg a.i./ha rate but only about 50% control at the 1.12 kg a.i./ha rate (Table 1).

The increased infection observed in the field with carbofuran use was also reproduced in the greenhouse, where greenbug transmission of the MDMV-A Hays isolate resulted in significantly greater infection percentages in carbofuran-treated plants than in the controls (Table 2). The other MDMV-A isolate also increased infection in insecticide-treated sorghum, but only experiment 1 results proved to be significantly different.

In mechanical inoculation tests in the greenhouse, treatment of sorghum with carbofuran significantly reduced the number of plants infected by both MDMV-A isolates (Table 3).

In greenbug transmission trials with MDMV-B, no infection was obtained in either insecticide-treated or untreated sorghum in any of three experiments. In mechanical inoculation trials with MDMV-B, the percentage of infection (average value for three experiments) for plants inoculated at 6 days was 24% for carbofuran-treated and 27% for untreated DK 18 sorghum plants. The average infection by MDMV-B for 12-day-old plants was 8% for both treatments.

DISCUSSION

The increased virus infection related to carbofuran treatment in our field studies appears to be similar to results of Onazi and Wilde (6), who observed enhanced infection of disulfoton-treated sorghum by MDMV. The insecticide-treated rows had the least number of greenbugs and an increase in the number of MDMV-A infected plants; the increase in MDMV-infected plants was not statistically significant, however. Significant increases in MDMV infection of insecticide-treated (aldicarb) sweet corn also have been reported (3). It has been suggested that the increased incidence of virus

Table 1. Effect of carbofuran on greenbug (*Schizaphis graminum*) infestation and incidence of maize dwarf mosaic virus strain A infection in sorghum plants under field conditions

Treatment	Average r at w	Average percent		
	3	4	5	infection
Test 1 ^x				
Carbofuran, 1.12 kg a.i./ha	176 a ^y	27 a	0	10 a
Untreated	372 b	35 b	0	4 b
Test 2 ^z				
Carbofuran, 2.24 kg a.i./ha	6 a	4 a	0	23 a
Untreated	296 b	220 b	0	10 b

^{*}Cultivar DK 18 planted 10 July 1985.

Table 2. Percentage of maize dwarf mosiac virus (MDMV) infection in carbofurantreated and untreated sorghum plants after greenbug (Schizaphis graminum) infestation in the greenhouse

Treatment	MDMV-A Hays experiment		MDMV-A experiment		
	1	2	1	2	3
Carbofuran	8.0	18.0	3.0	2.5	1.3
Untreated	4.0	7.0	1.0	1.5	1.0
LSD (0.05)	1.0	10.0	1.0	3.1	1.1

Treatments compared for the same time interval within a given test not having a letter in common are significantly different (P < 0.05).

²Cultivar DK 28 planted 8 July 1985.

Table 3. Percentage^z of carbofuran-treated and untreated sorghum plants infected at different ages by different isolates of maize dwarf mosaic virus (MDMV)

Treatment	Inoculated at 6 days		Inoculated at 12 days		
	MDMV-A Hays	MDMV-A	MDMV-A Hays	MDMV-A	
Carbofuran	30	61	13	50	
Untreated	47	66	18	57	
LSD (0.05)	5	2	4	3	

²Average value for three experiments.

diseases observed in aldicarb-treated fields is due to increased activity of aphids stimulated by sublethal doses of the insecticide (2). Another study using carbofuran showed no difference in infection between treated and untreated corn (5). The differences in results from these various studies may be attributed to different insecticide treatments, vectors, and virus isolates and strains being studied.

The reasons for the lack of transmission of MDMV-B are unknown. The minimal information available indicates that greenbugs have a lower transmission efficiency for MDMV-B than for MDMV-A (1). In addition, our MDMV-B isolate had been maintained mechanically and possibly could have had a change or loss in helper component (7). Loss of transmissibility of aphid-

transmitted viruses has been reported (4).

In this study, carbofuran-treated plants had less infection by MDMV than untreated controls. The reasons underlying this reduction in numbers of infected plants in mechanically inoculated, carbofuran-treated sorghum are unknown.

The results of these studies show that when viruliferous greenbugs are present, treatment of sorghum with the systemic insecticide carbofuran can cause an increase in MDMV infection under field and greenhouse conditions. These results are important because they document increased virus infection in field and greenhouse environments in response to insecticide application and the variation in response of virus isolates to methods of virus transmission in host tissue influenced by chemical treatment.

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