

RESEARCH NOTES

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Inhibitory Effect of Watermelon Mosaic Virus on *Meloidogyne javanica* (Treb) Chitwood Infecting *Cucurbita pepo* L.¹

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Interactions between root-knot nematodes and viruses were recently reviewed by Taylor (4). Compared to virus-free cowpea (*vigna sinensis* Endl.), cowpea mosaic virus-infected plants formed fewer galls on roots in response to infection by *Meloidogyne incognita* (Kofoid and White) Chitwood (2). On the other hand, tobacco plants (*Nicotiana tabacum* L.) infected with tobacco ringspot virus (TRSV) and *M. incognita* had more galls than did nematode-infected virus-free plants (4). Also, TRSV-infected bean plants (*Phaseolus vulgaris* L.) had more second-stage juveniles of *M. javanica* (Treb) Chitwood entering roots than did virus-noninfected plants, whereas tobacco mosaic virus (TMV) did not show similar influence on the nematode in tomato plants (*Lycopersicon esculentum* Mill.) (1). Tomato plants infected with TMV induced higher growth rates of the nematode than did virus-free plants, whereas TRSV in bean plants did not affect the rates (1). Similarly, Bird (1) found no synergistic effect between *M. javanica* and TRSV on bean root, but the effect on soybean (*Glycine max* (L.) Merr.) between *M. incognita acrita* and TRSV was reported by Ryder and Crittenden (3). Clearly, interactions between viruses and root knot nematodes on host plants are complex.

In Minas Gerais, Brazil, both *M. javanica* and watermelon mosaic virus (WMV) occur on zucchini (*Cucurbita pepo* L.). The purpose of our work was to investigate the effect of WMV on *M. javanica* infection and the concomitant effect of both organisms on zucchini plant growth.

Nematodes for inoculum were maintained in the greenhouse and collected as needed. Virus for inoculum was maintained on zucchini leaves. WMV crude sap from infected leaves was diluted with phosphate solution (0.01 M, pH 7.0) respectively to 5×10^{-1} , 1×10^{-1} , 5×10^{-2} , and 1×10^{-1} times. Two leaves per plant were inoculated with the sap by rubbing the leaf surface with carborundum (600 mesh). Mosaic symptoms appeared 3 days after inoculation with the crude sap.

The experiment was conducted in a greenhouse. In the first test, four *C. pepo* cv. Caserta seeds were planted in a 10-liter plastic bag filled with methyl bromide-treated soil (73% sand, 18% clay, and 9% cow manure). Thirty grams commercial fertilizer products at the rate of N-P-K = 4-14-8 and 20 g lime per bag were uniformly mixed with the soil. The plants were thinned to one seedling per bag after emergence. Four weeks later (four-leaf stage), the following treatments were made: 1) neither roots nor leaves inoculated (control); 2) roots inoculated with approximately 664 eggs and 1,624 juveniles of *M. javanica*, leaves not inoculated (N); 3) roots not inoculated, leaves inoculated with 10^{-1} dilution of WMV crude sap concentration ($V 10^{-1}$); 4) roots inoculated as in 2, leaves inoculated as in 3 (N + $V 10^{-1}$). Each treatment was replicated six times. Fifty-four

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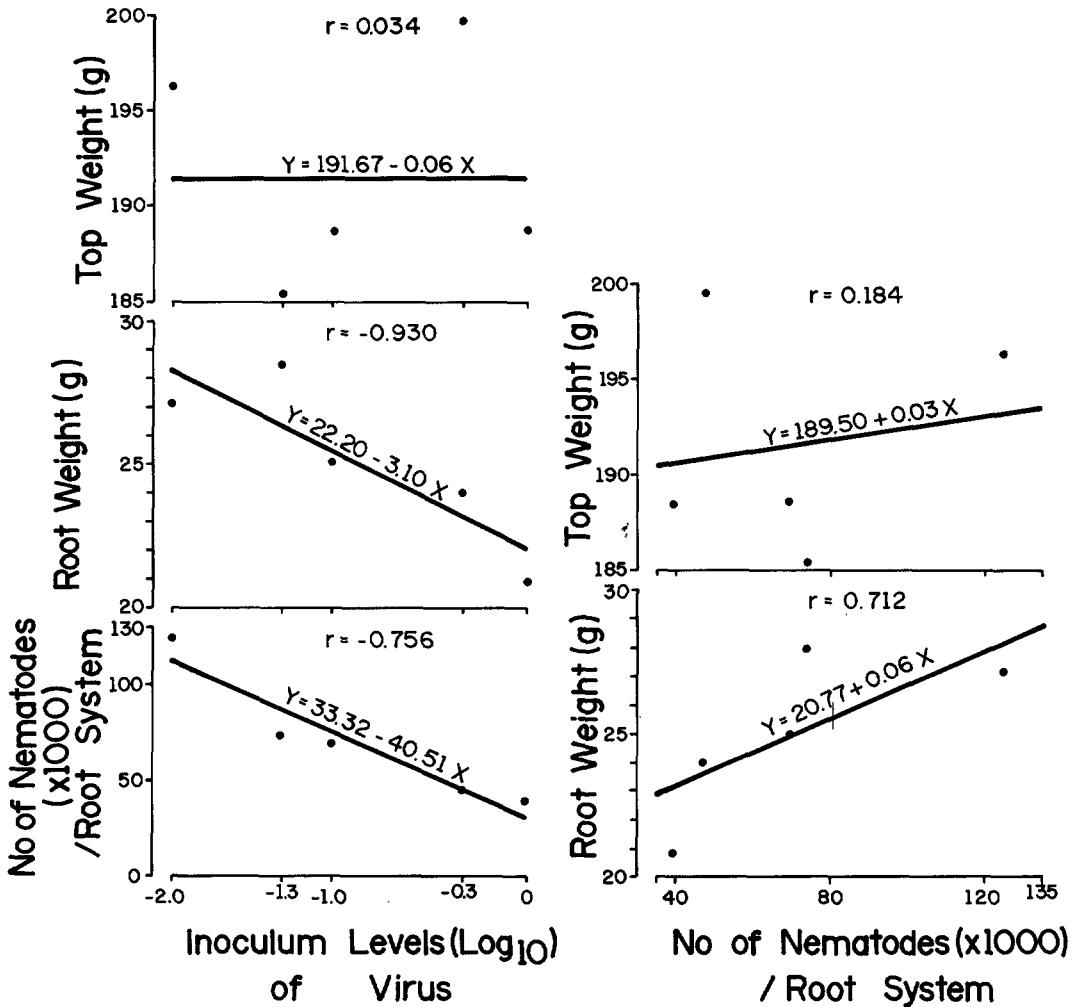


FIG. 1. Correlation among inoculum level of watermelon mosaic virus, number of *Meloidogyne javanica*, root weight, and top weight of *Cucurbita pepo*. Virus and nematode were inoculated at the same time on *C. pepo*.

days after inoculation, nematode numbers (eggs and second-stage juveniles) and fresh root weights were determined.

In the second test, each 20-liter plastic bag containing the same soil as in the first test was planted with five seeds (cv. Caserta) and thinned to three plants per bag. The following treatments were applied: 1) neither roots nor leaves inoculated (control); 2) roots inoculated with 3,102 eggs and 4,465 juveniles of *M. javanica*, leaves not inoculated (N); 3) roots not inoculated, leaves inoculated with 10^{-1} dilution of WMV crude sap ($V 10^{-1}$); 4-8) roots inoculated as in 2, leaves respectively inoculated with WMV at 10^{-2} , 5×10^{-2} , 10^{-1} ,

and 5×10^{-1} dilutions and crude sap (N + $V 10^{-2}$, N + $V 5 \times 10^{-2}$, N + $V 10^{-1}$, N + $V 5 \times 10^{-1}$, and N + $V 10^0$); 9) roots inoculated as in 2, 7 days later leaves inoculated as in 3 (N → $V 10^{-1}$); 10) leaves inoculated as in 3, 7 days later roots inoculated as in 2 ($V \rightarrow N 10^{-1}$). Each treatment was replicated seven times. Forty-five days after treatment, plants were harvested and weighed and nematode numbers (eggs and second-stage juveniles) were determined.

Nematode infection: In the first test, the roots inoculated with nematode only (N) contained higher final nematode numbers than the roots inoculated with nematodes and WMV (N + $V 10^{-1}$) (Table 1). The

TABLE 1. Effect of watermelon mosaic virus (V) infection of *Cucurbita pepo* on population development of *Meloidogyne javanica* (N).

Treatments		First test		Second test		
		Eggs and second-stage juveniles/root system	Root wt (g)/plant	Eggs and second-stage juveniles ($\times 1,000$)/root system	Top wt (g)/plant	Root wt (g)/plant
Pathogens*	Inoculum levels†					
Control			16.6 b		219.5 a	15.8 c
N alone		1,024 a‡	20.8 a	837 a§	233.8 a	23.6 ab
V alone	10^{-1}		15.7 b		191.9 b	
N + V	10^{-2}			125 b	196.2 b	27.1 ab
N + V	5×10^{-2}			73 c	185.2 b	28.3 ab
N + V	10^{-1}	492 b	23.3 a	69 c	188.6 b	25.0 ab
N + V	5×10^{-1}			47 cd	199.5 b	24.0 ab
N + V	10^0			39 d	188.6 b	20.9 bc
N → V	10^{-1}			63 cd	188.0 b	27.3 a
V → N	10^{-1}			12 e	195.7 b	16.8 c

* Control = no inoculation; N alone = only nematodes; V alone = only virus; N + V = both pathogens inoculated at same time; N → V = nematodes first, virus next; V → N = virus first, nematodes next.

† Inoculum levels of virus were 5×10^{-1} , 10^{-1} , 5×10^{-2} , and 10^{-2} dilutions of WMV crude sap (10^0). Each root system was inoculated with 2,288 nematodes in the first test and with 7,567 nematodes in the second test.

‡ Different letters in columns indicate differences ($P = 0.05$) according to Duncan's multiple-range test.

§ Data in this column transformed into \log_{10} for analysis.

overall low nematode infection in this test may be related to low temperatures (8–21 C) during fall and winter.

In the second test, the plants inoculated with nematodes only (N) were found to have significantly higher final nematode numbers in the roots than in any other treatments (Table 1). Final nematode numbers in the roots of plants inoculated with nematodes and WMV at the same time decreased with the increase in WMV concentration. The correlation coefficient (r) between the final nematode numbers and WMV sap concentrations was -0.756 (Fig. 1). On the other hand, in the three treatments using the same inoculum level (10^{-1} dilution) of virus and nematode, the final nematode numbers in the plants inoculated with WMV 7 days prior to inoculation with nematodes (V → N 10^{-1}) were lower ($P = 0.05$) than that inoculated with both organisms at the same time (N + V 10^{-1}), or than that inoculated with nematodes first, followed by WMV 7 days later (N → V 10^{-1}). The results indicate that WMV retarded the establishment of the *M. javanica* population in zucchini roots.

Plant growth: In the first test, root weights of plants inoculated with nematodes only (N), or with both organisms (N + V 10^{-1})

were higher ($P = 0.05$) than those of the control plants (control) or plants inoculated only with virus (V 10^{-1}) (Table 1), indicating that high root weights were induced by the nematodes forming heavy galls on roots. Similar results were also shown in the second test. In the five treatments (N + V), r between final nematode numbers and root weights was $+0.712$, whereas r between virus inoculum levels and root weights was -0.930 (Fig. 1). In the three treatments using the same level of virus inoculum (10^{-1} dilution) and nematode, root weights of plants inoculated with WMV first and nematodes next (V → N 10^{-1}) were lower than in the other two treatments (N + V 10^{-1} , N → V 10^{-1}) in which both root weights were no different. The results indicate that the reductions of root weights were related to the decrease in nematode numbers due to the inhibitory effect of WMV.

Top weights of plants inoculated with WMV only (V 10^{-1}) or with both organisms were less ($P = 0.05$) than those of the control plants (control) or plants inoculated only with nematodes (N) (Table 1). The result shows that WMV reduced normal plant growth. However, the top weights were almost independent of the WMV

inoculum levels ($r = +0.034$) and of the final nematode numbers ($r = +0.184$) (Fig. 1).

Neither synergistic nor additive effects on plant growth between the nematode and the virus pathogens were demonstrated.

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